

ASLP Dairy Project Publications



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animal

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Effect of pre-weaning feeding regimen on post-weaning growth performance of Sahiwal calves

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ABSTRACT:

The objective of the study was to assess the post-weaning growth response of Sahiwal calves reared on four different pre-weaning dietary regimens. The four diets were: (a) whole cow's milk, starter ration (SR; CP 52.0%, total digestible nutrients (TDN) 57.2%) and Berseem hay (H; Egyptian clover; CP 52.1%, TDN 56.3%); (b) whole cow's milk 1H; (c) milk replacer (MR; reconstituted to suppliers specification; Spray for) 1SR 1H; and (d) MR 1H. The protein and fat percentages of reconstituted MR were 2.22 and 1.84, respectively. Milk or MR were fed at the rate of 10% of the calves' body weight (BW) until 56 days of age, and then withdrawn gradually until weaned completely by 84 days of age. The average initial BW of calves in groups A, B, C and D were 56.361.0, 47.561.0, 40.461.0 and 30.361.0 kg, respectively. Initially, there were 12 calves in each group with six of each sex; however, one male calf died from each of groups B and C and were not replaced. During the post-weaning period, 13 to 24 weeks, the calves were fed as a single total mixed ration ad libitum based on maize, canola meal, wheat straw and molasses containing 16% CP and 70% TDN. Daily feed intake and weekly BW gains were recorded. The data were analyzed by MIXED model analysis procedures using the statistical program SAS. The intake of calves as a percent of their BW, feed conversion ratio and cost per kg of BW gain were not different ($P < 0.05$) across treatments. The daily gain at 24 weeks of age for the pre-weaning treatments A, B, C and D were 746633, 660633, 654633 and 527633 g/day and the final liveweights of calves were 11964.2, 10264.2, 9564.2 and 7564.2 kg, respectively. Gains were influenced significantly ($P < 0.05$) by pre-weaning treatments. The calves fed MR and H only during the pre-weaning period were unable to catch up post-weaning with calves on other dietary treatments. The calves fed whole milk from birth at the rate of 10% of liveweight together with concentrates had higher weaning weight and superior growth rate post-weaning as well. Thus, pre-weaning feeding was important for higher weaning weights and superior growth rates post-weaning.

Implications

The calves fed milk replacer (MR) and hay only during the pre-weaning period had slower growth rates post-weaning compared with calves on other dietary treatments. The calves fed whole milk together with concentrates had higher weaning weights and superior growth rate post-weaning as well. Total feeding cost was higher, but cost per kg body weight (BW) gain was lower in milk-fed calves than in MR-fed calves during the pre-weaning period. However, feed conversion ratio and cost per kg of BW gain was similar post-weaning. Thus, for better performance during pre- and post-weaning periods, the nutrition of calves is important from the day they are born.

Introduction

The growth potential of livestock remains underutilized because of underfeeding and poor management. Neglect of dairy calves during the pre-weaning or post-weaning periods results in decreased growth rates and a delay in the onset of puberty in the female calves. Calves in Pakistan are generally neglected because of their high feeding costs and low returns from their sale at weaning (Bhatti et al., 2009).

Hence, farmers are not motivated to raise healthy male calves of any breed of cow or buffalo (Ahmad et al., 2009). Raising replacement dairy heifers is costly and no commercial gain is attained until heifers reach lactating age (Greter et al., 2010). Strategies to reduce feeding costs during the pre-weaning period are needed to motivate the farmer to raise the young calves for meat or breeding purposes. Low cost feeding strategies include using milk replacers (MRs), weaning the calves at an early age and increasing concentrate feed intake from a young age (Hopkins, 1997; Hill et al., 2010). This will lead to decreased age at first calving through accelerated growth during the post-weaning phase (Brown et al., 2005). However, the growth performance of calves fed MRs during the pre-weaning period, is inferior to that of milk-fed calves on an equivalent protein and energy basis until 45 days post weaning (Lee et al., 2009).

Kuehn et al. (1994) reported that low-fat (15.6% on dry matter (DM) basis) MRs promote higher starter dry matter intake than do high fat (21.6%) MR fed from 14 to 42 days of age and result in higher liveweight gain of Holstein calves. This suggests that the replacement of energy provided by fat with that in concentrate led to faster growth. Little is known of how tropically adapted breeds such as the Sahiwal respond to different milk feeding regimes and their effect on concentrate intake and long-term growth performance post weaning. To develop an economical calf rearing protocol for this breed, the effects of feeding milk or MR at the rate of 10% liveweight during the pre-weaning period on their post weaning performance using a single total mixed ration (TMR) were investigated.

Thus, the objective of the study was to assess whether the poor growth response pre-weaning to milk replacer relative to that for milk was offset beyond weaning at 84 to 168 days of age (24 weeks) in Sahiwal calves fed a balanced TMR.

Material and methods

Three-month-old weaned Sahiwal calves procured from the Livestock Experiment Station, Bahadurnagar, Okara, were used for this post-weaning feeding trial. Before weaning, they had been a part of a two by two factorial pre-weaning feeding trial reported by Bhatti et al. (2011). The two factors were – liquid feed (milk or MR) and solid feed (starter concentrate ration with or without hay). Thus, the four dietary treatments were: milk with starter ration plus hay (A: Milk 1SR 1H), milk with Berseem clover hay only (B: Milk 1H),

MR with starter ration plus hay (C: MR 1SR 1H) and MR with Berseem hay only (D: MR 1H). The milk and MR were fed at 10% of liveweight, whereas the SR and H were fed ad libitum and intake measured. The amount of milk or MR fed was adjusted for body weight (BW) until 156 days of age, and then tapered in weekly increments to zero by 84 days of age.

Berseem clover hay (*Trifolium alexandrinum*) was fed to all the calves from 14 days of age until weaning. Starter ration was fed to groups A and C starting from 14 days of age until weaning at 84 days of age. During the pre-weaning period, initially there were 12 calves in each group with six of each

Table 1 Composition of total mixed ration

Ingredients	As fed (%)	Ingredients	As fed (%)
Maize grains	33.4	Mineral mixture	2.0
Soybean meal	4.0	Vitamin pre-mix	0.1
Maize oil cake	7.0	Sodium bicarbonate	0.5
Maize gluten (60%)	7.0	Calcium oxide	1.0
Canola meal	15.0	CP	19.5
Molasses	10.0		
Wheat straw	20.0	Total digestible nutrients	70

sex, however, one male calf died from each of the B and C treatments and were not replaced. Thus, 46 calves were assigned to the current post-weaning study, comprised of 12, 11, 11 and 12 calves from the original preweaning groups A, B, C and D, respectively, as described by Bhatti et al. (2011). During the post-weaning period, after 84 days of age, the calves were given a single TMR (Table 1) until 168 days of age. The experiment was conducted from mid-May to mid-August 2009. The ambient day time temperature during these months ranged from 25.18°C to 40.48°C.

Feeding and housing

Animals were housed in separate calf pens (14231123112cm:L3W3H). Calves were fed a TMR containing 16% CP and 70% total digestible nutrients (Table 1) ad libitum from 85 to 168 days of age and had free access to clean water. The composition of the TMR is shown in Table 1. The TMR and water were placed in plastic bowls attached to the pens. Calves were fed twice daily at 0900 and 1700h, and refusals were measured to calculate feed intake. During the day, calves were allowed access to an open shed to exercise for 2h.

BW measurement

The animals were weighed weekly using an electronic digital scale early in the morning before feeding. The calves were off feed at midnight before weighing the next morning. The experiment was terminated at the age of 168 days (24 weeks) for each calf.

Data recording

Daily feed intake, refusals and weekly liveweight were recorded. The data recorded were used to calculate total liveweight gain, averaged daily gain, averaged daily feed intake, total feed intake, nutrient intake and total feeding cost.

Statistical analysis

The data on weekly weight and TMR intake were analyzed using repeated measures analysis using MIXED procedures of SAS (SAS, 2002), with an AR(1) covariance structure as described by Littell et al. (1998). The effects of pre-weaning diets were tested in the post-weaning period. The preweaning treatments were: A: Milk, SR and H; B: Milk and H; C: MR and SR and H; D: MR and H only (explained in Bhatti et al., 2011). The calf was used as a random effect. The statistical model used for analysis was $Y_{ijklm} = \mu + \text{sex}_i + F_1 + F_2 + W + \text{sex} \times F_1 + F_2 \times W + \text{sex} \times F_1 \times F_2 + W \times \text{sex} + F_1 \times \text{sex} + F_2 \times \text{sex} + F_1 \times F_2 + W \times F_1 + W \times F_2 + W \times \text{sex} + F_1 \times F_2 \times W + \text{sex} \times F_1 \times F_2 + \text{sex} \times F_1 \times W + \text{sex} \times F_2 \times W + F_1 \times F_2 \times \text{sex} + F_1 \times W \times \text{sex} + F_2 \times W \times \text{sex} + F_1 \times F_2 \times W \times \text{sex} + \text{error}$ where Y_{ijklm} is the dependent variable, μ is the overall mean, sex_i is the sex of the calf where i is either male or female, F_1 is the fixed effect of factor 1 where j is either milk or MR, F_2 is the fixed effect of factor 2 where k is either SR and H or H only, W is the repeated measure of weeks, l is the interaction term, m is the random effect of calf and e_{ijklm} is the residual error.

For the other descriptive statistics (average growth rate, weight at the age of 24 weeks, total weight gain, total intake of TMR), the data were analyzed using MIXED procedures.

The statistical model was $Y_{ijk} = \mu + F_1 + F_2 + F_1 \times F_2 + \text{sex}_i + F_1 \times \text{sex}_i + F_2 \times \text{sex}_i + F_1 \times F_2 \times \text{sex}_i + \text{error}$ where μ is the overall mean, F_1 is factor 1 where i is either milk or MR, F_2 is factor 2 where j is either SR and H or H only, $(F_1 \times F_2)$ is the interaction term, sex_i is the random effect of calf and e_{ijk} is the residual error. The sex was included in the initial model, but its effect was not significant so it was dropped from the model during final analyses. The results are reported as least squares means. The means were declared significantly different at $P < 0.05$.

Results

Feed intake

Averaged daily TMR intake of calves in all groups ranged from 1.5 to 2.260.17 kg/day during the 13th week of age (days 85 to 92) and 3.7 to 5.0 kg during 24th week of age (Figure 1). Daily intake of TMR increased with increasing age ($P < 0.05$). Averaged daily intake of TMR remained the highest in group A and lowest in group D during the post-weaning period (Figure 1). The average daily intake of calves in groups B and C was not

significantly different ($P < 0.05$). There was no interaction between liquid feed source and solid feed source. However, there was an interaction ($P < 0.05$) between solid feed source with age, indicating that the response in post-weaning TMR intake to pre-weaning solid feeding regimes period was not linear with age.

The total TMR intake of calves during the post-weaning period was higher ($P < 0.05$) in group A followed by groups B, C and D (Table 2). However, there was no significant difference ($P < 0.05$) between the intake of calves in groups B and C on a fed basis. The milk-fed calves during the pre-weaning period consumed more TMR post weaning ($P < 0.05$) than the MR-fed calves (Table 2). Similarly, the SR-fed calves during the pre-weaning period consumed more TMR post weaning ($P < 0.05$) compared with calves fed hay only. There was no interaction ($P < 0.05$) among treatments for total TMR intake (Table 2).

Growth performance of Sahiwal calves post weaning

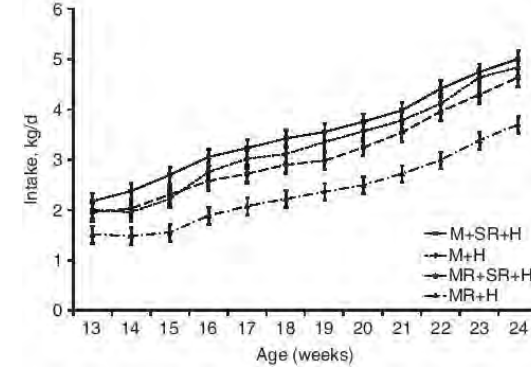


Figure 1 Effect of diet before weaning on the intake (kg/day) of total mixed ration (as-fed basis) post weaning in Sahiwal calves. M1SR1H: milk plus starter ration plus hay; M1H: milk plus hay; MR1SR1H: milk replace plus starter ration plus hay; MR1H: milk replace plus hay.

The TMR intake of calves (on a fed basis), as a percentage of their BW, ranged from 3.8 in group A to 4.86 in group C and was not affected ($P < 0.05$) by liquid feed source or solid feed source (Figure 2). However, the pre-weaning MR-fed calves tended to consume more TMR ($P < 0.07$) than milk-fed calves, whereas those fed hay before weaning tended to consume more TMR ($P < 0.09$) than concentrate-fed calves as a percentage of their liveweight during the post-weaning period. The incremental differences in TMR intake post weaning as a percentage of liveweight between the treatment groups increased ($P < 0.05$) with age (Figure 2). There was an interaction ($P < 0.05$) between source of solid feed before weaning and age, indicating that the addition of SR to H fed before weaning did not influence post-weaning intake.

Growth performance

Average daily BW gain of calves during the post-weaning period was highest in group A followed by groups B, C and D (Table 3). However, this did not differ ($P < 0.05$) between groups B and C. The pre-weaning milk-fed calves grew faster post weaning compared with MR-fed calves (Table 3). Similarly, SR-fed calves during the pre-weaning period grew faster during the post-weaning period than those fed hay only in the same period (Table 3). There was no interaction ($P < 0.05$) between liquid feed source and solid feed source or the two factors, liquid feed source seemed to have the most significant effect on BW gain post weaning.

The total weight gain during the post-weaning period, that is, from the 13th to 24th week of age, was 10 kg higher ($P < 0.05$) in calves fed milk than MR during the pre-weaning period (Table 3). Similarly, the calves fed SR before weaning gained 9.7 kg more post weaning ($P < 0.05$) than those fed hay only. There was no interaction ($P < 0.05$) between liquid feed source and solid feed source.

The final BW of calves at 24 weeks of age was 26.3 kg higher ($P < 0.05$) in calves fed milk before weaning than those fed MR (Table 3). Similarly, the calves fed SR before

Parameters intake	Main effects					Simple effects				P-values		
	Milk v. MR		SR v. hay		s.e.	Milk		MR				
	Milk	MR	SR + H	H		SR + H (A)	H (B)	SR + H (C)	H (D)			
TMR intake (kg)	267.8	216.3	265.4	218.8	8.0	282.5 ± 11.1	252 ± 11.6	246.8 ± 11.6	188.4 ± 11.1	***	**	ns
Intake (% of BW)	4.2	4.5	4.2	4.5	0.1	3.9 ± 0.1	4.4 ± 0.1	4.4 ± 0.1	4.5 ± 0.1	ns	ns	ns
Total feeding cost (PKR)	5356	4326	5308	4376	160	5650 ± 222	5040 ± 232	4936 ± 232	3768 ± 222	***	**	ns
Cost/kg liveweight (PKR)	90	88	90	88	2	90 ± 3	91 ± 3	90 ± 3	85 ± 3	ns	ns	ns

MR milk replacer; SR = starter ration; H = hay; TMR = total mixed ration; BW = body weight; PKR: Pakistan rupees (1US\$ ≈ PKR 87).

ns: non-significant, $P > 0.05$.

**Statistically significant, $P < 0.05$.

***Statistically highly significant, $P < 0.01$.

F1: factor 1, milk v. milk replacer.

F2: factor 2, SR + H v. H only.

F1 × F2, interaction of F1 and F2.

A, B, C and D refers to respective treatment groups.

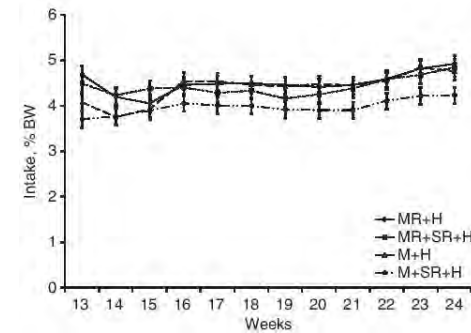


Figure 2 Effect of diet before weaning on the intake (kg/day) of total mixed ration (as-fed basis) post weaning expressed as a percentage of liveweight in Sahiwal calves. M1 SR1 H: milk plus starter ration plus hay; M1 H: milk plus hay; MR1 SR1 H: milk replacer plus starter ration plus hay; MR1 H: milk replacer plus hay.

weaning were 18 kg heavier ($P < 0.05$) than calves receiving only H before weaning. Similarly, there was no interaction ($P > 0.05$) between liquid feed source and solid feed source.

The post-weaning feed conversion ratio (FCR) of calves was not affected ($P > 0.05$) by any of the pre-weaning dietary treatments (Table 3). Feeding cost Total feeding cost of the calves during the 12 weeks post weaning was Pakistan rupees (PKR) 1030 (1US\$ PKR 87) higher in calves fed milk during the pre-weaning period than those fed MR (Table 2). Calves receiving SR before weaning cost PKR 932 more to feed post weaning than those fed H only. There was no interaction ($P > 0.05$) among the treatments.

The highest post-weaning total feeding cost during the 12 weeks was observed in calves receiving milk and SR during pre-weaning period and the lowest was in those fed MR and H only for the same period. However, the cost per kg BW gain post weaning did not differ ($P > 0.05$) among calves receiving any of the pre-weaning dietary treatments (Table 2).

Discussion

In the present study, the total TMR intake of calves post weaning fed milk during the pre-weaning period was higher than in MR-fed calves. Daily feed Intake is a function of BW of the animal. Khan et al. (2011) have concluded that post weaning feed intake of calves is governed by rumen volume, metabolic activity of rumen epithelium, rumen motility and feed quality. Higher TMR intake in the milk-fed calves, in the present study, was probably because of their greater digestive capacity (as a result of higher BW at weaning) than in MR-fed calves. The MR-fed calves during the pre-weaning period had lower weaning weights, and thus ate less than heavier calves at weaning. Khan et al. (2007) reported that depression in solid feed intake post weaning as a result of increased amount of milk feeding during pre-weaning period could be avoided if the calves are weaned gradually. Gradually weaned calves have no problem of depressed feed intake post weaning (Jasper and Weary, 2002). In the present study, the calves were gradually weaned from 9 to 12 weeks of age; thus, calves had adjusted to consuming solid feed during this period. This is perhaps another reason for the increased TMR intake post weaning by the calves fed milk during the pre-weaning period compared with the other treatments.

The daily BW gain of milk-fed calves during the pre-weaning period was higher than in MR-fed calves post weaning. The daily BW gain of growing animals is a function of the initial liveweight and nutrient intake. In the present study, the calves were fed ad libitum post weaning. Thus, nutrient availability was not the limiting factor, resulting in lower growth rate post weaning of MR-fed calves during the pre-weaning period. The higher average daily BW gain of milk-fed calves was because of their higher weaning weights. These calves continued to maintain the difference in their liveweight compared with the other groups (Figure 3).

Table 3 Least square means of post-weaning liveweight change and daily growth rate and FCR of Sahiwal calves as affected by different pre-weaning feeding regimens

Parameters	Main effects					Simple effects				P-values		
	Milk v. MR		SR v. hay		s.e.	Milk		MR		F1	F2	F1 × F2
	Milk	MR	SR+H	H		SR+H (A)	H (B)	SR+H (C)	H (D)			
Initial liveweight (kg)	52	35	49	38	0.8	56 ± 1	47 ± 1	40 ± 1	30 ± 1	***	***	ns
Final liveweight (kg)	110.8	84.5	106.2	88.3	2.2	119 ± 4.2	102 ± 4.3	95 ± 4.3	74.5 ± 4.2	***	***	ns
Total weight gain (kg)	59.4	49.4	59.2	49.5	1.8	63.0 ± 2.6	55.2 ± 2.7	55 ± 2.7	44.2 ± 2.6	***	***	ns
Daily BW gain (g/day)	705	588	702	590	24	746 ± 33	660 ± 35	653 ± 35	527 ± 33	**	**	ns
FCR	4.5	4.4	4.5	4.4	0.1	4.5 ± 0.1	4.6 ± 0.1	4.6 ± 0.1	4.3 ± 0.1	ns	ns	ns

FCR = feed conversion ratio; MR = milk replacer; SR = starter ration; H = hay; BW = body weight.

ns: non-significant, $P > 0.05$.

**Statistically significant, $P < 0.05$.

***Statistically highly significant, $P < 0.01$.

F1: factor 1, milk v. milk replacer.

F2: factor 2, SR + H v. H only.

F1 × F2, interaction of F1 and F2.

A, B, C and D refers to respective treatment groups.

Similar results have been reported by Jasper and Weary (2002), who reported that weight advantage of ad libitum-fed calves persists after weaning. They further argued that if this early opportunity of rapid growth by calves is missed, high levels of intake later in life may not allow for compensatory growth. Thus, our initial hypothesis that the MR calves may show compensatory growth if fed well after weaning could not be validated in this study with Sahiwal calves.

Similarly, the SR component of the pre-weaning diet was important, with SR-supplemented calves growing faster than comparable groups fed hay only. However, there was no difference in the growth performance of calves receiving MR+SR+H and milk+H only during the pre-weaning period. Presumably, the higher energy component of SR counteracted the superior nutrient availability in milk relative to MR. This study has confirmed that weaning weight, which was an indicator of pre-weaning dietary treatment, was the main variable influencing post-weaning growth performance. This is consistent with the observation that intake as percent of BW, the FCR and the feeding cost per kg of BW gain post weaning did not differ among calves receiving different pre-weaning dietary treatments. This indicates that, although the efficiency of feed utilization was similar in calves across all groups, calves with higher weaning weight maintained higher BW post weaning than all the other groups. Similarly, the calves receiving MR+H during the pre-weaning period, with lower weaning weights, could not catch up with the BW of calves receiving milk and SR even at the age of 24 weeks. This underlines the importance of pre-weaning feeding for early age at maturity.

The effects of pre-weaning dietary treatments on post-weaning growth performance of Holstein heifers were extended out to 600 days of age in the report of Moallem et al. (2010). In their study, the Holstein heifer calves fed whole milk (CP 525.9 and fat 529.4%, on DM basis) ad libitum during the pre-weaning period were heavier than those fed MR (CP 523.7 and fat 513%, on DM basis). The liveweight

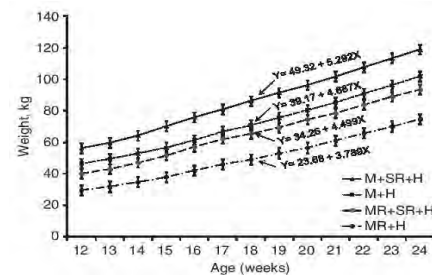


Figure 3 Effect of diet before weaning on liveweight change (kg) postweaning in Sahiwal calves. M+SR+H: milk plus starter ration plus hay; M+H: milk plus hay; MR+SR+H: milk replacer plus starter ration plus hay; MR+H: milk replacer plus hay

of milk-fed heifers, in their study, was 16.9 and 27.0 kg higher than that of MR-fed heifers at 300 and 600 days of age, respectively (P<0.04). Similarly with beef breeds, Christian et al. (1965) reported that heavier weaned Hereford calves reached slaughter grade of 'High Choice' in fewer days post weaning compared with lighter weaned calves. In this study, the heavier calves at weaning required more dry feed/kg of BW gain immediately after weaning, which is most likely due to higher body maintenance requirements.

Similar outcomes were observed by Robelin and Chilliard (1989). They reported that differences in MR intake (1380 v. 819 g/day on DM basis, respectively) up to 95 days of age resulted in a 25% heavier calf; however, subsequent growth rates up to 533 days of age were similar (806 v. 814 g/days), resulting in differential live weights at this age of 530 and 496 kg, respectively. However, Nocek and Braund (1986) reported that Holstein calves receiving either all-milk protein replacer (CP524.0 and ether extract 513.3%, on DM basis) or acidified milk replacer (CP523.0 and ether extract 56.9%, on DM basis) and weaned either abruptly or gradually reached 136 kg of live weight in approximately the same number of days (91 to 97).

Total feeding costs post weaning were higher in milk-fed calves than in MR-fed calves during the pre-weaning period; the same was true for the calves fed SR1Hv. This was because of the higher TMR intake in the respective groups. However, cost per kg BW gain post weaning was similar in calves receiving different pre-weaning dietary treatments. Thus, the higher total feeding costs post weaning in the calves fed milk or SR than those fed MR or H during the pre-weaning period were compensated with their superior growth rates in the same period. Conclusions Post-weaning growth performance of Sahiwal calves was established by the intake of nutrients before weaning.

Feeding whole milk (CP53.35 and fat 53.5%, on as-fed basis) from birth at the rate of at least 10% of live weight with concentrates lead to higher weaning weight and post-weaning growth rate, and hence may have a greater possibility for earlier puberty compared with feeding a milk replacer (CP52.22 and fat 51.8%, on as-fed basis) before weaning. Similarly, feeding the concentrate starter ration before weaning in preference to hay induced faster post-weaning growth rates. The cost per kg BW gain in Sahiwal calves was not different among treatment groups post weaning.

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Selecting an appropriate genetic evaluation model for selection in a developing dairy sector

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ABSTRACT:

This study aimed to identify genetic evaluation models (GEM) to accurately select cattle for milk production when only limited data are available. It is based on a data set from the Pakistani Sahiwal progeny testing programme which includes records from five government herds, each consisting of 100 to 350 animals, with lactation records dating back to 1968. Different types of GEM were compared, namely: (1) multivariate v. repeatability model when using the first three lactations, (2) an animal v. sire model, (3) different fixed effects models to account for effects such as herd, year and season; and (4) fitting a model with genetic parameters fixed v. estimating the genetic parameters as part of the model fitting process. Two methods were used for the comparison of models. The first method used simulated data based on the Pakistani progeny testing system and compared estimated breeding values with true breeding values. The second method used cross-validation to determine the best model in a subset of actual Australian herd-recorded data. Subsets were chosen to reflect the Pakistani data in terms of herd size and number of herds. Based on the simulation and the cross-validation method, the multivariate animal model using fixed genetic parameters was generally the superior GEM, but problems arise in determining suitable values for fixing the parameters. Using mean square error of prediction, the best fixed effects structure could not be conclusively determined. The simulation method indicated the simplest fixed effects structure to be superior whereas in contrast, the cross-validation method on actual data concluded that the most complex one was the best. In conclusion it is difficult to propose a universally best GEM that can be used in any data set of this size. However, some general recommendations are that it is more appropriate to estimate the genetic parameters when evaluating for selection purposes, the animal model was superior to the sire model and that in the Pakistani situation the repeatability model is more suitable than a multivariate.

Keywords: genetic evaluation model, Sahiwal cattle, genetic parameter estimation, cross-validation

Implications

This study is concerned with methods to assist in selecting the best dairy animals in developing dairy sectors such as Pakistan. Limited data are available in these sectors and so selection can be difficult. Genetic evaluation models can be used to help this process but models that are too simple or complex can lead to inaccurate or biased results. Based on the study outcomes, it is recommended to use a repeatability animal model for evaluating the first three lactations in which genetic parameters are estimated as part of the model fitting process.

Introduction

The genetic evaluation model (GEM) used in a country is dependent on the type of dairy system, the population of animals and the number of animals recorded for both performance and pedigree. Throughout the world there are varying levels of development in dairy sectors, and many of them have their own GEM. In developed dairy

sectors such as Australia and The Netherlands where the national herd size is ~1.5 million animals, about 45% of animals are being herd recorded (CRV, 2008; Australian Dairy Herd Improvement Scheme (ADHIS), 2011). In contrast, in developing dairy sectors performance records are seldom kept and therefore there is a vast difference in the options available for the genetic evaluation of animals. For example, in Pakistan there are ~2.5 million dairy cattle, but the most established progeny testing system in the country records <1000 milking animals per year. Furthermore, in developing dairy sectors, problems can be exacerbated due to the large environmental effects, poor management due to limited resources and poor data quality (Dahlin, 1998; Ilatsia et al., 2007). These problems have their implications and potentially reduce genetic gain. For example in developed dairy sectors, evidence shows genetic gains in milk production of ~2% per year (Hill, 2010), whereas looking at similar information from Pakistan we can see little to no increase (Khan et al., 2008). This difference in success is not confined to the Pakistani dairy system, but is common in low-input smallholder systems throughout the world (Rege et al., 2011).

The primary aim of any GEM is to select animals based on performance-recorded data as accurately as possible to maximize genetic gain in the population. In developed dairy sectors, complex GEMs such as random regression test-day models are used to account for as much variation as possible within the data (Interbull, 2009). Although a theoretically superior model, it requires sufficient recorded data to ensure the evaluation is accurate. In developing dairy sectors the number of recorded animals can often be very low and hence complex GEMs may not be feasible within their system. This can also occur in developed dairy sectors where there are animals being evaluated from small herds or where traits are recorded only on research farms. In these cases, simpler models involving less computation and requirements for recorded information may be helpful.

A particular problem with a low number of animals in an evaluation is to account for environmental or management effects for animals recorded from the same herd, year and season. These are generally fitted as fixed effects within the evaluation model and account for ~40% of the variation seen in milk and fat yield (Chauhan, 1987; Van Bebber et al., 1997). It is well established that ignoring fixed effects can lead to biased predictions of breeding values (Henderson, 1975a). However, it is also important to consider that when fixed effects are of little consequence, the resultant decrease in the contemporary group (CG) size may lead to an increase in prediction error variance. Furthermore, if records are obtained from closely related animals, then these records contribute little information to the evaluation (Van Vleck, 1987; Visscher and Goddard, 1993).

Therefore, the aim of this study was to assess the best GEM to predict breeding values in dairy cattle in Pakistan where there are limited data available. To address this aim, simulated datasets with known breeding values were used and compared with the estimated breeding values (EBVs) from various GEMs. Second, actual herd recorded data from an Australian progeny testing system was divided into subsets resembling the Pakistani progeny testing system. These subsets were then analysed using GEMs and breeding values were estimated and compared using cross-validation (CV).

Material and methods

Overall this study compares a number of different types of GEMs. The main factors considered are: (1) a multivariate (MV) v. repeatability (RP) model in accounting for production in the first three parities; (2) an animal (ANIM) v. sire (SIRE) model in modelling the random genetic effects; (3) different fixed effects models to account for effects such as herd, year and season; and (4) fitting a model with genetic parameters fixed v. estimating the genetic parameters as part of the model fitting process.

This was carried out by using historical data from Pakistan's Sahiwal progeny testing system to provide a basis of the herd structures and sizes. Subsequently, two methods were used for the comparison of models. The first method used simulated data based on the Pakistani progeny testing system. The second method used subsets of actual herd recorded data from the Australian dairy system to provide an indication of how the models compare based on actual industry data. Model comparisons were not carried out on the original Pakistani data set as it was only a single data set with small numbers of records per year and a small number of herds making CV less reliable.

Data

Pakistan milkyield data. Historical data from the Research Centre for the Conservation of Sahiwal Cattle, Jhang, Pakistan (RCCSC, <http://www.rccsc.com.pk/>), were used in this study as the basis for simulation and analysis. These records included both lactation and pedigree records from five major government Sahiwal herds involved in herd recordings since 1968. The herd size of these farms was between 100 and 350 milking animals. In total there were 29790 lactations from 310 sires and 6895 dams with an average number of lactations per dam of 4.3. The information available for each lactation record was the herd, date of calving, age at calving and in the majority of cases, the sire and the dam of the lactating animal.

Simulated data. Data were simulated based on genetic parameters estimated from a preliminary analysis of the RCCSC herd recorded data (see Supplementary material S1).

Specifically, additive genetic effects (breeding values, a) were generated for each of n animals and for three parities from a MV normal $N(0, G)$ distribution where

$$G = \begin{pmatrix} 2 & a_{12} & a_{13} \\ a_{21} & 2 & a_{23} \\ a_{31} & a_{32} & 2 \end{pmatrix} A$$

where A is the numerator relationship matrix based on the RCCSC pedigree, and where the additive genetic variances and covariances (a_{ij}) were based on the output of the preliminary analysis of the first three parities. Residual effects (e) were generated from a MV normal $N(0, R)$ distribution, where

$$R = \begin{pmatrix} 2 & e_{12} & e_{13} \\ e_{21} & 2 & e_{23} \\ e_{31} & e_{32} & 2 \end{pmatrix} I_n$$

where the residual variance-covariance matrix (R) was also based on the preliminary analysis, MV normal data were generated with the `rmvnorm` (Genz et al., 2013) function in R Version 3.0.2 (R Core Team, 2013). These additive and residual effects were summed to obtain phenotypes for the first three parities of each animal ($y = a + e$). Note that no McGill, Mulder, Thomson and Lievaart 1578 fixed effects were added to the simulated data (though fixed effect terms were included in the model fitting to assess the impact of CG size on the analysis, see below). Subsequently, a sample of second and third parity lactations were removed to mimic the culling and mortality levels seen in the original RCCSC dataset. This resulted in 1138, 921 and 698 first, second and third parities, respectively, the same as the original RCCSC dataset. This process was repeated 500 times to yield multiple simulated datasets to compare the GEMs in the study.

Actual herd recorded data. For assessing the GEMs on actual herd recorded data it would be difficult to obtain sufficient data sets from Pakistan to compare numerous models. So instead, historical test-day records were obtained from the ADHIS (<http://www.adhis.com.au>) and were used as a pool of data to draw subsets which resemble the general herd structure of the progeny testing records from Pakistan.

In total 178 dairy herds from Victoria with between 100 and 350 milking Holstein-Friesian animals each year from 1993 to 2002 were used to select five herds at random to represent the size of the Pakistan dataset. This was repeated 500 times and for each subset of data the test-day records were used to determine an adjusted 305-day lactation yield for the first three parities using the test-interval method (ICAR, 2009).

It is evident that Australian Holstein-Friesian animals do not truly represent the situation of the recorded Sahiwal population in Pakistan. However, for the purposes of this study by repeating the process of randomly selecting five herds of similar herd sizes to the Pakistan situation 500 times, we can assess the effect of different models with respect to fixed and random effects using CV. So although not truly representing the Pakistan situation, conclusions could be drawn from the Australian data by limiting the data included in its analysis. GEMs tested

$$y = Xb + Z_1a + Z_2pe + e(1)$$

where y is the vector of random variables of the recorded trait; b the vector of fixed effects with an incidence

matrix X relating observations to effects; the vector of additive genetic effects (or animal sire effects) with an incidence matrix Z_1 relating observations to random (polygenic animal sire) effects where $\sigma_a^2 = f\sigma^2$; A is the numerator relationship matrix and $f\sigma^2$ is the additive genetic sire variance; p is the vector of random permanent environmental effects with an incidence matrix Z_2 relating observations to permanent environmental random effects where $\sigma_{pe}^2 = f\sigma^2$; I is the identity matrix and $f\sigma^2$ is the permanent environmental variance; and the vector of independent residual effects where $\sigma_e^2 = f\sigma^2$. The MV model is:

$$y_i = X_i b_i + Z_i a_i + e_i \quad (2)$$

where the terms in the model (2) represent the same as in (1). However, there are now three traits indexed by i (that is parity one, two and three) and instead of a permanent environmental effect (p_e) for each animal there is a genetic (G) and residual (R) variance-covariance matrix such that:

$$G = \begin{pmatrix} \sigma_a^2 & a_{12} & a_{13} \\ a_{21} & \sigma_a^2 & a_{23} \\ a_{31} & a_{32} & \sigma_a^2 \end{pmatrix} \quad A \quad \text{and} \quad R = \begin{pmatrix} \sigma_e^2 & e_{12} & e_{13} \\ e_{21} & \sigma_e^2 & e_{23} \\ e_{31} & e_{32} & \sigma_e^2 \end{pmatrix} \quad I$$

assuming the data vector is stored in the form $y = (y_1, y_2, y_3)'$. For both the RP (1) and MV (2) model the random effects structure (Za) and the fixed effects (Xb) can be altered. In this study, the random effects were altered to compare the 'ANIM' model with the 'SIRE' model and the fixed effects structures (Table 1) were compared to see the impact of CG size and structure on animal evaluation.

The number of levels of each fixed effect was herd (five), year (upto 10) and parity (three). The number of levels of season was two, four or 12 as depicted within the brackets of Table 1. Age at calving was fitted as a second order polynomial effect.

In each of the simulated and actual herd recorded data runs, there were 20 models fitted and compared. That is, two model types (RP and MV), with two random effect structures (ANIM and SIRE) and five fixed effects structures (F1 to F5, from Table 1). All models were fitted using ASReml-R Discovery Edition 1.0 (Butler et al., 2009).

Table 1 Specification of fixed effect model structures

Model number	Fixed effects structure
F1	Herd \times parity + year \times parity + season[2] \times parity + AgeAtCalving \times parity + AgeAtCalving ² \times parity ¹
F2	Herd : year : parity + season[2] \times parity + AgeAtCalving \times parity + AgeAtCalving ² \times parity ¹
F3	Herd : year : season[2] \times parity + AgeAtCalving \times parity + AgeAtCalving ² \times parity ¹
F4	Herd : year : season[4] \times parity + AgeAtCalving \times parity + AgeAtCalving ² \times parity ¹
F5	Herd : year : season[12] \times parity + AgeAtCalving \times parity + AgeAtCalving ² \times parity ¹

The symbol ' \times ' indicates fields fitted with an interaction and ':' indicates concatenated fields which were fitted without the main effects.
¹The number of levels of season was two, four or 12 as depicted within the brackets.

The models were fitted with the estimation of genetic parameters included in the model fitting process, as well as fitted using fixed genetic parameter values. These fixed parameter values were obtained from the different analyses of: (1) the original RCCSC data that were used for the simulation study and (2) the entire Australian dataset of 178 herds. Details of these values can be found in Supplementary material S1 and S2.

Comparison of models

Convergence and estimation of genetic parameters. When genetic parameters are fixed, the model fitting process requires one non-iterative BLUP run to solve the generalized least square equations and therefore every model successfully returns estimates of fixed and random effects (e.g. breeding values). If genetic parameters are estimated as part of the model fitting process, the REML iteration process may fail to converge. Where parameters were estimated, the percentage of model fittings that converged was used to assess the robustness of

each model. In addition, the heritability (h^2), genetic correlation (rg) and other genetic parameter estimates of these models were compared to the true simulated values (Supplementary material S1) and ‘gold standard’ values obtained from the analysis of the ADHIS data set of 178 herds (Supplementary material S2).

Mean square error of prediction. For determining the ‘best’ model in each study, a mean square error of prediction (MSEP) was calculated and used to compare the ability of the model to estimate breeding values. In the simulation study, the simulated breeding values were compared directly with the EBVs predicted by each of the models. For the MV models, the EBVs were averaged over the three parities to yield a single EBV for comparison with the RP models. The MSEP was calculated for each model according to equation (3) (Mevik and Cederkvist, 2004).

$$MSEP_1 = \sum_{i=1}^n \frac{(\text{Estimated BV} - \text{Simulated BV})^2}{n} \quad (3)$$

where n is the number of records in the subset of data.

In the study using subset of actual herd recorded data from Australia, the true breeding values were unknown. Therefore, adjusted milkyields were calculated by subtracting the fitted fixed effects from the raw milkyields using the most complex model (MV-ANIM-F5) to the whole Australian dataset of 178 herds. These adjusted yields (y^a for the MV and y^{pe} for the RP) were compared to predicted sire effects (predicted yields; y^s for the SIRE models) or additive genetic effects (predicted yields; y^a for the ANIM models).

ACV procedure was conducted to assess the ability of the models to estimate breeding values with smaller subset of data. This was repeated ten times for each model in every run. In this procedure, 90% of the animals in the subset of the data for each run were selected at random and used to fit the model. The remaining 10% had their yields predicted using the model output. The MSEP for the analysis using actual herd recorded data was calculated for each of the 10 CV procedures using the following equation (Mevik and Cederkvist, 2004):

$$MSEP_2 = \sum_{i=1}^n \frac{(\text{Predicted Yield} - \text{Adjusted Yield})^2}{n} \quad (4)$$

An average of the 10 CVs was taken to be the MSEP2 for that particular model and run. For both the simulation method and the CV method using actual data, the model yielding the lowest MSEP2 for each run was considered to be the ‘best’ model for that particular dataset.

Sire ranking. The correlation and corresponding rankings of the EBVs of the sires evaluated was used as a secondary check on the differences between models. For each subset of data, the EBV of all sires with greater than five daughters was compared with the ranking according to: (1) the true EBVs for the simulation study; and (2) the EBVs from the ‘gold standard’ model output (MV-ANIM-F5) for the study on actual data. This is an important verification step as the sire ranking and selection is the primary outcome following the genetic evaluation process.

Results

Convergence

When genetic parameters were estimated, the results for both the simulated datasets and the subsets of actual herd recorded data (Table 2) show the RP model was the most robust model as it had the highest success rate of model fittings. With the MV models, the convergence rates are much lower than the RP models suggesting that these models fail to estimate the genetic covariance between parities one, two and three. Within the simulated datasets, the SIRE model was slightly more successful than the ANIM model in contrast to the results from the actual herd recorded data in which the ANIM model was more successful. Models that failed to converge were not included in subsequent analysis or calculations.

Table 2 Percentage of converged models for the 500 simulated datasets and 500 subsets of actual herd recorded data where genetic parameters were estimated as part of the model fitting process.

Table 4 Percentage of times from 500 simulated data sets that the specified model had the lowest MSE_{P1}

Fixed model ¹	Genetic parameters fixed				Genetic parameters estimated			
	Multivariate		Repeatability		Multivariate		Repeatability	
	Animal	Sire	Animal	Sire	Animal	Sire	Animal	Sire
F1	76.20	0.00	5.00	0.00	25.20	0.20	43.20	0.00
F2	10.40	0.00	0.80	0.00	6.20	0.00	10.20	0.00
F3	4.60	0.00	0.40	0.00	4.00	0.00	4.20	0.00
F4	2.40	0.00	0.00	0.00	2.80	0.00	1.40	0.00
F5	0.20	0.00	0.00	0.00	0.80	0.00	1.80	0.00
Total	93.80	0.00	6.20	0.00	39.00	0.20	60.80	0.00

This was calculated separately for models when genetic parameters were fixed and estimated.

¹Fixed model: F1 to F5 refer to the fixed effects structures described in Table 1.

Table 5 Average estimates of heritability (h^2) of the actual herd recorded data and their standard deviations for the repeatability models of 500 subsets of data

Fixed model ¹	Animal		Sire	
	h^2	s.d.	h^2	s.d.
F1	0.329	0.089	0.255	0.252
F2	0.311	0.090	0.205	0.181
F3	0.312	0.090	0.209	0.183
F4	0.316	0.093	0.218	0.185
F5	0.321	0.095	0.222	0.186

¹Fixed model: F1 to F5 refer to the fixed effects structures described in Table 1.

Simulated data

the 'gold standard' than that of the RP-SIRE. Considering only the RP models, the RP-ANIM-F1 model was the one with the closest estimate to the 'gold standard'. The average estimates of genetic parameters for the MV analysis can be seen in Table 6. The results for the F2 and F4 models are not shown here, but they were similar to those presented for F3. From Table 6 we can see that the h^2 estimates for the F1 are higher than that of F3 and F5. In addition, the estimates for the r_g between parities when using the F1 model are lower. Comparing these values

Table 6 Average estimates of genetic parameters (r_g , h^2) for parities one, two and three and their standard deviations as calculated from the F1, F3 and F5 multivariate models on actual herd recorded data

Fixed model ¹	Random model	Parity	h^2	s.d.	Correlation between parity X and Y	r_g	s.d.
F1	Animal	1	0.413	0.107	1 with 2	0.770	0.140
		2	0.355	0.106	2 with 3	0.873	0.132
		3	0.397	0.128	1 with 3	0.715	0.161
	Sire	1	0.648	0.301	1 with 2	0.677	0.217
		2	0.540	0.252	2 with 3	0.816	0.178
		3	0.550	0.255	1 with 3	0.557	0.264
F3	Animal	1	0.369	0.104	1 with 2	0.886	0.110
		2	0.300	0.109	2 with 3	0.974	0.140
		3	0.348	0.132	1 with 3	0.879	0.125
	Sire	1	0.296	0.122	1 with 2	0.801	0.196
		2	0.239	0.124	2 with 3	0.946	0.230
		3	0.287	0.160	1 with 3	0.803	0.237
F5	Animal	1	0.381	0.106	1 with 2	0.894	0.121
		2	0.318	0.117	2 with 3	0.973	0.135
		3	0.355	0.143	1 with 3	0.873	0.163
	Sire	1	0.294	0.124	1 with 2	0.782	0.407
		2	0.234	0.126	2 with 3	0.953	0.356
		3	0.291	0.161	1 with 3	0.797	0.291

¹Fixed model: F1 to F5 refer to the fixed effects structures described in Table 1. The results for the F2 and F4 models are not shown here, but they were similar to those presented for F3.

to each other, it seems that the h^2 of the MV-ANIM-F1 model are the closest to the 'gold standard' (0.429, 0.344 and 0.378). However, the values of the F1 model are very low compared with the 'gold standard' (0.945, 0.926 and 0.996). Contrastingly, the MV-ANIM-F5 model estimates of r_g are much closer to the 'gold standard' than that of the MV-ANIM-F1 but the h^2 estimates are slightly lower. Despite this, it seems that the MV-ANIM-F5 model estimates are the closest overall.

The greatest difference from these results is between the h^2 and r_g estimates from the ANIM and SIRE models. Not only do the estimated values differ, but the standard deviation values from the SIRE model are much higher than the ANIM. This again suggests that the ANIM model is more precise with datasets of this size. Model comparison. Using MSE_{P2}, Table 7 shows the percentage of times where each model was considered best when genetic parameters were fixed or estimated. When genetic parameters were fixed, the MV model (69.6%) was superior to the RP (30.4%), the ANIM model (92.0%) was superior to the SIRE (8.0%) and the fixed effects

models F3 (24.2%), F4 (23.8%) and F5 (23.8%) were considered superior more often than the F1 (10.4%) and the F2 (17.8%) models. When genetic parameters were estimated, the MV (44.4%) and RP (37.8%) models were similar, the ANIM model (82.2%) was still superior to the SIRE (17.8%) and models with increasing complexity of fixed effects were more frequently the best model (F1 to F5: 9.6%, 18.4%, 21.0%, 24.0% and 25.0%).

The overall outcome using actual herd recorded data shows that when genetic parameters are fixed or estimated, the MV-ANIM-F5 model is considered to be the superior model. This is in contrast to the results from the

Table 7 Percentage from 500 runs that the specified model had the lowest MSE_{P2} for the subset of five selected herds from the actual herd recorded data

Fixed model ¹	Genetic parameters fixed				Genetic parameters estimated			
	Multivariate		Repeatability		Multivariate		Repeatability	
	Animal	Sire	Animal	Sire	Animal	Sire	Animal	Sire
F1	5.8	0.4	4.0	0.2	2.8	1.0	5.6	0.2
F2	10.8	1.0	5.6	0.4	9.8	2.8	7.2	0.6
F3	14.8	1.0	6.6	1.8	9.4	2.8	7.4	1.4
F4	16.2	1.2	5.4	1.0	10.4	3.8	9.2	0.6
F5	18.2	0.2	4.6	0.8	12.0	3.6	8.4	1.0
Total	65.8	3.8	26.2	4.2	44.4	14.0	37.8	3.8

¹Fixed model: F1 to F5 refer to the fixed effects structures described in Table 1.

simulated data study which determined the MV-ANIM-F1 model to be superior when parameters were fixed and the RP-ANIM-F1 when parameters were estimated.

Sire ranking. Despite the differences in the MSE_{P2} the correspondence between the sire rankings from the output of each of the models varied very little for the herd recorded Australian data (see Table 8). These results show that the number of sires ranked in the top 10 sires from each model output had a narrow range with the lowest average across all models being 3.37 (MV-SIRE-F1) and the highest average value at 4.31 (MV-ANIM-F2 and RP-ANIM-F2). Furthermore, the correlation between the EBVs of the different tested models with the

Table 8 Mean number of corresponding sires with the 'gold standard' model in the top 10 breeding value rankings when calculated from actual Australian herd recorded data using both fixed and estimated genetic parameters

Random model	Fixed model ¹	Genetic parameters fixed				Genetic parameters estimated			
		Multivariate		Repeatability		Multivariate		Repeatability	
		Mean	s.d.	Mean	s.d.	Mean	s.d.	Mean	s.d.
ANIM	F1	3.39	1.38	3.39	1.39	3.56	1.39	3.59	1.42
	F2	4.31	1.43	4.31	1.42	4.00	1.38	4.15	1.35
	F3	4.28	1.46	4.30	1.45	3.99	1.34	4.13	1.35
	F4	4.19	1.52	4.22	1.43	4.04	1.38	4.11	1.34
	F5	4.26	1.42	4.13	1.45	3.94	1.32	4.11	1.34
SIRE	F1	3.37	1.41	3.43	1.44	3.42	1.37	3.49	1.40
	F2	4.18	1.43	4.11	1.38	3.92	1.40	4.03	1.38
	F3	4.17	1.44	4.11	1.39	3.95	1.38	3.99	1.37
	F4	4.18	1.44	4.07	1.5	3.98	1.41	4.01	1.38
	F5	4.19	1.48	4.14	1.42	3.86	1.37	3.99	1.39

¹Fixed model: F1 to F5 refer to the fixed effects structures described in Table 1.

'gold standard' EBVs ranged from 0.980 to 0.985 with standard deviations of ~0.01 for all models. These statistics demonstrate that sire rankings between the models compared were both highly correlated and showed little variation in selection outcomes. Discussion Data The primary aim of this research was to assess the best GEM to predict breeding values in Pakistan dairy cattle when there are limited data available.

Before discussing the results, it is important to first highlight some key assumptions that will affect breeding value estimation in any situation where data may be limited or of poor quality. A key problem is the accuracy of the pedigree information. Research shows that pedigree misidentification is common (Visscher et al., 2002; Weller et al., 2004; Sanders et al., 2006) and can reduce the accuracy of breeding values and hence reduce genetic gain (Sanders et al., 2006). This is likely to be an even greater problem in Pakistan, but in the short-term is

unavoidable. Therefore, for the purposes of this study it is assumed that the pedigree errors will have an equal effect on the different models tested. Keeping this in mind, the outcomes of this work are discussed below relating to convergence rates, estimation of genetic parameters, and finally the choice of model.

Convergence

From both the simulated data and actual data it was apparent that when using small datasets, a high number of MV model fittings failed to converge when genetic parameters were estimated. This suggests that these models may not be suitable and instead a RP model would be more appropriate, because fewer parameters need to be estimated. Comparing between the ANIM and SIRE model the rates differed slightly between the two datasets. However, this difference is more likely a reflection on the depth of pedigree rather than an implication for the model of choice.

The pedigree for the Australian data is more accurate and contains fewer gaps in parental information than the Pakistani pedigree used for the simulation. For this reason, the advantages of the ANIM model over the SIRE model could not be exploited with the simulated data, whereas in contrast, the Australian data could. Estimation of genetic parameters Results from both the simulated data and actual herd recorded data show that in some cases although model fittings may converge and yield genetic parameter estimates, they may yield biased genetic parameters or violate assumptions made. For example, the MV model r_{g} estimates, although close, are less than one and the variance components of the first three parities are quite different (Supplementary material S2). These values suggest that the RP model assumption, that each parity is genetically the same trait, is not correct. This is consistent with the literature which generally reports the three parities as separate traits (Weller, 1986; Schaeffer et al., 2000; Powell and Norman, 2006). Furthermore, the ANIM model would be more suitable than the SIRE model as the genetic parameter estimates are closer to the correct values and much more precise as shown by the lower standard deviations.

Looking further into the MV results from the actual herd recorded data (Table 6), estimates of h^2 in the F2 to F5 SIRE models were generally lower, by 20% to 30%, compared with both the ANIM models and the 'gold standard' using the whole Australian dataset (Supplementary material S2). This is presumably due to the inclusion of more complete relationships in the ANIM model as ignoring relationships that exist results in a reduction of estimates of genetic variance (Henderson, 1975b). In contrast to the h^2 estimates, the estimates of r_{g} were not so much affected by the random effects in the model (ANIM or SIRE) as expected (Don et al., 1988). However, the r_{g} estimates from the ANIM model were where genetic parameters are estimated a closer examination of the fixed effects models can be carried out. These results show clearly that the F1 model is superior (43.2%) more times than models F2 to F5 (10.2%, 4.2%, 1.4% and 1.8%) when using the simulated Sahiwal data (Table 4).

However, when using the actual herd recorded data the distinction is not as clear with the model superiority being very similar ranging only from 5.6% to 9.2% (Table 7). Therefore, as discussed earlier it is difficult to recommend a specific fixed effects structure for datasets of limited size.

This aligns with the generally accepted view that every dataset and structure is unique and hence it is difficult to make general statements about the most suitable model to analyse it (Henderson, 1975c). Consequently, we would refer to the general recommendation in the literature to keep the average CG size between 8 and 25 and to have no less than three records with each CG (Urgate et al., 1992; Van Bebber et al., 1997).

Conclusions

This paper aimed to select and recommend the best model to use for genetic analysis in Pakistan's dairy sector where limited data are available. Although a specific fixed effects model structure could not be chosen, broad recommendations can be given regarding the type of GEM to be implemented. The main outcome of this research suggests that applying a RP animal model where genetic parameters are estimated appeared to be the best GEM for the Pakistani Sahiwal progeny testing system.

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Pattern of partitioning of aflatoxins from feed to urine and its effect on serum chemistry in Nili-Ravi buffalo heifers

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Abstract

The objectives of the present study were (1) to monitor the pattern of excretion of aflatoxin M1 in urine after its conversion from aflatoxin B1 and (2) to observe the effects of different levels of aflatoxin B1 in feed on serum concentrations of key metabolites glucose, total protein, cholesterol and urea as indicators of metabolic status. Nili-Ravi buffalo heifers (n=12) of similar age and weight were randomly distributed to four groups. Animals in Groups A, B and C were offered a contaminated cottonseed cake-based concentrate at 0.5%, 1.0% and 1.5% of body weight, respectively. Control animals in Group D were fed with aflatoxin B1-free green fodder. Based on the level of contamination of the concentrate with aflatoxin B1 (554 mg/kg), Groups A, B and C consumed 953, 2022, 3202 mg of aflatoxin B1 daily. Feed samples were analysed at Romer Laboratories Pty Ltd, Singapore by high performance liquid chromatography. Aflatoxin M1 quantification in urine samples was conducted using a competitive enzyme-linked immunosorbent assay with kit supplied by Helica Biosystems, Inc., USA. Serum samples were analysed for concentrations of glucose, total protein, cholesterol and urea using clinical chemistry kits provided by Human Diagnostics (HUMAN, Biochemica und Diagnostica mbH, Germany). Carry-over rate of aflatoxin M1 in urine for Groups A, B and C was 15.51%, 15.44% and 14.04% of aflatoxin B1 while there was no detectable aflatoxin M1 in the urine of the control group (D). There was no significant difference in the concentrations of serum glucose, total protein and cholesterol between treatment groups. However, the concentration of serum urea was significantly higher ($P < 0.05$) in the group offered the highest level of aflatoxin B1-contaminated concentrate. This result suggests that mycotoxicosis may compromise protein metabolism and accretion in affected animals. This leaves open the possibility that high concentrations of aflatoxins in milk may ultimately affect the health status of human milk consumers.

Additional keywords: AFLB1, AFM1, mycotoxins, transferrate.

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Introduction

Aflatoxins (mycotoxins produced by *Aspergillus flavus* and *A. parasiticus*) are readily absorbed and distributed to almost all vital organs and body fluids after ingestion (Stubblefield et al. 1981). The microsomal cytochrome P450 system in liver facilitates both activation and deactivation of aflatoxin B1 (AFB1). Oxidation of AFB1 results in the formation of the biologically active metabolite, AFB1-8,9-epoxide (Kuilman et al. 2000). This metabolite can then react with RNA and DNA leading to hepatocellular carcinoma or with liver protein (Judah et al. 1993) to cause liver toxicity. Aflatoxin B1-8,9-epoxide is then converted into several less toxic metabolites such as aflatoxin M1 (AFM1), aflatoxin Q1 (AFQ1) and aflatoxin P1 (AFP1) after hydroxylation (Kuilman et al. 2000). The body has a mechanism to regulate toxicity through conjugation of AFB1 with glutathione, facilitated by glutathione S-transferases (Hayes et al. 1991). The kidneys, lungs, liver and mammary glands were found to sequester the highest concentrations of total aflatoxins (Stubblefield et al. 1983) while brain, gall bladder, bile, small intestine, heart, skeletal muscles, spleen, supra-mammary lymph nodes and tongue were also found to retain considerable amounts of aflatoxins. Truckness et al. (1983) found that the transfer of aflatoxin B1 and AFM1 to milk, plasma and red blood cells of the cattle is very rapid, reaching high levels within 1 h of dosing. Consistent with this rapid increase, the circulatory system is highly efficient at eliminating aflatoxin metabolites

through milk and urine. Stubblefield et al. (1983) found concentrations of AFM1 in kidney to be almost 40 times higher than the intact AFB1, showing extensive metabolism of the original feed contaminant in cattle. Thus urine is one of the major routes for excretion of AFM1 after its conversion from AFB1 in the liver (Nabney et al. 1967).

Pakistan is second in the world in buffalo milk (22.96 million tonnes) and meat (0.775 million tonnes) production after India (FAO 2011). Ingredients used as concentrate are often contaminated with fungi, which secrete mycotoxins that are then incorporated into the feed base (Sultana and Hanif 2009).

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Animal Production Science, 2014, 54, 1671–1675 <http://dx.doi.org/10.1071/AN14302> Journal compilation CSIRO 2014 www.publish.csiro.au/journals/an The pattern of secretion of AFM1 in buffalo urine may provide a means for measurement of mycotoxin likely to be stored in carcasses providing meat entering the human food chain. The objectives of the present study were (1) to determine the relationship between feed AFB1 status and urinary AFM1 contents and (2) to observe the effect of mycotoxins on key serum constituents in Nili-Ravi buffalo heifers.

Materials and methods

The Government Buffalo Research Institute, Pattoki (latitude: 31°50'N and longitude: 73°52'E) district Qasur of province Punjab in Pakistan provided Nili-Ravi buffalo heifers (n=12) of similar age (18.7–20.4 months) and liveweight (339–387 kg), which were randomly assigned to four groups offered with different levels of AFB1 naturally contaminated concentrate feed. Groups A, B and C were fed with contaminated concentrate at 0.5 (1.72 kg), 1.0 (3.65 kg) and 1.5 (5.78 kg) % of body weight, respectively; while Group D was kept as the control (animals were only fed with fresh green fodder, free of AFB1). Animals in each group were offered individually above mentioned contaminated concentrate and ad libitum AFB1-free green fodder (Berseem, *Trifolium alexandrinum*) for 10 days before the start of the experimental period of 5 days. The concentrate ration was highly contaminated with AFB1 (554 mg/kg). Total daily intakes of AFB1 for animals in Groups A, B, C and D were 953, 2022, 3202 and 0 mg over the 5 days, respectively. The AFB1-free green fodder was also available ad libitum during the 5-day test period and daily intake was recorded by weighing back feed residuals. Water was made available ad libitum and intake was measured on a daily basis. Total daily dry matter (DM) intake was calculated by adding the DM % of concentrate and green fodder. Total daily excretion of AFM1 was calculated by multiplying concentration of AFM1 (mg/L) by total urine production of that day.

Sample collection

Feed samples

A representative sample of green fodder was dried and preserved on Day 1 together with a representative concentrate sample. These were analysed for AFB1 at Romer Laboratories, Singapore.

Serum samples

Blood samples (10 mL) were collected by venipuncture using disposable syringes (19-gauge needles) and stored at 23°C for 2 h for serum to form. They were then centrifuged at 1200 g at 25°C for 20 min to collect the serum, which was stored at 20°C pending analysis. Concentrations of glucose, total protein, cholesterol and urea were determined by using clinical chemistry kits provided by Human diagnostics (HUMAN, Biochemica und Diagnostica mbH, Wiesbaden, Germany). Serum samples were analysed using a chemistry analyser (Microlab 300) provided by the ELI Tech Group, Paris, France.

Urine samples

Foley balloon catheters (24-gauge, Ningbo Greatcare Meditech Co. Ltd, Zhejiang, China) were passed through the urethra into the urinary bladder of all animals on Day 1 after the adjustment period of 10 days and kept there

for 5 days. These catheters were directed to airtight plastic bottles (20L). Urine samples (5mL) were collected every 24 h after mixing and stored at 4°C pending analysis. All animals were monitored continuously after catheter introduction.

Aflatoxin B1 analyses

Samples of green fodder and contaminated concentrate ration were sent to Romer Laboratories, for AFB1 analysis and were analysed by high performance liquid chromatography (HPLC). Samples were also analysed for AFB2, deoxynivalenol, fumonisin B1, ochratoxin A and zearalenone. Sample preparation and cleanup: A total of 2.5g of sample was ground. Ground sample was mixed well and then extracted with 100mL acetonitrile/water (84:16). After blending for 3min, it was filtered through folded filter paper. Tween 20 (33mL of 1% in PBS) was then added in 2mL of filtrate (acetonitrile/water sample extract). All diluted sample extract was applied to AflastarFit (immunoaffinity column) columns. The sample was allowed to pass through the column at the rate of 1–3mL/min. The column was then washed with 10mL of PBS. All excess liquid was removed and toxins were eluted from the column by applying two times 0.5mL methanol followed by two times 0.5mL of deionised water. After mixing, 100mL was injected into HPLC.

HPLC

HPLC analyses were performed using an HPLC series 1100 from Agilent Technologies (Waldbronn, Germany). Chromatographic separation of AFB1 was conducted by use of an Agilent Zorbax SB-Aq column (4.6mm × 150mm, 5µm). The mobile phase applied was water/acetonitrile/methanol mixture (5/1/1), including 100mM nitric acid and 0.3g potassium bromide per L. The flow rate was 2mL/min, column oven temperature 30°C, injection volume 100µL. A Kobra cell was used for post-column derivatisation, fluorescence detector settings were 360nm (excitation), 440nm (emission).

Aflatoxin M1 analyses in urine

AFM1 quantification in urine samples was conducted with a competitive enzyme-linked immunosorbent assay (ELISA) kit (Cat. No. 991AFLMO1Y-96 Helica Biosystems Inc., Santa Ana, CA, USA). Mean recovery of AFM1 in spiked samples (0.5 and 2.0ng/L) according to the manufacturer's specifications were 96.40% and recovery range was 78–111%. Carry-over rate of aflatoxin M1 in urine: The carry-over rate of AFM1 in urine was calculated by following the formula:

$$\text{Carry-over rate} = \frac{\text{Total daily AFM1 excreted}}{\text{Total daily AFB1 intake}} \times 100$$

Statistical analyses

Data were measured 5 times on each animal so they were analysed using a mixed model with Group*Day as fixed effects and Animal/Day as random effects. As data for serum (1672 Animal Production Science N. Aslam et al. concentrations of glucose, total proteins, cholesterol and urea were measured at only one time point, they were analysed by one-way ANOVA using a completely randomised design.

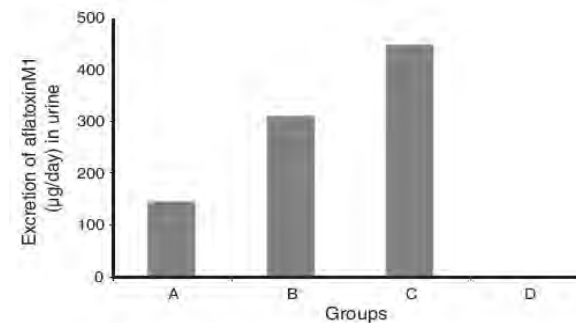


Fig.1. Total daily mean excretion of aflatoxin M1 in urine of buffaloes exposed to different levels of aflatoxin B1 ($P < 0.001$, s.e.d. = 30.07). Note: Animals in Groups A, B, C and D were exposed to 0, 53, 202, 320 and 0 mg/day, respectively.

Results

There was a significant ($P < 0.001$) difference in total daily excretion of AFM1 in urine among the four groups; higher levels of AFM1 in urine were associated with higher levels of consumption of AFB1 (Fig. 1). There were no significant differences in urinary excretion between days, with levels remaining constant over the 5 days of treatment (Fig. 2). A highly significant difference ($P < 0.001$) was observed in urinary AFM1 concentrations (mg/L) among Groups A, B, C and D. The mean concentrations of AFM1 (mg/L) were 14.37, 18.96, 24.17 and 0 for the animals fed with 953, 2022, 3202 and 0 mg/day of AFB1, respectively. There was no difference in the daily pattern of excretion of AFM1 (mg/L) over the 5-day experimental treatment period. A strongly significant ($P < 0.01$) difference in water intake was observed among all four experimental groups (Table 1). It was positively related with daily intake of the concentration. There was a highly significant ($P < 0.001$) difference in water intake from day to day. Mean water intake on Day 1 for all animals was 21.72 L, which was

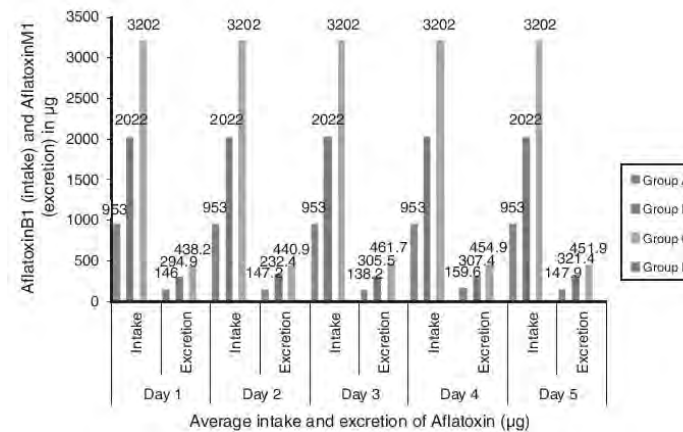


Fig. 2. Daily variation in excretion pattern of aflatoxin M1 in buffaloes exposed to various levels of aflatoxin B1 ($P > 0.05$; s.e.d. = 29.00). Note: animals in Groups A, B, C and D were exposed to 953, 2022, 3202 and 0 mg/day, respectively.

Table 1. Mean values for daily urine production, water intake, dry matter intake, carry-over rate, excretion of aflatoxin M1 and mean concentrations of serum constituents in buffaloes exposed to different levels of aflatoxin B1

Group	Urine production (L/day)	Water intake (L/day)	Dry matter intake (kg/day)	Aflatoxin B1 intake in feed (µg/day)	Aflatoxin M1 excretion in urine (µg/day)	Carry-over rate of aflatoxin M1 in urine (%) ^A	Glucose (mg/dL)	Total protein (mg/dL)	Cholesterol (mg/dL)	Urea (mg/L)
A	10.35	23.01	8.139	953	147.8	15.51	79.0	6.40	67.7	46.0
B	16.59	29.04	10.002	2022	312.3	15.44	65.7	6.17	88.0	62.7
C	18.67	33.35	12.252	3202	449.5	14.04	70.3	6.27	76.0	69.0
D	9.39	20.81	6.623	n.a.	0	n.a.	73.3	6.30	73.3	46.0
P-value	<0.001	≤ 0.01	<0.001	n.a.	n.a.	n.a.	0.744	0.990	0.617	0.030

^ACarry-over rate = aflatoxin M1 excretion in urine / aflatoxin B1 intake in feed × 100.

significantly ($P < 0.001$) lower than mean intakes for the subsequent days, i.e. 26.37, 27.68, 28.78 and 28.21 L for second, third, fourth and fifth day, respectively (s.e.d. = 1.368). Consequently urine production in animals from each group was significantly ($P < 0.001$) different (Table 1). There was no significant difference in urine production from day to day. A highly significant ($P < 0.001$) difference in total DM intake was detected between groups and days. Total DM intake was significantly ($P < 0.001$) lower for the control animals (6.623 kg/day) relative to Groups A, B and C (8.139, 10.002 and 12.252 kg/day, respectively). Total DM intake for all groups on Day 2 was 8.945 kg, which was significantly lower ($P < 0.001$) than the values for the other days. No interactions were apparent between groups and days for total AFM1 excretion per day and per litre of urine, daily water intake, daily urine production or daily total DM intake.

Difference in the mean blood concentrations of glucose, total protein and cholesterol among treatment groups were not significantly influenced by treatment (Table 1). Mean concentration of urea in blood serum was significantly higher ($P < 0.005$) in Group C exposed to the highest AFB1 concentration than all other groups.

Discussion

Deaths of several hundred calves in Australia (McKenzie et al. 1981), numerous animal deaths on a chinchilla farm in Argentina (Pereyra et al. 2008) and the death of 493 buffaloes in Landhi colony Karachi, Pakistan (Sultana and Hanif 2009) provide examples of the potential impact of acute aflatoxicosis in production animals. Nabney et al. (1967) reported a carry-over of AFM1 in urine of up to 5.94% in sheep. This result is clearly different from the results (14–15.5%) produced in the present study. The reason for this difference may relate to species differences as Nili-Ravi buffalo heifers were used in this experiment. Another reason may be the resistance of sheep to mycotoxins. AFM1 was carried over at the rate of 1.23–2.18% of AFB1 in the urine of humans fed with contaminated corn and peanut oil in another study conducted by Zhu et al. (1987) when average daily intake of AFB1 was 58 mg/day. The reason for this reduced carry-over rate may be the difference in physiology between humans and ruminants.

A total of 4.52% of the aflatoxin ingested was excreted through milk (0.18%), urine (1.55%) and faeces (2.79%) in a study with beef cattle (Allcroft et al. 1968). The apparent lower transfer in this study could relate to the single dose of aflatoxin administered to animals. The sensitivity of the analytical technique (ELISA) used in the present study relative to the methodology in use 40 years ago may also contribute to the differences. The mechanisms of transfer may also vary between the heifers used in this study and lactating cattle. Different factors may influence the variation in carry-over rate (0.3–6.2%) of AFM1 in milk (Creppy 2002).

Transfer of mycotoxins to the human food chain is particularly common in developing countries. In Cameroon for example, 35.5% and 45.5% of the urine samples of children suffering from kwashiorkor and Marasmic kwashiorkor were positive, having mean values of 0.109–2.840 mg/L and 0.109–0.864 mg/L, respectively (Tchana et al. 2010). Another reason for the difference in AFM1 concentration in urine may be the synergistic effect with the presence of other mycotoxins. The concentration used in the present study contained 50, 166, 230, 31.2 and 18 mg/kg of aflatoxin B2, deoxynivalenol, fumonisin B1, ochratoxin A and zearalenone, respectively. Various studies (Pozzi et al. 2001; Rajmon et al. 2001; Gelderblom et al. 2002) have reported synergistic effects of AFB1 with other mycotoxins at cellular and hepatic levels in different animals: similar effects may have occurred in the present study also. Mean concentrations of glucose and cholesterol observed in this study were all higher than the concentrations reported by Hagawane et al. (2009). Concentrations of glucose and cholesterol were found to be 50.06 and 26.76 mg/dL, respectively, in healthy lactating buffaloes. This most likely is the result of these animals being in full lactation. Differences in the expression of urea with increasing mycotoxin levels could result from a disruption of protein synthesis either within the rumen in the synthesis of microbial protein or in the post-ruminal gastrointestinal tract. The mycotoxins may also be acting in muscle tissue and liver hampering protein synthesis and causing deamination and greater excretion of urea into the urine. This mechanism deserves further investigation as it may be important in inhibiting growth or lactational efficiency.

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Transfer of aflatoxins from naturally contaminated feed to milk of Nili-Ravi buffaloes fed a mycotoxin binder

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Abstract

The objectives of this study were to observe the extent of transfer of aflatoxin B₁ in feed to the aflatoxin M₁ metabolite in milk in Nili-Ravi buffaloes and to evaluate the efficacy of a commercial mycotoxin binder (Mycofix, Biomin Singapore) incorporated into feed to minimise this transfer. Multiparous animals ($n=28$) were randomly distributed to four groups corresponding to two treatments each with two levels of aflatoxin B₁. Individual animals were exposed to naturally contaminated feed providing a total of 1475 mg/day (Groups A and B) or 2950 mg/day (Groups C and D) of aflatoxin B₁. Groups B and D were given 50 g of mycotoxin binder daily mixed with feed whereas Groups A and C were kept as controls. Feed samples were analysed by reverse phase high performance liquid chromatography for aflatoxin B₁ and milk samples were evaluated by enzyme-linked immunosorbent assay for the liver metabolite aflatoxin M₁. The mean value of total daily aflatoxin M₁ excretion for animals fed 2950 mg/day of aflatoxin B₁ (112.6 mg/day) was almost double ($P<0.001$) than the excretion in buffaloes fed 1475 mg/day (62.2 mg/day). The mean daily concentration of aflatoxin M₁ in milk of animals from both treatment groups supplemented with 50 g/day of mycotoxin binder was 76.5 mg/day, nearly 22 mg lower than those without binder at 98.3 mg/day (s.e.d.=5.99; $P<0.01$). The interaction of binder and treatment was not significant, i.e. the 50 g/day of binder was able to sequester aflatoxin B₁ with the same efficiency in groups fed with high and low concentrations of aflatoxin B₁. Carry over was (3.44%) lower ($P=0.001$) in animals supplemented with 50 g/day of mycotoxin binder than those fed no binder (4.60%). Thus buffaloes are highly efficient at transferring aflatoxins in feed to the aflatoxin M₁ metabolite in milk, whereas a mycotoxin binder is capable of alleviating without preventing this contamination risk.

Additional keywords: AFB₁, AFM₁, mycotoxin binder, Nili-Ravi buffaloes, transfer.
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Introduction

Aflatoxins (AF) are one of the principal mycotoxins of concern for human food safety (MacLachlan 2011). They are among the most acutely toxic and carcinogenic compounds known (Lilly 1965). The negative impact of aflatoxin B₁ (AFB₁) and aflatoxin M₁ (AFM₁) has been recognised by the International Agency for Research on Cancer (IARC 2002). Due to its genotoxicity and cytotoxicity (Hagawane et al. 2009), AFM₁ has now been categorised as a carcinogen for animals (IARC 2002). The mutagenic effects of AFM₁ in milk and milk products are potentially greater for children and infants due to their rapid growth and developmental processes (Sadia et al. 2012). However, mycotoxins exert antimicrobial, anti-protozoal and antifungal action directly in the rumen (Fink-Gremmels 2008). AFs, which are degraded in the rumen, are reduced to form the less active metabolite aflatoxicol (Pawłowski et al. 1977). The remaining undegraded fraction is absorbed in the digestive tract and converted in the liver into AFM₁ (Upadhyaya et al. 2010).

The maximum permissible level of AFM₁ in milk and milk products is 0.05 mg/kg in the European Union whereas the USFDA has suggested a level of 0.5 mg/kg (Berg 2003). The extent and rate of transfer of AFM₁ into milk from feed in Nili-Ravi buffaloes has not been reported. In dairy cattle, transfer of AFB₁ from feed to AFM₁ in milk may be as high as 3% (Veldman et al. 1992). AFM₁ appears in milk within 12 h of ingestion of AFB₁ and reaches a maximum concentration after 3 days. Similarly, it disappears from the milk within 4 days of removal of the source in feed (Diaz et al. 2004). The carry over of AFM₁ depends on milk yield and stage of lactation.

Furthermore, species differences, animal variability and health of mammary alveolar cell membranes may also contribute towards the efficiency of AFM1 transfer to milk (Masoero et al. 2007). Therefore, in countries that rely on the buffalo for their milk supply, for example Pakistan (Wasti 2013), it is important to investigate the extent of carryover to milk in buffaloes in the interest of consumer safety.

Several in vitro studies reporting on the effectiveness of sequestering agents such as activated carbons and aluminosilicates have shown positive binding effects on AFB1 in feed (Huwig et al. 2001) whereas in vivo studies have demonstrated the effectiveness of sequestering agents against AFB1 toxicity in animals and against the secretion of AFM1 in milk (Galvano et al. 1996). Charcoals or activated carbons are widely used for detoxification of mycotoxins (Diaz et al. 2004) but are highly variable in their effectiveness. This variation may be because activated charcoal is a relatively non-specific adsorbent and many essential nutrients are also adsorbed at equivalent efficiencies to the mycotoxins (Huwig et al. 2001).

As such, their practical application in animal feeds is questionable. In a more recent study, bentonites were reported as effective adsorbents for AFB1 (Vekiru et al. 2007). Given the importance of buffalo milk, the present study was conducted with the objectives of investigating the efficiency of transfer of AFM1 to milk in Nili-Ravi buffaloes and to evaluate the ability of a commercial mycotoxin binder to minimise this transfer.

Materials and methods

This study was conducted at the Buffalo Research Institute, Pattoki (latitude: 31°50'N, longitude: 73°52'E) in district Kasur of Punjab province, Pakistan. Multiparous Nili-Ravi buffaloes (Table 1) were randomly allocated to four groups corresponding to two treatments each with two levels of AFB1. A cottonseed cake-based concentrate feed was obtained from a local commercial source and was being used at the time of the study as the concentrate supplement for the buffalo herd across the Institute. Animals in low (A and B) and high (C and D) AFB1 groups were individually offered this mycotoxin contaminated cottonseed cake-based concentrate feed (2.5 and 5 kg/day; 88.2% DM) and corn (200 and 400 g/day; 91% DM), respectively. This provided a total of 1475 and 2950 mg/day of AFB1 to animals in low and high groups, respectively. Moreover, animals were offered either 80 kg/day (Groups A and B) or 70 kg/day (Groups C and D) of AF-free fresh cut berseem clover (17.8% DM).

Therefore, total daily DM intake for low and high groups was 16.62 kg and 17.26 kg, respectively. The AFB1 concentrations in feed for the low and high groups were 88.7 and 171.2 mg/kg of DM. Both concentrate feed and corn were naturally found to have these levels of contamination and were being fed already to animals. Groups B and D were given 50 g of a mycotoxin binder daily mixed with feed whereas Groups A and C were kept as controls. All data and sample collection were performed on individual animals. Experimental period was 10 days excluding 1 week for an adjustment period.

Table 1. Average milk production, lactation length and number, body weight and body condition score of animals in different experimental treatments

Treatment	Low aflatoxin intake		High aflatoxin intake	
	Without binder A	With binder B	Without binder C	With binder D
Number of animals	7	7	7	7
Average milk production (kg/day)	9.5	8.6	8.6	9.4
Months in lactation	1-4	1-4	1-4.5	0.5-4
Average bodyweight (kg)	510	480	535	550
Lactation number	2-6	3-6	4-7	3-7
Body condition score	2.62	2.5	2.94	2.84

Mycotoxin binder

The mycotoxin binder used in this study was a 50-50 mixture of commercial available products Mycofix Secure and Mycofix Plus of BIOMIN (Getzersdorf, Lower Austria, Austria). Mycofix Secure is composed of 100% of bentonite/dioctahedral montmorillonite. Mycofix Plus is composed of bentonite/dioctahedral montmorillonite, Biomin BBSH797 (Gen. sp. nov. nov., formerly Eubacterium), Biomin MTV (Trichosporon mycotoxinivorans DSM14153), phytophytic (Ascophyllum nodosum) and phytogenic (silymarin) substances. All these products are authorised by the European Union Commission for their safety for use in animals and humans.

Sample collection

Milk production of each animal was recorded daily at 0500 hours and 1700 hours. Milk samples were collected on Days 0, 2, 4, 6 and 8 of the experimental period. Milk samples (100 mL) from all animals from morning and evening milkings were mixed in proportion with the milk production to constitute a representative sample for each animal every day. Samples were then stored at 20°C until analysed.

Mycotoxin analyses

Samples of green fodder and concentrate fed to the animals were analysed for AF(B1, B2, G1 and G2), deoxynivalenol, fumonisin (B1 and B2), ochratoxin A and zearalenone by reverse phase high performance liquid chromatography following sample clean up. All samples were sent to Romer Laboratories, Bukit Merah, Singapore for analysis as described by Aslam et al. (2014).

Aflatoxin M1 analyses

Aflatoxin M1 was measured by direct competitive enzyme-linked immunosorbent assay (ELISA) using the AgraQuant AFM1 Fast ELISA kits supplied by Romer Laboratories Singapore, according to the method provided with the kits. The recovery of AFM1 in the assay was 93–119% and cross-reactivities with AFB1, B2, G1 and G2 were 88%, 27%, 11.5% and 4.7%, respectively.

Sample preparation/extraction

A 5-mL milk sample was incubated for 30 min at 4°C. The sample was then centrifuged at 3000 g at 4°C for 10 min. After centrifugation, the milk serum below the fat layer was diluted 20 times with double distilled water. Following this, 0.4 mL of the diluted milk serum was mixed with 0.1 mL of 100% methanol (4:1) and used in the ELISA.

ELISA assay procedure

One AFM1-specific antibody coated well was used for each standard (0, 100, 200, 500, 1000, 2000 ng/L) or sample. To each dilution well, 200 µL of the AFM1-specific monoclonal antibody-enzyme conjugate was dispensed. Then 100 µL of each standard or sample was placed into the appropriate dilution well. Each well was then mixed carefully. These solutions (100 µL) were then dispensed into the corresponding antibody coated microwell. Samples were incubated at room temperature (18–30°C) for 20 min.

Microwell strips were then placed into an automatic ELISA washer (ELx50, BioTek, Winooski, VT, USA) and washed five times and then drained using absorbent towel to dry residual solution. Enzyme substrate (100 µL) was dispensed into each well and incubated for 10 min in the dark. Stop solution (100 µL) was dispensed into each well. At this time, the colour changed from blue to yellow. Optical densities were recorded in a microwell plate reader (Multiskan EX, Thermo Scientific, Schaumburg, IL, USA) at a wavelength of 450 nm. Carryover of aflatoxin M1 into milk. The carryover of AFM1 into milk was calculated by following the formula:

$$\text{Transfer } (\%) = \frac{100 \times \text{Total Aflatoxin M1 in milk}}{\text{Total Aflatoxin in B1 in feed}}$$

Statistical analyses

All data were analysed using linear mixed models with AFB1 level in feed (high and low), mycotoxin binder (with and without), Day (1, 2, 3 and 4) and all interactions as main effects and animal/day as random effects. This use of animal/day as random effect identifies the individual animals as the experimental unit with repeated-measurement on several days. GENSTAT (16th edition) was used for all analyses.

Results

Mycotoxin content of feeds The concentration of AFB1 in the cottonseed cake-based concentrate for this study was 554 mg/kg. The concentration of AFB2, deoxynivalenol, fumonisin B1, ochratoxin A and zearalenone were 50, 166, 230, 31.2 and 18 mg/kg, respectively. The corn supplement used in the study also contained AFB1 (454 mg/kg), AFB2 (52 mg/kg), fumonisin B1 (275 mg/kg), fumonisin B2 (328 mg/kg) and ochratoxin A (9.2 mg/kg).

Transfer of aflatoxin M1 into milk

There was a difference ($P=0.001$) in carryover of AFM1 into milk of animals fed with and without mycotoxin binder. Carryover was lower (3.44%) in animals supplemented with 50 g/day of mycotoxin binder than those fed no binder (4.6%). However, there was no difference ($P=0.219$) in transfer in animals exposed to high and low levels of AFB1. Carryover in animals exposed to high (2950 mg/day) level of AFB1 was 3.82% whereas it was 4.22% in animals fed low (1475 mg/day) level of AFB1. No interaction was observed between high or low levels of AFB1 and presence or absence of mycotoxin binder ($P=0.109$). The values of transfer for individual groups are presented in Table 2.

Total aflatoxin M1 excretion in milk

The dynamic of AFM1 transfer into milk are shown in Table 2. There was a difference ($P<0.001$) in total daily AFM1 excretion between animals fed 1475 mg (Group A and B) and those fed 2950 mg (Group C and D) AFB1 per day. The mean for those fed 2950 mg was 112.6 mg/day, almost double the mean of those fed 1475 mg, which was 62.2 mg/day (s.e.d.=5.99).

Addition of the mycotoxin binder to the diet resulted in a decrease ($P<0.001$) in the excretion of AFM1 into milk. The mean daily excretion rate for the combined groups with the binder was 76.5 mg/day, nearly 22 mg/day lower than concentrations without the binder (98.3 mg/day; s.e.d.=5.99). The interaction between AFB1 intake and the presence of the mycotoxin binder on transfer to milk was non-significant, showing that the binder was able to bind mycotoxin with almost the same efficiency at both levels of AFB1 intake.

The effect of binder over a period of 5 days was different ($P<0.01$), showing a progressive decrease in concentration of AFM1 expressed in milk over the 5 days of the study in animals also offered the mycotoxin binder (Fig. 1). No such decrease was observed in the animals fed the mycotoxin without the binder (Fig. 1).

Aflatoxin M1 concentration in milk

A difference ($P<0.001$) was observed in concentration of AFM1 (mg/kg) in milk between the animals supplemented with low (1475 mg/day, i.e. Group A and B) and high (2950 mg/day, i.e. Group C and D) concentrations of AFB1. The concentration of AFM1 was 12.4 mg/kg for the animals from Groups C and D, almost twice that found in Groups A and B (6.9 mg/kg; s.e.d.=0.44). There was also a difference ($P<0.001$) in the concentration of AFM1 in milk between the animals supplemented with (8.0 mg/kg; Group B and D) and without (11.3 mg/kg; Group A and C) the mycotoxin binder (s.e.d.=0.44). The mycotoxin binder was equally effective ($P=0.051$) at suppressing transfer to milk irrespective of level of mycotoxin in the diet (Table 2). However, the binder was more effective ($P=0.017$) with time over the 5-day course of the trial. A similar decrease with time was not observed in the control groups ($P=0.461$).

Table 2. The effect of aflatoxin B1 intake and use of a mycotoxin binder on concentration of aflatoxin M1 in milk, total aflatoxin M1 excreted and calculated transfer of aflatoxin M1 into the milk of Nili-Ravi buffalo esn.a., not applicable; s.e.d., standard error of difference

Variables	Low aflatoxin intake		High aflatoxin intake		s.e.d.	Effect of aflatoxin	Significance level	
	Without binder A	With binder B	Without binder C	With binder D			Effect of binder	Effect of aflatoxin × binder interaction
Total aflatoxin B1 intake (µg/day)	1475	1475	2950	2950	n.a.	n.a.	n.a.	n.a.
Concentration of aflatoxin M1 (µg/kg)	8.1	5.6	14.6	10.3	0.44	$P<0.001$	$P<0.001$	0.051
Total aflatoxin M1 excreted (µg/day)	74.6	49.8	122.0	103.3	5.99	$P<0.001$	$P<0.001$	0.613
^A Transfer of aflatoxin M1 into milk (%)	5.06	3.37	4.14	3.50	0.45	0.219	0.001	0.109

^ATransfer of aflatoxin M1 into milk calculated as total aflatoxin B1 excreted/total aflatoxin B1 intake × 100.

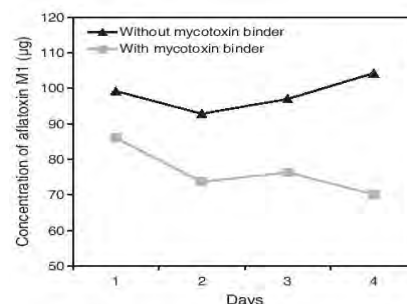


Fig.1. Changes in the daily concentration of aflatoxin M1 (µg) in milk of animals receiving 2950 mg (Groups A and B) or 1475 mg (Groups C and D) of aflatoxin B1. The triangles represent the means of Groups A and C without mycotoxin binder, whereas the squares represent the means for Groups B and D with 50 g of mycotoxin binder added to their daily diet (s.e.d. = 5.814).

Milk production

Milk production (kg/day) was not affected by level of inclusion of mycotoxin in the diet irrespective of the level of addition of the mycotoxin binder (Fig.2).

Discussion

Carryover of aflatoxin M1 into milk. Given the prevalence of inadequate feed storage facilities on small-holder farming operations in subtropical environments, the likelihood of fungal contamination of feed is high. Concentrate feed containing AFB1 at a concentration of more than 500 mg/kg DM is commonly used. Contamination levels may be higher in field conditions especially in peri-urban areas where kitchen and bakery wastes contaminated with fungi are routinely offered to animals in developing countries (Sultana and Hanif 2009). Concentrate feed used in the present study was not only contaminated with AFB1, but also contained high levels of AFB2, fumonisins, deoxynevalonol, ochratoxins and zearalenone. The influence of these co-contaminants on the metabolism and transfer of AFB1 into milk is not known and should be the objective of further study.

The milk AFB1 concentrations measured in this study of 8.1 or 14.6 mg/kg exceeded international standards of 0.5 mg/kg by up to 27-fold. The carry-over efficiency from feed to milk of 4.14% and 5.06% observed with high (2950 mg/day) and low (1475 mg/day) concentrations of AFB1 were similar to the 3.85% observed in Italian Friesian dairy cows (Pietri et al. 2009). In the Italian study, however, animals were offered only 97.3 mg/day of

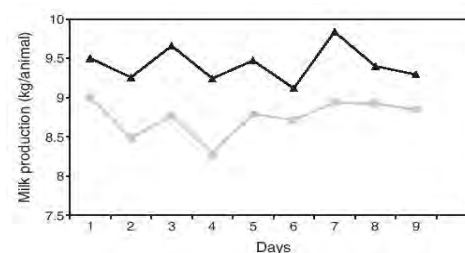


Fig.2. Averaged daily milk production for animals with mycotoxin binder (Groups B and D) represented by triangles and without mycotoxin binder (Groups A and C) represented by squares (s.e.d.=0.528).

AFB1, which theoretically should improve the efficiency of transfer. These transfer efficiencies are much higher than the 0.54% carryover reported for Holstein cows by Galvano et al. (1996). However, cows enrolled in that study were in late lactation, when the rate of transfer is reduced markedly (Veldman et al. 1992). With the study of Veldman a transfer of 6.1% and 1.8% for AFM1 was observed for early and late lactation dairy cows, respectively. The Nili-Ravi buffaloes used in the present study were in early lactation.

Bantaokul and Ruangwises (2010) reported a transfer of 2.35% in Holstein Friesian cows with milk AFM1 concentrations within the range of 0.035–11 mg/kg, which are comparable to the milk concentrations observed in

the present study. Moreover, Masoero et al. (2007) reported a transfer of 1.29% and 2.70% in low (21.2 kg/day) and high (41.8 kg/day) producing Holstein cows, which is again lower than the percent transfer of the present study. Clearly in order to compare the efficiency of transfer in buffaloes and cows, a controlled study using the same feed base needs to be conducted. Studies published to date show that stage of lactation, health, breed and nutritional status of animals all contribute to these differences. In addition, AFs are also excreted from the body through urine (Aslam et al. 2014) and faeces (Allcroft et al. 1968). Transfer of 15% of the total AFM1 ingested has been observed in the urine of Nili-Ravi buffalo heifers in a related study (Aslam et al. 2014). However, the excretion in faeces was likely to be less than 3% as demonstrated previously in a lactating cow following a single dose of 300 mg of mixed AF, 44% of which was AFB1 (Allcroft et al. 1968).

Effect of a mycotoxin binder on carryover of aflatoxin M1 in milk The effectiveness of mycotoxin binders in minimising AF transfer from feed to milk varies widely depending on dose, breed and species of animal studied. The addition of the same quantity of Mycofix Plus as used in the present study in the feed of Holstein Friesian cattle consuming only 5.6 mg of AFB1 per day decreased milk concentrations of AFM1 by 31% or 41% (Pietri et al. 2009). In the present study using the same concentration of a related but more advanced mycotoxin binder product at 300-fold higher doses of the contaminant in feed, 34% of the transfer was prevented. When the dose was increased further to 600-fold higher, the efficiency of binding was decreased to 16%. However, the fact that the efficiency of sequestration of the mycotoxin by the binder was not compromised when exposure levels were 300-fold higher in the present study, suggests that this product subtype is equally effective across a wider range of contamination levels.

The study by Pietri et al. (2009) with lower levels of contamination and the present study with higher levels of contamination showed no effect on milk yield, suggesting that the mammary epithelium is not damaged by the circulating concentrations of AFM1. The efficacy of various other carbon matrix-based mycotoxin binders has also been assessed:

Galvano et al. (1996), for example investigated the efficacy of the inclusion of three activated carbon products to reduce transfer of low levels (56.4–67.2 mg/day) of AFB1 into milk of Holstein Friesian animals. The reduction in transfer was 27%, 36% and 50% for the three products, respectively, which are comparable to the efficiency reported in the present study.

In another study, the efficacy of montmorillonite-based mycotoxin binders was assessed at inclusion levels of 0.2% and 1% of DM in the diet of animals offered 75 mg/kg of feed (Queiroz et al. 2012). The higher concentration of binders reduced milk AFM1 concentration by 17% whereas the lower concentration was ineffective. In another study Diaz et al. (1999) explored the efficiency of activated charcoal (0.25% of DM), MTB-100, an esterified glucomannan (0.05% of DM), calcium bentonite (1.25% of DM) and sodium bentonite (1.25% of DM) in reducing transfer of AFM1 in milk again in Holstein Friesian cows offered low levels of contaminant.

Sodium bentonite, calcium bentonite and mycosorb reduced AFM1 transfer to milk by 64.6%, 31.4% and 58.5%, respectively. Furthermore using an invitro model, Diaz et al. (2003) found that activated charcoals, sodium bentonites, calcium bentonite and esterified glucomannan were able to bind 5 mg of AFB1 in a solution with efficiencies of 99%, 98%, 98% and 97%, respectively. This study in essence simply assessed the maximum potential binding capacity of the matrix without assessing its bio-effectiveness in the animal. Perhaps the most comprehensive study has been provided by Diaz et al. (2004) who evaluated six different sequestering agents: SA-20, an activated carbon (AC-A); Astar Ben-20, a sodium bentonite (AB-20); MTB-100, an esterified glucomannan (MTB-100); Red Crown, a calcium bentonite (RC); Flow Guard, a sodium bentonite (FG) and Mycosorb, a sodium bentonite (MS), for their potential to reduce AFM1 transfer to milk. RC, MTB-100, MS, FG and AB were able to reduce the carryover of AFM1 by 31%, 59%, 50%, 65% and 61%, respectively. Again, these results are comparable to the efficacy of the mycotoxin binder used in the present study. Although mycotoxin binders are able to reduce the risk of incorporation of AFM1 into the human food chain, they do not

seem to be sufficiently effective to reduce contamination of milk to the minimum level of 0.05 mg/kg of milk set by the European Union.

Conclusion

The present study shows that the transfer kinetics of AFB1 from feed to AFM1 in milk is very similar in the Nili-Ravi buffalo to that observed in dairy cows. Furthermore, it was demonstrated that a commercial mycotoxin binder product is an effective sequestration agent for feed naturally contaminated with AF when fed to buffaloes.

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Perinatal Nutrition of the Calf and Its Consequences for Lifelong Productivity*

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ABSTRACT:

Provision of an optimal environment for the calf is critical to establishing the patterns of growth and development essential to allow the heifer to express its genetic potential for milk output and reproductive capacity during its productive life. Maternal nutrition during gestation is now recognised as a key genetic programming in utero and this influence is extended through the complexity of hormones, growth factors and immunostimulants incorporated into colostrum and milk consumed by the neonatal calf.

This natural process is most often disrupted as calves are weaned abruptly to maximise milk output for commercial exploitation. The key then is to accelerate the rate of maturation of the ruminal epithelium through the provision of concentrate starter rations and high quality forage, which promote VFA production. Management systems to promote these processes in Holstein Friesian cattle are well developed, however, little is known of these processes with buffalo and Bos indicus dairy cattle such as the Sahiwal. The development of methods to program the neonate to grow fast to puberty in these species will be important to improving their productivity for the dairy industries in tropical and sub-tropical environments in the future. (Key Words : Perinatal Nutrition, Calf, Buffalo, Productivity)

INTRODUCTION

The potential for growth and high health status in the newborn calf is largely influenced by the health and metabolic status of the dam. Much of the focus of cow management has been on the perinatal period as the calf prepares for delivery into a totally foreign environment in which placentally derived nutrition is replaced by the initial lacteal secretion from the mammary gland, colostrum. The composition of this secretion is extremely important in establishing the growth potential and life-long productivity of the calf. This then reverts to normal milk which acts as a source of dietary energy and protein through to the point when the calf is able to be weaned. This initial phase is most often termed the pre-ruminant period during which milk is passed directly into the abomasum through the reflex closure of the oesophageal groove. This initial period of development is termed the pre-ruminant phase and varies in duration from 14-21 days depending on the animal's ability to initiate the intake of dry feed. During the subsequent 3-6 weeks ruminal function develops and the calf derives more of its nutrient substrate from this source than from milk. At this point the animal is weaned and derives its nutrient substrate solely from dry feed through the activity of the newly established ruminal microbial population: this describes the ruminant phase of calf development.

This process is universal for bovine species although there are many variations in procedures used in different parts of the world.

PROGRAMMING DEVELOPMENT FROM CONCEPTION

Feeding cows to support the protein, energy, vitamin and mineral requirements of the growing conceptus is now recognised as just one factor that influences the life-long productivity of the calf. We are now aware of the importance of environmental factors on gene expression patterns and therefore development much earlier during embryogenesis. These so-called epigenetic mechanisms challenge the very basis of Darwinian evolutionary theory that the variability in populations occurs exclusively through random mutations. The mechanism through which this occurs is by altering gene methylation patterns (Khosla et al., 2001) which might not only regulate the growth potential of the calf but also alter the germline which may then persist as mutations across subsequent generations (Surani, 2001). Factors that initiate these changes which may persist differentially across different regions of DNA are influenced by changes in dietary components providing methyl groups for this mechanism. Synthetic xenobiotics (Danzo, 1998), the estrogenic molecules in plants the phytoestrogens and isoflavones, all of which can be found in the cow's diet contribute to these changes (Guerrero-Bosagna et al., 2005).

Thus at calving the growth potential of the animal may have already been compromised by factors other than the mere supply of nutrients substrate to support the growth process.

THE IMPORTANCE OF COLOSTRUM

It is generally considered that the newborn calf should receive 4L of colostrum in the first 12h, although up to 6L is often recommended for the first day (University of Sydney, 2007). Requirements will depend on the quality of colostrum usually determined by assessing its density associated with immunoglobulin (Ig) content: the presence of enzymes to form curd in the abomasum is also a rate limiting factor for Ig absorption (Gregory, 2003; Mastelloni et al., 2005).

The observation that over 100 hormones and growth factors have been identified in colostrum or milk (Koldovsky, 1995; Koldovsky, 1996) suggests that these secretions are complex biological fluids designed to extend the influence of the dam over developmental processes beyond the uterine environment. These include hormones of the hypothalamic-pituitary, the thyroid-parathyroid group, gastrointestinal regulatory hormones as well as growth factors. While the physiological significance of growth hormone regulatory peptides is most apparent the importance of the gonatropin regulatory peptide GnRH at this early stage is more obscure. However the developmental processes that these hormones regulate are most likely important when calves are born into a challenging environment in which ambient temperature varies significantly from that experienced in utero and when pathogen loads are high. The provision of key nutrients to support growth is also an integral function of milk with proteins, essential and non essential amino acids, lactose, fatty acids, vitamins and minerals all contributing to these requirements. Other non-nutritional factors include nucleotides, polyamines, enzymes as well as functionally important proteins such as lactoferrin (Blum and Hammon, 1999; 2000) serves specific roles in directing growth processes. Lactoferrin has also been extensively characterized in buffalo milk (Sharma et al., 1999).

Cellular components are also incorporated including mammary epithelial cells, erythrocytes macrophages, polymorphs, lymphocytes, plasma cells and epithelial cells (Uruakpa et al., 2002) while a number of the non-nutritional and bioactive molecules are sequestered from the circulation. These include growth hormone, prolactin, oestrogen, insulin and glucagon, all of which play a role in regulating protein and energy metabolism. It is often difficult to envisage a functional role for these protein hormones since they will be hydrolysed extensively in the abomasum prior to accessing functional receptors. However the key may reside in their co-secretion with the immunoglobulins, most notably IgG1.

These macromolecules comprise more than 90% of the protein content of colostrum and appear at concentrations 5-10-fold higher than in the circulation (Larson, 1992). Their sequestration coincides initially with higher oestrogen levels up to 1 month pre-partum and subsequently with the elevated corticosteroids, growth hormone and prolactin in the last week and then with depressed progesterone at 48h pre-calving.

The functional significance of these relationships is yet to be established. Equally intriguing is the mechanism that facilitates preferential uptake of IgG1, which is typically 10-fold higher than IgG2 in colostrum, but present in equivalent concentrations in the circulation. This imbalance provides a definitive characteristic of colostrum and is explained by the presence of specific receptors on the basal membrane of secretory epithelial cells which actively endocytose IgG and pass it to the secretory lumen of the alveolus (Butler, 1983; Barrington et al., 2001). It is important to note that this mechanism is only in place during colostrum synthesis and therefore IgG1 concentration falls markedly after the colostrum phase (Kemler et al., 1975). The immune status of the buffalo calf post-colostrum feeding is influenced also by the vitamin status of the dam: circulating Ig levels were increased by 80% in calves fed colostrum from their dams receiving bolus injections of vitamins A., D3 and E late in pregnancy (Sikka and Lal, 2005). VitE and selenium also limit the adverse effects of endotoxin from E coli infections associated with many calf rearing systems (Sharma et al., 2005).

Further protection against pathogenic bacteria and viruses is provided by the presence of antimicrobial proteins, lactoferrin and lysozyme. Lactoferrin is an iron-binding moiety that prevents microbial growth through depriving microbial of this essential mineral and by binding to bacterial cell membranes thereby compromising their permeability (van Hooijdonk et al., 2000), while lysozyme lyses bacterial cell walls (Lonnerdal, 2002). Interestingly these two proteins are capable of acting synergistically to enhance their bacteriostatic activity (Pakkanen and Aalto, 1997).

The immunoglobulins are accompanied by a range of protease inhibitors including trypsin inhibitor, f_{i2} -macroglobulin, f_{i2} -antiplasmin, antithrombin III, C1-inhibitor, inter- f_{i2} -trypsin inhibitor, bovine plasma elastase inhibitor and bovine plasma trypsin inhibitor, all of which serve to protect their functional integrity (Christensen et al., 1995).

Other multi-functional proteins are coming to light including a proline-rich polypeptide colostrin. This was originally found as a fraction accompanying sheep colostrum immunoglobulins which promoted T cell-tropic and maturational activity. It is also associated with the development of precognitive functions which inhibit pathological states centrally (Zimecki, 2008).

In addition to the role of the simple carbohydrate lactose in providing energy, more complex carbohydrates also add to the multi-functionality of colostrum. Sialyloligosaccharides are present in high concentrations for the first 12 h of lactation in the cow and are thought to be important in preventing infections acting against rotavirus, reovirus and *Helicobacter pylori* (Nakamura et al., 2003). Interestingly they also appear to be involved in the development of cognitive processes in the brain, with the supplementation of milk for piglets with sialic acid improving learning and memory in the piglet (Wan et al., 2007). There is little reason to suspect that suckling behaviour in both cows and buffalo is not influenced by these molecules, although bovine and human milk diverges in the concentration and composition of the sialyloligosaccharide content: this has implications for the development of infant formulae (Martin-Sosa et al., 2003). Interestingly buffalo milk gangliosides appear to have greater toxin binding and anti-inflammatory properties than cows milk suggesting some potential novel applications for the product from buffalo in the future (Colarow et al., 2003). This is also important for the survival of the buffalo calf in environments with high pathogen loads. Certainly buffalo milk is highly valued as an alternative to breast milk among Indian mothers (Kaushal et al., 2005).

The composition of colostrum changes rapidly and its provision to the calf during the first 24 h of life is critical to the calf's survival.

COLOSTRUM AND THE DEVELOPMENT OF THE GASTROINTESTINAL TRACT

The rich mix of hormones, growth factors, cytokines and nutrients in colostrum provide the ideal developmental mix to initiate digestive activity in the abomasum, small and large intestines. The initiation of the functional integrity of the intestinal epithelium is essential for the absorption of nutrients and bioactive molecules to direct developmental processes in the body. In particular the apical junctional complex plays an important role in maintaining the integrity of this epithelium and prevents access for pathogens to the circulation. A wider range of cytokines and growth factors influence tight junction integrity, with IFN- γ , TNF- α , HGF, TGF- β , IGF-I, IGF-

-II, VEGF, IL-1, IL-4 and IL-13 all decreasing barrier function while EGF, TGF- β , GDNF, neurturin, IL-10, and IL-17 have the opposite effect (Sawada et al.,

Table 1. The composition of colostrum and milk (Blum and Hammon, 2000; Klimes et al., 1986)

Nutrient	Colostrum (1 st milking day 1)	Mature milk (5-14 days <i>postpartum</i>)
Total protein (%)	17.12	3.57
Fat (%)	4.69	5.26
pH	6.31	6.43
Gross energy (MJ/L)	6.0	2.8
Crude protein (g/L)	133	32
Immunoglobulin G (g/L)	81	<2
Lactoferrin (g/L)	1.84	0.36 (at 4 th milking)
Transferrin (g/L)	0.55	0.21 (at 4 th milking)
γ -glutamyltransferase (μ kat/L)	509	52
Alkaline phosphatase (μ kat/L)	19	4
Aspartate aminotransferase (μ kat/L)	1.5	0.1
Tumour necrosis factor- α (μ g/L)	5	<2
Insulin (μ g/L)	65	1
Glucagon (μ g/L)	0.16	0.01
Prolactin (μ g/L)	280	15
Growth hormone (μ g/L)	1.4	<1
Insulin-like growth factor-I (μ g/L)	310	<2
Insulin-like growth factor-II (μ g/L)	150	ND

ND = Not determined.

2003). Clearly this is a closely regulated and functionally important property which can be influenced by the balance of these factors present in colostrum. A gain there is evidence to suggest that the intestinal epithelium in the buffalo calf is more resistant to some infections including paratuberculosis (Sivakumara et al., 2006).

NUTRIENT REQUIREMENTS FOR THE NEWBORN CALF

The calf requires nutrients for both maintenance and growth and it is important that the requirements for these two processes are combined. Environmental factors are extremely important in determining requirements with both extremes of heat and cold, high pathogen loads and physical and psychosocial stressors contributing to requirements.

The activation of the immune system and the role that maternal immunity plays in this process is important to animals calving in sub-optimal environments (Chase et al., 2008). As a guideline the metabolisable energy requirements for a 45 kg calf under thermoneutral conditions is 7.3 MJ/day. Since cow's milk contains 22.5 MJ ME/kg of solids they require 2.5 L of whole milk, while the equivalent with lower fat status milk replacer is around 3 L (Drackley, 2008). Others recommend higher intakes up to 10 and 12% of body weight per day to support growth (University of Sydney 2007). Milk requirements will be lower in the buffalo as milk fat content is higher than in cow's milk, although colostrum fat content is the same in each species (Ganovski, 1979).

Protein requirements for maintenance are low in the neonate (30 g/d for a 45 kg calf) and reflect rates of protein turnover in tissue. However the requirements for growth are approximately 6-fold higher than this, equating to 250-280 g of crude protein from milk replacer (Drackley, 2008). In general milk replacers containing up to 25% crude protein are recommended as long as dietary energy is not limiting.

Amino acid composition of replacers is also important with those most closely resembling the composition of cow's milk being most effective. Changes in energy requirements in cold and hot environments are required dramatic as body temperature is not buffered by the heat of fermentation in the undeveloped rumen. Maintenance ME increases by approximately 20% for each 10°C incremental decrease from 20 to -20°C (National Research Council, 2001). In contrast the effects of heat stress have not been published although older animals require an additional 20-30% ME (National Research Council, 2001); the ready availability of water on demand and shade for calves is well established yet not always adhered to in many countries.

The water content of body tissue is in the vicinity of 70% (Diaz et al., 2001), thus the constant availability of water is mandatory in any production system. Promoting the intake of dry concentrate feeds to enhance rumen

development is also dependent on constant water availability. Requirements for minerals and vitamins have also been documented (Council, 2001). Whole milk provides an adequate source of all minerals with the exception of iron and sometimes selenium and manganese. As most milk starters are supplemented with minerals and fat soluble vitamins these rarely compromise calf health and growth.

The only major concerns are with Vitamins A and E. NRC recommendations for Vitamin A are considered to be too high leading to potential toxicity, while those for Vitamin E are under-estimated (Drackley, 2008). While a state of deficiency will rarely be found, finding the correct dose for optimal growth represents a greater challenge.

DEVELOPING THE RUMEN

The general rule of thumb is to adopt strategies that allow calves to consume 700-900 g of concentrate ration by the time that they are weaned. Essentially this requires calves to commence intake of concentrates within 14 days post-partum (University of Sydney, 2007). Animals consuming 10% of their own body weight as milk per day will generally be consuming up to 300 g of concentrate by day 25. The microbial population accumulating in the rumen will ferment carbohydrate to form predominantly butyric acid and then propionic acid which in turn promote the differentiation of the ruminal epithelium to form the characteristic papillae (Heinrichs and Lesmeister, 2005).

Different sources of fermentable carbohydrate yield different responses, with corn and wheat based diets promoting ruminal development faster than oats or barley (Khan et al., 2008). Similarly processing can exert and influence the growth response: steam flaking of corn for example induced ruminal epithelial development faster than either dry rolling or leaving grain whole (Lesmeister and Heinrichs, 2005). These studies showed also that processing can influence the pattern of volatile fatty acids released.

Other alternative concentrate sources such as phalaris minor seeds have also been assessed as providing appropriate carbohydrate sources to support volatile fatty acid synthesis and rumen development (Kaure et al., 2006). Much has been written on the role of fibre and the so-called "tickle factor" in the diet of the pre-ruminant calf.

Results vary widely with concentrate particle size being more effective in some studies (Klein et al., 1987), while in others forages provided as specific particle size (8-19 mm) with concentrates gave superior results (Coverdale et al., 2004). In further studies pelleted diets yielded superior responses to mixed length fibre with other dietary ingredients held constant (Bach et al., 2007). Clearly our understanding of the development of the ruminal environment requires further investigation, although the role of the volatile fatty acids in this process is well established. The inclusion of cellulolytic enzymes as feed additives also provides a beneficial growth effect if included as a substitute for more conventional additives in buffalo calf diets (El-Kady et al., 2006).

CALF REARING: THE PAKISTANI EXPERIENCE

Pakistan, like many developing countries has an agrarian rural based economy. The livestock sector is a major contributor to the national (12%) and agricultural (50%) economy (Pakistan Economic Survey, 2006). This sector is growing quickly and provides a livelihood for more than 35 million people. The productivity of livestock for meat and milk is low, with improper calf management programming animals for a life of low productivity. High market prices for milk dictate that calves are weaned very early without appropriate quality milk replacers being used to meet the demand of the calf for growth and development.

Male calves are most often sold for slaughter or left to fatten on poor quality roughages. The slow growth of the female calves results in delayed puberty and age at first calving. Thus both the efficiency of milk and beef production are compromised.

FEEDING THE TRANSITION BUFFALO

Late pregnant or transition Holstein cows can be affected by a range of production diseases associated with their inability to cope with the metabolic demands of high production. These include hypocalcaemia,

hypomagnesaemia, ketosis, retained placenta, displacement of the abomasum and laminitis (Mulligan and Doherty, 2008). These are often associated with an imbalance in metabolites entering key biochemical pathways (Payne, 1972) which may lead to infertility.

Similar problems are associated with buffalo production, although the causes are most likely related to undernutrition. The pregnant buffalo needs to support the nutrient demands of both lactation and growth of the foetus. Yet the condition score of most small-holder buffaloes remains very low despite the need for additional nutrients to meet these demands. Very few studies have been conducted to investigate these relationships, but a positive relationship has been shown between live weight of the calving dam and the calf. At lower body weights (350-573 kg) calf birth weight increased by 1.8 g for each kg increase in weight of dam. This increment decreased to 5.5 g per kg live weight of dams weighing 576-815 kg (Usmani et al., 1987; Usmani and Inskip, 1989).

As with the cow the first 24 h post-calving are critical for the buffalo calf to absorb colostrum. In fact total protein and Ig levels are higher in the buffalo than in cross-bred cows (Singh and Ahuja, 1993). In this study 75% of Ig and 68% of colostrum protein were absorbed within 1 h of feeding a 7 h old calf. This rate of absorption declined rapidly after the first feed. However little other data are available on colostrum usage to account for high susceptibility to infection of buffalo calves.

CALF MORTALITY

Neonatal calf morbidity and mortality are major causes of economic losses in livestock production. It is roughly estimated that a calf mortality of 20 percent can reduce the net profit of an enterprise by 60% (Blood and Radostits, 1989). Ideally calf mortality should be less than 5 percent with growth rates of 0.5-0.7 kg/d (Blood and Radostits, 1989). Mortality rates for different countries employing different production systems are detailed in Table 2. Very high mortality rates of over 50% have been reported in buffalo calves in one month of age. Foot and mouth disease (FMD) and haemorrhagic septicaemia (HS) are endemic to Pakistan and account for up to 31 and 21.5% respectively of deaths in buffalo calves aged from 6-12 months (Ramakrishna, 2007). On other farms extreme mortality rates of up to 80% have been recorded (Tiwar et al., 2007). Although disease contributed to this statistic, the failure to provide colostrum, to deworm, to disinfect navels and to provide an adequate milk substitute and appropriate shelter and water all played their role in the etiology of these mortalities. Other causes of calf mortality include the greater susceptibility of crossbred and primiparous animals (Rao and Nagarkinkar, 1980). The failure to provide colostrum has also often been implicated (Afaq et al., 1992).

The giving of colostrum to friends is a custom found in some regions and as such the calf is inevitably deprived. Overall farmers in many regions consider calf rearing a very low priority, as the commercial value of this practice is not apparent to them.

SUCKLING AND HAND FEEDING OF BUFFALO CALVES

The average birth weights for buffalo and Sahiwal calves are 34.85±0.46 and 21.87±0.20 kg respectively (Ahmad, 1988). The method of provision of milk for these calves has been shown to influence their growth efficiency. For example calves reared by restricted suckling of their

Table 2. Incidence of mortality (%) among buffalo and cow calves in different countries

Age (days)	Mortality (%)	Country	Reference
Buffalo calves			
30	7.1	Pakistan	(Khan, 1994)
1-30	51.8	India	(Ramakrishna, 2007)
30	27.5	India	(Bhullar and Tiwana, 1985)
90	34.0	India	(Bhullar and Tiwana, 1985)
15	12.6	Vietnam	(Sharma et al., 1984)
16-30	29.9	Vietnam	(Sharma et al., 1984)
30	51.7	India	(Khera, 1981)
Cow calves			
30	11.0	Pakistan	(Khan, 1994)
15	32.5	India	(Veerapandian et al., 1993)
0	61.6	U.S.A.	(Bellows et al., 1987)
1-10	23.4	U.S.A.	(Bellows et al., 1987)
2	8.2	Ireland	(Mee, 1988)
14	56.4	Germany	(Fink, 1980)
4-7	1.2	Italy	(Mariani et al., 1986)
28	3.0	Norway	(Simensen, 1986)
90	8.7	Nigeria	(Umoh, 1982)
28	12.5-26.0	Libya	(Gusbia and Hird)
30	6.9	Iraq	(Maarof et al., 1987)

efficiency. For example calves reared by restricted suckling of their dams yielded better growth rates than if the milk was provided in a feeder or pail (552 vs. 370g/d) (Khan and Preston, 1992) and 500 vs. 350g/d in the study of Gaya et al., 1977). If adequate milk is provided these high growth rates are attainable: the provision of 15% of the milk production from Sahiwal cows or buffalo which equates to 10% of the calf's body weight has resulted in these growth rates (Ahmad, 1988). The training of buffalo calves to the use of automatic suckling units may compromise their growth performance by limiting intake (Rossi et al., 2004).

Given these problems it is most likely more effective to delay the weaning of buffalo calves reared in sub-optimal environments particularly as the buffalo dams display strong maternal instincts. Thus the provision of some milk to calves combined with the harvest of milk from a second milking each day for commercial sale or home consumption may provide the most effective means of rearing the calves of Sahiwal cattle and buffalo. This method also negates the need to use oxytocin to induce milk let-down: use of the calf is biologically more sustainable.

STARTER RATIONS FOR CALVES

Poor growth rates result from the limited milk supply, low protein and energy of fodders and concentrates that are available for calves. In order for the farmer to sell a high proportion of his milk, cost-effective milk replacers need to be developed. Starter rations containing 17% crude protein and total digestible nutrients of 75% have resulted in growth rates of up to 470g/d (Ahmad and Jabbar, 2000).

Similar results have been achieved through the substitution of milk with soybean milk containing 1% soybean oil (Matter et al., 2005). The levels of protein used in these studies are below recommendations for Holstein Friesian calves: thus productivity could be boosted further through the use of higher protein supplements. The importance of offering calves a soluble concentrate rich in protein and energy from calves should be considered.

FEEDING POST WEANING

Feeding strategies used post-weaning involve the use of low quality crop residues, straws and stovers characterized by high fibre and low crude protein. Numerous studies have been undertaken to improve their efficiency of utilization largely through their treatment or use in conjunction with strategic supplements (Sarwar et al., 2002).

Urea treatment of straw is popular as it increases the N content of roughages. The addition of molasses then provides a balance of N and energy for the rumen microbial population to utilize in digesting the insoluble carbohydrates. Wheat straw treated with varying levels of urea (0%, 2% and 4%) and molasses (2% and 4%) ensiled with 30% cattle manure (on dry matter basis) for different fermentation periods (20, 30 and 40 days) proved to be an ideal supplement providing linear growth responses with amount fed in buffalo calves (Sarwar et al., 2006). Similar results were achieved by Khan et al. (1992) who reported that crude protein increased by 18.4% to 22.2% in sugarcane bagasse ensiled with cattle manure for 30 and 60 days, respectively. However, the increasing cost of urea worldwide may make this option prohibitive.

Caution must be used in the evaluation of supplements. In one study sunflower meal was substituted for cottonseed meal at 0, 12, 24 and 36% on a nitrogenous basis to 11 month-old buffalo calves (Yunus et al., 2004). The sunflower meal yielded inferior responses both biologically and on a cost basis, suggesting that its use should be approached with caution. Maize has proved to be an effective concentrate for buffalo and Sahiwal calves in a number of studies. In comparing starter rations based on maize, oats and their combination normalized to 20% crude protein and 80% TDN, growth rates on the maize based diet were 18% higher (Rafique and Manzoor, 2000).

Supplementing with protein that is resistant to ruminal digestion can also yield excellent responses, such as has been achieved through the use of formaldehyde treated mustard cake (Chatterjee and Walli, 2003). Again caution should be used as if the basal diet consists of poor quality roughage more cost-effective supplements providing any source of N and energy to the rumen may yield similar responses.

CONCLUSION

The key to successful calf rearing commences with the appropriate feeding of the late pregnant or transition cow, as any metabolic disturbance will have consequences for the growth potential of the calf. Increasingly we are becoming aware of the key metabolic cues responsible for programming the growth and ultimately the production potential of the calf. Disturbingly most of the available literature pertains to the growth and development of the Holstein Friesian calf; in spite of the importance of the buffalo and the Sahiwal cow to the provision of dairy products for Asia, our fundamental understanding of these processes in these animals is poorly developed. Future research should focus on the nutritional regulation of these developmental processes in late pregnancy through to weaning; if this is not optimised then research on feeding of the weaner calf is compromised.

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A Review of Recent Developments in Buffalo Reproduction A Review

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ABSTRACT:

The buffalo is an important livestock resource in several countries of South Asia and the Mediterranean regions. However, reproductive efficiency is compromised due to known problems of biological and management origins, such as lack of animal selection and poor nutrition. Under optimal conditions puberty is attained at 15 to 18 months in river buffalo, 21 to 24 months in swamp buffalo and is influenced by genotype, nutrition, management and climate. However, under field conditions these values deteriorate up to a significant extent. To improve reproductive efficiency, several protocols of oestrus and ovulation synchronization have been adopted from their use in commercial cattle production. These protocols yield encouraging pregnancy rates of (30% to 50%), which are comparable to those achieved in buffaloes bred at natural oestrus. The use of sexed semen in buffalo heifers also showed promising pregnancy rates (50%) when compared with conventional non-sexed semen. Assisted reproductive technologies have been transferred and adapted to buffalo but the efficiency of these technologies are low. However, these latest technologies offer the opportunity to accelerate the genetic gain in the buffalo industry after improving the technology and reducing its cost. Most buffaloes are kept under the smallholder farming system in developing countries. Hence, future research should focus on simple, adoptable and impact-oriented approaches which identify the factors determining low fertility and oestrus behaviour in this species. Furthermore, role of kisspeptin needs to be explored in buffalo. (Key Words: Buffaloes, Reproduction, Developments, Techniques)

INTRODUCTION

Buffalo has a significant role in the agricultural economy of many developing countries by providing milk, meat and draught power. The world population of buffalo is estimated to be 199 million (FAO STAT, 2012) with more than 96% of the population located in Asia including 16.4% of Pakistan's contribution. In recent decades, buffalo farming has expanded widely in the Mediterranean and Latin America as well as, in Central/Northern Europe where several herds were introduced.

Dairy buffaloes have been used for milk production in India, Pakistan, some other South Asian countries, the Middle East and Italy; while dairy characteristics are being induced in the local population of Indo-Chinese Region and South America through crossbreeding with Pakistani Nili Ravi and Indian Murrah buffaloes. The milkyield increased from 700 to 2,000 kg/year in China through crossbreeding (Yan et al., 2007).

The buffalo can utilize poorer quality roughages, adapt to harsher environments and are more resistant to several bovine tropical diseases. Despite these merits, buffalo have relatively poor reproductive efficiency irrespective of their location throughout the world. Buffalo exhibit many of the known reproductive disorders including delayed onset of puberty, poor oestrus expression, longer postpartum ovarian quiescence, and most importantly lowered conception rates particularly when bred artificially (Gordon, 1996). However, higher fertility could be achieved through better feeding and management (Perera et al., 1987; Usmani et al., 1990; Qureshi et al., 2007). It appears that because buffaloes are located mostly in developing countries with meager resources, there is limited quality research in the area of basic physiology, health, management, nutrition and applied reproduction.

The objective of this review is to examine the major recent developments in buffalo reproduction. We discuss the impact of the various techniques as well as bottlenecks and possible future developments which will lead to improved reproductive performance in this species.

PUBERTY

Buffaloes usually attain puberty when they reach about 60% of their adult body weight (250 to 400) kg, but the age at which they attain puberty can be highly variable, ranging from 18 to 46 months (Jainudeen and Hafez, 1993). The factors that influence this are genotype, nutrition, management and climate. It could be attained under optimized conditions at 15 to 18 months in river buffalo and 21 to 24 months in swamp buffalo (Borghese, 2005). The delay in puberty, consequently delays conception and results in low reproductive efficiency and lengthening of the non-productive life. A major cause of delayed puberty may be poor feeding and management under field conditions.

OESTROUS CYCLE

In order to enhance reproductive efficiency of buffalo, a thorough understanding of the regulatory mechanisms involved in the oestrous cycle is required. The duration of the oestrous cycle in buffalo is similar to that in cattle, ranging from 17 to 26 days with a mean of around 21 days (Jainudeen and Hafez, 1993). However, there is a greater variability of the oestrous cycle length in buffalo compared to cattle, with a greater incidence of both abnormally short and long oestrous cycles. This may be attributed to various factors including adverse environmental conditions, nutrition and irregularities in secretion of ovarian steroid hormones (Kaur and Arora, 1982; Nanda et al., 2003).

In buffaloes, ovarian follicular dynamics during the oestrous cycle is similar to that in cattle. Studies from India (Taneja et al., 1996), Brazil (Baruselli et al., 1997) and Pakistan (Warriachand and Ahmad, 2007; Figure 1) have shown clearly that the majority of buffalo have two waves of follicular activity during their oestrous cycle. More investigations on the effect of follicle stimulating hormone and nutrition on number of follicular waves need to be studied in buffaloes.

Studies on oestrous behavior and endocrinology in buffalo (Roy and Prakash, 2009; Singh et al., 2000) indicate considerable variations in reproductive endocrine activity without external signs of oestrus (silent heat) are common. The low intensity of oestrus in buffaloes may be due to low circulating concentrations of 17- β oestradiol in comparison with dairy cattle (Seren et al., 1995).

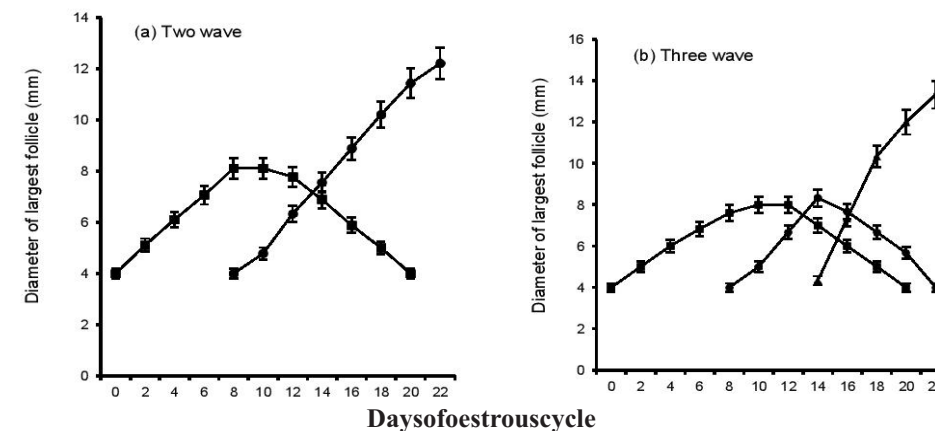


Figure 1. Observed patterns of development for the largest follicle during oestrous cycles. Shown are the average diameters of the first (■—■), second (●—●) and third (▲—▲) sequentially largest follicles in buffaloes that had (a) two (n=9) or (b) three (n=3) waves of follicular development respectively (Warriachand and Ahmad, 2007).

Furthermore, tying up the animals as per normal husbandry practices in many developing countries restricts the ability of buffalo farmers to observe heat signs (Warriachand et al., 2009). Buffaloes also tend to show heat signs

during the night when farmers are not observing their animals (Unpublished data). Season is another extrinsic factor that influences the characteristics of oestrous behaviour. In the tropics, high ambient temperature reduces sexual activity during the day (Jainudeen, 1977) and shortens the oestrous period (Gill et al., 1973) with the incidence of silent oestrous more common during the hot summer season. These adverse effects of heat stress make oestrous detection much more difficult in buffalo. Oestrous detection could be significantly improved through the introduction of a teaser bull or an androgenized female (Chohan et al., 1992).

The interval between standing oestrous and ovulation, which is very important for artificial insemination, was 30 hours in buffaloes (Warriach et al., 2008). Under field conditions, the am-pm rule of insemination originally developed for cattle (Trimberger, 1948) is generally followed in buffaloes. To follow this rule, the buffaloes should be bred 12 h after the detection of standing oestrus. However, onset of heat signs instead of onset of standing oestrus has been erroneously considered as the land mark with buffaloes often being inseminated, earlier than required. This early breeding is potentially responsible for lowered fertility, and can be explained by the fact, there is an interval of about 8 to 10 h between onset of heat signs and onset of standing oestrus. This indicates buffaloes should be inseminated 12 h after the detection of standing oestrus (detection by bull/teaser) or alternatively 18 to 24 h after the onset of heat signs. In order to confirm this approach, investigations are required on the timing of insemination in relation to standing oestrus and pregnancy rate.

SYNCHRONIZATION OF OESTROUS CYCLE

Various studies using protocols for synchronization based on progesterone and gonadotropin releasing hormone (GnRH) administration together with prostaglandin to induce luteolysis during breeding season have yielded quite promising conception rates ranging from 30% to 50% (Table 1). However, some buffaloes do not respond to treatment, especially during the low breeding season. There could be several reasons for this, but among the most likely is the animal's follicular status at the beginning of treatment. The ideal time of treatment can be established by determining ovarian activity by ultrasound (De Rensis and López-Gatiús, 2007). The presence of the dominant follicle and an active corpus luteum (CL) indicate the success of synchronization. Protocols for buffaloes with limited follicular and luteal activity, remain to be refined, but they most likely will be devised around the strategic timing of administration of reagents currently used in synchronization protocols while ensuring that the supply of dietary energy and protein are not lacking.

Reference	Treatment	Pregnancy rates (%)
Warriach et al., 2008	GnRH+PGF2 α +GnRH	36
Naseer et al., 2011	CIDR	37
Paul and Prakash, 2005	GnRH+PGF2 α +GnRH	33
Berber et al., 2002	GnRH+PGF2 α +GnRH	56.5
Neglia et al., 2003	GnRH+PGF2 α +GnRH	36
Chohan, 1998	PGF2 α (cloprostenol)	53
Rao and Rao, 1983	PRID	41

SEXED FROZEN SEMEN

Semen sexing has been successfully used for producing living offspring in bovine species (Seidel et al., 1999). In buffalo, a difference in DNA content between X and Y sperm was found, and based on this difference it has been further demonstrated that processing buffalo semen was feasible (Lu et al., 2007). In a recent study, promising pregnancy rates (50%) were achieved when inseminating a dose of sexed semen containing 4 million spermatozoa (Gaviraghi et al., 2013). In order to expand use of this technology, there is a need to further refine this protocol for buffalo breeds of commercial significance such as the Nili-Ravi buffalo of Pakistan.

ASSISTED REPRODUCTIVE TECHNOLOGIES

The first successful embryo transfer in buffalo was performed in the United States of America (Drost et al., 1983). Subsequent successful transfers have been reported from many other countries. However, the success rate is much lower in buffaloes, due to their inherently low fertility and poor superovulatory response (Misra et al.,

1990). The average yield of transferable embryos is less than one per superovulated donor. The buffalo ovary has a smaller population of recruitable follicles at any given time; an average of 12,000 primary follicles has been reported (Danell, 1987), compared to the average in the ovary of the cow, which has an average of 133,000 (Erickson, 1966). This technique comprises a series of carefully integrated sequential steps including donor selection, donor treatment, recipient selection, insemination of the donor, embryo recovery, embryo handling and evaluation, embryo transfer, and recipient care. The technology has to be refined to account for the lower, less responsive follicle population in the buffalo.

Assisted reproductive technologies have been introduced to overcome the inherent reproductive problems, fast propagation of superior germplasm, and to reduce the generation intervals. These technologies provide an excellent source of embryos for carrying on basic research in developmental physiology, farm animal breeding, and for commercial application of the emerging biotechniques like cloning and transgenesis. During the past two decades, considerable advances have been made in our understanding of buffalo reproductive physiology, however, previous reviews (Palta and Chauhan, 1998; Gasparrini,

2002) and more recent studies suggest that the rate of transferable embryo yield remains at a plateau (Manjunatha et al., 2009). Results have been quite variable between laboratories and are most likely related to differences in embryology, metabolism, and culture requirements among buffalo breeds. Further studies are also needed to improve the cryopreservation of in vitro embryo production embryos.

FUTURE RESEARCH

The hypothalamo-pituitary-gonadal axis is the regulatory system for reproduction in mammals. A newly discovered neural peptide, kisspeptin, has opened a newer area in reproductive neuroendocrinology. As shown in a variety of mammals, kisspeptin is a potent endogenous secretagogue of GnRH, and the kisspeptin neuronal system governs both the pulsatile GnRH secretion that drives folliculogenesis, spermatogenesis and steroidogenesis, and the GnRH surge that triggers ovulation in females (Okamura et al., 2013). Role of kisspeptin needs to be explored in buffalo.

CONCLUSIONS

Buffaloes are an important livestock resource for many countries. Most buffaloes are kept under the smallholder farming system in developing countries. Future research should focus on simple, adoptable and impact oriented approaches which identify the factors limiting fertility and oestrus behaviour in this commercially significant species.

Despite the inherited problems in buffalo, low progress has been made in the application of assisted reproductive techniques. Artificial insemination is practiced commercially; embryo transfer, in vitro embryo production, and nuclear transfer remain in the realm of experimentation. If the costs are reduced, these latest techniques offer the opportunity to accelerate the genetic gain in the buffalo industry with the proviso that they are used in conjunction with efficient national progeny testing and sire evaluation programs.

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Genetics Selection Evolution



Strategic test-day recording regimes to estimate lactation yield in tropical dairy animals

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ABSTRACT:

Background:

In developing dairy sectors, genetic improvement programs have limited resources and recording of herds is minimal. This study evaluated different methods to estimate lactation yield and sampling schedules with fewer test-day records per lactation to determine recording regimes that (1) estimate lactation yield with a minimal impact on the accuracy of selection and (2) optimise the available resources.

Methods:

Using Sahiwal cattle as a tropical dairy breed example, weekly milk records from 464 cows were used in a simulation study to generate different shaped lactation curves. The daily milk yields from these simulated lactation curves were subset to equally spaced (weekly, monthly and quarterly) and unequally spaced (with four, five or six records per lactation) test-day intervals. Lactation yield estimates were calculated from these subsets using two methods: the test-interval method and Wood's (Nature 216:164-165, 1967) lactation curve model. Using the resulting lactation yields, breeding values were predicted and comparisons were made between the sampling regimes and estimation methods.

Results:

The results show that, based on the mean square error of prediction, use of Wood's lactation curve model to estimate total yield was more accurate than use of the test-interval method. However, the differences in the ranking of animals were small, i.e. a 5% difference in accuracy. Comparisons between the different test-day sampling regimes showed that, with the same number of records per lactation (for example, quarterly and four test-days), strategically timed test-days can result in more accurate estimates of lactation yield than test-days at equal intervals.

Conclusions:

An important outcome of these results is that combining Wood's model for lactation yield estimation and as few as four, five or six strategically placed test-day records can produce estimates of lactation yield that are comparable with estimates based on monthly test-day records using the test-interval method. Furthermore, calculations show that although using fewer test-days results in a decrease in the accuracy of selection, it does provide an opportunity to progeny-test more sires. Thus, using strategically timed test-days and Wood's model to estimate lactation yield, can lead to a more efficient use of the allocated resources.

Background

Breed improvement and selection in dairy systems of developing countries is a challenge because field conditions are restricted by limited resources and infrequent milk recording. In these situations, frequent test-day (TD) recording throughout the entire lactation for genetic evaluation purposes is difficult and impractical [1]. This lack of data availability highlights the need to optimise the contribution of each collected record to the genetic evaluation process [2]. Therefore, in such situations, there is a need to develop an efficient TD sampling regime and subsequent genetic evaluation system that optimises selection outcomes given the current resources for TD recording. In dairy systems, genetic test-day models (TDM) provide a solution to the lack of data since they can effectively use fewer records [3-5]. However, TDM require accurate estimates of many genetic parameters calculated from large datasets [6, 7], which are difficult to obtain in developing country scenarios and hence can cause inaccurate results [3, 8]. Therefore, an approach in which lactation yield is first estimated and

subsequently used for breeding value estimation may be more appropriate. There are numerous methods to estimate lactation yield based on TD records. The test-interval method (TIM) is the reference method to calculate lactation yield [9] and is particularly useful in developing dairy sectors. Other methods involve the use of mathematical lactation models to predict milk yield. TD data from Sahiwal cattle, a tropical dairy breed from Pakistan [10], have been used in a number of studies that compare different lactation models. These studies indicated that suitable models include the inverse polynomial function proposed by Nelder [11], the incomplete gamma-type function proposed by Wood [12] and the WilMink model [13-16]. Various studies have investigated which of each of these models is the most appropriate in different tropical or sub-tropical conditions [11, 17-20]. Although some results of these studies are conflicting, it is clear that the most suitable model is the Wood model because of its ability to fit different shaped curves and its relative ease to describe a characteristic of the lactation curve [20-22]. Furthermore, we recently reported that the Wood model is more robust than others when fitting lactation curves in frequent and irregular test-days sampling regimes (TDSR), which are common in developing country scenarios [23]. For these reasons, the Wood model was used for data modelling and simulation of datasets for this study, although it is expected that other models of similar complexity would give similar results.

Previous studies have clearly shown that as milk recording frequency decreases, the accuracy of the lactation yield estimates also decreases [24-27]. Despite this, it is possible to record milk yield monthly or even just four or five times throughout a lactation and still estimate lactation yields sufficiently accurately to rank cows for selection based on milk production [25-27]. Some studies considered using unequally spaced sampling regimes with more TD around the peak of lactation or timed according to the visits of an AI (artificial insemination) technician. These studies found that, although the lactation yield estimates were less accurate when TD were unequally spaced [28], they did provide an opportunity to assess more bulls with the same resources while maintaining the reliability of the resulting sires' estimated breeding values (EBV) [27]. What has not been directly considered is whether a few, four, five, or six TD strategically timed throughout lactation would have an effect on the accuracy of EBV for lactation yield.

Therefore, the aim of this study was to investigate whether using fewer, but more strategic TD sampling approaches, and the Wood lactation model would improve the accuracy of lactation yield estimates and EBV for milk production. Milk records from Sahiwal cattle were used to simulate lactation curves as an example of the production in tropical dairy breeds. The simulated data were then used to compare different TDSR with only four, five or six sampling days within one lactation. Then, comparisons of EBV between sampling and estimation methods were assessed to determine the most efficient approach to estimate breeding values given the resources available.

Methods

Raw data

Sahiwal cattle were used as an example of a typical tropical dairy breed. Sahiwal lactation records collected between 2005 and 2010 from the Livestock Production Research Institute (LPRI), Bahadurnagar Okara, Pakistan, were used. The raw data consisted of 839 lactations with weekly TD records from 464 dams from 82 sires, with an average of 5.65 daughters per sire. The dams ranged in age from three to over ten years, with approximately 25% of lactations from cows in their first lactation, 45% from the second to fourth lactation, and the remaining 30% from the fifth lactation and above. Using these TD records, Wood's [12] lactation curve model was fitted to each lactation. This model is defined as follows in its original nonlinear form (1) and its linear form (2):

$$W(t) = at^b e^{-ct} \quad (1)$$

$$W(t) = \exp(k + b \ln t - ct), \quad (2)$$

where $W(t)$ is the model-based lactation yield at time t (days in milk (DIM)), $k = \ln a$, and where k , b , and c specify the shape of the lactation curve.

Analysis of raw data

To obtain estimates of the various components of the Wood model for the simulation study, the raw Sahiwal TD

records were fitted using a nonlinear mixed model. This was done by the `nlme()` function of R Version 3.0.2 [29], using the methods documented in Pinheiro et al. [30]. In this case the model fitted was:

$$y_{it} = \exp(k_i + b_i \ln t - c_i t) + \varepsilon_{it}, \quad (3)$$

where f is the index of the particular cow, with $i=1, 2, \dots, c$, and n is the number of cows in the dataset. The linear form (3) of the Wood model was used here because of its closer approximation of parameters (k_i, b_i, c_i) to a multivariate normal distribution [22], as required for the model assumption to be met. The Wood model can be developed further as a nonlinear mixed model with additive (linear) sub-models for k_i, b_i and c_i such that:

$$\begin{aligned} k_i &= k + k_{MOC} + k_{Year} + k_{Age} + K_{i,G} + K_{i,E} \\ b_i &= b + b_{MOC} + b_{Year} + b_{Age} + B_{i,G} + B_{i,E}, \\ c_i &= c + c_{MOC} + c_{Year} + c_{Age} + C_{i,G} + C_{i,E} \end{aligned} \quad (4)$$

where k, b and c are overall fixed parameter intercepts, $k_{MOC}/Year/Age, b_{MOC}/Year/Age$ and $c_{MOC}/Year/Age$ are the fixed effects of each parameter for month of calving (MOC), year of milking (Year) and age (Age) of the cow at calving, $K_{i,G}, B_{i,G}$ and $C_{i,G}$ are cow-specific polygenic random effects, and $K_{i,E}, B_{i,E}$ and $C_{i,E}$ are cow-specific "environmental" random effects. These random components will have multivariate normal distributions such that:

$$\begin{pmatrix} K_{i,G} \\ B_{i,G} \\ C_{i,G} \end{pmatrix} \sim N \left(\begin{pmatrix} 0 \\ 0 \\ 0 \end{pmatrix}, \begin{pmatrix} \sigma_{K,G}^2 & \sigma_{KB,G} & \sigma_{KC,G} \\ \sigma_{KB,G} & \sigma_{B,G}^2 & \sigma_{BC,G} \\ \sigma_{KC,G} & \sigma_{BC,G} & \sigma_{C,G}^2 \end{pmatrix} \right),$$

and

$$\begin{pmatrix} K_{i,E} \\ B_{i,E} \\ C_{i,E} \end{pmatrix} \sim N \left(\begin{pmatrix} 0 \\ 0 \\ 0 \end{pmatrix}, \begin{pmatrix} \sigma_{K,E}^2 & \sigma_{KB,E} & \sigma_{KC,E} \\ \sigma_{KB,E} & \sigma_{B,E}^2 & \sigma_{BC,E} \\ \sigma_{KC,E} & \sigma_{BC,E} & \sigma_{C,E}^2 \end{pmatrix} \right), \quad (6)$$

where i indexes the cow, with $i=1, 2, \dots, c$, and n is the number of cows in the dataset. First, considering the polygenic terms ($K_{i,G}, B_{i,G}$ and $C_{i,G}$), it will be assumed that $f^*G = (K1.G, K2.G, \dots, c, Kn.G) \cdot \mathbf{E}$, $f^*G = (B1.G, B2.G, \dots, c, Bn.G) \cdot \mathbf{E}$ and $f^*G = (C1.G, C2.G, \dots, c, Cn.G) \cdot \mathbf{E}$ have the following distributions: $f^*G \sim N(0, \mathbf{Z} \mathbf{K} \mathbf{Z}^T + \mathbf{I}_n)$, where \mathbf{Z} links the g phenotypes h (k_i, b_i, c_i) to the animal pedigree records, and \mathbf{A} is the numerator relationship matrix. In the current situation, it was assumed that only one lactation is simulated per cow, so $\mathbf{Z} = \mathbf{I}_n$. It is also assumed that the "environmental" components $f^*E = (K1.E, K2.E, \dots, c, Kn.E) \cdot \mathbf{E}$, $f^*E = (B1.E, B2.E, \dots, c, Bn.E) \cdot \mathbf{E}$ and $f^*E = (C1.E, C2.E, \dots, c, Cn.E) \cdot \mathbf{E}$ are all independent and their covariances are assumed to be 0, i.e. $f^*E \sim N(0, \mathbf{I}_n)$.

Putting these together, equation (5) yields:

$$\begin{pmatrix} \mathbf{K}_G \\ \mathbf{B}_G \\ \mathbf{C}_G \end{pmatrix} \sim N \left(\begin{pmatrix} \mathbf{0}_n \\ \mathbf{0}_n \\ \mathbf{0}_n \end{pmatrix}, \begin{pmatrix} \sigma_{K,G}^2 & \sigma_{KB,G} & \sigma_{KC,G} \\ \sigma_{KB,G} & \sigma_{B,G}^2 & \sigma_{BC,G} \\ \sigma_{KC,G} & \sigma_{BC,G} & \sigma_{C,G}^2 \end{pmatrix} \otimes \mathbf{A} \right) \quad (7)$$

(assuming $\mathbf{Z} = \mathbf{I}_n$), and equation (6) yields

$$\begin{pmatrix} \mathbf{K}_E \\ \mathbf{B}_E \\ \mathbf{C}_E \end{pmatrix} \sim N \left(\begin{pmatrix} \mathbf{0}_n \\ \mathbf{0}_n \\ \mathbf{0}_n \end{pmatrix}, \begin{pmatrix} \sigma_{K,E}^2 & \sigma_{KB,E} & \sigma_{KC,E} \\ \sigma_{KB,E} & \sigma_{B,E}^2 & \sigma_{BC,E} \\ \sigma_{KC,E} & \sigma_{BC,E} & \sigma_{C,E}^2 \end{pmatrix} \otimes \mathbf{I}_n \right) \quad (8)$$

Simulated data

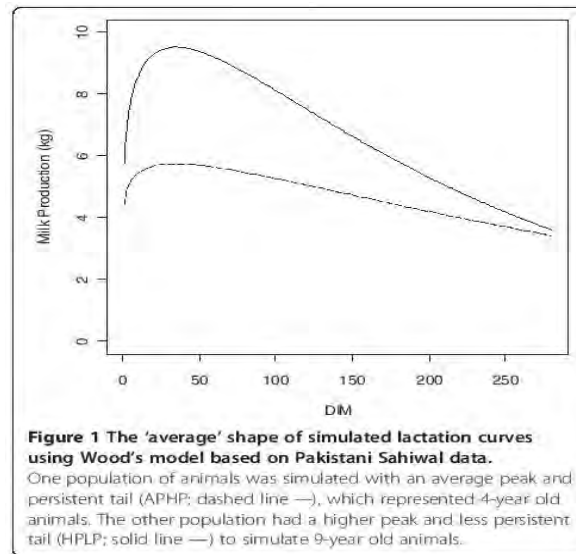
From the initial `nlme()` model output that fitted the Wood model to the raw Sahiwal lactation records, estimates of the fixed effects of k, b , and c and the variance and covariance matrix of the combined random effects ($K_{i,G} + K_{i,E}, B_{i,G} + B_{i,E}$ and $C_{i,G} + C_{i,E}$) were obtained. This variance-covariance matrix was split up to resemble these separate random cow-effects ($K_{i,G}, B_{i,G}$ and $C_{i,G}$) and the random "environmental" effects ($K_{i,E}, B_{i,E}$ and $C_{i,E}$), such that simulated lactation curves yielded realistic curves. For the purposes of the simulation, non-zero covariances were used in equation (7) but all covariances in equation (8) were set equal to 0 (see Additional file 1). These two variance-covariance matrices and the relationship matrix (\mathbf{A} , based on the LPRI

pedigree) were used with the $\text{rmvnorm}()$ [31] function of R Version 3.0.2 [29] to generate random effects drawn from multivariate normal distributions (equations (7) and (8)) for a simulated population. These were then added to the estimates of fixed effects parameters (from the Sahiwal lactation data) to yield realistic simulated values for k_i , b_i and c_i according to equation (4).

The outcomes of this simulation were values for k_i , b_i and c_i for a population of Sahiwal cattle (where $n=464$) that calved in January 2006 at 4 years of age. The resulting lactation curves had a general shape that had an average peak of production and high persistency (APHP; a slowly declining curve). A second set of data was simulated in which only the fixed effect of age was changed to 9 years to generate lactation curves that on average had a higher peak and a less persistent tail (HPLP). The fixed effects of 4 and 9 years of age were selected for the simulations since they yielded lactation curves that were required to be different in shape, yet still typical of Sahiwal cows. Plots of the average simulated lactation curves that highlight differences between these fixed effects are in Figure 1.

To ensure that the simulated datasets were realistic to the Pakistani situation, a random error term ϵ_{it} was added to each simulated day of milking (according to equation (3)) to take into account the important environmental variation that occurs in milk production due to daily variation in nutrition, management and other factors. Note that the random errors were taken as independent, although in reality they may be serially correlated with an autoregressive error structure.

To calculate the true simulated lactation yield (Y) for an animal i within the herd, the yield on each DIM ($t=1,$



2,3,...,280) was summed according to the following equation;

$$Y_{i,TRUE} = \sum_{t=1}^{280} \exp(k_i + b_i \ln t - c_i t) + \epsilon_{it}. \quad (9)$$

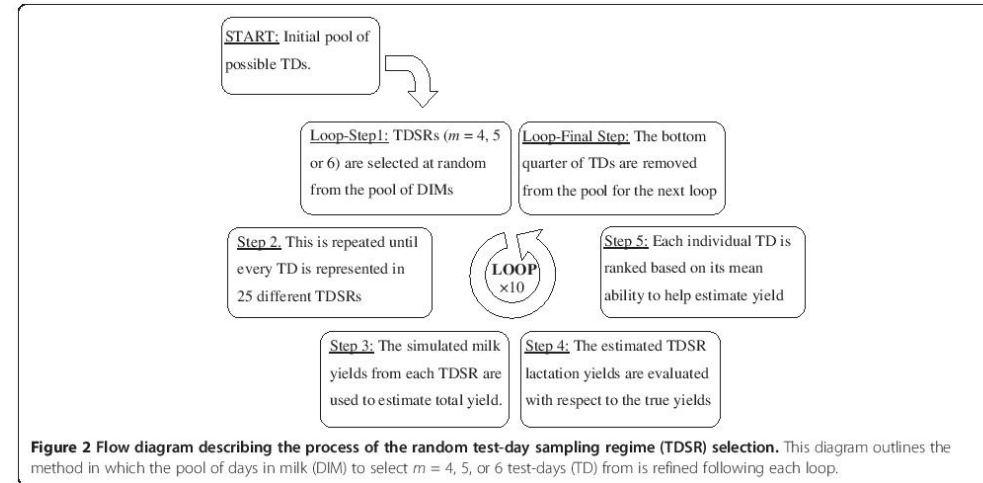
This value $Y_{i,TRUE}$ was calculated for each animal and is considered the 'true' lactation yield from the simulations. This was done separately for the simulated datasets for which animals were 4 (APHP) and 9 (HPLP) years old. An assumed lactation length of 280 days was used here since the average lactation length in Sahiwal cattle in Pakistan is 235 days [10] and only a small proportion of animals produce milk for more than 305 days [32].

Test-daysampling regimes tested Standard lactation yield estimation methods use the TIM based on TD records collected at equally spaced weekly, monthly and quarterly intervals. By sub-setting the simulated data to represent these recording regimes, lactation yields were recalculated ($Y_{i,TIM,WKL}$, $Y_{i,TIM,MON}$, $Y_{i,TIM,QTR}$) and used for comparison with other TDSR and estimations using Wood's model. The main aim of this study was to investigate the possibility of using fewer (m) TD per lactation (where m equals to 4, 5 or 6), strategically timed

throughout lactation to estimate lactation yield. In order to determine the ‘ideal’ TDSR, numerous TD combinations were tested and compared.

The number of possible TDSR within a single lactation is very large. For example, from a lactation of 280 DIM, if each combination of five ($m=5$) randomly selected TD was tested, there would be over 1.6×10^{12} possible combinations. Since it is unrealistic to compare each of these possible TDSR, a method was developed to refine the selection process of the TDSR to be tested based on previous results. A diagram of this process is in Figure 2, and details are given below.

The process of TDSR comparison was carried out over a number of loops by selecting combinations of m ($=4, 5$ or 6) TD from the pool of DIM. In the first loop, the pool of TD included all DIM ($1, \dots, 280$). In subsequent loops, the pool of DIM was reduced by one quarter based on the



results from the previous loop (see Figure 2). The steps to compare TD within each loop were as follows:

- Step 1: Select m TD at random from the pool without replacement. This will make up one TDSR for comparison (TDSR _{j}). This was repeated until every TD from the pool was represented at least once in a selected TDSR _{j} .
- Step 2: Repeat ‘Step 1’ until each TD in the pool is represented in at least 25 different TDSR _{j} , to ensure that there are enough repeated measurements to justify valid comparisons when TD are evaluated. Fewer repeats could have been used (for example 5), but for the purposes of this study, 25 were used to allow for greater certainty in the assessment of the contribution of each DIM to the lactation yield estimate.
- Step 3: For each randomly selected TDSR _{j} , the corresponding TD yields in the simulated population were used to estimate lactation yields for each cow. This was done by fitting Wood’s lactation curve model using the nonlinear mixed effects model by the nlme() function of R Version 3.0.2 [29]. The fitting process and lactation yield estimation followed the processes outlined by Raadsma et al. [22]. This resulted in a lactation yield estimate for each cow, using Wood’s model based on TD records from the TDSR _{j} ($Y_i, \text{WOOD}, \text{TDSR}_j$).
- Step 4: For every TDSR _{j} , the mean square error of prediction (MSEP) was calculated using the following formula:

$$\text{MSEP}_{\text{WOOD}, \text{TDSR}_j} = \frac{1}{n} \sum_{i=1}^n (Y_{i, \text{TRUE}} - Y_{i, \text{WOOD}, \text{TDSR}_j})^2, \quad (10)$$

where n is the number of simulated lactation yields.

Step 5: The 25 different TDSR _{j} that contained each TD were averaged to yield a single average MSEP for each TD. Using this value, each TD from the pool was ranked from the highest MSEP (most inaccurate) to the lowest.

Step 6: One-quarter of the TD with the most inaccurate average MSE P were removed from the pool. This reduced the pool of TD for the next loop to theoretically include TD that on average allow for more accurate estimation of lactation yield. This loop process can theoretically continue until the number of TD in the pool is less than the number of TD (m) per TDSR. It was expected that the MSE P would initially decrease as the size of the pool decreased but then either reach a plateau or increase again, i.e. decrease in accuracy. For the purposes of this study, the looping process was repeated ten times, when the pool had only 20 DIM remaining.

Comparisons

The methods used to estimate lactation were compared in three ways: (1) MSE P between true and estimated lactation yield, (2) comparison of EB V based on true lactation yield and estimated lactation yield and (3) number of sires that could be theoretically tested for a given number of test-day recordings. More detailed descriptions of these comparisons are given below.

First, the simulated lactation yields were compared directly with the various estimates of lactation yield using the MSE P. Simulated lactation yields (Y_i , TRUE) were compared with the estimates obtained with methods based on fewer TD records. This included the estimation methods already described (Y_i ; TIM; WKL; Y_i ; TIM; MON; Y_i ; TIM; QTR; Y_i ; WOOD; TDSR_j) and additionally, the TIM at the different TDSR_j (Y_i ; TIM; TDSR_j) and the Wood method at the equally spaced sampling regimes (Y_i , WOOD, WKL, Y_i , WOOD, MON, Y_i , WOOD, QTR). Using the estimates of lactation yield, MSE P were recalculated (similarly to that described in equation (10)) and used to compare the accuracy of the different lactation yield estimation methods.

Second, the predicted EB V from each method were compared with the 'true' EB V based on the simulated data. EB V for each cow were calculated based on the lactation yield estimates using the TIM (EB V_i, TIM, WKL, EB V_i, TIM, MON, EB V_i, TIM, QTR, EB V_i; TIM; TDSR_j) and the Wood method (EB V_i, WOOD, WKL, EB V_i, WOOD, MON, EB V_i, WOOD, QTR, EB V_i; WOOD; TDSR_j). These EB V were recalculated in ASReml-R Discovery Edition 1.0 [33], using the A matrix based on the LPR pedigree data. The EB V based on the simulated data using the true lactation yields (Y_i , TRUE) were also calculated (EB V_i, TRUE). EB V of the different lactation yield estimation methods were compared by using the number of cows in the top 100 EB V that corresponded with the 'true' top 100 EB V (Top 100). This enabled an assessment of the similarities between methods to estimate lactation yield in terms of the cows that would be theoretically selected.

Lastly, the number of sires that could theoretically be progeny-tested was used as a comparison to determine which method could use allocated resources most efficiently. According to the method outlined by Ducloset al. [27], the theoretical reliability (R) of a sire can be calculated as:

$$R = \frac{dh^2}{4 + (d-1)h^2 + 4(\sigma_{\Delta}^2/\sigma_p^2)}, \quad (11)$$

where h^2 is the heritability of Y_i , TRUE, d is the number of daughters and σ_{Δ}^2 is the phenotypic variance of Y_i , TRUE.

The $f\Delta^2$ represents the increase in the residual variance due to the TDSR recording protocol and can be calculated as the variance of the differences between the estimated yield under the TDSR recording protocol, Y_i , ESTIMATE, and yield under a full recording protocol, Y_i , TRUE, $f\Delta^2$

$$\sigma_{\Delta}^2 = \text{Var}(Y_{i,\text{ESTIMATE}} - Y_{i,\text{TRUE}}). \quad (12)$$

Then, using these equations and simple algebra, an expression can be developed to determine the theoretical number of daughters necessary to prove a sire with a specific reliability level. Then, the number of sires that could possibly be proven given a limited number of resources can be calculated and compared according to the method used to estimate lactation yield.

In Pakistan, the current Sahiwal progeny testing system (not including government farms) records data monthly from approximately 30 private farms, which we assumed, had 2.5 milking animals each. This means that 7500 (30 x 25 x 10) TD records can be collected within a given lactation. Thus, the number of possible proven sires is equal to:

where m is the number of TD recorded per lactation and d is the number of daughters necessary (calculated using equation (11)) to prove a sire with a given reliability (R) and $f \geq 2f \phi$ based on the method of lactation yield estimation.

Results

TDSR selection and comparison were done separately for the two lactation curves shapes (APHP and HPLP) and form recorded TD (where m is equal to 4, 5 or 6) within a lactation. This allows for comparison of the key outcomes and practical applications of these results based on both shape of the lactation curve and the number of TD recorded. Test-days sampling regimes tested Selection of the TDSR to be compared within this study was done using a process that removed DIM with each loop in order to reduce the number of possible combinations of TDSR while maintaining the DIM that contributed to accurate estimates of lactation yield using Wood's model. We can test the efficacy of this process by examining the trend of the median MSE P of all the TDSR tested within each loop (Figure 3). These plots show that, in general, the median MSE P decreased with each subsequent loop. However, this was not the case when the shape of the curves were APHP and $m=4$ TD, for which a small increase in the median MSE P was observed (Figure 3a).

Test-days sampling regimes tested

Selection of the TDSR to be compared within this study was done using a process that removed DIM with each loop in order to reduce the number of possible combinations of TDSR while maintaining the DIM that contributed to accurate estimates of lactation yield using Wood's model. We can test the efficacy of this process by examining the trend of the median MSE P of all the TDSR tested within each loop (Figure 3). These plots show that, in general, the median MSE P decreased with each subsequent loop. However, this was not the case when the shape of the curves were APHP and $m=4$ TD, for which a small increase in the median MSE P was observed (Figure 3a). The general downward trend of the median MSE P with each loop shows that, overall, the accuracy of the TDSR improved with an increasing number of loops. However, this does not necessarily mean that the TDSR with the lowest MSE P will be the last loop because the TDSR were chosen at random from the remaining pool of TD in each loop. Thus, it is possible to randomly choose the TDSR with the lowest MSE P in the earlier loops, although this is not very likely because there are more DIM to choose from.

Comparison of lactation yield estimates

The key question in this study was how lactation yields estimated with Wood's model [12] using fewer TD records compare with estimates from the recommended TIM method [9]. The plots in Figure 4 show the distribution of the MSE P values for the TDSR using four, five or six TD to estimate lactation yield. Lower values of MSE P indicate more accurate estimates of lactation yield.

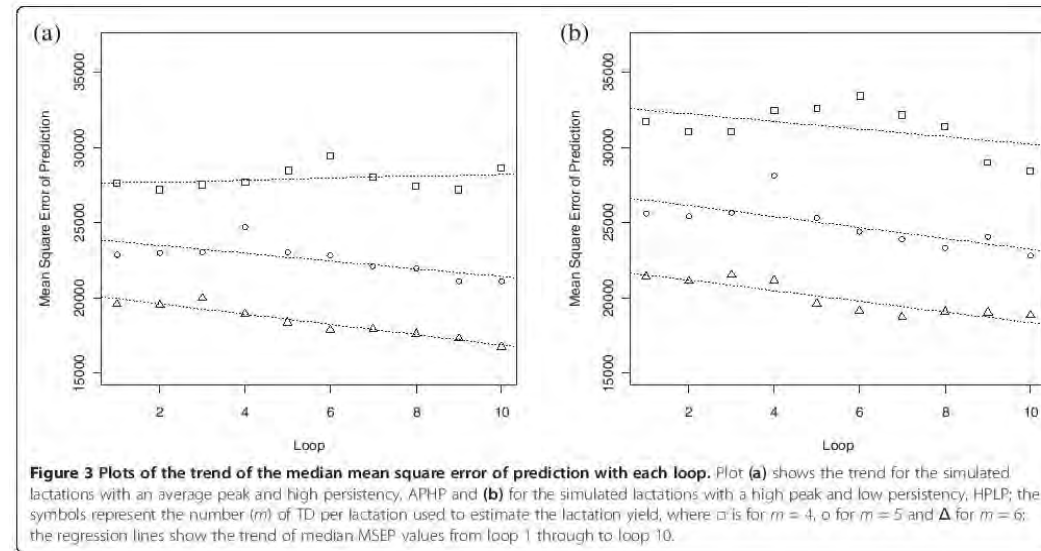
The TDSR method based on six TD per lactation resulted in comparable estimates of lactation yield as the TIM with monthly records, which had MSE P values of 12385 (APHP) and 13587 (HPLP), as shown in Table 1. This is indicated in Figure 4a and 4b by the location of the left-tail of the dashed MSE P curve ($m=6$), which indicates that a small proportion of the TDSR tested had MSE P values that were less than or equal to the MSE P for the TIM monthly estimates. Similarly, the position of all three probability density plots show that the use of Wood's model with m TD (where $m=4, 5$ or 6) to estimate lactation yield produced more accurate estimates than the TIM with quarterly records (MSE P values; 43621 for APHP and 71631 for HPLP) but less accurate estimates than TIM with weekly records (MSE P values; 2936 for APHP and 3112 for HPLP).

Comparison of estimated breeding values

The accuracy of lactation yield estimates, measured by the MSE P, is an important parameter to compare different methods of estimation. However, it is not easy to apply a measure when dealing with animal selection to determine the correspondence of the ranking of EBV calculated using the alternate methods compared with the 'true' EBV rankings based on the simulated lactation yields. Figure 5 shows the distribution of the number of cows in the top

100EBV that corresponded with the 'true' top 100EBV. In this figure, values closer to 100 are considered more accurate.

In both Figure 5a and 5b, the probability density plots show that there was a large proportion of TDSR that had greater Top 100 values than the estimates with the TIM based on quarterly records (Table 1; APHP- 73, HPLP- 77) and to a lesser extent the monthly records



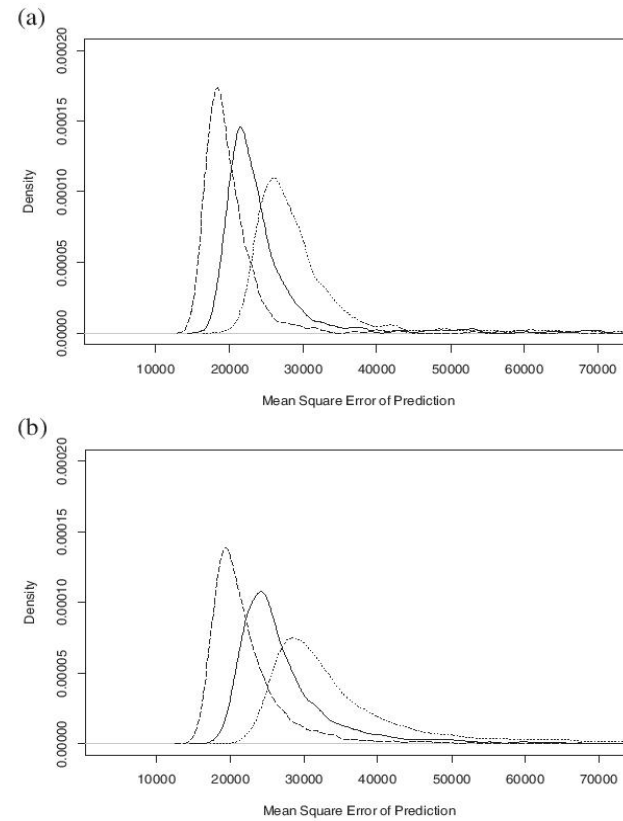
(Table 1; APHP- 85, HPLP- 88). As the number (*m*) of TD recorded per lactation increased, the proportion of TDSR that had a greater correspondence with the 'true' EBV rankings also increased compared to estimates obtained based on monthly records. This was even truer when the graph shape showed a rapid decline (HPLP) as compared to a slow decline (APHP). The median values for the probability density plots are in Figure 5 and Table 1. Results in Table 1 show that, as the frequency of recording increased, the median MSEP decreased. Similarly, as the median MSEP decreased the correspondence between the EBV rankings and 'true' EBV rankings increased. The values of the Top 100 corresponding animals show that using four, five or six TD, strategically timed throughout lactation, allowed for the selection of animals that aligned better with the 'true' EBV than the 'TIM Quarterly' regime and in some cases the 'TIM Monthly' regime.

Another measure that can be used to compare methods is the correlation of the resulting EBV with the 'true' EBV. For the APHP curves using four, five or six TD, these correlations were equal to 0.882, 0.905 and 0.923, respectively. For the HPLP curves, correlations were equal to 0.931, 0.947 and 0.959. These values concur with the trends seen in the Top 100 values but give a more direct indication of the potential amount of genetic gain lost because of using fewer TD per lactation.

Comparison of sires tested

By applying selection index theory, the EBV results can be further extrapolated for application within a progeny testing situation. Table 2 shows the bias and σ of the different methods to estimate lactation yield. The σ is the standard deviation of the deviations between true and estimated lactation yields using equation (12), whereas the bias is the mean of the differences between true and estimated lactation yields, $(Y_i, ESTIMATE - Y_i, TRUE)$. The resulting values of σ were used in equation (11) to calculate the number of daughters necessary to prove a sire with a reliability of 50%. Subsequently, assuming that a given amount of TD recording resources was available, the number of possible sires that could be tested was determined using equation (13). Results show that accuracy of the lactation yield estimate tended to increase as the number of TD per lactation increased. Hence, with the more accurate methods to estimate lactation yield, fewer daughters were required to prove a sire with a given reliability. However, the results also indicate that, with limited resources, the more frequent TD sampling reduced the number of sires that could be proven.

'Ideal' test-daysamplingregimeUsingtheMSEPvaluesfromthesimulationstudy,anidealsamplingregimefor collecting TD records can be recommended. The accuracy of lactation yield estimates and subsequent EBV predictions with the TDSR evaluated varied greatly. To develop possible recommendations for sampling regimes, a criterion was established to subset the TDSR into those that were accurate enough for selection and those that were not. For this study, the



criterion was set such that the estimation method had to have over 80 animals in the top 100 ranked EBV which corresponded with the 'true' EBV ranks. Of all the TDSR tested with $m=4$ TD within a lactation, 18.1% of the APHP and 63.9% HPLP shaped curves met this criteria. Figure 6 shows the distribution of the sampling days (one to four) for the TDSR that met these criteria. Similar figures could be produced for $m=5$ and $m=6$ as well, but for the purposes of the discussion we will focus on $m=4$. Figure 6 shows that it was necessary to have the first TD early in lactation, around the peak of lactation (where the average peak occurs at 34 DIM for APHP and 35 DIM for HPLP).

Using Kolmogorov-Smirnov goodness of fit tests [34], the distributions from Figure 6 were shown to differ significantly from the distribution of $m=4$ order statistics from a uniform $U(0, 280)$ distribution (all $P < 10^{-11}$). Therefore, the results from this study suggest that strategically timing TD sampling throughout the lactation yielded more accurate estimates of lactation yield than random (uniform) sampling.

Discussion

This study investigated methods to estimate lactation yield more accurately in a developing dairy sector where resources for data collection and progeny-testing are limited. The focus was to investigate estimation methods using fewer, yet more strategic, TD sampling regimes and to propose a methodology to progeny-test bulls for milk production when milk recording is limited. The two key comparisons were: (1) the TIM with Wood's lactation curve model and (2) TD recording regimes with equal intervals (weekly, monthly and quarterly) with TD recording regimes with four, five or six records per lactation more strategically placed throughout lactation.

Table 1 Comparison of different estimation methods and sampling protocols

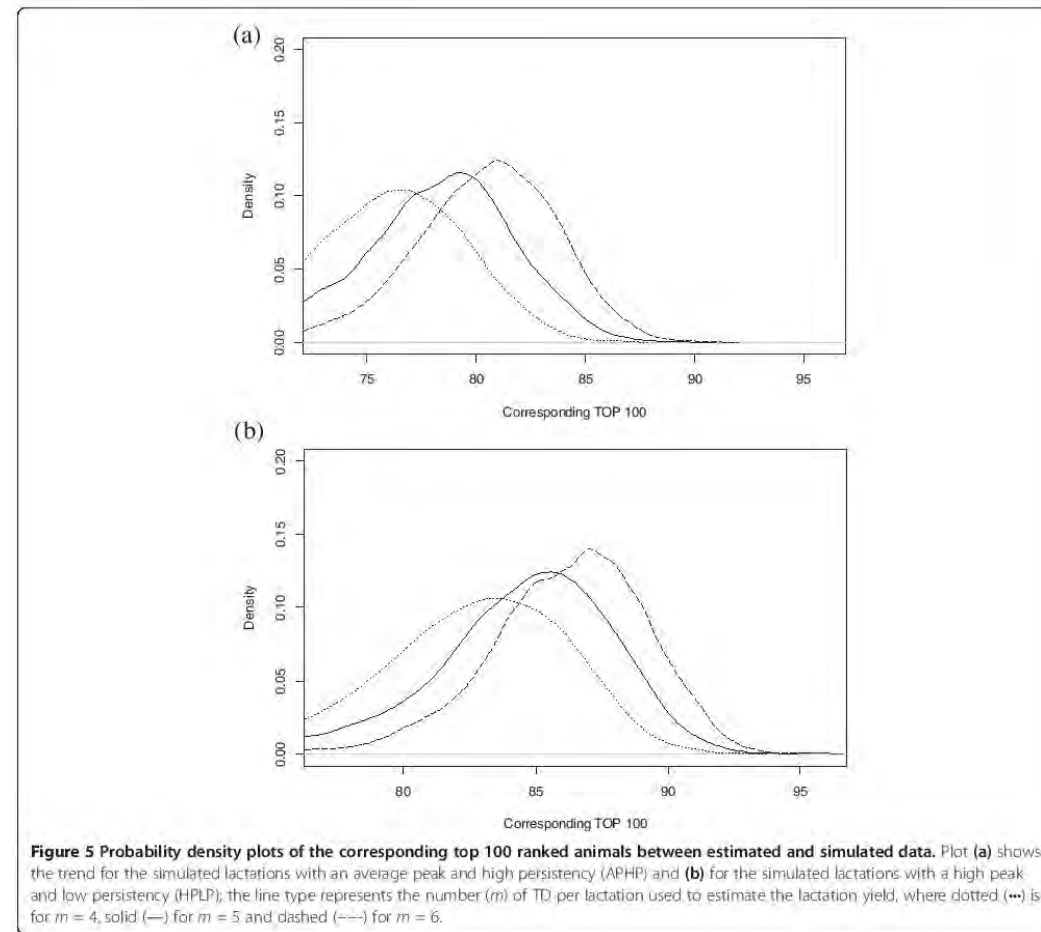
Sampling protocol	Average peak and persistent tail		High peak and less persistent tail	
	Top 100 _{Med}	MSPE _{Med}	Top 100 _{Med}	MSPE _{Med}
Test-interval method				
Weekly ^a	96	2936	96	3112
Monthly ^a	85	12385	88	13587
Quarterly ^a	73	43621	77	71631
6 Test-days/lact ^b	82	26829	87	32229
5 Test-days/lact ^b	81	30649	86	35479
4 Test-days/lact ^b	81	35767	85	47048
Wood model estimation				
Weekly ^a	96	2613	96	2945
Monthly ^a	87	10530	90	12098
Quarterly ^a	73	31334	82	34508
6 Test-days/lact ^b	82	18511	87	20666
5 Test-days/lact ^b	81	21654	85	24964
4 Test-days/lact ^b	81	25306	84	30324

^aFor the equally spaced estimation methods (weekly, monthly, quarterly) only one regime was tested and this value is reported here; ^bfor the different TDSR ($m = 4, 5$ and 6 test-days/lactation), the values presented here are the median top100 and MSPE values for all the TDSR tested that have at least 80 animals in common in the top 100 compared to the 'true' EBV.

The MSE_{Med} results (Table 1) show that in all cases, the TIM had higher MSE_{Med} than the corresponding recording regime using the Wood model with differences ranging from 5% (HPLP-Weekly) to more than double (for HPLP-Quarterly). This suggests that estimating lactation yield with the Wood model is more accurate than with the TIM. Therefore, if genetic gain was predicted based on the accuracy of EBV from this analysis, using the Wood model would be superior. However, the differences between the estimation methods are not as large based on the corresponding Top 100_{Med} values, which were either the same or differed by only one or two animals ranked in the top 100, with a maximum difference of 5 for HPLP-Quarterly. This suggests that the TIM and Wood model methods ranked the animals similarly for selection purposes and hence genetic progress would be essentially the same, regardless of the method.

Although the selection outcomes of the TIM and Wood model estimation methods may be the same, if we extrapolate this information further, the difference in the accuracy of the estimation will have an effect in the long term. Based on theoretical calculations, the Wood model estimation method can prove more sires (Table 2) than the TIM with the same TD sampling regime. Therefore, for a given TD sampling regime, we would expect the Wood model method to yield more accurate lactation yield estimates and hence use the available resources more efficiently. Nonetheless, it should be noted that a nonlinear model other than the Wood model may also be appropriate. The main consideration here was to adopt a model with relatively few parameters, bearing in mind the relatively small number of observations (m) per lactation.

With regard to the number of TD recorded per lactation, the results in Table 2 show that as the number of records within lactation decreased, the residual standard deviation (σ^2) of the bias in the estimation method increased consistent with other studies [26,28]. Using this σ^2 value and equations (11) and (13), we can determine the theoretical number of daughters required to prove a sire to attain a reliability (R) of 50% (Table 2). These results show that as σ^2 increases, more daughters are required to prove a sire with a given reliability. Despite this, recording fewer TD per lactation provides an opportunity to record more cows with the same resources allocated to the progeny-testing system (Table 2). Thus, recording regimes with fewer TD per lactation and using the Wood model method to estimate lactation yield are the most efficient in terms of use of the resources. The implications of this to the overall outcome of the progeny testing program is a greater pool of progeny-tested sires to select from, which means that genetic gain can be increased by increasing selection intensity. Duclos et al. [27], using similar calculations, also concluded that more animals can be tested by using fewer TD records in a lactation, without affecting the reliability of the bulls' EBV.



The novelty of this study lies in the testing of different strategically placed TDSR, with the aim of finding TDSR that provide the most accurate lactation yield estimates and the highest accuracy to select animals. The purpose of the loops and selection process for the TDSR was to find these ideal recording regimes. The results of this study show that more DIM are required in the earlier portion of the lactation than in the latter portion. This agrees with the idea discussed in previous research that suggests the earlier portion of lactation is more difficult to model than the latter portion [27,28]. Looking at all the TDSR tested and their lactation yield estimates, Figure 3 shows the median MSE from each loop. The general trend of both these plots (for APHP and HPLP) is that the median MSE decreased with each loop. This suggests that as the loop process continues, it is testing TDSR that allow for more accurate lactation yield estimates and hence is more likely to find the ideal TDSR.

Despite the positive outcomes of the loop process and the selection of superior TDSR, a number of issues must be considered. First, in reality, each cow has a different recording regime since not all cows will give birth at the same time. Furthermore, data collection occurs at different farms at different times. Therefore, even if one ideal TDSR was found, it would be difficult to implement that precise recording regime. Therefore, a more realistic outcome of this research is to develop recommendations about possible ranges of TD sampling times that yield good estimates of lactation. Figure 6 shows the distribution of the four TD of the TDSR tested with over 80 animals in the top 100 ranked EBV which corresponded with the true EBV ranks. The distribution and intersection of these curves indicate that there is a range for each TD, which, if followed, allows for adequate lactation yield estimation. The ranges shown in Figure 6 suggest

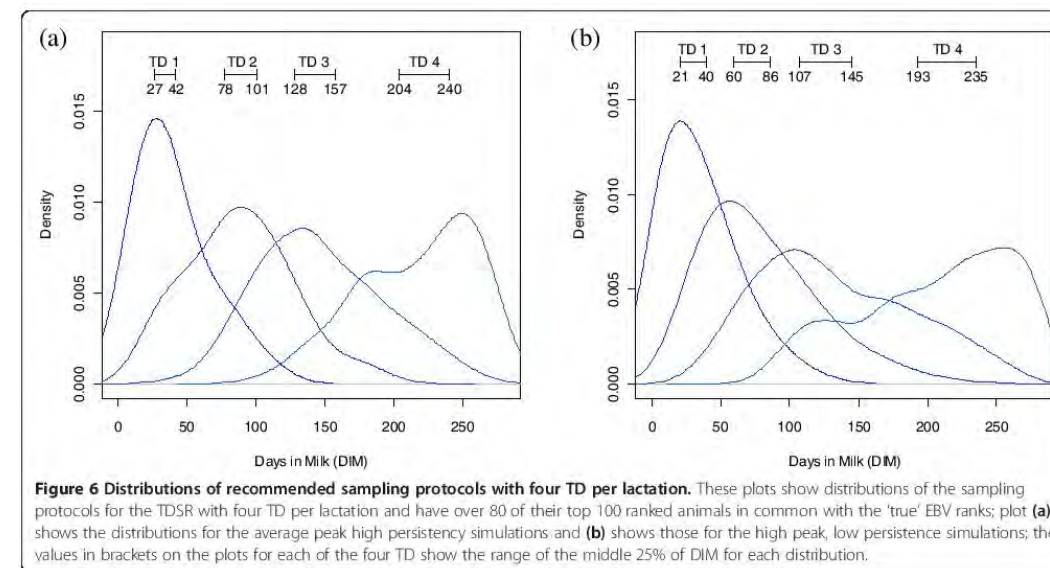
Table 2 Comparisons between estimation methods and sampling protocols based on the number of possible sires tested

Sampling protocol	Average peak and persistent tail			High peak and less persistent tail		
	Average bias ($\pm\sigma_{\Delta}$) [†]	Number daughters	Possible sires tested	Average bias ($\pm\sigma_{\Delta}$) [†]	Number daughters	Possible sires tested
Weekly	-9 (± 53)	16.3	11.2	-5 (± 56)	15.3	11.9
Monthly	-18 (± 110)	19.3	35.4	-29 (± 113)	16.8	40.7
Quarterly	-64 (± 199)	28.0	66.9	-177 (± 201)	20.9	89.6
6 Test-days/lact	5 (± 163)	23.9	52.4	61 (± 169)	19.2	65.3
5 Test-days/lact	7 (± 174)	25.1	59.8	46 (± 182)	19.8	75.8
4 Test-days/lact	10 (± 188)	26.7	70.2	76 (± 202)	21.0	89.3
Weekly	2 (± 51)	16.3	11.2	7 (± 54)	15.3	12.0
Monthly	14 (± 102)	18.7	36.4	21 (± 108)	16.6	41.1
Quarterly	33 (± 174)	25.1	74.8	31 (± 183)	19.9	94.3
6 Test-days/lact	20 (± 134)	21.1	59.1	19 (± 142)	17.9	70.0
5 Test-days/lact	23 (± 145)	22.1	67.9	21 (± 156)	18.5	81.1
4 Test-days/lact	27 (± 156)	23.1	81.1	25 (± 171)	19.2	97.6

The values reported in this table are the median values calculated from the TDSR tested that have at least 80 animals in common in the top 100 compared to the 'true' EBV; [†]the 'Average Bias' of each method is presented here with its residual standard deviation (σ_{Δ} , calculated using equation 12); the 'Number Daughters' reports the theoretical number of daughters that would be required given the σ_{Δ} of the estimation method to prove one sire with a reliability of 50% and a heritability of 0.2 (calculated using equation 11); the 'Possible Sires Tested' shows the predicted number of sires according to equation 13 that could theoretically be proven (with a reliability of 50%) given the required number of daughters.

that, although the frequency of TD in the later stages of lactation was not as high, it is important to have TD both pre-peak and post-peak lactation. Other studies have reported similar results, which suggest that the first TD should be recorded early in lactation [26] and post-peak sampling is important in the estimation procedure [28]. There is evidence to suggest that unequal intervals between TD lead to more biased estimates than equidistant records [27]. However, this study has completed a more thorough and direct comparison of these differences and found that unequal TD sampling intervals strategically placed throughout the lactation can provide less biased lactation yield estimates. This simulation process used in this study assumed a lactation length of 280 days and an approximate heritability for cumulative milk yield of 0.2 (see Additional file 1).

These values were used to ensure that the simulated lactations were similar to those of the Sahiwal population in Pakistan. If a longer lactation length (for example, 305 days) was used, the only difference, if any, could be slight changes in the recommended time frames from which one should take TD samples (Figure 6). This is because, although the overall length of the lactation would be longer, the key characteristics of the curve (peak, inflection) would not change and so the ideal



TD sampling times, which presumably revolve around these characteristics, would also not change. With regard to heritability, if for example, a higher heritability was used, partitioning of variation in the raw estimates of the parameters of the Wood model between polygenic random effects and cow specific “environmental” random effects would be different (see Additional file 1). This would lead to simulated lactation curves that would be more similar than the lactation curves simulated in this study. The implications of this could lead to lower MSPE values in all ITDSR, but the general comparative differences and recommendations would ultimately be expected to be the same.

It can be argued that the use of TDM or daily milk yields would be beneficial in developing progeny-testing systems since it would allow for the inclusion of unfinished lactations and handle the analysis of lactations with few records [25]. Several publications suggest that TDM can supersede selection based on even completed lactation yields [35, 36] because with improved statistical methods, both environmental and genetic effects [4, 37] are better accounted for and any yield more precise definitions of contemporary groups and stage of lactation [38-40]. However, for these methods to be effective, accurate estimates of genetic and phenotypic parameters are required [3, 8] which are difficult to obtain in developing countries [3] because in many cases field recording is inefficient and poor [41]. Research on data from Pakistan shows that TDM could be used [42] but this was based on a limited dataset. In the future, as more TD data become available electronically, the use of a fixed regression TDM could be a viable option. Furthermore, if TD are strategically placed, as suggested in this study, it could aid in the estimation of the parameters that describe the lactation curve shape in the fixed regression TDM. However, due to the current level of recording and electronic data entry, this study did not consider a TDM suitable for the Pakistani situation and instead looked at various approaches for which lactation yield is first estimated and subsequently used for breeding value estimation.

Conclusions

The results of this study show that using Wood's model to estimate lactation yield is more accurate than the TIM, although selection outcomes in terms of the ranking of EBVs were very similar. Results also show that using few TD records (say four, five or six TD within one lactation) that are more strategically placed throughout lactation can produce more accurate estimates of lactation yield than a quarterly recording regime and have the potential to be as accurate as a monthly recording regime. Lastly, although using fewer TD causes an increase in the residual standard deviation for the lactation yield estimate, they provide an opportunity to progeny-test more sires and thus for a more efficient use of the allocated resources. Although this study was based on data from Sahiwal cattle in Pakistan, these recommendations can be applied to any dairy breed with similar lactation curve characteristics.

Additional files

Additional file 1: Variance matrices used for simulation.
This file contains a description of the variance matrix values for the genetic and environmental effects used for the simulation of lactations in this study.

Competing interests

The authors declare that they have no competing interests.

Authors' contributions

DM and PT conceived the study and developed the preliminary design of the study. This was modified with input from both JL and HM. DM carried out all aspects of the study including simulation, data analysis and drafting of the manuscript. PT was heavily involved in the analysis process and the preparation of the manuscript. HM was involved in the design of the simulation study and contributed to the manuscript's intellectual content. JL aided in the interpretation of results and in the preparation of the manuscript. All authors read and approved the final manuscript.

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Participation of Women in Dairy Farm Practices under Small Holder Production System in Pakistan

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The objective of the study was to investigate the participation of women in dairy farm practices in the small-holder production system in Pakistan. A questionnaire was designed to collect the data from female farmers (n=200) of districts Bhakkar and Jhelum in the Punjab province. Our results indicate that the participation of women in dairy farm practices was significantly higher ($p < 0.05$) in Bhakkar as compared to district Jhelum. The present study suggested that, in order to maximize the small-holder dairy farm productivity, a strong extension program should be implemented to enhance the skills and knowledge of women.

Key Words: Women, Dairy farm practices, Smallholder

INTRODUCTION

Livestock are considered a key asset for rural livelihoods and offer significant opportunities for improving household incomes. Women traditionally play a major role in conducting various livestock management activities all over the world. Pakistani women have a significant role in agriculture and livestock rearing. Nearly 65.9% of Pakistan's population are living in rural areas that are directly and indirectly linked with the agricultural related sector for their livelihood (Farhana et al; 2008). Women comprise half of the rural population and contribute 60 to 80% of labour in the animal husbandry (Younas et al., 2007). Women not only perform normal household chores such as cooking, cleaning, mending clothes and raising children (Kazmi, 1999), but also participate in rearing of livestock and carry out various dairy farm practices. These practices include feeding and watering, fodder cutting, cleaning animals and their sheds, caring for sick animals, calf rearing, milking and the processing of dairy products like ghee, butter and yogurt. Some of these activities, like fodder production, are generally considered the responsibility of men, but in many cases the women are also involved.

The participation of women in dairy farm practices varies by region, age, culture and social status and are changing rapidly in some parts of the country. The existing information regarding participation of women in dairy farm practices is very limited. Therefore, the present study was aimed to investigate the participation of women in dairy farm practices in the small-holder production system within the districts of Bhakkar and Jhelum. Additionally, possible factors affecting their participation in dairy farm practices were investigated. It is anticipated that the information generated from this study will be helpful in identifying the extension needs and areas where women can improve dairy production by enhancing their skills and knowledge.

MATERIALS AND METHODS

A dairy extension project (no. LPS/2010/2007, funded by ACIAR) is working in Pakistan aimed at strengthening the dairy value chains in Pakistan through improved farm management and more effective extension services. A questionnaire was designed to collect data from project working areas, 92 female farmers from Bhakkar and 102 from Jhelum. These two districts provide a contrast between an undeveloped arid region poorly served by irrigation and state livestock services (Bhakkar) and a more advanced region where farmers have access to extensive irrigation and support from the state livestock veterinarians (Jhelum). Eight villages were selected from Bhakkar and nine from Jhelum.

Statistical analysis

Participation of women in dairy farm practices under smallholder production system in comparison to Bhakkar and Jhelum was analyzed using Chi-square test. All the analysis was carried out with the Statistical Package for

Social Sciences (SPSS-13.0) A p-value of 0.05 was regarded as significant.

RESULTS

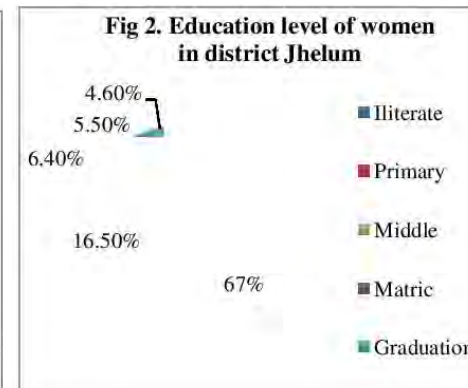
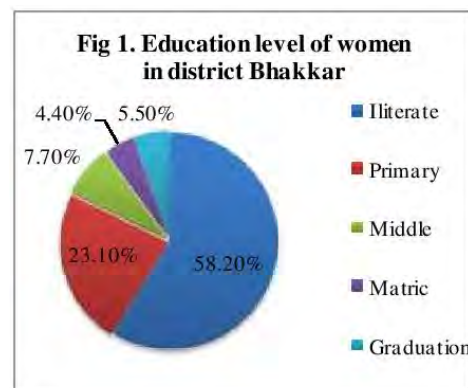
Participation of women in dairy farm practices was significantly higher ($P < 0.05$) in Bhakkar as compared to district Jhelum. A comparison of female participation in various dairy farm practices between the two districts (Table 1).

Table 1. Comparison of participation of women in various dairy farm practices between district Bhakkar and Jhelum

Livestock management activities	Districts	
	Bhakkar (%)	Jhelum (%)
Shed Cleaning	71 (78.3)	80 (73.1)
Fodder cutting from field	19 (20.7) a	17 (15.7) b
Feed and watering	58 (64.1) a	22 (19.4) b
Care of calf	67 (73.9) a	32 (28.7) b
Milking	71 (78.3) a	22 (19.4) b
Milk sale	64 (69.6) a	61 (56.5) b
Care of sick animals	37 (40.2) a	10 (9.3) b
Value addition of milk	71 (78)	98 (89.9)

a, b means with different superscript within rows are significantly different ($P < 0.05$)

Socio-economic status and cultural norms are affecting women's participation in dairy farm practices in the small-holder production systems of both Bhakkar and Jhelum. In Jhelum 15.6% of women are not participating in dairy farm practices due to traditional cultural barriers and 24% do not contribute because of their relative affluence. In contrast, in district Bhakkar only 4.4% are prohibited from participating because of cultural barriers while 9% are not participating because of their relative affluence. The education level for women was observed to be almost the same in both Bhakkar and Jhelum (Fig 1 and Fig 2).



DISCUSSION

The present study reveals that the participation of women in dairy farm practices was significantly higher in Bhakkar than in district Jhelum. Women contribute exclusively in various dairy farm practices such as routine husbandry and nutritional management. The present study proposes that the provision of appropriate extension services to women in these activities can significantly improve the productivity of animals. The working competencies of the rural women can be strengthened and upgraded by providing training on livestock rearing practices (Iftikhar et al., 2007). Trained women will help to increase dairy production and enhance household incomes. Furthermore, to enhance the social standards and participation of women in economic activities, there is a need to provide them with quality education and knowledge of the latest

technical advances in the fields of agriculture and livestock production (Khalida, 2009). Social mobilization and agricultural productivity can be advanced through active involvement of women in the development of land, livestock, education, extension, financial and employment resources (FAO, 2010-11).

Factors that limit participation of women in dairy farm practices are socioeconomic status and cultural norms. Ideology, religion, economics and culture are the limiting factors in terms of the provision of gender specific services and opportunities (Moser, 1989). From a cultural perspective of Pakistan the role of women has always been misconceived. Constraints relating to women include cultural values, normative patterns and customs, most of which are without religious and ethical sanction (Khan, 2012). Present study shows that from Jhelum 15.6% women are not taking part in dairy farm practices due to cultural barriers and 24% due to their high economical status. The women from affluent families do not work themselves but they hire laborers, whereas most of the rural and tribal women from disadvantaged communities do most of the on farm work themselves (Rangnekar et al, 1992). The findings of Rathod et al. (2011) are also consistent with the earlier that these women can afford labor which in turn reduces their participation in livestock management activities. Land holding is an important determinant of their economical status. Although the average land holding in Bhakkar (9 acres) is more than in Jhelum (6.9 acres) the poorer fertility and lack of irrigation water combined with more hot weather are major factors limiting production in Bhakkar compared to Jhelum. The present study indicates that the education level was almost the same in the two districts. The overall literacy rate (aged 10 years and above) in Pakistan is 57.7% (Pakistan Economic Survey, 2010-11). It is important to know that equal opportunities (schools and colleges) are available for females in both of the districts, which was consistent with the results of our survey.

The present study suggests that, in order to maximize the dairy farm productivity a strong extension program needs to be implemented to transfer adoptable technologies and enhance the knowledge and skills of women in all aspects of livestock management practices including husbandry, nutrition, calf rearing, health and value addition to milk. Additional surveys of this nature will help to identify other regions of Pakistan in which women need to be the focus of specialist workshops. This will then lead to rapid improvements in their prosperity of village communities across the country.

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PRODUCTION AND REPRODUCTION PERFORMANCE OF NILI- RAVI BUFFALOES UNDER FIELD CONDITIONS OF PAKISTAN

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ABSTRACT

The objective of the longitudinal study was to evaluate the production and reproductive performance of buffalo on small-holder dairy farms in Pakistan. The data were collected from 207 farms located in the districts of Okara and Bhakkar. Milk production and reproductive parameters were recorded on a weekly basis by trained extension workers in specifically designed herd books for 18 months from November 2007. Preliminary results indicate that the average milk production per lactation was higher ($P < 0.05$) in buffaloes (1226.63 ± 43.50 lit) than in cows (1027.04 ± 44.88 lit). The percentage of oestrus detection, A.I., natural service and pregnancy rate for buffaloes were (0.38%, 7.29%, 19.17%, 78%), respectively, during the whole year. We conclude that neither the Nili-Ravi buffalo nor cows are attaining their potential for milk production. The low reproductive efficiency of these animals is mostly likely related to both the poor technical acumen and the nutritional status of animals. Provision of appropriate extension services for these farmers will improve the productive and reproductive performance of buffaloes.

Keywords: Buffaloes, Production, Reproduction

INTRODUCTION

The world population of domestic buffaloes, *Bubalus bubalis*, has been estimated to be more than 150 million (Bhat, 1992) or one-eighth the population of cattle, with the numbers steadily increasing. Two main types of domestic buffalo are the river buffalo and the swamp buffalo. The Nili-Ravi breed of buffalo is classed as a river type and is the best milk producer among the other breeds of buffaloes in the world. They have a wedge shape, massive frame, small curly horns, and walleyes. They often have white markings on the forehead, face, muzzle and legs and white switch of tail (buffaloes with such markings are highly desired and popularly called "PanjKalian") (Warriach et al., 2008).

Pakistan like many developing countries has an agrarian rural based economy. Livestock is a major contributor to the national (12%) and agriculture (50%) economy (Pakistan Economic Survey, 2006). Milk is the key livestock product. Pakistan is ranked the 4th largest milk-producing country in the world. Seventy percent (70%) of the milk and fifty percent (50%) of total meat produced in Pakistan comes from buffalo (Usmani et al., 1987). An important point considering milk constituents are its high values in buffalo than cows: milk fat 6.5-8.0% versus 3.5-4.0% and solids-not-fat 9.0-10.5% vs. 7.5-8.5% (Gordon, 1996). Thus the price of buffalo milk tends to be higher than the milk produced by dairy cattle. Furthermore, buffalo utilize poorer quality roughages, adapt to harsher environments and are more resistant to several bovine tropical diseases (Gordon, 1996).

Despite these merits, buffalo has relatively poor reproductive efficiency that varies little with location throughout the world. Buffalo exhibit many of the known reproductive disorders and have delayed onset of puberty, poor oestrus expression, longer post partum ovarian quiescence, and most importantly lowered conception rates particularly when bred artificially (Gordon, 1996). It appears that because buffalo is populated mostly in developing countries with meager resources, it remained neglected or

underutilized in terms of quality research in the area of health, management, nutrition and reproduction. The role of extension has been to provide research-based education and information to the production sector. Services to the dairy sector are being provided by government agencies and a range of NGOs, and virtually all services providers who interact with the farmers are veterinarians or para-veterinarians who performed vaccination, treatment and A.I. Limitations in the extension service and the research/extension interface are considered to be bottlenecks in the development of the dairy sector in Pakistan. Keeping in view these facts the major objective of this study is to evaluate the production and reproductive performance of buffaloes on small-holder dairy farms under field conditions of Pakistan.

MATERIALS AND METHODS

Dairy Project Background:

In 2007, a 2 ½ year dairy project “Improving dairy production in Pakistan through improved extension services” was started in the two contrasting environments of districts Okara (well developed) and Bhakkar (less well endowed) with the objectives of increasing dairy production through improved extension services. Small dairy farmers having 3- 10 (buffalo and/or cattle) for production are the main target group in the project. Improved extension services as well as veterinary services are being provided to the farmers by already existing agencies (Livestock & Dairy Development Department, Punjab, National Rural Support Program and Idara-e-Kissan) in both of the districts.

Longitudinal survey:

The data were collected from 207 farms located in the districts of Okara and Bhakkar. Milk production and reproductive parameters were recorded on a weekly basis by trained extension workers in specifically designed herd books. Milk was measured with weighing scales. One year data of milk production from (n=222) and (n=163) buffaloes and cows respectively, were collected for analysis. Reproductive parameters were tracked from (n=385) animals for analysis.

Farmers and extension worker trainings:

The project emphasized on comprehensive interdisciplinary educational program of discussion group meetings, workshops and trainings of both farmers and extension workers separately Table 1. Basic husbandry, nutrition, forages, health and calf management were the first areas addressed during the first six months of the project.

Statistical analysis

Data was collected on a weekly basis and entered into a data base containing all the information about each of the farm. A single lactation of milk production was calculated from each animal based on the weekly production as well as the duration of the particular animal's lactation. Then these estimates of the total milk production during the lactation were compared between the different breeds and species of animals using an ANOVA. The reproduction data values are based on observations from the field and counts of observations made by farmers and field workers. These values are simply reported as percentage values of the observations and no statistical analysis was carried out.

RESULTS

Average milk production per lactation was higher ($P < 0.05$) in buffaloes (1226.63 ± 43.50 lit) than in cows (1027.04 ± 44.88 lit). Average milk production per lactation in buffaloes and various breeds of cows have been presented in Fig 1. The percentage of reproductive parameters like oestrus detection, A.I, natural service, dry and pregnancy rate for buffaloes during the whole one year have been presented in Fig 3.

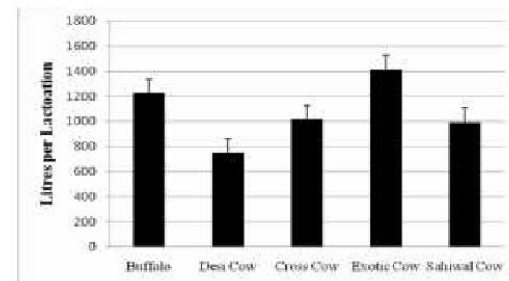


Fig 1. Average milk production per lactation was higher ($P<0.05$) in buffaloes than in cows

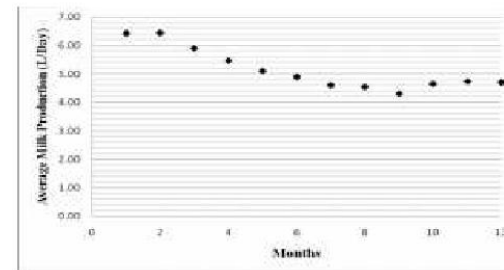


Fig 2. Average milk production per lit/day of dairy animals in field conditions

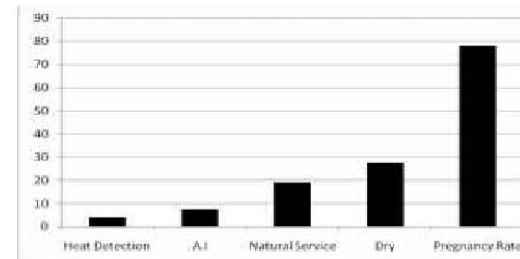


Fig 3. Reproductive parameters in buffaloes around the year in field conditions

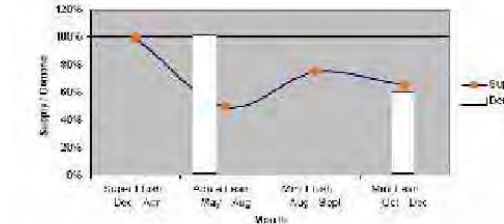


Fig 4. Analysis of Milk Marketing Chain- Umm e Zia, 2007

Table 1. Topics and visitation schedule of the training of farmers and extension workers during first year of the project.

Visiting order	Farmers trainings	Extension workers trainings
1.	Basic husbandry management practices	Ultrasonography in animal reproduction
2.	Fodder demonstration plot training	Basic farm management principles
3.	Herd health and calf management	Herd health and calf management
4.	Animal health and nutrition module	Animal health and nutrition module
5.	Calf nutrition and health	Calf nutrition and health

DISCUSSION

The present study demonstrates that in buffaloes, average milk production per lactation was higher than in cows under field conditions. Milk production potential is about double the present level of milk production (Iqbal and Ahmad, 1999). This is evident by comparing the maximum milk production observed in the elite herds maintained at Government research stations with those on private livestock farms. Several researchers have reported data on maximum milk production potential of the breeds. Shah (1991) reported that Nili - Ravi buffalo have a production potential of 5000 lit per lactation of 305 days. Similarly, Husnain and Shah (1985) reported that maximum breed potential observed in the elite animals of Sahiwal is up to 6500 lit of milk per lactation of 305 days. A possible short term strategy for increasing the milk production on any farm is by the better management of the existing feed resources through balanced feeding. Past research has shown that this yield increase of 30% are achievable if this is carried out (Burkiet al., 2005), as well as significant reductions in production costs (Mahmood et al., 2004). However, for long term success and sustainable growth there is a need to plan and implement a simple and effective selective breeding regime to allow for an increase in milk production for the entire country.

This present study shows a gradual decline in average milk production per day of dairy animals during the year. This is shown in Fig 2.

In Pakistan, this decline coincides with an increase in demand during the hot summer months. This is due to the fact that most of the buffaloes calving takes place during July-September (Ahmad et al., 1981) and very few calving occurs during February- June. Furthermore, milk production is associated with the availability of green fodder and is at its maximum between January and April and hits a low from May to August. Alternatively, milk consumption is low during the winters and is at its peak during the summer due to higher intake of consumer intake of milk products such as lassi, yogurt, and ice cream (Fig 4).

In the present study, oestrous detection in buffaloes was observed in only 0.38% of the small-holder farmers. This could be due to a number of factors. One of the most likely reasons for the low oestrous detection rates could be due to husbandry practices such as the practice of keeping dairy animals tied up. In doing this, the animals are unable to show as many signs of oestrous as they could if they were kept free and untied. Furthermore, buffaloes have more tendencies to show heat signs during night and a negligible number of the farmers observed the oestrous signs during the night time (Warriach et al. unpublished data). Season is also one of the other extrinsic factors that influence the oestrous behaviour symptoms. In the tropics, high ambient temperature reduces sexual activity in the day time (Jainudeen, 1977) and shortens the oestrous period (Gill et al., 1973), and the incidence of silent oestrous is more common during the hot summer season. These adverse effects of heat stress make oestrous detection more difficult and may exert a considerable influence on the oestrous behaviour in buffaloes.

In conclusion, preliminary results indicated that neither the Nili-Ravi buffalo nor cows are attaining their potential for milk production. The low reproductive efficiency is mostly likely related both to poor technical acumen and nutritional status of animals. Provision of appropriate extension services for these farmers will improve the productive and reproductive performance of buffaloes. It is hoped that the data generated from this longitudinal survey will be helpful to devise better strategies for improved extension services. In order to optimize the dairy production of smallholder farmers and will have a ripple effect for the others to follow.

Acknowledgements:

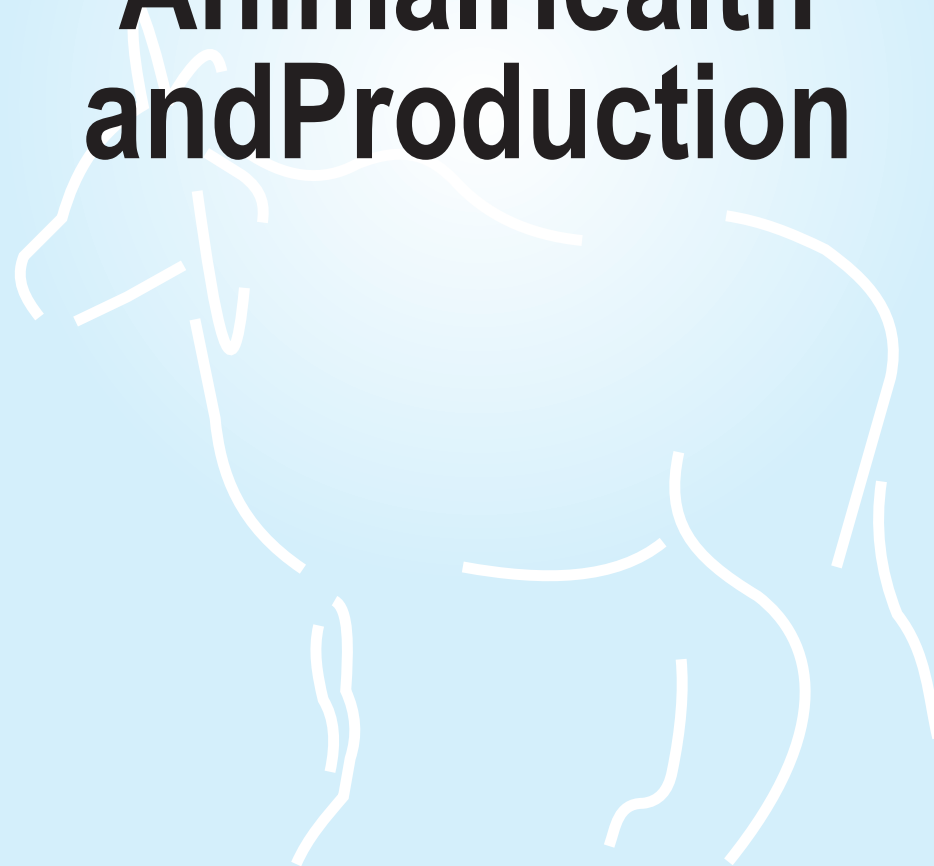
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**Tropical
Animal Health
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Effect of diet on preweaning performance of Sahiwal calves

Shaukat Ali Bhatti & Muhammad Faisal Ahmed & Peter Charles Wynn & David McGill & Muhammad Sarwar & Muhammad Afzal & Ehsan Ullah & Musarrat Abbas Khan & Muhammad Sajjad Khan & Russell Bush & Hassan Mahmood Warriach & Ahrar Khan

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Abstract

The objective was to study the growth potential of Sahiwal calves given milk or milk replacer with or without concentrates. For this purpose, forty-eight Sahiwal calves were divided into four groups of 12 animals each with equal sex ratio. In each group, the calves were offered either milk or milk replacer (MR) at a rate of 10% of their body weight adjusted weekly. In addition to this, calves were fed either a starter ration plus Egyptian clover hay (SR+H) or hay only (H) until the end of trial. The milk or MR was withdrawn gradually from day 56 until animals were weaned completely by day 84. Calves offered milk grew faster than those offered MR (357 ± 9 vs. 162 ± 9 g/day; $p < 0.05$) and displayed higher weaning weights (51.6 ± 0.8 vs. 35.2 ± 0.8 kg; $p < 0.05$). The calves offered SR+H grew faster (311 ± 9 vs. 208 ± 9 g/day; $p < 0.05$) and displayed higher weaning weights (48.7 ± 0.8 vs. 38.1 ± 0.8 kg; $p < 0.05$) than those fed H alone. Calves offered milk plus SR+H showed the highest growth rate and weaning weights (401 ± 13 g/day and 56.3 ± 1 kg, respectively). The lowest growth rate and weaning weights were observed in calves offered MR and H only (115 ± 13 g/day and 30.3 ± 1 kg, respectively). Calves offered the MR had a higher number of scour days than those offered milk (13.5 vs. 3.3). The feeding of whole milk in combination with the starter ration and hay resulted in superior growth rates, higher weaning weights, and healthier calves than the other feeding regimens.

Keywords

Calf nutrition. Milk replacer. Concentrates. Growth

Introduction

High feeding costs coupled with a high mortality in preweaned buffalo and cow calves during the preweaning period is a major limitation to productivity in the Pakistan dairy system. This leads to a reduction in the number of heifer and bull calves raised for breeding and meat production purposes. The cost of feeding whole milk to calves, through to weaning under the traditional feeding system ranges between 7–8 and 10–12,000 Pakistani rupees (US\$94 and 130, respectively) for a cow and buffalo calf, respectively (Bhatti et al., 2009) and still may go higher with increasing prices of whole milk per liter. The market price for a weaned male buffalo or cow calf is usually less than half the milk feeding cost to weaning. In commercial enterprises, male calves are most often sold on day one or two after milk according to their growth requirements (Ahmad et al., 2009). This is to conserve milk for human consumption or to sell at a higher price than a calf would return. However, the female calves are kept as the future dairy replacements (Ahmad et al., 2009), but they are still not fed appropriately to reach puberty in minimal time and therefore lifetime productivity is compromised.

Different alternatives are available to reduce the feeding cost of the calves during the preweaning period without compromising their growth rate and health.

These include weaning the calves at an early age or substituting milk replacers for whole milk. Milk replacers are generally cheaper and offer an alternative to whole milk feeding to weaning. These practices are common place in the management of calves from temperate breeds such as Holstein-Friesians in contemporary dairy systems. However, comparable growth responses to similar feeding regimes in calves of tropical breeds like Sahiwal

(Zebu cattle), which is famous for its heat tolerance and tick resistance the world over (Berman, 2011), are less well documented.

Sahiwal cow originated and developed in Pakistan and has been imported to 29 countries of the world for crossbreeding in different agroecological zones including Australia, tropical Latin America and Africa (FAO, 2007).

The objective of the study was to compare the effects of feeding whole milk or milk replacer with or without concentrates on the growth and health status of Sahiwal calves.

Materials and methods

Animals

Forty-eight 3-day-old calves born during the months of January and February, 2009 were procured from the Livestock Experiment Station (LES), Bahadurnagar, Okara. The calves received fresh colostrum at the rate of 10% of their body weight within 6 h of calving that continued today 3. The animals were housed under hygienic conditions with ample clean straw as bedding and their navel cords were disinfected before being transported from the LES to the University of Agriculture Farms at Faisalabad (UAF).

Housing and management

Calves were then housed in separate calf pens which were 142 × 112 × 112 cm (length × width × height) in size. The wooden slatted floor of the pens was raised 23 cm. Paddy rice straw was used as bedding and replaced on a daily basis. Animals had access to water ad libitum in buckets that were cleaned daily. The present study was conducted in the winter–spring months of January to April. In order to ensure the thermoneutrality of calves at night when the ambient temperature fell below 20 °C, they were housed together in an adjacent indoor facility.

Feeding

The calves were randomly allocated to four treatment groups in a two by two factorial arrangement. In each treatment group, there were 12 calves (six males and six females). Factor one was liquid diet, either whole milk or milk replacer. The second factor was the solid diet, either starter ration (SR; Table 1) plus berseem hay (H) (Egyptian clover: *Trifolium alexandrinum*; CP: 16.1%) or H only, offered ad libitum from day 8 of age until weaning.

Whole milk was obtained fresh from Sahiwal cows on a daily basis. Milk replacer (MR: Sprayfo®) was procured from Sloten, B. V., (Deventer, Netherland) and reconstituted to the manufacturer's recommendations. The milk and MR consisted of 11.49 and 11.11% dry matter, 3.35 and 2.22% protein 3.20 and 1.84% fat and 4.29 and 4.00% lactose, respectively. Milk or MR was offered at the rate of 10% of the calf's body weight with animals weighed weekly and feed adjusted accordingly. The daily milk offered was divided equally and fed at 0900 and 1600 h at a temperature of 40 °C. The milk was fed through nipple feeders fitted in feeding buckets.

Table 1 Composition of calf starter ration

Ingredient (%)	Calf starter ration
Maize grains	20.0
Canola meal	25.8
Wheat bran	11.2
Maize oil cake	20.0
Maize gluten 30%	20.0
DCP	2.0
Salt	1.0
TDN	78.0
Dry matter %	97.83
CP %	19.6
CF %	4.95
EE %	3.7
Ash	4.5
Energy kcal/kg	4,046

Thus, the treatments were: milk with SR+H (Group I), milk with H only (Group II), MR with SR+H (Group III), and MR with H only (Group IV). Milk or MR was offered until day 63 of age, and then tapered to zero by day 84. The calves were weighed weekly in the morning before feeding. Records were also maintained on the occurrence of calf scours in each animal and any medical treatments that were given.

Economics of production

The costs of whole milk and MR were PKR 30 and 19 per liter and of SR and H were PKR 20 and 7 per kg, respectively (1 US\$. PKR 87). The cost of feeding for each animal each day was calculated and used for the statistical analysis.

Statistical analysis

The recorded information on weekly growth and liquid and solid feed consumption was analyzed using repeated measures analysis. This was conducted using the MIXED Procedures of SAS (SAS, 1996) with an AR(1) covariance structure as described by Littell et al. (1998).

The effect of calf was considered to be random. The statistical model used for analysis was where Y_{ijk} is the dependent variable, \bar{f} is the overall mean, sex_i represents the sex of the calf and i indicates either male or female, F_1 is the fixed effect of factor one where $j = e$ is either milk or milk replacer, F_2 represents the fixed effect of factor two where $k = e$ is either the SR+H or H only, W is the repeated measure of weeks l , while d sex F_1 F_2 W T_{ijkl} represents the effect of interaction of sex, factor one, factor two and weeks, c_{lm} represents the random effect of calf m and e_{ijklm} is the residual error.

For the other descriptive statistics (average growth rate, weaning weight, total weight gain, total intake of milk, milk replacer, hay and starter ration), the birth weight was used as a covariate. The data were analyzed using MIXED procedures. Sex was also included in the model.

However, sex had a non-significant effect ($P=0.07$), so it was dropped from the model in the final analysis.

The statistical model was: where \bar{f} is the overall mean, F_1 is the liquid feed either milk or milk replacer, F_2 is the solid feed either SR+H or H only, $(F_1 \cdot F_2)$ is the interaction of liquid feed and solid feed, BWT is the birth weight of calf taken as a covariate, c_{lm} is the random effect of calf m and e_{ijklm} represents the residual error.

One animal died from group I and group II each during the experiment, but this is accounted for in the AR(1) covariance analysis structure. The results are reported as least square means. The means were declared significantly different at $P < 0.05$.

Results

Birth weights

The birth weights of Sahiwal calves were 22.7 ± 0.9 and 20.3 ± 0.9 kg in males and females, respectively. Intake of milk or milk replacer Total intake of whole milk was significantly higher ($P < 0.05$) than the intake of MR in the calves over the 84 day preweaning period (Table 2). Figure 1 shows the changes in consumption of milk or MR during this period. Intake of starter ration and hay The total intake of SR was not different ($P > 0.05$) in calves fed milk or MR (Table 2), but tended to be higher in the former. The SR consumption was negligible (< 200 g/day) until day 42 of age in all calves and then increased gradually (Fig. 3). The intake of starter ration was similar in calves fed either milk or MR until day 63. However, from day 70 the SR intake was higher in calves fed milk than those offered MR. By day 84, the SR intakes were 943 and 693 g/day in calves offered milk and MR, respectively. The total hay consumption was not different ($P > 0.05$) in calves in all the treatment groups (Table 2). Hay intake was negligible (< 200 g/day) in all the calves until day 42 (Fig. 4) and increased gradually to more than 400 g/day by 63 days post-partum. From day 70, the calves offered milk plus hay only consumed more hay than the other three groups, reaching 1 kg/day by weaning at day 84. Daily hay intake in other groups did not differ significantly today 84, ranging from 653 to 785 g/day.

Averagedailygrowthrate

AveragedailygrowthrateofSahiwalcalvesofferedmilkwas195g/dayhigher($P<0.05$)thanthoseofferedMR (Table 3). The SR + H-fed calves displayed 103 g/day higher growth rate ($P<0.05$) than calves offered the berseemhayonly(311• }8vs.208• }8g/day).Thehighestgrowthrate

Table2 LeastmeansquaresoffeedconsumptionofSahiwalcalvesoffereddifferentdietarytreatmentsduringpreweaningF1 milkvs.MR,F2SR+Hvs.H,MRmilkreplacer,Hhay,SRstateration

Intake	Milkvs.MR		SR+Hvs.hay		F1	F2	F1.F2
	Milk	MR	SR+H	Hay			
Milk/MR(l)	217.5±1.7	184.5±1.7	209.9±1.7	192.0±1.7	0.0001	0.7	0.8
SR(kg)	25.0±2.1	22.1±2.2	23.6±2.1	0	00.36	N/A	N/A
Hay(kg)	21.3±1.2	18.4±1.2	17.8±1.2	21.9±1.2	0.08	0.001	0.4

F1milkvs.MR,F2SR+Hvs.H,MRmilkreplacer,Hhay,SRstateration

(401±13g/day)wasobservedincalvesfedmilkandSR+Handthelowest(115±13g/day)incalvesgivenMR andhayonly.Weeklyweightsofthecalvesondifferentfeedingregimensreflectedsimilarrends(Fig.2).

Weaningweights

WeaningweightsofSahiwalcalvesofferedmilkwere17kghigher($P<0.05$)thanthoseofferedMR(Table3). TheSR+H-fedcalveswere1kgheavieratweaningthanthoseofferedberseemhayonly.Thehighestweaning weight(56±1kg)wasobservedincalvesofferedmilkplusSR+Hfollowedbythecalvesofferedmilkwith berseemhay(Table3).Thelowestweaningweight(30±1kg)wasobservedincalvesgivenMRandberseemhay only.

EfficiencyofmilkorMRutilization

TheefficiencyofutilizationofmilkorMR(FCR)wascalculatedinthisstudy,bydividingliveweightgain(kg) bythetotalvolumeofmilkorMRconsumed(liters)whenoferredSR+Horthayasthesupplement.This calculation doesnotaccountfordifferencesinhayandSRconsumedand,therefore,isonlyindicative.Theconversionratio ofmilkto liveweightwassignificantlyhigher($P<0.05$)thanforMR(7.4±0.8and15.8±0.8lofmilkandMR, respectivelyconsumedperkgofliveweightgain).Therewasnodifferenceinmilkconsumedperkgoflive weightgainincalvesofferedSR+Horthayonly(8.1±1.1vs.6.8±1.1perkg liveweightgain, respectively).

However, less MR was required per kg live weight gain in SR + H fed calves than calves offered hay only (11.1±1.1 vs.20.2±1.1, respectively).

Incidenceofcalfscour

NumbersofscourdayswerehigherincalvesofferedMRthanthoseonmilkonly(13.5vs.3.3).ThecalvesfedH onlyhadmorescourdays thanthose fedSRplusH(10.1 vs. 6.2).Economics of feeding Total feeding cost (includingbothliquidandsolidfeed)washigher($P<0.05$)inmilk-fedcalvesrelativetothoseofferedMR(Table 3).ThetotalfeedingcostwashighestincalvesfedmilkplusSR+HandlowestincalvesofferedMRplushay only.Overall,theproductioncostperkilogramoflive-weightgainwashigherinMRfedcalves thanforthosefedmilk.

Fig.1 Intake(l/day)ofmilkormilkreplacer(MR)inSahiwalcalvesofferedmilkplusstateration(SR)andhay(H),milk plusH,MRplusSRandH,MRplusHduringpreweaning

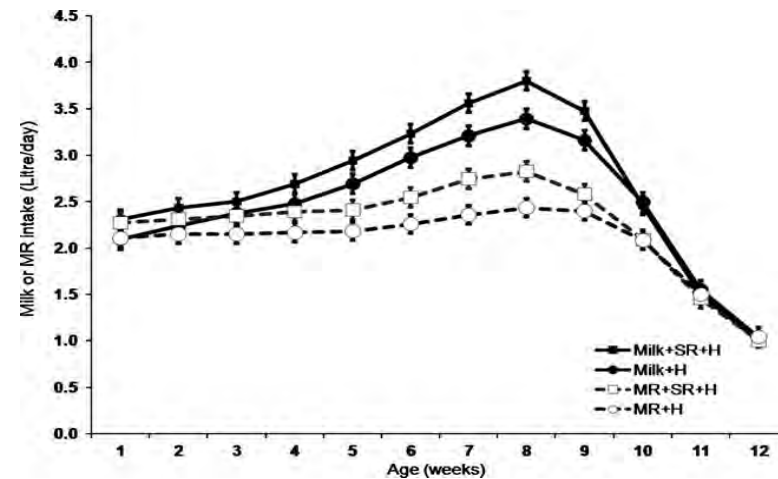


Table 3 Growth performance and feeding cost of Sahiwal calves offered different dietary treatments during preweaning

Intake	Milk vs. MR		SR+H vs. hay		F1	F2	F1.F2
	Milk	MR	SR+H	Hay			
Birth weights (kg)	21.5±0.6	22.6±0.6	20.6±0.6	0.9	0.01	0.75	
Weaning weights (kg)	51.6±0.8	35.2±0.8	48.7±0.8	38.1±0.8	0.0001	0.0001	0.66
Total weight gain (kg)	30.0±0.8	13.6±0.8	26.1±0.8	17.5±0.8	0.0001	0.0001	0.66
Daily growth rate (g/day)	357±9	162±9	311±9	208±9	0.0001	0.0001	0.67
Total feeding cost (PKR)	6,935±32	3,842±32	5,878±32	4,898±32	0.0001	0.0001	0.44
Feed cost/kg live-weight gain (PKR)	236±15	323±15	232±15	327±15	0.0005	0.0005	0.005

F1 milk vs. MR, F2 SR+H vs. hay, H hay, MR milk replacer, SR starter ration. The total feeding cost is calculated from the feed costs: whole milk PKR 30 per liter, milk replacer PKR 19 per liter, starter ration PKR 20 and hay PKR 7 per kg. (1 US\$. PKR 85)

Discussion

The calves grew faster and had higher live-weight when fed milk compared with MR. This was because of higher intake of total nutrients in calves offered milk than MR. The intake of milk or MR was not influenced ($P > 0.05$) in calves fed SR+H or H only. However, it was numerically higher in calves on SR+H than on H only (210±1.7 vs. 192±1.7). The calves offered SR had better live weight gain than those offered hay only. This is because of increased supply of nutrient to calves offered SR than H only.

Calves were offered starter ration from the second week of their age. The SR consumption was negligible (<200 g/day) until the sixth week of their age in all calves and then increased gradually (Fig. 3). Similar results are reported by Khan et al. (2011a) in Holstein calves given higher quantities of milk (approximately 20% of body weight of calves). The intake of hay and starter ration in their study was negligible (<200 g/day) until the fifth week of the age of calves and then increased gradually.

In the present study, the intake of starter ration was not affected by either milk or MR until the ninth week of their age (Fig. 3). However, from the tenth week, the SR intake was higher in calves fed milk than on MR. In the 12th week, the SR intakes were 693 and 943 g/day in calves fed MR and milk, respectively. This was contrary to our initial assumption that calves on MR may consume less SR than milk-fed calves during the initial weeks of their age but will consume more and thus will be equal in performance to the milk-fed calves in the later age. The MR fed calves could not catch up their growth and were not at par with other groups even during later weeks of their age. The tendency to consume more starter ration in calves on milk may be due to the more developed rumen as a result of greater supply of energy and nutrients from milk (Shen et al. (2004) reviewed by Khan et al. (2011b)) compared with

Fig.2 Weekly weight (kg) of Sahiwal calves offered milk plus starterration (SR) and hay (H), milk plus H, milk replacer (MR) plus SR and H or MR plus H during preweaning

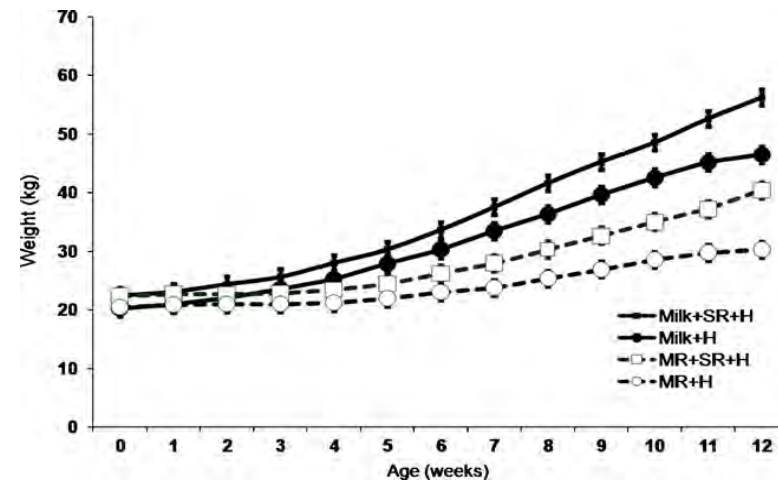
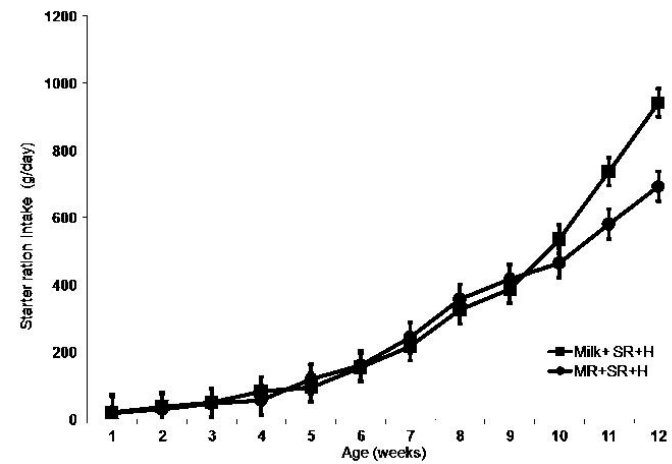


Fig.3 Intake (g/day) of starterration in Sahiwal calves offered milk plus starterration (SR) and hay (H) or milk replacer (MR) plus SR and H during preweaning

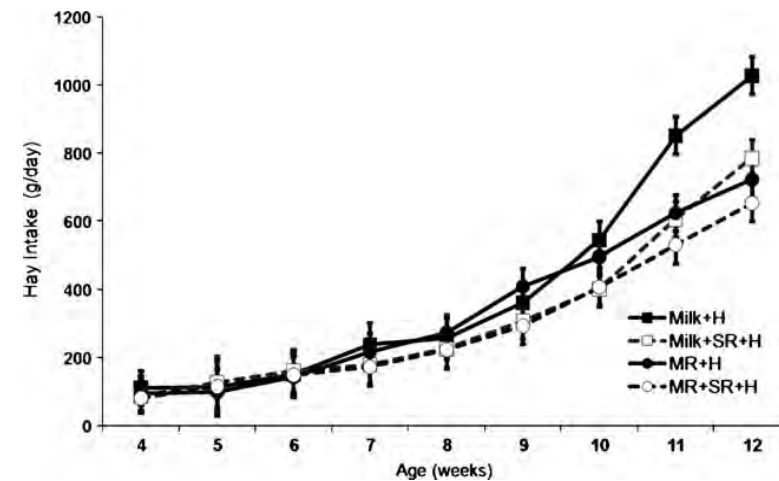


those on milk replacer. Ad libitum feeding of milk discourages solid feed intake whereas limited milk feeding encourages solid feed intake by calves (Khan et al., 2007; Khan et al., 2011b). In the present study, the calves were offered limited quantity (10% of their body weight) of milk rather than ad libitum. The calves offered higher quantity of milk (15.20% of their body weight) per day showed lower solid feed intake (Khan et al., 2007). This may have been the second reason of higher intake of SR by calves fed milk than milk replacer. Higher incidence of calf scour in MR fed calves may be another reason for their poor performance and less SR intake than milk fed calves.

The calves were offered SR and hay in the second week of their age but they started nibbling or eating SR or hay in the fourth week. They started eating a reasonable quantity of SR or hay in the ninth week. It seems that the Sahiwal calves with a lower birth weight than Holstein-Friesian calves remained mainly dependent on milk or milk replacer than on solid feed until the eighth week of age. Weaning of Sahiwal calves at an earlier age (eighth week) may affect their subsequent growth rate. However, a separate study is needed to draw a final conclusion. Calves of temperate dairy breeds like Holstein-Friesian can be weaned at the age of day 28 (Quigley et al., 2006). By that time, they start consuming solid feed to support their growth. However, Sweeney et al. (2010) reported that Holstein calves weaned at an earlier age (day 19) had poorer gain than later weaned calves. Results from our study suggest that Sahiwal calves, fed milk at the rate of 10% of their body weight, may show poorer weight gains post weaning if weaned earlier than day 56 than those at the age of day 84. On a high milk feeding plan, weaning

age of Sahiwal calves may possibly be reduced without compromising postweaning weight gain; however, further research is required to be carried out to determine if this is accurate.

Fig.4 Intake (g/day) of hay in Sahiwal calves offered milk plus starter ration (SR) and hay (H), milk plus H, milk replacer (MR) plus SR and H or MR plus H during preweaning



Growth rate of calves offered MR was lower than for those offered milk. This was because of a lower intake of nutrients in calves offered MR. The reconstituted MR was offered to calves at equal volumes with milk rather than on an isocaloric or equal protein basis. Ideally, the reconstituted milk should have had similar nutrient densities to make a comparison. However, in a recent study with Holstein calves weaned at day 49, Lee et al. (2009) have reported that the weaning weights were lower in MR-fed calves than on whole milk (64 compared to 72.2 kg, respectively), despite similar DM intake and gross composition of both MR and whole milk.

They described that better weaning weight of calves on whole milk was probably because of the better bioavailability of nutrients and some unknown growth factors from the whole milk. Increasing fat in milk or milk replacer increased body weight gain during the first month of the life of calves (Jaster et al., 1990) but did not increase the lean tissue gain (Hill et al., 2008). At a similar energy intake, increasing protein content of MR resulted in increased average daily gain in male Holstein calves (Blome et al., 2003). Lee et al. (2008) has reported no difference in the performance of Holstein calves fed varying levels of protein in energy in the MR-fed Holstein calves. However, in the first study, the general appearance and overall performance of calves was poor in all groups compared with reported figures with milk in the literature.

Hill et al. (2011) have reported that supplementation of MR with a blend of butyrate, medium chain fatty acids and linolenic acid reduced the incidence of scour and clostridium sickness in young Holstein calves.

Low fat MR are reported to promote higher starter DM than high fat MR during preweaning period (Kuehn et al., 1994; Cowles et al., 2006). However, results of the present study are contrary to these findings. In the present study, weaning weight, SR and hay intake were higher in calves fed milk than MR. Weaning weight of Sahiwal calves at the Livestock Production Research Institute, Bahadurnagar, Okara has been reported as 45 kg (Anonymous, 1989). This weaning weight is less than that obtained in the present study offered whole milk (Table 3). Higher weaning weights in the present study were probably due to higher milk consumption and improved housing facilities such as raised calf pens and a clean environment. The weaning weights of Holstein calves most often are higher than the weaning weights of Sahiwal calves in this study. Khan et al. (2007) have reported weaning weight of female Holstein calves as 65 kg at 50 days of age. The higher weaning weight of Holstein calves in their study, was due to their higher birth weights (44.6 ± 1.3 kg) and greater milk consumption (4.63 l/day) than Sahiwal calves (21.4 ± 0.9 kg and 2.6 ± 0.05 l/day, respectively).

There were higher numbers of scour days in the calves fed MR than milk. This may be a reason in the lower performance of calves fed MR than milk. These results are contrary to the work of Lee et al. (2008) who reported no difference in scour days in Holstein calves fed either milk or MR. The reason for higher incidence of diarrhea

in MR fed calves, in the present study, could not be established.

Total feeding cost (including both liquid and solid feed) was higher ($P < 0.05$) in milk-fed calves than on MR (Table 3). This was due to higher price and intake of milk than MR by the calves. The total feeding cost was highest in calves fed milk plus SR and lowest in calves on MR plus hay only. The total feeding cost of calves on milk plus hay and MR plus SR were not different. The cost to gain 1 kg live weight in calves had an opposite response and was higher in MR fed calves than on milk. Although calves on milk plus SR had the highest total feeding cost, they required the lowest money to gain 1 kg of live weight gain.

This was despite the fact that the cost of veterinary medicine used for treatment of scours was not included in the cost calculations of MR fed calves. During the preweaning period, the total feeding cost of MR fed calves was about 45% less than that of the total feeding cost of milk fed calves. However, the cost to gain 1 kg live weight gain was 37% higher in MR fed calves than on milk. The calves on MR and hay only had about 53% less total feeding cost than on calves fed milk plus SR but required about 78% more money to gain 1 kg of live weight. Thus, in the present study, feeding MR to Sahiwal calves did not seem economical compared with whole milk feeding.

Conclusions

Milk-fed Sahiwal calves grew better and were healthier than those on MR during preweaning period. Milk replacer did not support growth as well as whole milk fed calves at equal volumes. Feeding milk and starter ration was more economical than milk replacer for having accelerated growth in Sahiwal calves. The addition of calf starter ration to the preweaning feeding regimen was beneficial in helping the animal to grow at a faster and more economical rate. Under traditional preweaning feeding plan, weaning of Sahiwal calves at less than 56 days of age may not be possible without compromising on their growth rate. However, further studies are required to draw a final conclusion.

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MODIFICATION OF LACTATION YIELD ESTIMATES FOR IMPROVED SELECTION OUTCOMES IN DEVELOPING DAIRY SECTORS

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SUMMARY

Animals are extremely important to the livelihood of smallholder dairy farmers in developing countries. However, due to limited resources, investment in long-term genetic improvement is rare and herd recording is minimal. Therefore, obtaining adequate performance records for genetic evaluation and selection is difficult and hence it is important to optimize these selection outcomes from any data that are collected. The aim of this study is to determine a robust and efficient method for estimating lactation yield for low-producing dairy cattle and their subsequent genetic evaluation. Using Sahiwal cattle as an example, simulated datasets, based on lactation data from Pakistan, were used to compare different methods of lactation yield estimation (i.e., test-interval method, and three nonlinear models). Furthermore, these estimates were analysed to explore their implications on the subsequent estimated breeding value (EBV) ranking and selection outcomes. Utilising these results, different test-day sampling schedules were compared to investigate possible recording regimes involving few records that can accurately estimate lactation yield without significantly affecting selection. Results indicate that the lactation models proposed by Wood (1967) and Wilmink (1987) yield similar selection outcomes to the recommended test-interval method. These results provide opportunities for further research into test-day scheduling which could reduce the number of records required and have considerable implications on progeny testing systems of low-producing dairy cattle and developing dairy sectors.

INTRODUCTION

Breed improvement and selection in developing dairy systems can be challenging as field conditions are generally constrained by a lack of infrastructure for regular test-day recording. For this reason, regular twice daily recording of milk yield for entire lactations is not feasible (Khan et al. 2008). The limited resources and data exacerbate the need to utilize each record efficiently to maximize their contribution to the evaluation process (Bajwa et al. 2002). Therefore, for any developing dairy sector there is a need to develop genetic evaluation systems which optimize selection outcomes given the current resources for test-day recording.

There are numerous methods for genetically evaluating milk production based on test-day records. In developed nations complex methods such as test-day models are commonly used. These models require accurate estimates of genetic and phenotypic parameters based on many daily milk yields from large populations of animals which are unlikely to be available in a developing dairy sector (Ilatsis et al. 2007). For this reason, simple methods such as a two-step approach can be used. This is where test-day records are first used to estimate lactation yield and then these values are used as the phenotype for genetic evaluation. Methods of lactation yield estimation from test-day records are well researched. In a developing country scenario, the Test-Interval Method [TIM] (Sargent et al. 1968) is recommended by the International Committee for Animal Recording (ICAR 2009). Other approaches involve fitting a mathematical model to lactation data and using the model output to estimate yield. Many models have been proposed for describing the lactation curve of dairy animals (Dongre et al. 2011). A handful of studies have investigated the ability of lactation curve models to depict Sahiwal cattle lactation data. Kolte et al.

(1986) found that the inverse polynomial function proposed by Nelder (1966) was the superior fitting model, followed by the gamma function proposed by Wood (1967). Contrary to this, Rao and Sundaresan (1979),

reported that Wood's (1967) function was the most appropriate. The Wood (1967) model is one of the most widely accepted lactation models and is commonly used in research (Swalve 2000). Similarly, the Wilmink (1987) model is frequently used within test-day evaluations to model the lactation curve of dairy cattle (Naranchuluu *et al.* 2011).

This current study is concerned with Sahiwal cattle in Pakistan and will focus on how different lactation models behave when fitted to the lactation characteristics of this particular breed. Specifically, this study aims firstly to determine which lactation model is the most robust at modelling the lactation curve of Sahiwal cattle at different test-day recording schedules. The second aim is to discuss what implications this may have on the future of test-day sampling in Pakistan and how it can be used to improve their current progeny testing program.

METHODS AND MATERIALS

Lactation Estimation Models and Methods. The lactation estimation methods used within this study were:

1. The test-interval method (TIM) described in Sargent *et al.* (1968) which is based on an approximation of the area under a curve
2. The inverse polynomial model proposed by Nelder (1966): $\text{yield}_i = \frac{1}{a + b * \text{dim}_i + c * \text{dim}_i^2}$
3. The gamma function proposed by Wood (1967) $\text{yield}_i = a * \text{dim}_i^b * e^{-c * \text{dim}_i}$
4. The lactation model proposed by Wilmink (1987): $\text{yield}_i = a + b * \text{dim}_i + c * e^{-0.05 * \text{dim}_i}$

where a , b and c are different parameters to be estimated separately with each model and dim_i are the days in milk ($i=1, \dots, 280$) for a lactation length of 280 days.

Data. Weekly test-day Sahiwal lactation records from 839 lactations from 464 dams, collected during 2005-2010 from the Livestock Production Research Institute (LPR I), Bahadurnagar Okara, were used as the basis for data simulation in this study. Data were simulated using three different lactation models (Wood, Wilmink and Nelder). Variance and covariance matrices of the parameters (a , b and c) and a residual variance of each of these models was determined based on the raw Pakistan data. Using these variance structures and the pedigree relationship matrix (A), phenotypic lactation yields were simulated for entire lactations for all the dams in the population. This was repeated 100 times for each of the simulation models to yield three batches of one hundred datasets for comparison. Data were simulated using three different lactation models because it allows for a more thorough comparison of lactation yield estimation methods as it gives an indication of their robustness across different lactation curve shapes.

Model Comparison. Four lactation yield estimation methods were used to fit and calculate the lactation yield for every dam for each set of simulation data. These included the recommended TIM as well as three lactation models, Wood, Wilmink and Nelder, fitted and estimated using the nonlinear mixed effects (NLME) model function in R Version 2.13.0 (R Development Core Team 2008) following a similar process outlined by Raadsma *et al.* (2009). This was carried out for four different test-day scheduling regimes (weekly, monthly, five test-days; random selection and five test-days; stratified selection). For each method, the percentage of models which successfully converged was recorded as well as the lactation yield estimates. The lactation yield estimates were compared with the true simulated lactation yield and summed to calculate a mean square error (MSE) of estimation for each simulated dataset. The MSE was then used to directly compare between the lactation yield estimation methods. Lastly, the lactation yield estimates for each simulated data set were used to calculate estimated breeding values (EBVs) for each of the animals in the dataset using ASReml-R Discovery Edition 1.0 (Butler *et al.* 2009). The output of this analysis allowed further comparison between models to determine if the lactation yield estimation method had any effect on the ranking and subsequent selection of animals.

RESULTS AND DISCUSSION

The robustness of each of the lactation models for fitting Sahiwal test-day data can be determined by comparing the percentage of success rates of each model's ability to be fitted to the different simulated datasets (Table 1). These results show that overall the Wood model is superior to the Wilmink and Nelder models as it generally has

higherratesofsuccess,mostimportantlywhenfittingdatafrombotharandomandstratifiedselectionoffive test-day records. This has an important practical implication, as in the field conditions of Pakistan, test-day recording is likely to be irregular and infrequent.

Table1. Percentage of lactation yield estimation models that were successfully fitted to each set of simulated lactation data at each of the four different test-day recording regimes (weekly, monthly, 5 test-days: random sample and 5 test-days: stratified sample).

Data Simulation Model	Fitted Model	Test-Day Recording Regime			
		Weekly	Monthly	Random	Stratified
Wood	Wood	100	100	82	92
	Wilmink	100	100	76	83
	Nelder	100	100	70	72
Wilmink	Wood	100	100	88	86
	Wilmink	100	100	74	83
	Nelder	100	100	78	75
Nelder	Wood	97	98	60	67
	Wilmink	94	100	69	82
	Nelder	75	71	82	83

Using the MSE values from each of the lactation yield estimation methods we can directly compare between models for the same simulated lactation. The average MSE values across lactation yield estimates can be seen in Table 2. These are represented for only two of the data simulation methods (Wood and Wilmink). The results from the Nelder simulated data are not reported here as the number of failed models caused unreliable values. From Table 2 the MSE values show that the Wilmink and Wood models were superior to the TIM and Nelder methods. Furthermore, the Wilmink model has a lower average MSE than the Wood model in both sets of simulated data (5,124,550 vs 5,327,934 for the Wilmink simulated data and 5,234,436 vs 5,235,715 for the Wood simulated data). This suggests that the Wilmink model is superior to the Wood model in its ability to accurately estimate lactation yield on different types of lactation data.

Despite the differences in the MSE seen in Table 2, the important outcome of this analysis relates to the animals in the top proportion of the population that would be selected for breeding and how they compare with the true (simulated) superior animals. For the different methods of lactation yield estimation, using the Wood simulated data, the average number of corresponding animals with the true top fifty superior animals were; TIM 39.2±2.22, Wood 39.7±2.27 and Wilmink 39.6±2.28. For the Wilmink simulated datasets, the results were very similar; TIM 36.8±2.33, Wood 37.1±2.38 and Wilmink 37.1±2.41. The results show that the average number of corresponding animals with the true top fifty were all within one animal of the other estimation

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methods. This suggests that these methods of estimating lactation yield, for a given test-day scheduling regime, are each capable of selecting the superior animals from a given population.

Table2: Average Means Squared Error values (±st.dev) for four different methods of lactation estimation (TIM, Nelder, Wilmink and Wood) when calculated using monthly records from two methods of data simulation (the Wilmink and Wood models)

Model used for lactation yield estimate	Model used for data simulation			
	Wilmink		Wood	
	Average MSE (±sd)		Average MSE (±sd)	
TIM	6,102,273	(±385,457.5)	6,143,607	(±377,174.1)
Nelder	5,962,774	(±413,556.1)	5,696,461	(±387,866.5)
Wilmink	5,124,550	(±311,253.8)	5,234,436	(±347,630.8)
Wood	5,327,934	(±342,708.3)	5,235,715	(±368,825.4)

CONCLUSIONS

The benefit of modelling test-day yields is the ability to subsequently estimate lactation yield on fewer records. This then provides an opportunity to record more animals fewer times which will help to improve the accuracy of evaluations as well as increase the population of animals from which selection can take place. The outcomes of this study show that although the Nel model is capable of fitting and modelling low producing dairy cattle like the Sahiwal, it is unreliable with different lactation curves and test-day sampling regimes. The results from the other lactation model tested, the Wood or Wilmink, show that they are both robust in different scenarios with the Wood model better fitting irregular and infrequent test-day recording regimes. Despite this, both the Wood and Wilmink models provide an opportunity to further investigate their use in estimating lactation yield in Sahiwal cattle and the possibility of reducing the number of required test-day records whilst maintaining the accuracy of selection outcomes.

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Factors affecting milk price under smallholder dairy production system in Pakistan

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Abstract

The smallholder dairy farmers all over the world are facing diverse problems to make their businesses profitable. The production cost is growing with the increasing rate of input costs, while the milk prices are not equally supporting them. Due to these reasons small-holders are converting their dairy farms to other profitable businesses. The purpose of this study was to better understand the factors that affect milk price for the smallholder dairy producer to help them derive more profit from their businesses. The major factors which have a direct influence on market rates of milk include geographic location of the farm, nature of business, literacy of farmer, season, circumstances of the milk buyer, terms and conditions for selling milk, and nature of primary business.

A baseline survey data was collected during 2011 from registered farmers (n=523) in five districts of Punjab, Pakistan. Data were analysed using uni-variate and multivariate analysis. The results show that education level (p=0.029), geographic area (p<0.001), season (p<0.001) and milk buyer (p<0.001) affect significantly the milk price, whereas the other factors like nature of the contract and primary business did not affect the milk price significantly. We conclude that if smallholder dairy farmers are educated about the factors and circumstances which influence milk pricing, they can then derive greater profits from their produce.

Keywords: Milk price, contract, season, education, geography

Introduction

Milk price is a major issue that affects profitability of dairy farmers and must not be ignored. The smallholder dairy farmers in developing countries like Pakistan are facing problems to understand the cost of production and market prices for their livestock and dairy enterprises. If dairy farmers can better understand those factors that affect milk price and find options to better market their milk, they absolutely be able to gain a better price for what they produce.

In Pakistan, within the livestock sector, milk is the single most important commodity with a value that exceeds the combined value of wheat, rice, maize and sugarcane (Economic Survey of Pakistan, 2013-14). Most of the milk volume is produced by more than 8.5 million smallholder farmers owning less than 10 animals kept on small areas of one to five hectares (Khan et al, 2010). Milk acts as an exclusive source of animal protein thereby improving household nutrition as well as providing money to purchase goods for household consumption, paying the utility bills and for the education of children. Research on diverse factors that affect milk price are generally limited to simple comparisons which provide little information that can be used to help farmers make better decisions. The smallholder dairy producers are faced with daunting challenges in the areas of infrastructure, financial security, quality assurance, price regulation, trained manpower, and seasonality (Umme Zia, 2007). Pakistan is gifted with outstanding dairy breeds of buffaloes (Nili-Ravi & Kundhi) and cattle (Sahiwal, Cholistani & Red Sindhi). The consumer's preference for buffalo milk as compared to cattle milk is based on taste and its higher butterfat percentage. The total volume of buffalo and cattle milk produced during 2013-14 was 31,252,000 & 18,027,000 tonnes, of which 25,001,000 & 14,421,000 tonnes respectively were consumed by the human population (Economic survey of Pakistan, 2013-14).

The base prices of milk in Pakistan are set by the government as well as private agencies but there are many other players who have a vibrant role in deciding milk prices. There is also seasonal variation in supply and demand that affects milk prices. Profits vary across dairy farms, and various studies have investigated factors that affect milk prices (Moreki J.C., 2013, Jonas H., 2012, Sikawa et al., 2011, Omoro et al., 2009, Lucy S.K., 2009, Khan et al., 2008 & Takashi K., 1997). These studies have concluded that factors like location, education level, season, milk contracts and primary farming activity have a significant effect on the pricing of milk. However, there has been very little research carried out in Pakistan on this major area. Therefore, the main objective of the present study was to determine the most critical factors that significantly affect milk price under small-holder dairy production system in Pakistan. The results and conclusions from this study will provide farmers with a better understanding of the decision-making processes that will lead to more profitable dairy businesses.

Materials & Methods

Data collection

The Australia-Pakistan Agriculture Sector Linkages Program (ASLP) dairy project has been working with small-holder dairy farmers since 2007 to enhance their earning through improved extension activities and market linkages. A questionnaire was developed to obtain data on location, level of education, season, milk marketing channels, milk selling contract and primary farming activities. A copy of the questionnaire can be found as Appendix 1. The small-holder dairy farmers (n=523) registered with the project were interviewed to study different factors affecting milk price during 2011. The wide geographic distribution of farmers is representative of the predominance of small holder farmers of Pakistan.

Statistical analysis

Data on both buffalo and cow milk were analysed statistically using uni-variate and multivariate analyses to determine the effect of each factor on milk price. Data were log transformed to normalise variance and factors were analysed independently in a uni-variate analysis using GenStat (VSN International, London).

Results

To assess the factors affecting milk price in buffaloes and cows, the analysis was conducted in two stages. Firstly, the effect of each factor on price was considered in isolation, in separate univariate analyses, although species (cows vs. buffalo) and the interaction with species was fitted using a linear mixed model. Farm number was added to the model as a random effect to account for any clustering. Secondly, all these factors were considered simultaneously in a multivariate model. As well as the interaction of species with each of the factors, an interaction term was included between geography and season, to allow for district-specific effects. Non-significant terms were removed from the model using a backward elimination process. In all models, because of the non-normality and unstable variance, the milk price data were log-transformed. Model-based means were back-transformed, with approximate standard errors calculated.

Univariate analysis: The table 1 shows the univariate associations, as well as individual interactions with species (Sp: Buffalo & Cow). The factors like a defined contract among the seller & buyer, contract free, geography, season and whom the milk sell to, are significantly related to price (all $P < 0.05$), with those terms with a significant interaction with species indicating different effects on cows and buffaloes. In addition, the education level of farmer tended to be associated with farm profitability ($P = 0.080$) and species differences in milk price due to the effect of primary farming activity ($P = 0.095$).

Multivariable analysis: When putting the variables education, geography, season and sell to in a combined multivariable model, all variables with the exception of education ($P = 0.822$) were significant. Then considering interactions among stall variables, only that between geography and season was significant ($P = 0.006$).

The final form of model fitted to the data was;
 $\log_e \text{Price} = \text{constant} + \text{Education} + \text{Species} + \text{Geography} + \text{Species} + \text{Season} + \text{Species} +$
 $\text{Geography} + \text{Season} + \text{Sell To} + \text{Farm No} + ?$

Geography×Season: There was a general trend of higher prices in summer compared with winter, but this varied with location. Milk prices were significantly higher for buffalo milk in both winter and summer in Kasur ($P < 0.001$), and Pakpattan ($P = 0.029$), but not significantly different for Okara, Jehlum and Bhakkar (all $P > 0.8$). The highest prices were obtained in Jehlum, in both summer and winter, and lowest prices obtained in Okara in winter and Pakpattan in summer irrespective of whether the milk was collected from buffalo or cows.

Discussion

The results obtained from this study revealed that species, location of farms, season and the type of marketing system affect milk prices significantly ($P < 0.001$). It is clear that the price in Jehlum district remained highest as compared to others and the reason may probably be that this location is near the big cities where the price obtained in Pakpattan district remained lowest due to the relative remoteness of this area from the target commercial milk outlets. The results are in accordance with the report of Dairy Co (2012) which shows that location and farm size will have an impact on the type of contracts offered for individual farmers. Similarly, the results of a study conducted by Sikawa et al. (2011) showed that education is a vital factor that assists farmers to make better informed decisions like linking with associations that attract better prices. This is consistent with the present results which show that education has a significant effect on the price of milk. Similarly, the results of another study conducted by Meral et al. (2012) on factors influencing packed and unpacked milk consumption of consumers, age, education level, professional status, marital status, household size, income, the type of commercial outlet, milk preferences, and milk price also relates with the results of the current study. Therefore, education of the smallholder dairy farmers might bring a change to their understanding of these factors resulting in higher milk prices for all small-holder dairy farmers in Pakistan.

The retailers appear to dominate marketing chains and exploit their suppliers by offering low prices and binding contracts as well as keeping additional returns for themselves when prices are seasonally high due to lower milk supply (Jonas H., 2012). These findings are closely related to the results of the current study that show if there is a monopoly exploited by middlemen the milk price is lowered. Similarly, this season has higher influence as prices rise in summer and decrease in winter.

The results from the current study also show that when farmers sell milk directly to consumers and not to middlemen they have a better chance to attract a higher milk price similar to that reported by Omoro et al. (2009). This is also directly proportional to the distance of the farm from the consumer market with higher prices being attracted from milk transported longer distances to the market place also reported by Stephen et al. (2009) and Lucy S. K. (2009). The results of this survey study indicated that where there is a defined contract among the seller and buyer, the milk price is more and vice versa which is comparable to the findings of the report published by Dairy Co (2012), which showed that the milk buyers prefer to collect milk from those farms which are located in their collection area and also have easy contract conditions.

Conclusion:

It is concluded that there are many factors that have a direct or indirect effect on milk price under small-holder dairy farming system in Pakistan. The farmers should be well aware of these factors to increase the profitability of their dairy farms. There is a need to explore other factors that can affect milk selling price. In this regard, awareness about these factors is necessary to educate farmers through implementation of effective extension strategies.

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Table 1: This shows the uni-variate associations, as well as individual interactions with species (spp. Buffalo & Cow).

Predictor			P-value	Group	Buffalo	Cow
		Mean	SE	Mean	SE	
Contract_Free×Sp	0.043	Contracted	34.40	0.55	34.88	0.38
Contract_Free	0.797	Free arrangement	31.06	0.65	30.17	0.42
Education×Sp	0.275	None	33.58	0.74	28.33	0.93
Education	0.080	Primary+Middle	34.40	0.41	29.84	0.45
		Matric	35.66	0.53	31.79	0.60
		Intermediate	35.41	1.17	30.39	1.40
		University	34.61	1.18	31.56	1.61
Geography×Spp.	<0.001	Bhakkar	34.92	0.59	30.17	0.60
		Jehlum	39.33	0.71	34.33	0.79
		Kasur	36.49	0.66	30.94	0.74
		Okara	33.48	0.44	28.62	0.57
		Pakpattan	30.54	0.61	28.70	0.69
Primary Farming						
Activity×Spp.	0.095	Cropping	34.99	0.42	29.99	0.45
Primary Farming						
Activity 0.846		Dairying	34.33	0.45	30.60	0.49

Cropping+Dairying			34.74	1.11	31.37	1.44
		Other	35.73	1.43	31.06	1.77
Season×Spp.	0.027	Winter	33.75	0.30	29.90	0.33
Season <0.001		Summer	35.69	0.32	30.85	0.34
SellTo×Spp.	0.497	Bigmilkcompany	35.48	1.21	30.85	1.14
SellTo <0.001		Dodhi	33.31	0.30	29.37	0.35
		Neighbour/Villager	39.77	0.72	34.16	0.82
		Other	32.72	1.96	30.88	2.41
No.ofMilking						
Animals×Spp.*	0.953		0.0002	0.0039	0.0008	0.0097
No.ofMilking						
Animals0.942						
No.ofdodhi's×Sp*	0.115		-0.0009	0.0013	0.0018	0.0020
NDodhis	0.655					

* For the quantitative predictors, values shown are regression coefficient (and standard errors) on logarithmic scale.

Here are some comments on significant or suggestive associations.

- " Defined Contract Free: When the farmers made a Contract with the buyer than they can get higher prices than Free Arrangement, but the difference is greater in Cow than Buffalo.
- " Education: The level of education that farmers achieve in their career has a little effect on price but there is a significant difference among species.
- " Geography: range from 30.54Rs/L (Pakpatan) to 39.33Rs/L (Jhelum) for buffalo milk, and 28.62Rs/L (Okara) to 34.33Rs/L (Jhelum).
- " Primary Farming Activity: Combined Dairying Cropping is associated with increased prices in both species; 'Other' is highest in Buffalo, but difficult to interpret, not knowing what this category contains.
- " Season: Higher prices in summer than winter, but a greater differential for buffalos.
- " Sell To (Buyer): Neighbour/villager provided highest prices in both species (Buffalo: 39.77Rs/L; Cow: 34.16Rs/L).

Table 2: The level of significance for the final multivariate model, as well as model-based means, are shown in table below:

Predictor			P-value	Group	Buffalo	Cow
		Mean	SE	Mean	SE	
Education×Species	0.087	None	36.27	1.05	30.54	1.25
		Primary+Middle	35.80	0.72	32.10	0.71
		Matric	34.95	0.87	29.20	1.08
		Intermediate	35.13	0.63	30.66	0.58
		University	35.27	1.13	32.66	1.47
Geography×Species	<0.001	Bhakkar	35.02	0.74	30.14	0.75
		Jhelum	39.73	0.95	34.40	0.96
		Kasur	37.30	0.78	31.72	0.89
		Okara	33.92	0.68	28.90	0.72

		Pakpatan	31.91	0.77	30.14	0.81
Season×Species	0.020	Winter	34.50	0.59	30.54	0.61
		Summer	36.49	0.62	31.50	0.63
SellTo <0.001		Bigmilkcompany	36.56	1.07	31.94	0.98
		Dodhi	33.72	0.36	29.49	0.44
		Neighbour/villager	39.02	0.67	34.12	0.69
		Other	32.92	1.73	28.76	1.53

Predictor			P-value	Group	Winter	Summer
			Mean	SE	Mean	SE
Geography×Season	<0.001	Bhakkar	32.46	0.71	32.52	0.72
		Jehlum	36.93	0.92	37.08	0.93
		Kasur	32.72	0.75	36.16	0.83
		Okara	30.17	0.63	32.52	0.68
		Pakpatan	30.48	0.76	31.56	0.79

Appendix 1: Pakistan Small-holder dairy production systems longitudinal survey

- a) When you sell your milk, what price do you generally receive in summer and winter?
(Please only fill in the relevant rows and the milk quality specifications, e.g. fat% if there are any)

Summer	Rs/kg	Quality Specifications:
Buffalo		
Cow		
Buffalo/cowmix		

Winter	Rs/kg	Quality Specifications:
Buffalo		
Cow		
Buffalo/cowmix		

- b) How much milk do you generally sell and consume each day in...

Consume (kg/day)	Sell (kg/day)
Winter	
Summer	

- c) To whom do you currently sell milk?

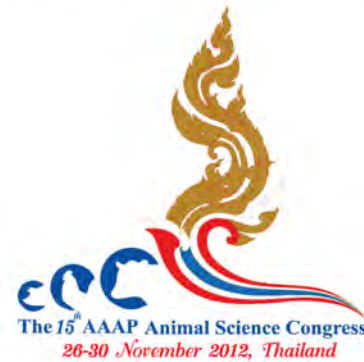
Neighbor or villager	Dodhi	Big milk company	Other
Free arrangement	Contract	Free arrangement	Contract
Free arrangement	Contract	Free arrangement	Contract



**Improving Smallholder and Industrial Livestock Production
for Enhancing Food Security, Environment and Human Welfare**

**Proceedings
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The 15th AAAP Animal Science Congress



Developing a Feed Calendar for Pakistan's Small-Holder Dairy Farmers

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Providing adequate year-round nutrition to Pakistan's small-holder dairy cattle and buffalo poses a major challenge to improving animal productivity and profitability. In response, a region-specific feed calendar has been developed by a Nutrition Focus Group (NFG) formed as part of an Australian Centre for International Agricultural Research (ACIAR) research project titled „Strengthening dairy value chains in Pakistan through improved farm management and more-effective extension services. (ACIAR LPS/2010/007). The aim of the calendar is to promote „best practice. in relation to growing and harvesting forages with an emphasis on combining self-explanatory photos and images with descriptive text in an easily understood format. Detailed notes have also been compiled to enable extension workers to adequately train farmers in the use of the calendar. Information will be provided specifically for buffalo and cattle requirements with respect to winter and summer environments under rain fed and irrigated conditions. The availability and feeding of concentrates and by-products will also be incorporated. Members of the NFG are drawn from university and government research institutions across Pakistan. It is envisaged the calendar will provide a valuable resource for small-holder farmers as the format will be able to be adapted to a range of environments within Pakistan as well as other developing countries.

Key Words: Buffalo, Cow, Forage, Concentrate, Byproducts, Rainfed, Irrigated

INTRODUCTION

Providing adequate year-round nutrition to meet animal requirements poses a major challenge to improving animal productivity and profitability in developing countries. The Pakistan small-holder dairy cattle and buffalo industry is no exception. With an average of 5 litres of milk produced per cow per day across lactation (Warriach et al., 2012), severe underfeeding is a major contributor. In addition to the lack of quantity provided, very poor quality (nutritional value and digestibility) is also a feature. An important explanation to this underfeeding problem lies in the difficulty in producing forage in vastly different winter and summer environments which support specific species. Winter can be very cold (<0°C) whereas summer is extremely hot (>45°C) (Pakistan Meteorological Department, 2012). Small-holder farmer knowledge of the agronomic practices required to grow and harvest forages as well as animal requirements at different stages of the production cycle are also limited and a contributor to underfeeding. In an bid to improve the provision of nutrition for dairy animals within Pakistan, a region-specific feed calendar was developed by a Nutrition Focus Group (NFG) formed as part of an Australian Centre for International Agricultural Research (ACIAR) research project titled „Strengthening dairy value chains in Pakistan through improved farm management and more-effective extension services. (ACIAR LPS/2010/007). The aim of the calendar is to promote „best practice. in relation to growing and harvesting forages with an emphasis on combining self-explanatory photos and images with descriptive text in an easily understood format. This paper reports on the development and distribution of this feed calendar for Pakistan's small-holder dairy farmers.

MATERIALS AND METHODS

Formation of Nutrition Focus Group (NFG)

In order to access relevant information relating to the establishment, growth and utilisation of forages across Pakistan, a „think-tank“ of key researchers was assembled to form the Nutrition Focus Group (NFG). Members

from university and government institutions with experience in forage growing and feeding larger ruminants were recruited to the group based on their reputation for achieving solutions through applied research. Regular meetings were conducted to prioritise research initiatives aimed at improving the feeding of small-holder dairy animals as well as to undertake major group activities such as the development of the feed calendar.

Development of the feed calendar

Calendar content

Initial meetings of the NFG detailed the feeding management needs of small-holder dairy producers in Pakistan and determined the content to be included in the calendar. It was agreed the information be presented on a monthly basis and be divided into five broad areas, with consideration given to forage species, time of year (winter rabi season and summer kharif season) and locality within Pakistan. The subject areas were:

- Agronomic practices for each forage species . forage plot preparation and management (includes seed quality, sowing, fertilization, harvesting and irrigation)
- Yield at harvest for each forage species (note: some species are multi-cut).
- Animal requirements for different production stages (includes dry/transition as well as early, peak and late lactation for cattle and buffalo).
- Husbandry and management practices aimed at improving overall productivity (includes forage conservation, provision of water to animals, feeding concentrates and reproductive management).
- The availability and feeding of concentrates and by-products.

Collection of information

Each member of the NFG was assigned a subject area to collect information. Sources of information included published previous research relevant to Pakistan's four provinces (Punjab, Sindh, Balochistan and Khyber-Pakhtunkhwa) as well as reports and other available technical notes. An emphasis was placed on sourcing information from within Pakistan.

Calendar design

The aim was to appeal to small-holder farmers so the information presented is in the local language, Urdu, and where possible images and diagrams are used to illustrate key messages. Initially, utilising a calendar template from another country, such as within Africa, was considered. However, despite several attempts this was not possible as a realistic model chart could not be found. Ultimately, a consultant was engaged to design the final product.

Distribution and feedback

Before mass distribution occurred, several prototype calendars were distributed to NFG team members as well as the Agricultural Sector Linkages Program (ASLP) Dairy Project team to distribute to farmers within their local network. Feedback from consultation with these farmers led to some minor „cosmetic. changes relating to message clarity to assist with the rate of adoption

The calendars were then distributed to a broad network of farmers within the villages serviced by the ACIAR LPS/2010/007 project in Punjab province. These being: Bhakkar, Jhelum, Kasur, Okara and Pakpattan. Detailed notes were compiled to enable extension workers to adequately train farmers in the use of the calendar at the time of distribution. This enables farmers to familiarise themselves with how to read the calendar and fully utilise the information provided. Repeat visits from these trainers provided follow-up information where required. Feedback is being currently sought from these farmers in order to gauge utilisation and make further improvements if required.

An example of the calendar appearance is presented in Figure 1. The top third of the page provides agronomic

information on the sowing, management and harvesting of forage species suited to grow at that time of year. This information includes pictures for each forage species. The bottom part of the page is divided into two sections. The left-hand-side presents a small table containing recommended rations for milking animals at different stages of lactation based on available ingredients whilst the remainder of the page illustrates a management to husbandry message. For example, in December (Figure 1-left) the message relates to harvesting maize whilst in June (Figure 1-right) the message relates to feeding supplements (concentrates, hay, haylage and silage) during summer when feed resources are limiting.



Figure 1 An example of calendar pages for December (left) and January (right)

RESULTS AND DISCUSSION

Small-holder dairy farmers in Pakistan now have access to a comprehensive feed calendar which can be used to provide guidance on the provision of forage for animals year-round. This is especially important during the annual feed gaps in winter and summer. Strategies such as fodder conservation as well as the feeding of concentrates and by-products were included to complement available forage resources. Utilisation of the calendar is being carefully monitored by project staff and extension workers associated with the ACIAR project. This support is necessary during the implementation period of the calendar to ensure queries associated with interpreting the information are answered appropriately and in a timely manner. Information provided on when to plant specific species and how to manage forage growth and harvesting is dependent on time of year. However, exact dates will be influenced by climatic events such as the timing of the break in season which may vary considerably between seasons and years (Pakistan Meteorological Department, 2012). It is therefore imperative small-holder farmers are instructed to use the calendar as a guide in combination with previous knowledge they possess themselves. It is envisaged regular feedback will be sought from the small-holder farmers utilising the calendar to ensure improvements can be made in subsequent versions where required. Having the basic format established will also facilitate the incorporation of new knowledge into future versions. The next step to improve productivity of the small-holder dairy farmer in Pakistan is the development of a handbook on ration formulation. This will provide a valuable tool for farmers wishing to increase milk production through the feeding of rations which contain ingredients such as concentrates, forages and by-products (Hussain et al., 2010). It is envisaged the calendar will provide a valuable resource for small-holder farmers as the format can be adapted for a range of environments within Pakistan as well as other developing countries.

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Relationship of Body Condition Score on Cyclicity and Pregnancy Rate in Nili-Ravi Buffaloes

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The objective of the study was to establish the relationship of body condition score (BCS) on cyclicity and pregnancy rate in Nili-Ravi buffaloes. The study was conducted at the government Livestock Production Research Institute, Bahadumagar, Okara in Punjab. Buffaloes (n=113) were assigned for BCS 1-5 scale using an Edmonson chart. Cyclicity and pregnancy status were determined twice at two week intervals by using transrectal ultrasonography. Blood samples were analyzed for progesterone using radioimmunoassay. Results indicated that BCS has a significant relationship with cyclicity ($p < 0.05$) and pregnancy rates ($p < 0.01$) in buffaloes. The data generated from this study will be helpful in gaining a better understanding of nutritional relationships with reproductive status in buffaloes.

Key Words: Body condition score, Cyclicity, Pregnancy rate

INTRODUCTION

Pakistan is dominated by an agricultural rural based economy. Livestock are the backbone of the agricultural sector contributing to the national (12%) and agricultural (50%) economy (Pakistan Economical Survey, 2006). Buffalo contribute 62% (Zia 2011) of Pakistan's 62.9 million milking herd and 67% of milk production (Afzal 2007). However, buffalo have relatively poor reproductive efficiency and exhibit many reproductive disorders including delayed puberty, poor oestrus expression, long postpartum ovarian quiescence, and most importantly lowered conception rates particularly when bred artificially (Gordon, 1996). Thus the production and reproductive potential of the Nili-Ravi buffalo has not been fully exploited. This is exacerbated by the fact that buffalo are generally found in developing countries with meager resources and where there is a lack of quality research in health, nutritional and reproductive management. BCS provides a method of assessing the adequacy of nutrition being supplied to an animal (Schroder, 2005; Walter, 1993) and can be set as a standard for each reproductive stage (Moran 2005; Schroder, 2005). It is especially applicable in the field where there are limited resources as it is a simple observation made on the animals (McDonald, 2002). Yet, despite the close genetic relationship between domestic cattle and buffalo, little of this information is applied to dairy buffalo in Pakistan and no data are available on the influence of body condition score on ovarian cyclicity and pregnancy status.

Thus the aim of the present study is to establish the relationship between BCS and ovarian cyclicity and pregnancy rate in buffaloes. This will help dairy farmers, particularly the small holders, to better understand the possible reasons for low reproductive efficiency of buffaloes. This relationship will provide information upon which to base the decision as to whether to breed or depending on BCS.

MATERIALS AND METHODS

This experiment was conducted at the Livestock Production Research Institute (LPRI) Bahadar nagar, near district Okara, Pakistan (The trial was based on 113 multiparous (lactating and non-lactating) Nili-Ravi buffaloes. All animals were permanently housed at LPRI. For the period of the trial all buffalo were managed and fed similarly. All buffalo were allowed to graze on pasture for 6-8 hours of the day and housed in animal sheds overnight where they were offered green chopped fodder (berseem) and wheat straw. The lactating herd were offered concentrate at milking time (evening and morning) roughly according to the milk production.

Assessment of body conditions score:

The body conditions scoring system used in the current study was based on a 1 (thin) to 5 (obese) scale with 0.5 intervals using the Edmonson chart developed for use in Holstein Friesian cows (Castellano, Bentsen et al. 2010). Similar conditions attributes could also be identified in buffalo and therefore the method was considered appropriate for body conditions scoring buffalo. Using this system all of the buffalo (n=113) were assigned a BCS by each of the four independent investigators who agreed for each animal within one unit (0.5). Inaccuracies associated with allocating BCS to pregnant animals were avoided, as the points used for assessment were not affected by changes in body shape due to pregnancy.

Assessment of pregnancy and cyclicity:

Pregnancy status was confirmed through ultrasound using a portable B-mode ultrasound machine and a 7.5 MHz transrectal probe (Shenzhen Well. D Electronics Co., Ltd, China, Model number: WED-9618 V). Pregnant animals (n=82) were body conditioned and allowed to return to the herd to reduce the risk of interfering with the pregnancies. After BCS, non-pregnant (n=31) animals were subjected to further examination for any uterine abnormality and ovarian cyclicity. Size of the ovary, presence of follicles, corpus luteum and cysts were examined to monitor the ovarian cyclicity of the animals. Those animals which had no detectable pregnancy were subjected to the same assessment of BCS, pregnancy and ovarian cyclicity two weeks later. This was to confirm early pregnancies not previously detected. This also allowed the operator to detect changes in ovarian structures to refine the assessment of the ovarian cyclicity of the animals. Seventeen buffaloes were found to be cyclic and 14 non-cyclic.

Statistical analysis:

Relationship of the BCS on ovarian cyclicity and pregnancy rate was analyzed using a logistical regression model using BCS and days postpartum as an explanatory variable in the final model. Among the non-cyclic buffaloes, data from two buffaloes were excluded, one having a tumor and the other having a structural adhesion. For the second screening two animals could not be returned for second examination due to local management problems.

RESULTS

Higher body conditions score has a significant ($P < 0.05$) correlation with higher pregnancy rates in buffaloes. The relationship between body conditions score and probability of pregnancy rate has been shown in figure 1 and table 1. Similarly, higher body conditions score is also significantly ($P < 0.05$) correlated with ovarian cyclicity during the breeding season in buffaloes. The relationship between BCS and probability of ovarian cyclicity in buffaloes is shown in figure 2 and table 2.

DISCUSSION

The present study demonstrated that the BCS can provide an accurate assessment of reproductive performance in buffaloes as in cattle where it has been proven as a consistent and reliable predictor of nutritional status (Domecq, 1995; Kunklen, d.). BCS was positively associated with pregnancy rates when insemination was synchronized and performed within a short fixed period postpartum (Burke et al., 1996) which is in agreement with the present study. This is supported by the evidence that, when animals are in negative energy balance or have a low BCS, circulating levels of leptin in the body are too low to activate the HP axis to permit cyclicity (Popa 2008; Meza-Herrera 2009; Castellano, Bentsen et al. 2010). Therefore, as shown in the current study, animals with low BCS and nutritional status are less likely to be cycling and are, consequently, less likely to be pregnant (Spicer 1990). The present study clearly indicates that the probability of ovarian cyclicity and pregnancy was significantly higher in those animals with higher BCS than those with low BCS. For every unit increase in BCS, a 13 percentage unit increase in pregnancy rates was observed, which suggested that signs of behavioral estrus were stronger and fertility was improved as BCS improved (Thatcher et al 1999). For the present study, both the probability of ovarian cyclicity and pregnancy approached one as BCS approached 3. Such high probabilities would not be expected for a normal population of buffalo or cattle at BCS 3. These abnormally high probabilities for cyclicity and pregnancy at BCS 3 were obtained in the current study may be due to the low number of animals in this BCS category.

CONCLUSION

BCS is a highly practical method for determining nutritional status of buffaloes, especially in the developing countries with limited resources like Pakistan where farmers can not afford expensive techniques and expertise. Such a strong correlation can influence the decision as to whether to breed or not thus minimizing the economic loss associated with the failure to conceive. It will thus be an important additional tool for the farmer to manage infertility in the milking herds.

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Comparison of Traditional Prostaglandin and CIDR Based Synchronization Protocol on Oestrous and Fertility in Buffaloes in Low Breeding Season in Pakistan

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A growing and emerging area in poultry research is the finding and application of medicinal herbs as natural growth promoters to enhance immunity and growth performance due to serious criticism on the use of antibiotics. In present study we examined the potentials of feeding dried garlic powder to male broiler chicks (Cobb-500). One hundred and sixty, one-day-old broiler chicks were randomly allocated to four replicated ($n=4$) treatments; GE-3, GE-2, GE-1 and GE-0 that received garlic @ 3, 2 and 1 g/kg of feed, respectively and GE-0 served as control. Birds were reared in floor pens (10 birds/pen) in an open sided house where optimum environmental and managemental conditions were maintained during the experimental period of 42 days. Birds in all pens had unlimited access to fresh drinking water and feed and were vaccinated against Newcastle (ND), Infectious Bursal (IBD), and Infectious Bronchitis (IB) diseases. On day-42, three birds from each pen were randomly selected for blood collection to determine serum lipid profile using Blood Biochemical Analyzer, antibody titer using H and ELISA kits and were killed to find out dressing percentage. Garlic powder did not affect feed intake. However, feed efficiency, body weight gain and dressed weight was significantly ($p<0.05$) improved by birds in group GE-3 and GE-2. GE-3 had higher ($p<0.05$) antibody titer against ND (455.2 ± 24), IB (5.50 ± 0.7) and IBD (4.25 ± 0.6) compared to other groups except GE-2. A significant reduction in serum cholesterol (120 ± 0.2), low-density lipoprotein (83 ± 0.1) and triglycerides (50 ± 0.2) and increase in high-density lipoprotein (29 ± 0.1) was observed in GE-2 to other groups. The difference between GE-2 and GE-3 was however insignificant. These findings demonstrated that garlic powder can potentially be used in broiler production to improve the immune status, growth performance and to optimize lipid profile.

Key Words: Garlic powder, Immunity, Growth performance, Lipid profile, Broilers

INTRODUCTION

The buffalo is an integral part of the Pakistan dairy system. According to the Pakistan economics survey (2011-12) the population of buffaloes is 32.7 million providing 29,565 thousand tonnes of milk annually. Buffaloes however suffer from a variety of reproductive problems. These problems include a long prepubertal period, seasonal breeding and silent oestrus. These problems alone or collectively contribute towards prolonged inter calving intervals. Domestic buffaloes have an inclination towards seasonal breeding (Qureshi, Habib, Samad, Lodhi, & Usmani, 1999) with a suspension in sexual activity during summer in many parts of the world (Hafez, 1955; Shah, 1990). Buffaloes are very prone to thermal stress (Pandey & Roy, 1966) causing hyper-prolactinemia and low luteinizing hormone (LH) concentrations; they have fewer sweat glands (Acharya, 1990; Cockrill, 1993; Heranjal, Sheth, Wadadkar, Desai, & Rao, 1979). The hyperprolactinemia induced by high ambient temperature is also associated with increased photoperiod causing changes in pineal gland secretory activity; (Sheth, Wadadkar, Moodbidri, Janakiraman, & Parameswaran, 1978; Wettemann & Tucker, 1974). In temperate regions where buffalo are found such as Italy where feeding management is very good a distinct seasonal reproductive pattern also prevails and according to Borghese (2005) and Zicarelli (1997b) seasonality is influenced by photoperiod through changing melatonin secretion which reduces fertility in a seasonal manner (Morgan & Williams, 1989). Hyper-prolactinemia is a cause of subfertility and ovarian inactivity by suppressing gonadotropin secretion and ovarian steroid synthesis decreasing progesterone in the blood supply (Hafez, Jainudeen, & Rosnina, 2000; Razdan, Kaker, & Galhotra, 1981; Reiter, 1991, 1998; Roy & Prakash, 2007; Wettemann & Tucker, 1974). Low levels of circulating LH due to high ambient temperature and photoperiod cause poor follicle maturation and thus a low level of oestradiol (Heranjal, et al., 1979; Palta, Mondal, Prakash, & Madan, 1997). The role of melatonin in reproduction is poorly understood in buffaloes. Melatonin secretion is

related with the season and as with all species the Italian Mediterranean buffalo produces high levels during shorter days (Borghese, Barile, Terzano, Pilla, & Parmeggiani, 1995; Parmeggiani et al., 1993; G. Presicce, 2007; G. A. Presicce, 2007). Management and nutrition are also important contributory factors for summer anoestrus (Kaker, Razdan, & Galhotra, 1982; Qureshi, Habib, et al., 1999; Qureshi, Samad, Habib, Usmani, & Siddiqui, 1999; Zicarelli, 1997b). The reason for 36.5% of breeding Egyptian buffaloes not being pregnant in the study of Heranja et al. (1979) was nutritional deficiency. In summer the hot ambient temperature decreases feed sources (Mudgal, 1979) and intake (Jainudeen, 1990), thus causing loss in body condition score and further deterioration of reproductive condition. Management plays an important role in heat detection. Buffaloes are not renowned for detectable oestrous expression (Jainudeen, 1990), which is often silent (Chaudhry, 1990) and mostly occurs during nighttime: the problem is exacerbated by hot summers. The major role of hormones in the expression of oestrus and the fact that buffaloes respond well to exogenous hormones (De Rensis & López-Gatius, 2007) suggest that the use of hormones to control fertility and decrease intercalving intervals (Madan M. L., 1983; Perera, 2011) may be a viable way of minimising the problem of long intercalving intervals. Various hormones are used in different combinations in cattle and buffalo so far; progesterone based protocols are considered to be the most effective in controlling summer anoestrus (Barile et al., 2001; Naseer, Ahmad, Singh, & Ahmad, 2011; Neglia et al., 2003; Singh, Singh, Sharma, & Nanda, 1984). High circulating progesterone increase ovarian follicle turnover and also sensitizes the hypothalamus and pituitary to gonadal activity (Baruselli, 2001). In this study the efficacy of a controlled internal drug release (CIDR) device was compared with a prostaglandin (PG) administration protocol extensively used in Pakistan in the Pakistani summer. These methods were compared without any addition of GnRH and oestradiol in the interest of keeping the protocols simple and cheap for small-holder farmer to adopt.

MATERIALS AND METHODS:

This study was conducted at government and small private dairy farms in Okara and Kasur districts of Pakistan during the low breeding season (June to August). Nili Ravi buffaloes (n=120) were randomly selected from government (n=70) and small rural dairy farms (n=50) with an average of 9 months postpartum anoestrus (Min: 2.47, Max: 24 months). The animals ranged in age from 4 to 8.6 years and were fed under normal small-holder farm conditions. Rectal palpation was carried out and the animal's history of any reproductive disorders was collected. Data on age, parity, BCS, per day milk production and presence of calf was also collected. All the animals were reproductively sound and healthy. Oestrous response rate was calculated by confirmation of oestrous, but as a part of the protocol all the animals in CIDR group were inseminated and pregnancy rate was calculated dividing pregnant divided by inseminated. Two animals lost CIDR and 3 animals died (2 in CIDR and one in PG group) after insemination.

Experimental design:

Buffaloes were randomly allocated to two treatment groups (n=60 per group). Group one was treated with a CIDR for 7 days, prostaglandin (PG) on day 6 with the CIDR being removed on day 7. The CIDR devices were provided by Pfizer Australia containing 1.38 mg progesterone and the PG source was prostaglandin F₂α (Utalysse, Pfizer). The animals were subjected to rectal palpation to check signs of oestrus. Oestrus was confirmed with uterine tone 2+ on scale of 1-5 and mucus and vulvular swelling. Time of artificial insemination (TAI) was carried out twice at 48 and 60 hours after removal of the CIDR device. Group two was treated with PG injection on day 0 and was bred on confirmation of heat. Those animals without any heat signs were re-injected with PG injection on day 11 and were bred in the next 3-4 days if on heat. Pregnancy diagnosis was carried out after 40 days via transrectal ultrasonography (Honda; Model: HS-1500; 7.0 MHz).

Statistical analysis:

Data for oestrus response and pregnancy rate between the two treatments, the impact of management (tied and untied) and the presence of a calf was compared using a Chi square analysis. The effect of body condition score (BCS) and parity on the oestrus response and pregnancy rate was confirmed with a Mann Whitney statistical test. The effect of age and daily milk production on the oestrus response, and the duration of anoestrus post-partum in days were compared using Anova and a t-test. A probability level with P value <0.05 was considered significant.

RESULTS

The results for oestrous behaviour and pregnancy rates with the two treatments are given in Table 1. When the CIDR was used with PGF2 α (Treatment 1) oestrus was detected in 84.5% of animals, but only in 23.3% of animals receiving a PGF2 α regime only (Table 1). This did not lead to a higher pregnancy rate (CIDR 19.6%; PGF2 α regime 30.8%; $P=0.296$). Body conditions scored did not vary significantly between animals expressing oestrus and those that did not irrespective of treatment. However animals that expressed a calf were in significantly ($p=0.006$) better condition than those that did not at the time of joining (Table 1). Similarly there were no differences in age between animals expressing oestrus and those that did not. However animals tended to be younger if they delivered a calf ($p=0.057$; Table 1). There was also no difference in the parity of animals expressing oestrus nor was there an association with success at delivery of a calf (Table 1). The restraint of animals by tying them to a specific object resulting in less than optimal water and feed availability did not affect either oestrus expression or success with delivery of a calf (Table 1). Similarly milk production was not associated with either oestrus expression or a successful pregnancy (Table 1).

	Oestrous expression			Pregnancy success			Test used
	Yes	No	P-value	Yes	No	P-value	
Treatment 1 CIDR: number of treated animals (%)	49/58 (84.5)	9/58 (15.5)	0.000	11/56 (19.6)	45/56 (80.4)	0.296	Chi-Square test
Treatment 2 PGF2 α : number of treated animals (%)	14/60 (23.30)	46/60 (76.70)		4/13 (30.8)	9/13 (69.2)		
BCS across treatments (1-5)	2.90	2.80	0.335	3.20	2.80	0.006	Mann Whitney
Parity across treatments	3.00	3.10	0.712	3.60	2.90	0.141	
Age (years) across treatments	8.30	9.00	0.253	9.80	8.00	0.057	Anova or T-test same results
Milk production (Kg) across treatments	5.10	4.10	0.160	5.20	5.20	0.986	
	74(61.5)	52(43.4)					

DISCUSSION

Postpartum anoestrus is a major cause of infertility and long inter-calving intervals. This study demonstrated that oestrus expression was higher in animals receiving a protracted regimen of progesterone release using a CIDR device. This is because high progesterone in the blood supply suppresses GnRH neurons in the tonic center of the hypothalamus. However LH secretion still remains relatively high along with tonic FSH release which helps in the development of a cohort of follicles. The high P4 prevents follicles from reaching the preovulatory stage. Furthermore high P4 primes the brain for estrogen and on CIDR removal a surge of GnRH occurs, which increases FSH and LH secretion from the anterior pituitary. The change in the size of the follicle due to FSH and LH release also increases E2 production (Senger, 2005). Therefore the CIDR causes a high percentage of animals displaying anoestrus response relative to those receiving PG only. The PG is only effective in the presence of a functional CL, while in summer the buffalo has small inactive ovaries without any palpable structures and so they do not respond to PG. The pregnancy percentage was lower in CIDR group than PG group animals and there were no significant differences across treatments. This may be due to the failure of ovulation after the GnRH surge, or an inappropriate timing of AI following a delay in ovulation by 30 hours after standing heat observed in buffaloes (Kanai & Shimizu, 1983; Warriach, Channa, & Ahmad, 2008). The relatively high pregnancy percentage in the PG groups suggests that heat detection was accurate and this option may be an effective method for small holder dairy farmers to use system. Only one animal (1/35) in PG group was detected on heat on government farms where teaser bull was used twice a day for heat detection along with visual observation.

Suspected animals were rectally palpated for confirmation of heat. Therefore the current 48 and 60 hours TAI, which was originally estimated for cattle/cows, needs further research for Buffaloes. There is also a need for investigations on detecting heat and AI using CIDR devices in small-holder rural dairy systems where the animals are under frequent observation. The farmer can detect animal on heat without any additional time and cost. Body condition score has no influence on oestrus response but has a significant effect on pregnancy rate. This further accentuates the need for good nutrition and shows the efficacy of these treatment protocols in well fed animals.

CONCLUSION

This study shows that the CIDR device is more effective in synchronizing oestrus than the use of PG. The high oestrus response and low pregnancy rate in CIDR group requires more research on the use of TAI techniques with the CIDR device to optimise the timing of AI.

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The Influence of Improved Colostrum Management and Milk Feeding Regimen on Serum Protein and Weight Gain in Sahiwal Calves in Pakistan

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Provision of adequate colostrum to newborn calves is essential for calf health and future animal productivity. The ideal combination of colostrum and milk ration and its subsequent effect on Sahiwal calf performance and immune system initiation was investigated in this study. Sahiwal calves (n=32) were allocated to colostrum and milk ration groups that represented traditional Pakistani and widely accepted improved husbandry practices. It was found that appropriate colostrum feeding increased serum protein concentration significantly ($p < 0.01$), while milk ration and calf starter intake had the greatest influence on overall live weight gain to weaning ($p < 0.05$ for both parameters). By improving provision of colostrum and milk, Pakistani farmers will be able to raise dairy calves in a manner that is cost effective while also increasing the future productive potential of their milking herds.

Key Words: Sahiwal, Calf, Colostrum, Pre-weaning, Pakistan, Productivity

INTRODUCTION

Newborn calves are born with a non-functional immune system and rely on the passive transfer of antibodies via colostrum from their dams in the first 24 hours after birth in order to initiate immune system development (Weaver et al. 2000). Provision of colostrum to calves is known to reduce morbidity and mortality due to the decreased chance of microbial infection, and the failure of passive transfer of colostrum immunoglobulins to the newborn calf is a major economic concern for cattle producers across the globe (Godden 2008). The rate of mortality in Sahiwal heifer calves to 30 days of age is estimated to be above 11% in Pakistan (Wynn et al. 2009), more than twice that of calves in American dairy farms (Waltner-Toews, Martin & Meek 1986). Sahiwal and buffalocalves are often deprived of colostrum because of husbandry practices in Pakistan that prevent free access to the dam, and prohibit the suckling of the calf until the placenta is expelled. This is often accompanied by restriction to milk fed pre-weaning. It is hypothesised that traditionally raised calves are immunocompromised as a result of this colostrum deprivation. Morbidity and mortality of calves can be reduced by making improvements to traditional husbandry practices (Perez et al. 1990), including provision of adequate colostrum in the 24 hours after birth (Nocek, Braund & Warner 1984). Calf performance and weight gain is also enhanced by an increase in the volume of milk fed to calves (Huber et al. 1984). The objective of this study was to evaluate the effects of improving calf management in Pakistan by increasing the volumes of colostrum and milk fed to calves in order to positively influence immunological development and weight gain to weaning.

MATERIALS AND METHODS

Colostrum group and serum protein

Newborn Sahiwal calves (20 male and 12 female) were used in a 2x2 factorial experiment conducted at the Livestock Production Research Institute (LPRI), Bahadurnagar, Pakistan. Calves were randomly allocated to a colostrum group ('Traditional' or 'Improved') to encompass the first 24 hours post birth, and were housed as a group separate from their dams. Colostrum was hand milked from the calf's dam, weighed, and fed to the calf. Colostrum intake for 'Traditional' calves was as per general Pakistani practice, with only one quarter of the udder being milked. The Improved group reflected ideal calf rearing practices with all four quarters of the udder being milked. Blood was collected by jugular venipuncture at 48 hours of age and centrifuged (1200xg) to separate the serum. Serum protein concentration was used as a measurement of immunological development and was determined using a hand-held refractometer (Milwaukee MR series, accuracy +/- 0.02g with automatic temperature control)

Milk ration and growth rate

At 24 hours of age, calves were removed into individual crates and randomly allocated to milk ration groups. The 'Low' group reflected common calf rearing practices in Pakistan and milk was fed at a rate of 7.5% calf body weight per day. The 'High' group was fed at a rate of 12.5% calf body weight per day. Calves were fed pooled fresh milk twice daily via nipple feeders and weighed every seven days on a standing scale four hours after their morning milk feed. Milk rations were adjusted weekly after weighing the calves to reflect the new body weight. Calves were fed their specific milk ration until they reached six weeks of age when they were then gradually weaned over three weeks using a step-down method (Khan et al. 2007). Upon reaching eight weeks of age, milk was completely withdrawn from the diet. Calf starter pellets were offered *ad libitum* from four weeks of age until weaning, and intake was recorded three times per week over a 24-hour period. Calves remained in the trial until three months of age, upon which they were returned to the LPR herd.

Statistics

An ANOVA was used to analyse the effect of the main factors on calf serum protein concentration, weight gain per day, and final weaning weight. The effect of covariates was investigated by way of simple linear regression. The level of significance used in this study was 0.05.

RESULTS

Serum protein

Colostrum intake for 'Traditional' calves averaged $2.56L \pm 0.90L$, and for 'Improved' calves averaged $4.39L \pm 0.93L$. Serum protein concentration was significantly influenced by colostrum group ($p < 0.01$). This is a direct result of improved colostrum intake, with total volume of colostrum ingested also having a significant effect ($p < 0.01$). Litres of colostrum ingested per kilogram of calf body weight also influenced serum protein concentration significantly ($p < 0.01$). The quality of the colostrum (grams of protein per litre of colostrum) had no effect on serum protein concentration. Failure of passive transfer (FPT) of immunity was classified as a serum protein concentration less than 5.5 g/dL (Tyler et al. 1996). Calves in the 'Traditional' group experienced a FPT rate of 25%, while 'Improved' calves all acquired successful passive transfer.

Liveweight gain

Overall weight gain to weaning was most significantly influenced by starter pellet intake per day ($p < 0.05$). Milk ration group also had a significant effect ($p < 0.05$), with calves in the 'High' group gaining $32.5kg \pm 5.61kg$, and the 'Low' group gaining $27.0kg \pm 7.28kg$ to weaning. Colostrum group and the interactions between the colostrum group and milk ration did not have a significant effect on overall weight gain.

DISCUSSION

As anticipated, the traditional methods used to raise Sahiwal calves in Pakistan resulted in inferior liveweight gain and poor passive immunity. Provision of colostrum in volumes greater than those currently fed to newborn calves will significantly increase calf serum protein concentration and therefore improve immunocompetence (Hopkins & Quigley 1997). Current research shows that a minimum of 4L of colostrum provided within 12 hours of calving is required for newborn dairy calves; however, this recommendation was derived from studies with Bos taurus breeds (Weaver et al. 2000). Sahiwals (*Bos indicus*), may be able to achieve successful passive transfer with lower volumes. Increasing the volume of milk fed each day to calves pre-weaning will result in enhanced weight gain. Heavier calves at weaning have the potential to reach puberty faster than lighter calves, provided adequate nutrition is maintained post-weaning (Boggset al. 1980). This could lead to more lactations per animal and therefore reward the farmer with a higher overall milk yield.

CONCLUSION

Improvements to colostrum and milk feeding practices resulted in an enhancement of the functional immune

status of Sahiwal calves and an increase in live weight gain to weaning. These two husbandry practices can be combined to improve the efficiency of calf rearing both for financial gain and improved calf welfare. Farmers are therefore likely to spend more time generating income from their animals rather than simply keeping them for sociological reasons.

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Effect of Improved Extension Services on Adoption Rates and Production of Small Holder Dairy Farmers in Pakistan

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The objective of the study is to demonstrate the effect of improved extension services on adoption rates and production of small holder dairy farmers in Pakistan. During the first phase of the project a simple approach of extension targeting male farmers was used. Whereas in the second phase of the project a whole family approach targeting all the family members was utilized. The effect of improved extension services on adoption rates and production data was collected every month from the farmers (n=538). These farmers were working in districts Kasur, Okara, Pakpattan, Jhelum and Bhakkar within Punjab province. Preliminary results indicated higher adoption rates when extension services were provided using the whole family approach compared to the simple approach during the first year of both phases. Improved farm practices had significant effects on the milk production of farmers. The average milk production was higher in animals from the farmers who had provided free access to water and feeding to their animals compared to those who kept their animals under the traditional practices of tethering animals. The data generated from this study will be helpful to devise better strategies for improved extension services in order to optimize the dairy production of small holder farmers and will have a ripple effect for the others to follow.

Key Words: Extension services, Small holder farmer, Dairy

INTRODUCTION

Pakistan, like many developing countries, has an agrarian rural based economy. Livestock is a major contributor to the national (12%) and agricultural (50%) economy (Pakistan Economic Survey, 2006). The livestock sector has been recently declared as one of the fastest growing sectors and provides improved livelihoods for more than 35 million people with farmers/households deriving 30 to 40% of their income from livestock. Milk remains the major contributor to income derived from livestock. The value of milk alone exceeds the combined value of wheat, rice, maize and sugarcane in the country. Milk is produced under different production systems namely, rural subsistence small holding, rural market oriented small holding, rural commercial farms and peri-urban dairying. It is estimated that around 70% of the dairy households in Pakistan still operate under conditions of subsistence by maintaining herds of three or four animals (Burki et al., 2005). The productivity of livestock is still lagging behind its potential level. In order to meet the requirements of a rapidly growing population, dairy production needs to be increased. This can be done by adopting modern techniques of dairy farming. New technologies developed by researchers are disseminated among the farmers through an effective extension program.

The role of extension has been to provide research-based education and information to the production sector. The most important management areas on a dairy farm are feeding and forages, udder health, reproduction, calf raising, and herd health (Dahl et al., 1991a). Problemsolving in these areas requires a broad base of knowledge and expertise, and often the implementing agency must organize a multidisciplinary team of extension specialists or other professionals to assist producers (Dahl et al., 1991b). Services to the dairy sector are being provided by government agencies and a range of NGOs, and virtually all services providers who interact with the farmers are veterinarians or para-veterinarians who perform vaccination, treatment and A.I. Limitations in the extension service and the research/extension interface are considered to be bottlenecks in the development of the dairy sector. In particular, the style of communication between farmers and extension staff, the information available to extension staff, the number of skilled extension staff and a failure to consider problems and solutions in a

whole-of-farmsystemscontextareimportantlimitations.Thusthemajorobjectiveofthisstudyistodemonstrate theeffectofimprovedextensionsservicesonadoptionratesandfarmeconomics ofsmallholderdairyfarmersin Pakistan.

MATERIALSANDMETHODS

In2007,anAustralianCentreforInternationalAgriculturalResearch(ACIAR)researchprojectLPS/2005/132 wascommencedwiththeaimofincreasingdairyproductionthroughimprovedextensionsservices.Smalldairy farmershaving4-10(buffaloand/orcattle)forproductionwerethemaintargetgroupforthisproject.Duringthe firstphaseoftheprojectasimpleapproachofextensiontargetingmalefarmerswasused.In2011,ACIAR extendedthisresearchprojectforanadditionalfiveyears.Theprojectiscurrentlyworkinginfivedistrictsof Punjab(Okara,Pakpattan,Kasur,Jhelum,Bhakkar)andtwodistricts(ThattaandBadin)ofSindhprovince. Duringthissecondphaseoftheprojectawholefamilyapproachtargetingallfamilymemberswasutilized.A numberofinnovativewaysofextensionhavebeenadoptedincludingtheuseofvideopractices,demonstration plots,problembasedlearning,stagedrama,radioandTVshowstoimprovetheeffectivenessoftheprogram. Benchmark data were collected on whole farming systems from 228 farmers during the first phase and 292 farmersduringthesecndphaseoftheproject.Subsequently,attheendofeveryyeardatahavebeencollectedto monitortheimpactoftheseinitiativesontherateofextensionmessageadoption.Inordertoanalyze thefarmeconomics,datawascollectedfrom10primaryleadingfarmers,twofromeachofourprojectdistrictof Punjabannually.

Theprojectplacedemphasisonacomprehensiveinterdisciplinaryeducationalprogramofmeetings,workshops andtrainingsofbothfarmersandextensionworkers.Basichusbandry,nutrition,andcalfinmanagementwerethe initialsubjectsaddressedduringbothphasesoftheproject.Adoptionratesbetweenthewholefamilyapproach andsimpleapproachafteroneyearofbothphaseswereanalyzedusingaChi-squaretest.Similarly,adoption ratesofvariousmodulesatthestartandafteroneyearofprojectphase-IIwereanalyzedusingaChi-squaretest. Comparisons of various average monthly incomes of small holder farmers were analyzed using a t-test. StatisticalanalysiswascarriedoutusingSPSS(Version10.0)with $P < 0.05$ regardedassignificant.

RESULTS

Adoptionrates(50%)weresignificantlyhigher($P < 0.05$)bymorethanthree-foldwhenawholefamilyapproach wasimplementedcomparedtothesimpleapproach(14%)afteroneyearofbothphases(Figure1).Comparison ofadoptionratesofvariousmodules(animalthusbandry,basicconceptsofnutritionandcalfnutrition)atthestart andafteroneyearofthesecondphaseisshowninFigure2.Untyinganimalsandgivingfreeaccesstowater togetherwithtwicedailycleaningofsheds werereadilyadopted,howeverinvestingininfrastructureintheform ofshedconstructionwasnotaseasyforfarmers(Figure2a).Offeringfodderadlibitum,feedingconcentrateand Improvedextensionsserviceshavesignificantlyincreased($P < 0.05$)thefarmincomeofsmallandmineralmixes wereperceivedasbeingeasyandofdirectbenefittoproductivity:incontrastfeedingcottonseedcakewasnot (Figure2b).Irrespectiveofthefmessageforcalfrearingfeedingcolostrum,offeringconcentrateandsufficient milkandwaterwereperceivedasbeingreadilyadoptablemessages(Figure2c).Overalltherewasabout\$ US/monthincreaseintheincomeofsmallholderdairyfarmer(Table1)

DISCUSSION

Toourknowledgethisisthefirstreportwhichclearlydescribestheeffectofimprovedextensionsserviceson adoption rates and farm economics of small holder farmers in Pakistan. Higher adoption rates (50%) were achievedwhenweintroducedawholefamily(male,femaleandchildren)approachcomparesimpleapproach (14%) relative to the traditional male only approach to extension. An effective extension program with participationbythewholefamilyishighlydesirabletoenhancefarmproductivity.Manyorganizationsaimingat improvingsmallholderdairyinfailtoappreciatethisfactandignorewomenandchildrenintheirtrainingand skills development programmes. Women normally cannot leave their home and families for a few days to participateintrainingprogrammesandusuallyrequirefemaletrainersforeffectivecommunication.Thusduring thesecondphaseoftheprojectwearrangedparallelsessionsforwomenandchildrenco-ordinatedbywomen trainers.Thisapproach,alongwithtrainingofthemalefarmersresultedinhigheradoptionrates.

In order to demonstrate the role of children in rural communities this project initiated a buffalo calf raising competition among the children of farmers (10-16 years) in Pakpattan district. These results demonstrated that under field conditions the average daily buffalo calf live weight gain (431 gm/day) was comparable to many controlled studies in various leading research institutes of Pakistan (Bhatti et al., 2009; Iqbal and Iqbal, 1992). These findings clearly indicated that we can effectively enhance animal productivity through the active involvement of children in our extension program.

In the present study, adoption rates of various modules at the start and after one year of project phase-II are significantly higher. Possible reasons of high adoption rates other than the whole family approach are the implementation of the innovative ways of extension like video practices, demonstration plots, problem based learning, staged drama, radio and TV shows. Innovative methods of extension played a significant role in order to achieve higher adoption rates. While devising training programmes, one should keep in mind that "Seeing is believing" and "Farmers don't have ears, they only have eyes."

The present study clearly demonstrated that improved extension services resulted in significantly increased farm income for small holder dairy farmers. There was an average increase of about \$US 100 in monthly income after one year of effective extension. Farmers were shown to have adopted basic husbandry and improved nutrition practices. Providing ad-libitum access to water and feeding resulted in the increase of approximately 1 lit/animal/day (Warriach et al., unpublished data). However, there is a need to investigate further the effects of various adoption rates on the productivity and farm economics of small holder farmers. In conclusion, these preliminary results indicate that improved extension services have a significant impact on higher adoption rates resulting in an increase in farm incomes of small holder farmers.

ACKNOWLEDGEMENTS

The author sincerely thanks the cooperating small dairy holder farmers and their families, extension workers, research assistants for participation and contribution to this project; ACIAR for funding the project No. LPS/2010/007.

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Table 1. Comparison of various average monthly incomes of small holder farmers

Survey	No. of milking animals	Land (Acers)	Dairy income/ month (Rs)	Crops income/ month (Rs)	Income from other sources/ month (Rs)	Total income/ month (Rs)
2011	2	6.7	7161a	5172a	4710a	17043a
2012	2.2	6.7	12758b	9358b	5400b	27516b

a,b Means with different superscripts within a column are significantly different (P < 0.05)

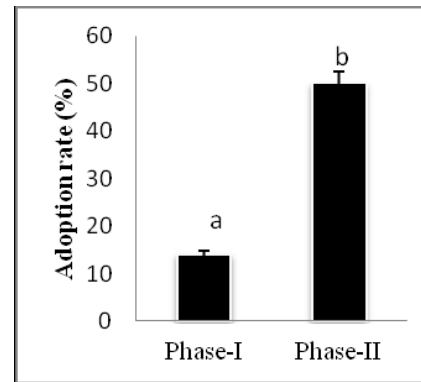


Figure 1. The difference in adoption rates (%) of extension messages following the use of a traditional “male only” extension approach (phase 1) as compared with a whole family approach (phase 2).

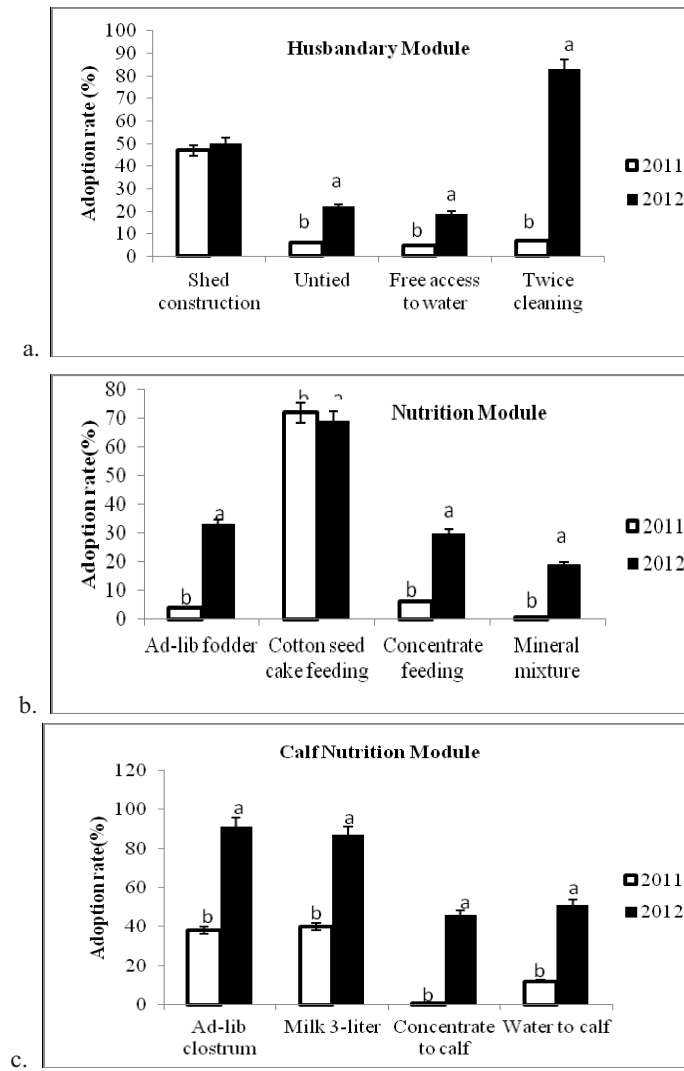


Figure 2. The difference in adoption rates (%) of specific extension messages following the use of a traditional “male only” extension approach (phase 1, 2011) as compared with a whole family approach (phase 2, 2012). 2a: watering, cleaning and shed construction; 2b: aspects of animal nutrition; 2c: aspects of calf feeding

Participation of Women in Dairy Farm Practices under Small Holder Production System in Pakistan

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The objective of the study was to investigate the participation of women in dairy farm practices in the small-holder production system in Pakistan. A questionnaire was designed to collect the data from female farmers (n=200) of districts Bhakkar and Jhelum in the Punjab province. Our results indicate that the participation of women in dairy farm practices was significantly higher ($p < 0.05$) in Bhakkar as compared to district Jhelum. The present study suggested that, in order to maximize the small-holder dairy farm productivity, a strong extension program should be implemented to enhance the skills and knowledge of women.

Key Words: Women, Dairy farm practices, Smallholder

INTRODUCTION

Livestock are considered a key asset for rural livelihoods and offer significant opportunities for improving household incomes. Women traditionally play a major role in conducting various livestock management activities all over the world. Pakistani women have a significant role in agriculture and livestock rearing. Nearly 65.9% of Pakistan's population are living in rural areas that are directly and indirectly linked with the agricultural related sector for their livelihood (Farhana et al; 2008). Women comprise half of the rural population and contribute 60 to 80% of labour in the animal husbandry (Younas et al., 2007). Women not only perform normal household chores such as cooking, cleaning, mending clothes and raising children (Kazmi, 1999), but also participate in rearing of livestock and carry out various dairy farm practices. These practices include feeding and watering, fodder cutting, cleaning animals and their sheds, caring for sick animals, calf rearing, milking and the processing of dairy products like ghee, butter and yogurt. Some of these activities, like fodder production, are generally considered the responsibility of men, but in many cases the women are also involved.

The participation of women in dairy farm practices varies by region, age, culture and social status and are changing rapidly in some parts of the country. The existing information regarding participation of women in dairy farm practices is very limited. Therefore, the present study was aimed to investigate the participation of women in dairy farm practices in the small-holder production system within the districts of Bhakkar and Jhelum. Additionally, possible factors affecting their participation in dairy farm practices were investigated. It is anticipated that the information generated from this study will be helpful in identifying the extension needs and areas where women can improve dairy production by enhancing their skills and knowledge.

MATERIALS AND METHODS

A dairy extension project (no. LPS/2010/2007, funded by ACIAR) is working in Pakistan aimed at strengthening the dairy value chains in Pakistan through improved farm management and more effective extension services. A questionnaire was designed to collect data from project working areas, 92 female farmers from Bhakkar and 102 from Jhelum. These two districts provide a contrast between an undeveloped arid region poorly served by irrigation and state livestock services (Bhakkar) and a more advanced region where farmers have access to extensive irrigation and support from the state livestock veterinarians (Jhelum). Eight villages were selected from Bhakkar and nine from Jhelum.

Statistical analysis

Participation of women in dairy farm practices under small holder production system in comparison to Bhakkar and Jhelum was analyzed using Chi-square test. All the analysis was carried out with the Statistical Package for Social Sciences (SPSS-13.0). A p-value of 0.05 was regarded as significant.

RESULTS

Participation of women in dairy farm practices was significantly higher ($P < 0.05$) in Bhakkar as compared to district Jhelum. Comparison of various dairy farm practices between both districts are shown in Table 1. Socio-economical status and cultural norms are significantly affecting women's participation in dairy farm practices in the small-holder production system in Bhakkar and Jhelum. From Jhelum 15.6% women are not participating in dairy farm practices due to cultural or religious barriers and 24% due to their relative affluence. Whereas, in district Bhakkar only 4.4% are prohibited from participating because of a cultural barrier while 9% are not participating due to their affluence. The education level for women was observed to be almost the same in both Bhakkar and Jhelum (Fig 1 and Fig 2).

DISCUSSION

The present study revealed that the participation of women in dairy farm practices was significantly higher in Bhakkar in comparison to district Jhelum. Women are exclusively taking part in various dairy farm practices like husbandry and nutritional management. The present study shows clearly that provision of appropriate extension services to women in the field of animal husbandry and nutritional is likely to significantly improve the production of animals. The women trained in livestock production activities will help increase milk and meat production at the national level and raise household incomes (Younus et al., 2007; Shehzad, 2004).

Factors causing lower participation of women in dairy farm practices are socio-economical status and cultural norms in Jhelum. Gender roles are shaped by ideological, religious, ethnic, economic and cultural factors and are a key determinant of the distribution of responsibilities and resources between men and women (Moser, 1989). Our survey data showed that from Jhelum 15.6% women are not taking part in dairy farm practices due to cultural barriers and 24% due to their high economical status. The latter 24% of economically advantaged women do not work themselves but they hire labourers, whereas most of the rural and tribal women do most of the farm work themselves. Land holding also affects their economical status. Although the average land holding in Bhakkar (9 Acres) is larger than in Jhelum (6.9 Acres) but the fertility of land is poorer. The lower soil fertility with less irrigation water and more hot weather are major factors limiting production in Bhakkar while holdings in Jhelum are highly fertile and productive.

The present study indicated that the education level was almost the same in the two districts. According to the Pakistan Social and Living Standard Measurement (PSLM) Survey 2010-11 the literacy rate for the population (10 years and above) is 58 percent. (Pakistan Economic Survey 2011-12). It is important to know that equal opportunities (schools and colleges) are available in both of the districts, which would seem to be the case given the results of our survey. The present study suggested that, in order to maximize the dairy farm productivity a strong extension program should be implemented to enhance the skills and knowledge of women. We propose to identify the remote areas where more women are taking part in livestock production. Following this, these women can be engaged with extension programs to ensure they know of the best practices for livestock health and production.

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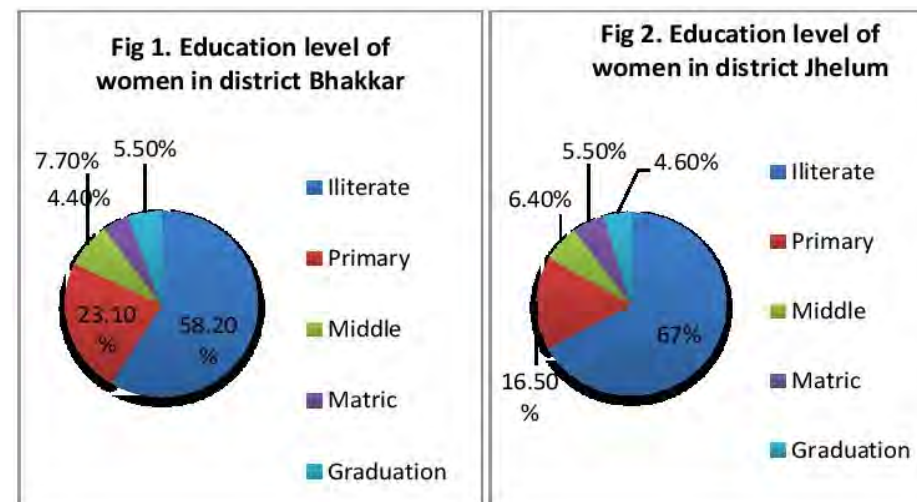
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Table 1. Comparison of participation of women in various dairy farm practices between district Bhakkar and Jhelum

Livestock management activities	Districts	
	Bhakkar (%)	Jhelum (%)
Shed Cleaning	71(78.3)	80(73.1)
Fodder cutting from field	19(20.7) ^a	17(15.7) ^b
Feed and watering	58(64.1) ^a	22(19.4) ^b
Care of calf	67(73.9) ^a	32(28.7) ^b
Milking	71(78.3) ^a	22(19.4) ^b
Milk sale	64(69.6) ^a	61(56.5) ^b
Care of sick animals	37(40.2) ^a	10(9.3) ^b
Value addition of milk	71(78)	98(89.9)

a, b means with different superscript within rows are significantly different ($P < 0.05$)



Marketing Milk from Small-Holder Dairy Farmers in Pakistan

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The dairy sector offers enormous scope for economic growth and poverty alleviation. A very high proportion (97%) of the milk produced goes to consumers through traditional value chains. These chains are a dominating link between millions of farmers and consumers. Development strategies, however, have not been able to tap the pro-poor potential that these chains offer, targeting instead high profile processors. These large scale processors service only the most accessible and larger farmers without considering the predominant population of small-holder farmers: in effect they are only interested in economies of scale. The study reported in this paper will describe the key attributes of traditional chains and then highlight the opportunities and risks associated with their evolution and their potential to assist the profitability of farmers contributing milk to them. The chains are characterized by the collection of milk on farm and then selling this to a primary milk vendor of *flhodi*. The milk is then sold to 2-3 larger milk vendors in a chain who collect and then transport the milk to commercial retail outlets in large urban centres. During these processes of transfer milk may be adulterated and diluted to ensure that each vendor is able to make a profit from their transactions. Milk is also sold either by volume or by weight: since the density of milk is greater than that of water there is a small incremental profit that accumulates with these transactions. The volumes of vessels used for the assessment of payments are also not standardized. In essence the farmer is rarely accorded full payment for his product, which is often of poor quality once marketed. However, importantly each of the vendors provides loans to those one step further up the chain thereby enabling each member to survive financially. The farmers receive cash advances for their product to enable them to continue to purchase resources required to feed and care for the animals. Thus the flow of production in one direction to market and the flow of cash in the other direction are equally significant to the sustainability of these chains.

KeyWords: Milk, Small-holder, Dairy farmers, Marketing chains

INTRODUCTION

In spite of a range of industrial development, agriculture still forms a major part of the back-bone of the Pakistani economy. Its contribution to the nation's GDP has decreased from 25.9 percent in 1999-2000 to 20.9 percent in 2010-11. The major agricultural pursuits include cropping, livestock, fisheries and forestry. While the cropping sector has shrunk over the past 12 years from 65.1 percent in 1990-91 to 37.5 percent in 2010-11, the contribution of livestock to the agricultural sector has increased from 29.8 percent to 55.8 percent in the same period (Hassan, 2010-11). Of the 46.44 million tonnes of milk produced in Pakistan 16.13 million tonnes come from cattle and 28.69 million tonnes from buffalo. This substantial milk harvest is generated from a milking herd of 31.7 million buffaloes and 35.6 million cattle (Farooq, 2011), which translates to a daily production of around 5 litres of milk per animal per day. In spite of this low productivity Pakistan remains as one of the top 5 milk producing countries in the world. With an ever rising consumption of 94 litres per person per year, the industry struggles to meet domestic demand let alone expand to export markets.

Most of this milk volume is generated from more than 8.5 million small-holder farmers owning less than 10 animals kept on small areas of 1-5 hectares: 94 percent of farms in Pakistan are less than 1 hectare in area (Khan et al., 2010). Although cropping activities have traditionally generated the predominance of family income, the challenge remains to increase livestock productivity to help the family budget and alleviate poverty in rural regions.

Both milk production and demand vary significantly over the course of the year. Fortunately the production curves of buffalo and cows complement each other well to provide a relatively constant supply of milk. Peak

productivity for buffalo is achieved post calving from September to December, while spring calving cows provide peak milk yields during the high consumption summer months from May to August. Maintaining this pattern of milk production requires the farmer to maintain feed supply during the annual periods of feed deficit at the height of summer and middle of winter. This is rarely achieved and so milk supply decreases to 55% of peak production in summer just when demand has increased by 60%. The link between the producer and the consumer is provided predominantly by informal milk marketing chains, with only 3% of the product being handled by commercial milk processing companies. The nature of the informal marketing network has been investigated here.

MATERIALS AND METHODS

Two districts of Punjab were chosen for these observational studies based on their contrasting climatic attributes as well as their relative economic development. Farmers in the Okara area are serviced by irrigation and have ready access to government veterinary services. Characteristically they generate income from both cropping and livestock enterprises. This region is close to large urban markets in Lahore. In contrast Bhakkar is a drier more isolated environment, where irrigation is limited by water supply most often from bores and government veterinary services are scarce.

The author has investigated the structure and dynamics of typical marketing chains in each region by interviewing participants from the initial milk collector operating at the farm gate through to the final milk vendor in urban Lahore. Limited milk samples have been analyzed for milk protein, fat, added water and mycotoxin M1 to assess milk quality at the shop-front.

RESULTS

It is important to note that a significant proportion of milk on small-holder dairy farms is consumed domestically, often with little entering these marketing chains. However with the advent of more effective extension programs a higher proportion of the family production enters these chains. The traditional chains remain in areas where transportation is limited by poor road infrastructure allowing only motor or bicycle access to the farm gate.

These marketing chains are characterized by a complex series of inter-personal interactions that dictate the flow of product from the farm to the consumer. They involve both financial transactions as well as the manipulation of the product to generate a profit for each operator in the chain. One such chain is illustrated in Figure 1.



Figure 1 Description of a typical milk value chain

The first step: the dhodi

In this case milk from over 800 farmers was collected by small milk collectors, known colloquially as dhodis, servicing 10 farmers each using motor bicycles. The farmer and the dhodi are dependent on each other with the farmer providing the product and the dhodi extending payments 2 weeks in advance to allow the farmer to pay for the upkeep of his animals. These payments extend from PKR 2,000 to 10,000 per fortnight depending on milk volume. Anecdotal evidence shows that the dhodi in some instances delivers pharmaceuticals and other essential products to isolated families and of course is a major disseminator of information throughout the community. The product leaves the farm intact containing the normal fat (6% in buffalo and 3.5% in cow) and protein (3.2-3.5%) levels. The quality of milk leaving the farm is assessed typically by taste and smell only with no assessment for cell count or microbial safety. The price paid for the milk will vary according to supply and demand as well as local factors such as the intervention of milk buyers from the larger companies. The price differential between the dhodi and the corporate buyer is most often around PKR 2. However the price offered by the dhodi is dictated by the medium milk collector. Milk is collected in containers with 44.7 kg which when adjusted for milk density becomes 49 litres. If the milk fat is as high as 6% the volume ends up as 53 litres. The adjustment from 44.7 to 53 litres provides a lucrative margin for the small collector of PKR 265 working on the basis of PKR 32 per litre for milk.

The second step: the medium milk collector

The milk from around 10 small collectors is aggregated by medium milk collectors, who assess milk quality by measuring fat content only. Commercially fat is the most important component down the marketing chain. The medium collector is paid a fixed commission on milk volume, which is fixed and not dependent on the prevailing milk price. These collectors in turn provide loans of up to PKR 0.1 million to allow dhodis to service or replace the mode of transport, the motor bicycles. They collect around 600 litres and do not have to transport the milk anywhere.

The third step: the large milk collector

At this point the volumes of milk are getting larger with more than 5,000 litres being collected from 10 small collectors. The large collector requires a form of transportation for the milk which is usually a truck while milk is being collected in containers of around 160 litres. The merchant now needs to employ at least 6 staff with a truck which might well be on the road collecting and distributing for 18 hours daily for 365 days of the year. Ice is now being added to containers at about 10% by weight of volume.

The fourth step: the retailer

The typical retail outlets sell around 400 litres per day. Again the hours of operation are long with retail outlets being open for 17 hours per day; ice is often used as power outages are common over the summer months during the peak milk consumption period. Some of the milk may be converted into fermented products like yoghurt and lassi, both of which are gaining in popularity and add to the profitability of the shop. Preliminary analyses of milk samples purchased from retail outlets in Lahore have suggested that milk protein content can be as low as 1.5% with milk fat at around 3.1% and water added to dilute the original product by 1:2 (N. Aslam, unpublished results). In the limited analyses conducted to date aflatoxin M₁ is expressed at levels 5-10 times acceptable world standards (N. Aslam, unpublished results).

DISCUSSION

Clearly the structure of traditional milk marketing chain has been honed from centuries of evolution in which the most limiting resources have been transport and refrigeration: in essence each member of the chain is quick to pass on the product to the next link as quickly as possible while at the same time generating a small profit margin for each litre of milk transacted.

In return the chain provides a flow of finance in the opposite direction of finance the activities of those one step closer to the source of milk, the cow or buffalo. The very complexity of the structure provides significant

employment opportunities for relatively under-educated merchants. With improvements in transportation and an increasing demand by the consumer for a high quality product that meets acceptable food safety standards, the whole structure of these traditional means of milk marketing may be threatened. The consequences for village communities remain to be seen.

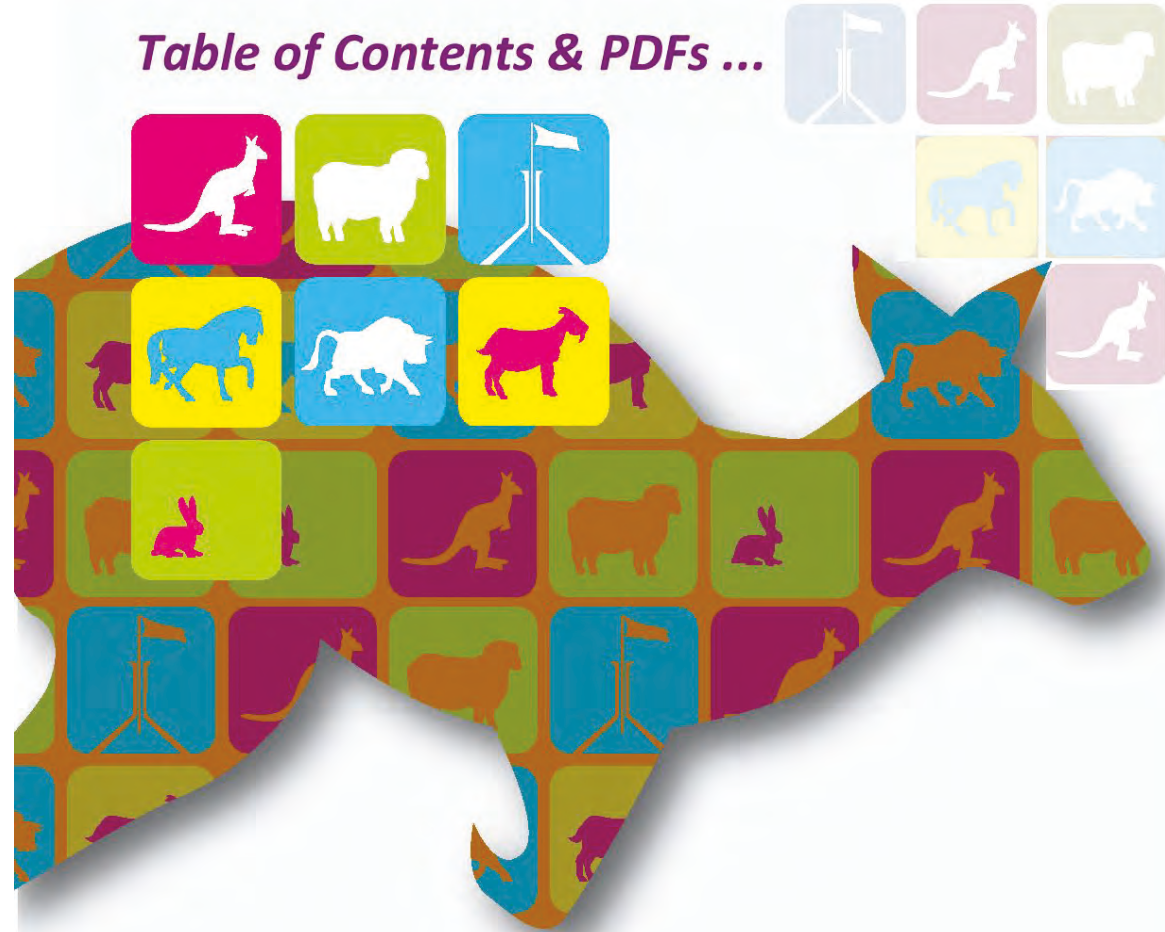
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Improved Calf Rearing Practices Under Smallholder Farming Systems in Pakistan

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Abstract

The objective of the study was to evaluate the improved calf rearing practices under smallholder farming systems in Pakistan. In the 2011, a survey was carried out across 523 smallholder dairy farms from five districts (Okara, Pakpattan, Jhelum, Kasur, and Bhakkar) of the Punjab through personal interview. Data on aspects such as calving season, calf husbandry, colostrum feeding and nutrition were collected and analyzed. In the second survey in 2013, improved extension services were provided to the farmers for a two-year period. A follow-up survey of 427 of the same farmers was conducted in order to capture the impact of the intervention on adoption rates of calf rearing practices. The results from the first survey show that calving rates were significantly higher ($P < 0.001$) in buffalo during July to September and in March in cattle compared to other months of the year. The majority of the smallholder farmers (60%) did not offer colostrum to their calves immediately after birth and instead waited for the expulsion of the placenta. Most of the farmers (55%) weaned their calves off milk at 6 months while 39% left their calves to suckle up to one year of age. In the second follow-up survey, 94% of trained farmers adopted some form of improved colostrum feeding which was significantly higher ($P < 0.001$) than observed in the control group (20%). The adoption rate of correct colostrum feeding before placenta expulsion was significantly higher ($P < 0.001$) in the trained farmers (89%) compared to the untrained control group (21%). Of these, only 58% offered colostrum ad libitum, although this was still significantly higher than observed in the control group (9%). The outcomes of this study clearly indicate that by providing appropriate extension services to smallholder dairy farmers in Pakistan calf rearing practices throughout the country can be greatly improved.

Keywords: calf rearing, extension services, smallholder farmers

Introduction

Pakistan has a agrarian rural-based economy like many developing countries with livestock a major contributor to the national (11.9%) and agricultural (55.4%) economy (Pakistan Economic Survey 2012). Dairy is a major component of the livestock sector in both numbers of cattle and buffalo (38 and 33 million respectively) and income generation (Habib et al 2007). Milk is produced under different production systems namely, rural subsistence smallholding, rural market oriented smallholding, rural commercial farms and peri-urban dairying. It is estimated that around 70% of the dairy households in Pakistan still operate under conditions of subsistence by maintaining herds of three or four animals. Under these smallholder farming systems, on an average there are 5-8 cows and buffalos per herd including 3-4 lactating and 2-4 heifers/calves. Animals are fed under a fodder cut and carry system with mixed grazing in some areas. Smallholders also grow seasonal fodders for their animals and mix it with crop residues, with wheat straw being very popular throughout the country.

Calves play a significant role in the development of any dairy sector, as the rearing of healthy calves through the provision of appropriate nutrition and health management practices will result in a more productive herd (Mehmood 1991). Generally, calves in Pakistan are neglected because of their high feeding costs and low returns from sale at weaning age (Bhatti et al 2009). Calf nutrition and feeding management is the most ignored area in both husbandry and research (Wynn et al 2009). Higher mortality losses in buffalo calves (52%) are reported

from the region due to poor colostrum and feeding management of calves (Ramakrishna 2007). Mostly calves are deprived of colostrum due to human consumption or the custom of giving colostrum to friends to make sweets (Wynn et al 2009). Calf management starts from the late trimester of pregnancy because a cow's nutrition and health status influence calf health at birth. After birth, the calves are more susceptible to diseases compared to the adults because of low immunity.

For this reason there is a need for the proper management of all aspects of calf rearing, including husbandry, nutrition and health, to sustain a healthy and productive dairy herd. Feeding colostrum immediately after calving is the only source of immunoglobulin for passive immunity particularly in ruminants, for which no exchange of immunefactors occurs in utero (Larson et al 1980). However in Pakistan, many farmers do not feed colostrum in a timely manner (Ahmad et al 2009). Farmers usually wait for the expulsion of the placenta due to their adherence to traditional practices, and that leads to a lower immunity level and high mortality before weaning. Therefore, the objectives of the present study were (1) to determine the ongoing calf rearing practices and (2) capture the impact of improved extension services on calf feeding.

Materials & methods

Dairy project

The Australian Centre for International Agricultural Research (ACIAR) funded a research project (LPS/2010/2007) with the aim of strengthening the dairy value chains in Pakistan through improved farm management and more effective extension services. The target groups of the project were smallholder dairy farmers having a herd of 4-8 (buffalo/cattle) mixed animals. The present study was conducted in five districts of Punjab (Okara, Pakpattan, Kasur, Jhelum and Bhakkar). A baseline survey was conducted to monitor the ongoing farming practices and identify problems, followed by 2 years of extension activities and a follow-up survey was carried out to capture its impact. The climate of the study districts in summer ranges from (24-33°C), winter (11-23°C) and average rainfall was (15-115mm) (Climate of Pakistan, 2011).

Survey 1

In 2011, a baseline survey was conducted from (n=523) registered (working with project) smallholder dairy farmers through personal interview. It was a generalized survey containing several questions regarding whole farm practices. It includes ongoing calf rearing practices such as calving season of buffalo and cattle, colostrum feeding, calf husbandry and nutrition. The baseline survey data was analyzed to assess the existing management practices, to evaluate the extension needs of dairy farmers, and to determine what farmers perceive as important factors for rearing calves.

Extension services

After the baseline survey was conducted, we developed a wider range of extension material comprised of simple, adoptable and significant impact oriented messages. Comprehensive extension material on calf husbandry, nutrition and health were disseminated to the farmers. We have adopted a whole family approach of extension in which we trained all the family members (male, female and their children). Training was provided to the same registered farmers on a monthly basis throughout the duration of project. Every month one fact sheet was delivered by our trained extension workers. A number of innovative extension techniques have been adopted, including the use of video practices, problem-based learning, and role plays, calf rearing competitions and direct farmer counseling in order to disseminate these extension messages.

Survey 2

In the first step, we filtered one extension message out of many other key messages regarding calf rearing through a preliminary survey (n=462) from all the stakeholders (farmers, extension workers and academia) involved in various project activities, in order to segregate one simple, adoptable and significant impact oriented message. We followed this process because it was very difficult to capture the adoption rates of all extension messages. Ad libitum colostrum feeding before expulsion of placenta was chosen as the most important message.

In 2013, we carried out a follow up survey from the same group of registered farmers (n=427) and a control group of farmers (n=105) from the same villages in order to compare the impact of improved extension services on adoption rate of calf rearing practices. This control group had never attended project training sessions.

Statistical analysis

Logistic regression was used to assess the simultaneous effect of both surveys on ad libitum colostrum provision. Analyses were conducted using GenStat Release 6. In the second study, 96 farmers could not be interviewed due to their unavailability. The data of these missing farmers were excluded from all the analysis. Impact of improved extension services on adoption of colostrum feeding practices were compared using 2-test for proportion test (SPSS version 10.0).

Results

Survey 1

Calving rates were significantly higher ($P < 0.001$) in buffalo during July to September and during March in cattle compared to other months of the year. Various calf husbandry practices under the smallholder dairy production system are presented in Table 1. The majority of the smallholder farmers (60%) did not offer colostrum to their calves immediately after birth and instead waited for the expulsion of the placenta which can occur 24 hours post-partum. Most of the farmers (55%) weaned their calves off milk at 6 months while 39% retained calves on their mother up to one year of age. Some farmers started offering green fodder and concentrate to their calves from one month of age with many starting this procedure at 2 months of age (Figure 1).

Districts	No. of farmers	Calves tied up continuously (%)	Calves housed with adult animals (%)	Calves tied part of day (%)	Dehorning (%)	Nivel disinfection (%)
Kasur	76	57 (75)	29 (38.1)	7 (9.2)	6 (8)	6 (7.8)
Pakpattan	91	78 (85.7)	40 (43.9)	7 (7.6)	5 (5.4)	7 (7.6)
Jhelum	101	77 (76.2)	36 (35.6)	15 (13.8)	1 (0.9)	9 (8.9)
Bhakkar	106	101 (95.2)	78 (73.5)	2 (1.8)	32 (30.1)	27 (18.1)
Okara	149	112 (75)	61 (40.9)	28 (18.7)	32 (21.4)	26 (17.4)

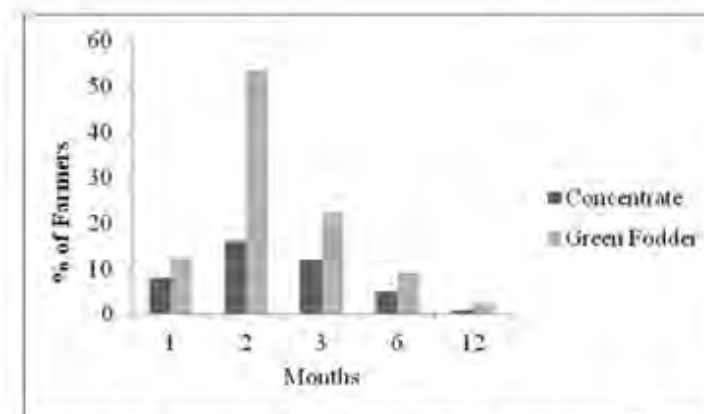


Figure 1. Age at which farmers start offering green fodder (■) and concentrate (■) to their calves in Punjab.

Survey 2

The proportion of trained farmers who used improved colostrum feeding practices was 94% which was significantly ($P < 0.001$) higher than the use of these practices (20%) in the control untrained group (20%) of farmers. Of these trained farmers 89% fed colostrum before placenta expulsion which was significantly higher ($P < 0.001$) than the 21% use of this practice in the control farmers. Of the trained farmers

Table 2. Impact of provision of extension services on adoption of improved colostrum feeding practices

Discussion

To our knowledge this is the first report which clearly describes the impact of improved extension services on adoption rates of calf rearing practices under smallholder farming systems in Pakistan. A higher adoption rate (89%) of colostrum feeding before placenta expulsion was achieved when we introduced a whole family approach. An effective extension program with participation by the whole family is highly desirable to enhance farm productivity.

Many organizations aiming at improving smallholder dairying fail to appreciate this fact and ignore women and children in their training and skills development programs. Women normally cannot leave their home and families for a few days to participate in training programs and usually require female trainers for effective communication and for cultural reasons. This approach, along with training of the male farmers, resulted in higher adoption rates. The present study demonstrated that the highest number of calvings occurred in buffalo during July to September and during March in cattle. This finding is consistent with the earlier report (Hassan et al 2010). The factors responsible for most of the calving occurs during these specific months may be due to greater fodder availability, seasonal adaptability, genotype and management. Factors affecting calving patterns of various breeds need to be investigated.

The present study captured the various ongoing calf rearing practices under the smallholder farming systems in the Punjab. It shows that most of the smallholder farmers (60%) do not offer the colostrum to their calves immediately after birth and wait for the expulsion of the placenta. This finding is in agreement with a previous study where up to only 20% of calves received colostrum within 2-3 hours of birth (Ahmad et al 2009). Local mythology dictates that if farmers feed colostrum immediately after birth, it may lead to diarrhea in the newborn and retention of placenta in the dam. However, in ruminants, transplacental transmission of antibodies does not occur and the newly born calves are passively immunized by colostrum feeding alone.

The neonatal digestive system can absorb antibodies for up to 24 hours (Bush and Staley 1980). As the intestinal cells mature, they lose the capability for absorption, so early colostrum feeding is critical and preferably within 12 hours of calving. The present study also demonstrated that the majority of farmers tied up their calves continuously for the whole day within the confines of the larger herd.

Most likely this leads to chronic stress. The housing management of the livestock seriously affects their health and productive performance, especially in calves which are more prone to diseases as their immunity level is low (Tiwar et al 2007). The majority of farmers do not disinfect the navel cord after birth and this is a common cause of navel cord infection in calves. In particular, the navel cord is a source through which pathogenic agents can enter the body and cause serious infections in newborn calves.

All these poor calf-rearing management practices contribute to high calf mortality, which can reach 60% on some smallholder buffalo farms (Ahmad et al 2009). Calf mortality has a relation with other key management practices such as the provision of a clean environment and proper high quality feeding management: if the farmer manages all aspects of health related to calf rearing satisfactorily, then calf mortality can be reduced significantly.

This study clearly demonstrates that by providing appropriate extension services to smallholder dairy farmers in Pakistan calf rearing practices can be greatly improved.

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Economic Comparison of Feeding Buffalo and Cow Milk to Kundhi Buffalo Calves in Pakistan

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Calves play a significant role in the development of every dairy sector. Rearing healthy calves through provision of appropriate nutrition and health management practices ensures a productive herd. Generally, calves in Pakistan are neglected because of their high feeding costs and low returns from sale at weaning age (Wynn et al. 2009). Profitable calf rearing strategies are needed to encourage the farmers to raise calves for dairy, meat or breeding purposes.

Feeding buffalo milk to calves is more costly than feeding cow milk because it has a higher milk fat content which is preferred by consumers in Pakistan and hence, it receives a better price. The objective of this study was to compare the economics of feeding buffalo and cow milk to Kundhi buffalo calves in Pakistan. Sixteen calves after parturition were randomly divided into two groups and housed in individual pens.

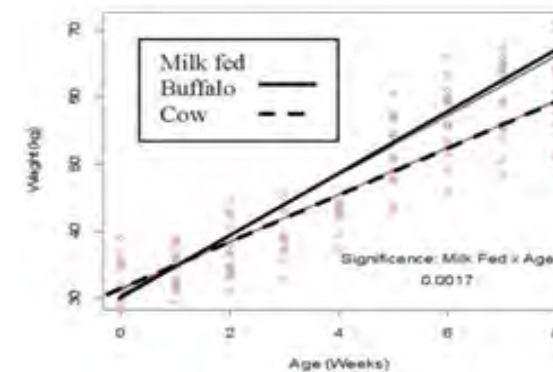
All the calves were offered colostrums ad libitum. Calves in each group were offered either cow milk or buffalo milk at a rate of 15% of their body weight. This was adjusted weekly with the maximum amount offered set to five litres per day. Ad libitum calf starter ration was offered from the third day to both groups. The milk was gradually withdrawn from day 42 until weaned completely at day 56. Milk samples were collected weekly in the morning and evening and analyzed for fat and protein content. Weight gain was measured weekly along with body measurements to track growth performance and health status.

Results show that calves offered buffalo milk for the 8-week duration of the study have significantly ($P < 0.05$) better growth performance than calves fed cow milk in the pre-weaning period (Figure 1). Therefore, to maximize growth performance buffalo milk would be preferred. However, this study also shows that buffalo calves can be successfully reared on cow milk. At weaning the average liveweight of the calves fed buffalo milk was 65.0 ± 4.1 kg and those fed cow milk was 58.7 ± 5.3 kg. The corresponding difference in feed costs was 1850 Pakistani rupee less for the cow milk fed group which would not significantly alter the profit of the enterprise.

Therefore it is recommended that feeding cow milk is a viable option for small-holder dairy farms who are trying to lower costs whilst maintaining a profitable growth rate for buffalo calves.

Wynn, P.C., Warriach, H.M., Morgan, A., McGill, D.M., Hanif, S., Sarwar, M., Iqbal, A., Sheehy, P.A. and Bush, R.D. (2009)

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Factors Affecting the Quality of Colostrum in Buffalo

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Calves play a significant role in the development of every dairy sector. Rearing healthy calves through provision of appropriate nutrition and health management practices ensures a productive herd. Generally, calves in Pakistan are neglected because of their high feeding costs and low returns from sale at weaning age (Wynn et al 2009). Profitable calf rearing strategies are needed to encourage the farmers to raise calves for dairy, meat or breeding purposes.

Colostrum contains a large amount of serum immunoglobulins and it is the primary source of immunity which can be absorbed by the calf in the first 24 hours of life. Newborn calves are born without a measurable concentration of serum immunoglobulin (IgG) which is critical for proper immune function (Barrington et al 2001). It is generally accepted that calf mortality is caused by improper colostrum feeding management in this first 24 hours post-partum. Colostrum quality, assessed by measuring density, depends upon many factors including the time from the previous lactation (dry period) and feeding level and parity of the dam. There is limited data available on the quality of colostrum from buffalo (e.g. Arumughan and Narayan 1981).

The objective of this study was to evaluate the factors affecting the quality of colostrum in buffalo and the relationship between this parameter and live weight gain to 8 weeks of age. A total of 16 pregnant buffaloes in the last trimester were selected from a number of different commercial dairy farms in the Hyderabad area of Sindh, Pakistan. All buffaloes were kept under a similar feeding and housing system. Information on the length of the dry period and parity of the buffalo was collected from the records maintained at the farm. Colostrum quality was analyzed using a colostrometer which predicts the IgG content based on viscosity. Serum protein levels were also measured on the third day of age with a refractometer. The relationship between colostrum density, the duration of the dry period and parity of the dam was analyzed using linear regression.

The results from this study show that the colostrum immunoglobulin content assessed with a colostrometer ranged from 60 g/L to 120 g/L with an average of 92.5 ± 17.1 g/L. The corresponding dry periods from these buffaloes ranged from one to four months with an average of 3.0 ± 0.72 months. The correlation between dry period and colostrum quality was high with a value of 0.74. The regression analysis showed that the length of the dry period had a significant ($P < 0.05$) effect on the colostrum quality (R^2 value of 0.55 and residual standard error of 1.95) and that each additional month a buffalo was dry yielded an increase in colostrum quality of 17.6 g/L. The results also show a dry period of at least three months is required to ensure a good colostrum quality of over 90 g/L. In contrast, the parity of the 16 buffaloes measured in this study (average 4.38 ± 1.20) did not have a significant effect on colostrum quality ($P > 0.05$). However, the relationship between colostrum quality and live weight gain over 8 weeks was not significant. Therefore, it was not surprising that the duration of the dry period and live weight gain over 8 weeks were not significantly correlated.

Therefore, the conclusion from the study is that farmers should ensure the dry period of their pregnant buffaloes is long enough to maximize colostrum quality in the ensuing lactation to improve calf survivability.

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Factors Affecting the Milk Price in a Small-holder Dairy Production System of Pakistan

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There are numerous factors that contribute to milk price in the Pakistan dairy production system. Buffalo (Bubalus bubalis) milk is most often preferred because of its high (>6%) fat content. Factors that influence milk price received on farms are seldom analysed, which makes it difficult for farmers to make management decisions that will improve their profitability. The aim of this study was to determine factors affecting buffalo milk price in small-holder dairy production systems in Punjab province Pakistan. Dairy farmers (n=523) from seven districts of Punjab & Sindh were surveyed on different factors affecting buffalo milk price. Questions were asked related to locational, seasonal, marketing and educational factors that impact their dairy enterprise. Data were log transformed to normalise variance and factors were analysed independently in a univariable analysis.

Table 1: Factors that influence the price (PKR/Litre) received on farm for buffalo milk.

Predictor Group	Mean	SE	P value	
Location/district	Bhakkar	34.96	0.50	<0.001
	Jhelum	39.13	0.55	
	Kasur	36.49	0.51	
	Okara	33.52	0.35	
	Pakpattan	30.55	0.47	
Education	none	33.34	0.60	0.029
	primary+middleschool	34.51	0.34	
	matriculation	35.64	0.43	
	intermediate	35.58	0.95	
	university	34.68	0.92	
Season	winter	33.75	0.31	<0.001
	summer	35.77	0.33	
Milk buyer	big milk company	35.95	1.00	<0.001
	Small buyer/dodhi	33.31	0.24	
	neighbour/villager	39.81	0.55	
	other	32.88	1.59	
Milk contract	supplied under contract	34.67	0.44	0.400NS
	supplied to free market	34.92	0.31	
	other	31.19	2.68	
Primary farm activity	cropping	34.91	0.33	0.703NS
	dairying	34.45	0.36	
	dairying+cropping	35.11	0.89	
	other activities	35.43	1.22	

Location, educational achievement, season and type of milk buyer all influence milk price significantly. Factors that did not influence milk price received included the relative importance of cropping compared to livestock to the enterprise and whether milk was produced under contract. The results have shown that there is scope for improvement relating to the process of education: thus the further refinement of outreach training programs is warranted. The higher price for milk in summer reflects both increased demand and a scarcity of supply. The mechanism of marketing milk through traditional marketing chains also deserves further attention.

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Transfer of Aflatoxin from Highly Contaminated Feed to Milk and Effect of Mycotoxin Binder on Transferrate in Nili-Ravi Buffaloes

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Fungal contamination leads to production of mycotoxins that provide a major limitation to dairy production worldwide and a key health hazard for consumers. This experiment was conducted to: a) observe the extent of carryover rate of aflatoxin B1 in feed to the aflatoxin M1 metabolite in milk in Nili-Ravi buffaloes and b) evaluate the efficacy of a commercial mycotoxin binder (Mycofix®, Biomin Singapore) incorporated into feed to reduce this transfer. Multiparous buffaloes (n=28) were randomly distributed to four groups corresponding to 2 levels (high and low) of aflatoxin B1 each with 2 treatments (with and without mycotoxin binder) in a factorial design. Animals were offered mycotoxin contaminated concentrate ration and corn providing a total of 1475 µg/day (groups A and B) or 2950 µg/day (groups C and D) of aflatoxin B1. Groups B and D were given 50 g of mycotoxin binder daily mixed with feed while groups A and C were kept as controls. Feed samples were analyzed by HPLC (Romer Labs, Singapore) for aflatoxin B1 and milk samples were evaluated by ELISA (AgraQuant® Aflatoxin M1 Fast ELISA kits) for the liver metabolite aflatoxin M1. The total daily mean aflatoxin M1 concentration in milk for animals fed 2950 µg/day of aflatoxin B1 was significantly (P<0.001) higher (112.6 µg) than those fed 1475 µg/day (62.2 µg; SED=5.99). The mean daily concentration of aflatoxin M1 in milk of animals from both treatment groups fed with 50 g of mycotoxin binder was significantly lower (76.5 µg) than those without binder at 98.3 µg (SED=5.99; p<0.001). The interaction of binder and treatment was not statistically significant, i.e. the 50 g of binder was able to sequester aflatoxin B1 with the same efficiency in groups fed with high and low concentrations of aflatoxin B1. Effect of aflatoxin B1 intake and mycotoxin binder on milk production and carryover rate into the milk is given below (Table).

Table 1: The effect of aflatoxin B1 intake and use of a mycotoxin binder on milk production, concentration of aflatoxin M1 in milk, total aflatoxin M1 excreted and calculated carryover rate of aflatoxin M1 into the milk of Nili-Ravi buffaloes

Variables	Low aflatoxin intake		High aflatoxin intake		Significance level (SED)		
	Without binder A	With binder B	Without binder C	With binder D	Effect of aflatoxin	Effect of binder	Effect of Aflatoxin x binder interaction
Total aflatoxin B1 intake (µg/day)	1475	1475	2950	2950	N/A	N/A	N/A
Milk production (kg/day)	9.3	8.9	8.4	10.2	P=0.831 (0.71)	P=0.357 (0.71)	P=0.181 (1.0)
Concentration of aflatoxin M1 (µg/kg)	8.1	5.6	14.6	10.3	P<0.001 (0.44)	P<0.001 (0.44)	P=0.051 (0.63)
Total aflatoxin M1 excreted (µg/day)	74.6	49.8	122.0	103.3	P<0.001 (5.99)	P<0.001 (5.99)	P=0.613 (8.48)
**Carryover rate of aflatoxin M1 into milk (%)	5.06	3.37	4.14	3.50	N/A	N/A	N/A

**Carryover rate of M1 into milk calculated as total aflatoxin B1 excreted / total aflatoxin B1 intake × 100

The addition of Mycofix® to the diet of buffalo has a clear inhibitory effect on mycotoxin transfer to milk, which is similar to the efficacy of this binding agent on controlling mycotoxin transfer to cows' milk (Pietri et al 2004)

Pietri A., Bertuzzi T., Piva G., Binder E.M., Schatzmayr D. and Rodrigues I. (2009). Inter. J. of Dairy Sci. 4, 34.

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VOLUME 1

Utilisation of conserved forage to improve livestock production on smallholder farms in Asia and Africa

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Abstract.

Ruminant livestock are essential to the livelihoods of smallholder farmers in many developing countries. Livestock production on these farms is characterised by low milk production, low live weight gain and poor reproductive performance because of poor nutrition. Access to high quality forage has been identified as key to improving livestock health and productivity. Conservation of surplus forage as hay or silage provides the opportunity to ensure livestock have access to high quality forage year-round. This paper reports on forage conservation in select countries in Asia and Africa.

Keywords: Forage conservation, silage, hay, smallholder, livestock.

Introduction

Livestock are an important livelihood strategy in most developing countries (Otte et al. 2005; Otieno et al. 2006; Rao et al. 2009) by providing meat, milk, manure, hides, draught power and collateral (financial asset, insurance). Smallholder cattle farming is the major source of livelihood for over 54% of poor people in sub-Saharan Africa (IFPRI 2007) and over 70% of the population in Pakistan is directly involved with livestock as a primary source of food and/or income (Devendra and Thomas 2002; Thomas et al. 2002). Furthermore smallholder livestock farms are the major meat and milk producers in developing countries of Asia and Africa. Despite their importance, livestock are usually undernourished due to lack of feeds of sufficient quality and quantity; the consequences of which are low production, increased disease susceptibility, high mortality rates and reduced fertility (Xu et al. 2005). This is typical of livestock production systems on smallholder farms in developing countries where traditionally farmers have been livestock keepers/users.

Lack of high quality forage, which is influenced by seasonal supply and competition with neighbouring farmers for the available forage resource is the major challenge faced. The majority of livestock are part of integrated crop/livestock systems and low quality crop residues represent a significant proportion of the total annual diet, particularly where livestock are tethered for all or most of the year. In regions dependent on grazing, access to high quality feed is seasonal and dependent on rainfall and temperature. Opportunities for improvement involve forage production and conservation as well as efficient use of local feed resources. This paper outlines experiences with forage conservation and using conserved forage to improve livestock production on smallholder farms from some countries of Asia and Africa with a diverse range of environments and situations.

China

The Qinghai-Tibetan Plateau is one of the three most important livestock production areas in China. It has a harsh environment and insufficient feed supply during the long cold season (November to June). With an area of 129.3 million ha, it accounts for 32.5% of China's total grassland area and supports about 41.5 million sheep and 13 million yaks. Year-round survival of the Tibetan livestock is largely dependent on the native alpine pastures, which are characterized by high altitude (>3,500m), very low annual average temperature (-5 to 1°C), a short growing season (from June to September), low productivity, without an absolute frost-free period, and extensive seasonal variation in feed supply. Animals in the traditional grazing system often suffer from poor nutrition, health-related problems, low production and low fertility, and losses of up to 30% have been reported during some extended cold seasons (Xue et al. 2005). Moreover, severe degeneration of the native alpine pasture has extended the gap between forage supply and animal feed demand in recent decades.

Conservation of forage and feed supplementation are important strategies to sustain animal farming systems in the cold season. Hay of cultivated forage, generally oat (*Avena sativa*), and straw residues from crops, such as oat and barley (*Hordeum vulgare*) have been used as winter supplements by local farmers since the China Reform of 1978 (Wu 1996). In the agro-pastoral areas of the plateau, haymaking is a universal and traditional way for fodder conservation. In purely pastoral areas of the plateau, herders normally buy oat hay or highland barley straw from the agro-pastoral zone. However, only limited amounts of oat hay are provided to lactating and weak animals throughout winter or to the whole herd during a severe snow disaster (Cai and Wiener 1995) when losses due to starvation frequently exceed 20% of a household's herds or flocks (Zhang and Zhang 2002); and most animals are not given any supplements during the cold season (Long et al. 1999). Local herders also harvest grasses from fenced winter pastures and conserve this as hay. Research has shown that supplying oat hay or barley straw to yak herds during winter time could improve calving rate by 19 to 25%, and considerably reduce body weight loss compared to unsupplemented yaks (Long et al. 1999). Silage with high nutritive value and palatability has been made by local farmers from either native grasses or crops. However, temperature during harvest in most regions is only 10 to 15°C and commercial inoculants do not produce well fermented silage (Zhang and De 2007). Local farmers have been asked to spray several indigenous inoculants e.g. diluted yak milk yoghurt, onto the wilted and chopped forage at ensiling in order to make high quality silage in this unique environment. Research has shown that feeding goat silage to pregnant Tibetan sheep grazing pastures increased ewe body weight and lamb birth weight (Hua et al. 2012). Another study showed that supplementation of native grass (*Elymus nutans*) silage to finishing Tibetan sheep is an economical and efficient way to improve daily weight gain on the Tibetan Plateau (Feng et al. 2009). Forage conservation or feed supplements in the growing season or in the winter/spring is a promising technology for many areas, and especially for those areas where snow storms frequently occur.

Indonesia Smallholder farming, with average areas of 0.5 ha, is the predominant system in Indonesia. Smallholder farms use a mixed cropping strategy, involving first sowing subsistence food crops (mainly paddy rice) in the rainy season, followed by cash crops (e.g. peanut, bean, maize) in the early dry season. As part of their farming system, farmers also raise a small number (2-5) of cattle. Indonesia's traditional beef production sector makes an important contribution to the country as the second largest contributor to meat supply (19%) after broiler chickens, as well as providing employment and income for over 4.57 million rural families.

Smallholder farmers rely on natural grass as the major component of cattle feed using the 'cut and carry' feeding system, which is time-consuming for farmers. High quality forage has become less available because previously uncultivated land is now being used for crops. When forage is scarce and of low quality e.g. during the dry season, cattle are under nutritional stress and low quality straw and locally purchased, available, cheap agricultural waste and by-products are often fed. Only a small number of farmers practice forage conservation. Various approaches to fodder conservation have been trialed and a number of forage conservation technologies including silage, urea-ammonia treatment of straw and hay production have been delivered to the smallholder farmers in Indonesia. Both silage and urea-ammonia treatment of straw pose problems for farmers and are not widely used. A survey of 96 smallholder farmers in the district of Bulukumba, South Sulawesi showed that while the majority of farmers (56.25%) understand the principals of forage conservation, a sizeable proportion (43.75%) do not know how to conserve forage effectively.

Hay is the most promising technology that smallholder farmers have for cattle or buffalo feed, mainly in Java and some outer islands. Farmers commonly collect dry straw, mainly rice straw, and store it in a stall near their house. Priyantietal.(2012) reported that about 74% and 80% of farmers in lowland and upland East Java, respectively collected rice straw from their own and other fields, while 24% of farmers purchased from other farmers or agents. Farmers also collect peanut, mungbean, and corn stover. Farmers often fed the straw plus other supplements such as rice bran, tofu waste, or tree legumes to meet animal nutrient requirements. This is in line with current findings that supplementation strategies improve the utilization of low quality forage by animals. For example, Syahniaretal.(2012) reported that supplementation with tree legumes at a level of 2.8g DM/kg BW per day was sufficient to meet the maintenance energy requirements of Ongole cows fed rice straw and bran. Moreover, Marsetyoetal.(2012) suggested that addition of Gliricidia or a mixture of copra meal and rice bran increased feed digestibility and live weight gain of Balicalves given Elephant grass hay or corn stover as a basal diet. Such integration between animal and crop production is a traditional practice in the villages but could be improved through education on how to create more nutritious diets.

There has been no economic evaluation of the impact of forage conservation practices on beef cattle smallholder farmers. The only observation in Central Sulawesi and South Sulawesi on forage management indicated that conserved forages, mainly hay making of straw (rice, peanut, mungbean, corn stover), have led to a substantial labour savings by decreasing time feeding from 4-6 hours down to 1-2 hours/day. With better feeding management, the body condition of cattle improves, which leads to a better price when sold. The additional income is used to purchase goods such as motorbikes that are then used to take children to school, and for carrying forages, thus saving even more time, or in some cases, allowing smallholder to take off-farm work. Haymaking has led to an increase in the price of straw which also potentially provides a source of additional income to farmers.

Kenya

Kenya has a national dairy cattle herd estimated at about four million (MoLD 2011) and about 80% (Friesians, 1733 Ayrshires, Guernseys, Jerseys and their crosses with local zebu) are found on small-scale farms and produce about 80% of the marketed milk. Kenya experiences a bimodal rainfall pattern and the distribution of livestock feed closely follows this pattern resulting in periods of feed shortage between the rainy seasons. The situation is worsened with frequent droughts. Therefore, there is a need to conserve the excess feed produced during the rainy season to stabilize feed supply throughout the year.

Forage conservation is done as standing hay (particularly in the Arid and Semi-Arid Lands (ASALs)), baled hay or silage. There are certain feeds available on these farms which would be more suitable to conserve as silage than as hay; maize stalks, sugarcane (*Saccharum officinarum*) tops, Napier grass (*Pennisetum purpureum*) and sorghum (*Sorghum bicolor*) stover. However, the major limitation to the use of these methods by small-scale farmers is the lack of simple and appropriate technologies for hay baling and ensiling.

In Kenya, drying as the means of conserving grass species has been a common practice, but currently, ensiling of various forages is being promoted by several government agencies and other development partners. Production of high quality hay, by harvesting at full maturity, increases yield of grass per unit of land. The technology of ensiling forage in nylon bags has been taken up in some smallholder production systems but there are various constraints to adoption. These include the high cost of the bags, lack of suitable choppers and inappropriate storage resulting in losses of ensiled material to rodents. Ensiling using large plastic bags placed horizontally can be an alternative for medium and large scale farms, but small scale farmers find this a challenge. For the smallholder farmers, tube silage is recommended especially where the farmer has a small quantity of material to ensile. One tube can hold up to 500kg, however small tubes are also available and the size chosen largely depends on the quantity of material to be ensiled. Where a farmer has a larger quantity of forage to ensile the above ground (silage bun) method of making silage is preferred.

Smallholder farmers in Kenya are increasingly making tube silage, a low cost procedure which requires high quality fodder, a silage tube (polythene tube 1000' gauge), molasses and polythene sheet gauge 500. One tube may contain from 350-500kg of silage or compacted material and therefore becomes difficult to move. When the tube is full and tied at both ends, the farmer has a large cylindrical airtight bag in which the forage mixture ferments, turning into silage. The technology works with a wider range of green fodder. The ensiled material will

be ready for use in the dry season after 3 months and can be conserved for even 30 years without losing quality provided the airtight seal remains intact.

Pakistan

Pakistan has an estimated 160 million ruminant livestock, comprising buffalo, cattle, goats, sheep and camels, but is not self-sufficient in milk, meat and other products of animal origin. More than 90% of all ruminant livestock are integrated with small-scale mixed farming systems (Devendra and Thomas, 2002; Thomas et al. 2002). Pakistan is the fourth largest milk producer in the world (34 billion litres per annum), but productivity is low due to poor nutrition, poor management and neglect of health problems which results in late maturity, extended calving intervals and low milk production (Farooq, 2011). More than half (51%) of total revenues from dairy sales are accrued by small holders having 1-4 animals, with each of these household generating a monthly income of US\$60 to \$240. Approximately 60% of the milk produced by small holders is consumed at home, and the remaining 40% is marketed (FAO 2011).

Fodder scarcity is the core constraint, with current supply 40% less than demand and there are two periods of severe scarcity each year; May to June and October to November. Farmers rely heavily on low quality crop residues (Thomas et al. 2002); approximately 40 million tons (46% of the total animal feed resources) of crop residues are fed annually. Yields of traditional forage crops are less than half of a quarter of potential for improved hybrid varieties when grown with traditional agronomic practices. Improved varieties grown with recommended farming practices have a yield increase of around 75% (Din 2008).

Silage technology was introduced into Pakistan almost two decades ago by different government and international agencies, and also private sector companies like Nestlé Pakistan and Pioneer Seeds. However, despite heavy inputs in terms of time and money, uptake of silage technology has been less than hoped for in traditional livestock feeding and production systems. This is because, in many parts of the country, small holder farmers are still able to feed crop residues and agricultural by-products during the dry season. However, more recently maize and sorghum silages have become more popular with a key group of small holder farmers that have the necessary skills and are able to routinely produce well-fermented silage. Berseem silage making is also getting acceptance in the farming community though the high moisture content at the time of silage making can be problematic.

Silage making systems include Little Bag Silage (LBS), where forage is ensiled in strong plastic bags with a capacity of 5 kg of chopped green forage (Lane 1999). LBS allows conservation of available fodder in small quantities over a long period of time and is suitable for the small farms producing only a couple of bags over a 100 day growing season. Silage making in small (1.2m x 0.7m) drums was extensively promoted by Pakistan Dairy Development Company (PDDC) to a group of model small-holder farmers during 2007-2008 with training provided. Chopped maize, sorghum and berseem were ensiled successfully and produced excellent results when fed to livestock. Milk production remained steady throughout the year, was better than for neighbouring farmers, and with surplus silage being available for sale. Farmers are now prepared to buy silage, indicating a willingness to feed silage, but a lack of awareness on how to make it. Small holders could now engage in entrepreneurial activity and produce silage as a marketable commodity.

Rwanda

Agricultural production in Rwanda is based on mixed crop-livestock production (Rutamu 2004) and feed resources remain a major constraint to livestock development. Grazing lands are sharply shrinking because of encroachment of crop cultivation with increasing human pressure (Mutimura and Everson 2011). As land holdings are small, with over 60% of households cultivating less than 0.7 ha, livestock owners practise zero-grazing (cut and carry). Napier grass (*Pennisetum purpureum*), introduced in the 1970s, is adapted to the cut-and-carry system, and it constitutes 95% of animal feed resources (Staal et al. 1997; Kamanzi and Mapiye 2012). Herbage production is high but its nutrient concentration is too low for dairy cows. Other feed resources like fodder trees (e.g. *Leucaena* sp., *Calliandra* sp.) are used to supplement the Napier grass. Grasses including *Chloris gayana*, *Brachiaria* sp. and *Cenchrus ciliaris* are also used by large and small-holder farmers (Lukuyu et al. 2009), as are crop residues such as sweet potato vines, banana leaves and banana pseudostems, stovers and straws. Except for Napier grass (*Pennisetum purpureum*) which is conserved as silage, other grasses e.g. *Chloris*

gayana, *Cenchrus ciliaris*, *Brachiaria* sp. are conserved as hay (Mutimura et al. 2007, unpublished). Farmers rely heavily on these feeds as a strategy to cope during the dry season (Kamanzi and Mapiye 2012).

Forage conservation has been challenging for farmers in Rwanda, especially smallholder farmers who have limited land size. For example, when ranking feed resources in central and southern plateau areas of Rwanda, Kamanzi and Mapiye (2012) found that conserved feed was the feed resource least used by smallholder dairy farmers. However, in peri-urban areas, dairy farms (small or large) use silage to feed their dairy cows (Nyiransengimana and Mbarubukeye 2005). Lack of technical skills has been identified as the cause why farmers fail to adopt forage conservation and this stops farmers from coping with feed shortages, especially during the dry season (Lukuyu et al. 2009). Except for isolated research results, Rwanda has very limited experience with the treatment of crop residues to improve quality, especially digestibility and intake. However, a recent experience in Rwanda proved that ammoniated straw-based rations for feedlot beef production is economically feasible even when using indigenous Ankole steers (Mpangwa et al. 2010).

A seed bank of improved forage has been developed in Rwanda to increase quality forage production. The use of improved forage was adopted by at least 5,000 dairy farmers, and subsequently imports of protein-rich concentrates were reduced by 30% (Ndabikunze 2004). About 50 farmers are generating income from sales of hay in the Eastern Province of Rwanda while more than 200 farmers in the Western Province get income from sales of fresh Napier grass to dairy farms who use it to make silage (Mutimura and Ebong 2010, unpublished data). This technology has been coupled with the use of conserved crop residues as roughage to satisfy ruminant feed supply. Different technologies for conserving forage/feeds are available in the country, and improved forage cultivation and conservation, including hay and silage making technologies have been disseminated to smallholder and commercial dairy farmers. Farmers' capacity to apply the introduced technologies has been enhanced through a series of training workshops and practical on-farm training sessions (Ndabikunze 2004).

Discussion

Growth in the agricultural sector in developing countries has been identified as the key to poverty alleviation (Cervantes-Godoy and Dewbre, 2010). Improving livestock productivity through improved nutrition is a critical component of this growth. Access to more and better quality forage provides the resource needed for improved livestock nutrition on smallholder farms. Given the constraints of land, water and the need to produce food for human nutrition this will require access to improved, high yielding and adapted forage varieties combined with better management to achieve potential yields. In addition, access to high quality forage year round by smallholder farmers will require production of surplus forage that can be conserved as either hay or silage, and stored for use when forage quality or quantity is limiting. Smallholder farmers still need to be encouraged to adopt better forage production, conservation and feeding technology to improve animal production. This will require targeted extension services with clear goals. Forage conservation systems will need to be tailored to the specific needs of the different farming systems, be low cost and provide a safe storage method that are without risk of damage, e.g. from rodent attack. In areas where hay making is difficult due to wet weather or low temperatures, and for certain crops, e.g. maize, the preferred means of conservation will be silage (Mannetje 2000). Farmers making silage will need additional skills because a number of factors influence fermentation quality. Fermentation quality is important because poorly fermented silage is unpalatable and, even if high in energy and protein will only support low intakes. As a result milk production or live weight gain will be low.

Conclusion

Despite a diverse range of environments and situations, experiences in production of high quality forage, forage conservation and developing local feed resources have been proved as an efficient way to increase livestock productivity, and the livelihoods of smallholder farmers, in developing countries.

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Effect of body conditions score on milk production and reproductive in buffalo

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ABSTRACT

The objective of this study was to determine the effect of BCS on milk production and reproductive disorders in Nili-Ravi buffalo under field conditions of Pakistan. The data was collected from 55 small holder farms from districts Kasur (n= 26) and Okara (n= 29) respectively. Data regarding milk production and reproductive disorders were collected by trained veterinarian. The result showed that buffaloes having high BCS (4, 5) have significantly ($P > 0.05$) higher milk production as compared to low BCS (1, 2). Similarly, data showed that reproductive disorders were significantly higher ($P > 0.05$) in low BCS (1, 2) as compared to high BCS (4, 5).

Keywords: Nili-Ravi, body conditions score, milk production, reproductive disorders

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Establishing A Successful Dairy Community Development Program: The Challenges And Pitfalls

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Abstract

The awarding of a research grant designed to investigate the limitations to dairy animal productivity in socioeconomically disadvantaged communities in a country totally foreign to the successful applicants presents many challenges. This paper highlights some of the key challenges that have been arisen throughout the 6 years of our dairy extension program awarded through the Agriculture Sector Linkages Programme established as a co-operative agreement between the Pakistani and Australian governments. Successful sustainable programs depend on the identification of the right partner organizations and staff to conduct the program and then the keeping of accurate records to measure and report on the success of farmers to both sponsors and host organizations.

Introduction

Pakistan is heavily dependent on an agrarian rural based economy, with livestock contributing 50% to the country's economy (Pakistan Economic Survey, 2006). The predominance of villages are populated with farmers owning 3-10 dairy animals, with much of the milk consumed domestically, while a small proportion is sold into either the local community or to milk buyers operating in a marketing chain culminating in the sale of milk in large cities. However, Pakistan's standing as the world's 4th largest milk producer relies on production of less than 6 litres per day per animal from over 50 million buffalo and cattle. Our aim was to improve the productivity of the national herd.

Materials and Methods

Forming the right partnerships as an Australian dairy scientist funded to investigate ways of "revolutionising" the commercial acumen of these farmers through co-operation with organizations already working in the field with them, it was difficult to know where to start. Governmental institutions like the state or federal Livestock Ministries are the most likely choices on offer from governments. The advantage of working with a large bureaucracy like the state Livestock Ministry which employ hundreds of veterinary officers servicing the needs of millions of dairy farmers is that there is a structure to work with. Furthermore, there is scope for extension of their activities as only about 20% of small-holder farmers receive any assistance at all.

The disadvantages are that the organization is most often hampered by inertia based on a career structure for the veterinary extension staff that offers no incentive to excel. Some officers can wait for as long as 20 years for promotion beyond base grade. There is no mechanism for promotion based on performance. Thus engaging these staff in a new performance based paradigm presents a real challenge.

The other issue relates to the management of finance, which once sent to the ministry to fund the project, becomes difficult to access because of the bureaucratic process of approval of expenditure. Delays in fund release can cause major delays in progress with program inception. Working with NGO's is an ideal objective, but their activities can change markedly over a short period: therefore there is no continuity in support. Similarly, smaller co-operative companies have their own commercial objectives which often do not align with the welfare of farmers.

Co-operation with other aid agencies

Some countries are washed with funding from a range of international agencies most often with a motive to service the political agendas of countries funding the organization. Their objectives most often overlap, and yet because their funding is dependent on the delivery of a work program decided at the point of signing of contracts, there is little incentive or scope for co-operation or sharing of resources between projects. In some cases, the program can be designed by one consultancy and then implemented by an entirely independent organization which might not be able to deliver effectively.

Shifting objectives

The political agendas of governments can change and therefore the emphases of projects can also change in line with these. A sudden push for a “pro-poor” approach to aid, for example, means that flexibility needs to be built into every project to meet the needs of a never-changing political environment.

Choosing the right staff

The rule of thumb is to choose young resilient expatriate staff to immerse directly into the environment in which the project is to be implemented. Their decision-making processes will not be influenced by previous project management experience. They then need to have an advisory group who visits on a frequent basis to provide some guidance with program direction and also to assure senior bureaucrats in partner organization that back-up assistance is never far away. Then newly trained local graduates, who will be compliant with the aims of the project, need to be chosen carefully. Having the right staff in place in both the operational team and among the partner organizations is critical to success. Once local staff are trained, expatriate staff need to withdraw to oversee progress with the project from afar. These staff will also need to visit the project team on a frequent basis to monitor progress and offer further suggestions.

Keeping good records

The key to success is to measure the rate of success in the targeted farming communities through accurate record keeping. This involves the development of a comprehensive survey that will provide the baseline for on-farm productivity and community development. This must be constructed carefully as the survey can be conducted only once. Thereafter, accurate on-farm record keeping is required to measure progress. The database needs to be maintained constantly and analysed at fixed times to ascertain responses to the extension activity. There is a need for constant reviewing of the project approach to ensure that the financial investment is reaping the return demanded by the funding agency.

Results and Discussion

Project sustainability: the key to success

Results can only be evaluated after the sustainability and outcomes of the project can be measured. Success is dependent on developing a working relationship with farmers and collaborating partners based on trust. Rather than offering incentives to farmers to ensure their engagement, a more effective mechanism is to initiate a project with sentinel farmers who are the innovators in any community. These leaders will soon demonstrate the virtues of the project if it is implemented correctly and others will then follow. Engagement with women and children as well as the local school teacher is a critical component for success with extension. Above all, it is essential that the project develops to keep all partners happy. This might mean adjusting the project aims to accommodate the wishes of local industry leaders. In the end, it is critical that a cadre of workers in the partner organization is left with funding provided from within their organization or from farmers to carry on the project after external aid funding ceases. Persistence combined with flexibility together with accurate data recording and reporting will always pay off in the long run.

Conclusion

Successful extension activities require total involvement of project staff with the communities they are working with. The nature of extension material has to vary to ensure maximum engagement of the whole family. At an organizational level it is essential that an established network of extension staff is in place to ensure that success with the project extends well beyond the period of funding for the program. The leaders of the program must be working cohesively and preferably are from the current generation of young graduates who are the future of the industry we are assisting.

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New Extension Model for Small Holder Dairy Farmers in Pakistan

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Abstract

The objective of the study is to demonstrate the effect of improved extension services on adoption rates of small holder dairy farmers in Pakistan. Preliminary results indicated significantly higher adoption rates when extension services were provided using the whole family approach compared to the simple approach after one year of both phases. In conclusion, this study indicates that improved extension services have a significant impact on adoption rates of small holder farmers.

Key words: Extension services, Dairy, Adoption rate, Small holder farmer

Introduction

Pakistan, like many developing countries, has an agrarian rural based economy. Livestock is a major contributor to the national (12%) and agricultural (50%) economy (Pakistan Economic Survey, 2006). It is estimated that around 70% of the dairy households in Pakistan still operate under conditions of subsistence by maintaining herds of three or four animals (Burki et al., 2005). The productivity of livestock is still lagging behind its potential level. In order to meet the requirements of a rapidly growing population, dairy production needs to be increased. This can be done by adopting modern techniques of dairy farming. New technologies developed by researchers are disseminated. Thus the major objective of this study is to demonstrate the effect of new extension model on adoption rates of small holder dairy farmers in Pakistan.

Materials and Methods

Australian Centre for International Agricultural Research (ACIAR) research project was commenced with the aim of increasing dairy production through improved extension services. Small dairy farmers having 4-10 (buffalo and/or cattle) for production were the main target group for this project. During the first phase of the project a simple approach of extension targeting male farmers was used. During the second phase of the project a whole family approach targeting all family members was utilized. A number of innovative ways of extension have been adopted including the use of video practices, demonstration plots, problem based learning, stage drama, radio and TV shows to improve the effectiveness of the program. Benchmark data were collected on whole farming systems from 228 farmers during the first phase and 292 farmers during the second phase of the project. Subsequently, at the end of every year data have been collected to monitor the impact of these initiatives on the rate on the rate of extension message adoption.

Statistical analysis

Adoption rates between the whole family approach and simple approach after one year of both phases were analyzed using a Chi-square test. Similarly, adoption rates of various modules at the start and after one year of project phase-II were analyzed using a Chi-square test.

Results and Discussion

To our knowledge this is the first report which clearly describes the effect of improved extension services on adoption rates of small holder farmers in Pakistan. Higher adoption rates were achieved when we introduced a

whole family (male, female and children) approach compared to simple approach relative to the traditional male only approach to extension. An effective extension program with participation by the whole family is highly desirable to enhance farm productivity. Many organizations aiming at improving small holder dairying fail to appreciate this fact and ignore women and children in their training and skills development programmes. Women normally cannot leave their home and families for a few days to participate in training programmes and usually require female trainers for effective communication. Thus during the second phase of the project we arranged parallel sessions for women and children co-ordinated by women trainers. This approach, along with training of the male farmers resulted in higher adoption rates.

Conclusion

Preliminary results indicate that improved extension services have a significant impact on higher adoption rates of small holder farmers in Pakistan.

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Dairy enterprise and whole farm performance in mixed farming systems: Punjab, Pakistan

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Introduction:

The share of agriculture to GDP in Pakistan is 21%, more than half of which is from livestock. Milk is the major product of livestock. Pakistan is the third largest milk producer in the world with gross annual production of 47M tonnes. 40 million of the country's rural population are dependent on livestock for their livelihood. Punjab province has the largest share of milk production. It is important to understand the economics of dairy production in Pakistan's complex mixed farming systems.

Material & Methods:

- This study used farm level data from a longitudinal survey in irrigated Okara and partially rain fed Bhakkar districts of Punjab (Table 1, Figure 1). The survey recorded milk production per animal whereas feed quantities, health costs, crops cultivated and herd composition were for each farm from January 2007 to December 2009.
- Whole-farm and enterprise gross margins have been calculated for animal, fodder, crops and vegetable enterprises.
- Labour being permanent, with no significant opportunity costs, has been taken as a fixed cost to estimate operating costs. The value of land managed and livestock owned were assumed to be the major farm assets, with a 9% finance cost per annum for estimating net profit or disposable household income.

Table 1: Mean physical and economic attributes for farms in Okara and Bhakkar districts of Punjab, Pakistan. Standard deviations (SD) in parentheses.				
Measure	Okara		Bhakkar	
Total sample size (n)	115		97	
Total land (Acres)	9.04	(7.08)	9.47	(9.00)
Herd size (hd)	10.93	(5.16)	10.50	(5.58)
Milking cows and buffaloes (hd)	3.71	(1.92)	3.77	(2.23)
Total milk production (kg)	3,238	(1,846)	3,288	(2,604)
Milk prices (Rs/kg)	22.99	(2.59)	21.14	(3.01)
Milk average variable cost (Rs/kg)	24.13	(19.19)	19.67	(20.05)
Milk GM (Rs)	13,896	(38,130)	28,026	(41,638)
Livestock activity GM (Rs)	-29,292	(220,241)	-11,625	(151,877)
Total farm gross margin (Rs)	405,771	(425,326)	271,399	(299,857)
Operating profit (Rs)	278,240	(380,650)	132,630	(236,049)
Net profit (Rs)	-218,093	(299,866)	-276,542	(267,778)
Return on assets (Ratio)	4.30	(7.60)	1.87	(6.81)

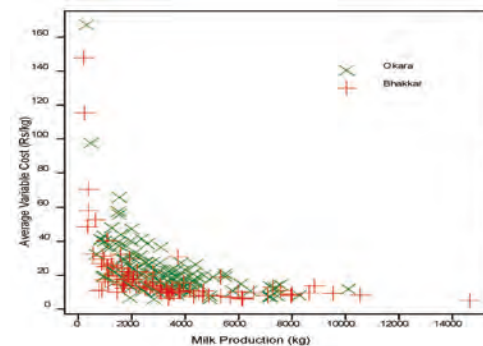


Figure 2: Relationship between Milk Production (kg) and Average Variable Cost (Rs/kg)

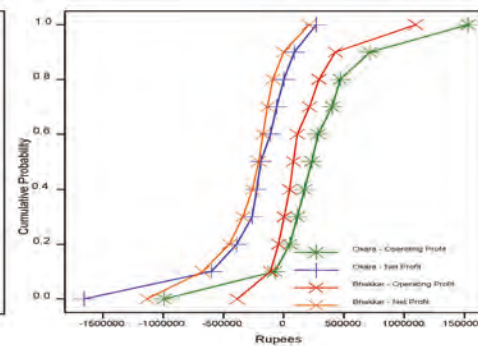


Figure 3: Cumulative Distribution Function (CDF) for Operating and Net Profits

Conclusion:

- Overall losses from livestock enterprises are possibly due to low productivity and directly related to nutrition and management practices. Our study confirms this premise of the ACIAR project.
- Actual cost of capital, in particular to the small dairy holder, may be much higher than the assumed (9%), as they borrow from traditional money lenders.

Question 1. Does integrated mixed farming limit farmers from capitalising on advantages of becoming specialized producers?

Question 2. Are the better production efficiencies we found with larger numbers of milking animals also associated with greater milk marketing efficiencies?



Figure 1: Map of Pakistan and Punjab

Results:

- 40% farmers in Okara and 30% in Bhakkar had average variable costs (Rs/kg) higher than farm gate milk prices of Rs 23 and Rs 21 per kg respectively and were thus making losses. Milk average variable costs per kg had an inverse relationship with production (Figure 2, Table1).
- Milk gross margins were slightly positive although overall livestock activity gross margins were negative. The whole-farm gross margin that included crops was positive, mitigating the negative effects of livestock in both the districts.
- Farm operating profits, after removing labour cost from whole farm gross margin estimates, was positive for most sample farms. Net profit showed losses when finance costs were included, though return on assets was higher for Okara than Bhakkar (Figure 3, Table1).



Identifying producer, middlemen, retailer and consumer issues from a pro-poor value chain perspective: A dairy case study from Punjab of Pakistan

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Abstract:

Informal fresh unpackaged milk value chains (movers of 95% production) in Pakistan, link millions of rural producers and urban consumers and provide a livelihood to thousands of middlemen (colloquially known as dhodhi) and retailers. Not much is known about the way these chains operate, apart from their general structure and a negative public perception of end product quality following adulteration of milk as it passes from farm to the consumer. This paper looks at a specific chain from rural producer to urban consumer using quantitative and qualitative methods, taking a case study approach. It describes the information sharing mechanism and the unwritten rules under which the chain operates amid low profit margins and dated technology. Financial advantage is gained by changing units, dilution and lowering price, where both producers and consumers are at the losing end. Milk is diluted with ice which affects its microbial quality and lowers protein and fat concentrations. Yet the consumer still prefers fresh milk as its price is lower and fat content possibly higher (4 to 5%) than the alternative packaged milk (3.5%). The chain is “pro-poor” as it offers small financial incentives through cash advances, loans and income generation/employment opportunities: margins for each operator are low and their distribution inequitable. Poor rural producers and urban consumers are linked by these chains but milk quality and the pricing mechanism along the chain require review.

Keywords: milk, value chain, producer, collector, retailer, consumer, gadvi, kilogram, litre, Rs

Introduction:

In developing countries, 2.6 billion live on less than \$2 a day. A billion of them live in rural areas and depend on agriculture and livestock for their livelihoods. Another three quarters of a billion are urban based. Poverty is pervasive in both rural and urban environments closely linked to malnutrition. Effective value chains are essential in meeting the evolving needs of rural and urban poor. These typically represent small scale production and marketing systems, which offer the means to increase access to animal sourced foods for poor consumers, and present opportunities for poor producers and marketers: thus they have a pro-poor value chain focus (Baker, 2008; Echeverría, Solh, Seré, & Hall, 2011; Otte, et al., 2012).

Collins (2009) defines “food value chains as systems driven by interaction of their technical (production, processing, transport, etc.), economic (profitability), information-related (communication) and governance (human relationships) subsystems”.

A value or marketing chain is a network of multiple firms/actors connecting the original producer to the end user. These firms or personnel integrate across organizational boundaries through the provision of products and services that add value for their customers (Lambert & Cooper, 2000). Fresh produce chains require physical transport, storage and some processing that is customized to the requirement and profitability of consumer segments (Anderson et al., 2007; Kohls & Uhl 2002; Schaffner, Schroder, & Earle, 2003).

Product exchange takes place only if an arbitrage opportunity exists. This means an opportunity to make profit by moving produce to areas with higher demand and at higher prices. While the product exchanges hands, its ownership, price, and the level of risk borne by each handler also changes. Income and its distribution is gauged

by mapping the chain. These chains cater to differentiated product markets where competitive advantage is gained only by lowering prices, quality or quantity attributes of the product (Gunderson, Wysocki, & Stern, 2009; Kaplinsky & Morris, 2001; Kula, Downing, & Field, 2006; Schaffner, et al., 2003).

The traditional open markets function with minimal flow of information between various tiers of the chain. Price is a highly effective communication signal to induce change. Accurate and open sharing of information among participants can help businesses succeed but the lead firms often behave opportunistically by either distorting information or disclosing it in an incomplete manner (Boehlje, 1999; Boehlje & Schiek, 1998; Williamson, 1991). In these markets official rules are either weak or poorly implemented. Local traders and retailers often located close to the consumer, impose their own rules to control the key processes along the chain dubbed as governance. In the absence of formal contracts, safeguards are set in the form of investment hostages. Product quality aspects are imposed upon the chain members upstream but with limited information sharing that often leads to antagonistic relations (Altenburg, 2006; Purcell, Gniel, & Gent, 2008).

This study examines a specific rural-urban informal fresh un packaged milk value chain in Punjab of Pakistan. The study provides an insight into technical, economic, information and governance that have an impact on a poor value chain for whole milk, and identifies possible avenues for policy intervention to improve outcomes for the producer and consumer.

Why milk and Pakistan's dairy industry?

With 181 million inhabitants (33% below poverty line), Pakistan is the sixth most populous nation in the world. The country has 2% population growth rate with a strong rural-urban migration trend. Current estimates suggest that 37% of the country's population is urban with projection that by 2050, nearly 56% of the people will be living in cities (Mazhar, 2011-12; United Nations Department of Economic and Social Affairs, 2012). Milk has a major share in the food basket of an average Pakistani household. Half of the household's total budget is spent on food and one quarter of that is spent on milk and milk products. With an average consumer price inflation (CPI) of 17% in the last four years, with food having 40% weighting on this index, price escalation poses a serious risk to average Pakistani households. A food price rise of 10 to 30% can push 4 to 10 million Pakistanis below the poverty line (Carrasco & Mukhopadhyay, 2012; Khan, 2011-12; Pakistan, 2010-11).

Nearly 80% of the milk is produced by rural producers and 95% of this milk goes to consumers through informal value chains. The industry generates significant income and employment generation opportunities not only for farmers and hired labourers in country's crop-livestock farming system, but also for milk traders and retailers in the final markets (Staal, Pratt, & Jabbar, 2008; Zia, et al., 2011).

The livestock sector contributes 11.6% to national GDP and is considered the most important sector to help alleviate poverty in a country where one third of the population lives below the poverty line (Amjad, 2010; Farooq, 2011-2012; Naveed & Ali, 2012). Milk is the major product of livestock and its market value exceeds that of the combined value of major cash crops (Afzal, 2010). Of the country's 28 million households, 8.8 million households or 31% rear buffalo (33 million) and cattle (37 million). Almost 90% of these households have 1 or less animals. The dairy animals have low milk yields compared to other milk producing countries. The average farm size in the country is 6.4 acres (Garcia, Khan, & Hemme, 2004; Pakistan Agricultural census organization, 2010).

Milk as a product, its attributes and handling:

Milk is regarded as being nature's most complete food. The type of animal (breed and species) and its diet can lead to differences in the colour, flavour, and composition of milk. The average percentage composition of cow and buffalo milk is as follows:

Table 1: Cow and buffalo milk composition

Species	Water	Fat	Protein	Lactose	Ash
Cow	87.2	3.7	3.5	4.9	0.7
Buffalo	82.8	7.4	3.6	5.5	0.8

Data Source: FAO (Fellows & Hampton, 1992)

The price and quality is usually dependent upon three factors namely milk-fat, protein content and microbial quality. Milk is a perishable commodity and spoils very quickly unless ambient temperature is maintained that prevents microbial contamination. Dilution of milk with water (or ice) will reduce the quality by diluting the key measures of butterfat, protein and microbial quality (Harding, 1995).

Methods:

This research was carried out in two stages during September 2011. Irrigated Okara and drained Bhakkar districts of Punjab were chosen based on access to and prior economic analysis of farm data to estimate the cost of milk production in these districts (Godfrey, Behrendt, Nordblom, & Wynn, 2011). Four questionnaires were redesigned for producers, middlemen, retailers and consumers to capture quantitative and qualitative data in an informal milk value chain. Quantitative data collected included physical (volumes) and financial (costs and margins) flows as well as technology and infrastructure (transport, storage, cooling and processing) aspects. Qualitative data included information on pricing and the frequency and mode of payment for milk and a set of key rules by which the chain is governed. The consumer's preferred attributes for fresh milk were ascertained as well.

Face-to-face interviews were conducted with each respondent, which took between fifteen minutes and an hour depending on the respondent's place in the chain. The difference in time taken depended on the level of complexity and number of questions asked: for example, the questionnaire for the consumer was brief compared to the ones used for chain members upstream. All questionnaires included an explanation of the purpose of the research to all respondents. In the first stage, a purposive sampling method was used to ensure the sample adequately represented the particular interest of the study (Patton, 2002; Robson, 2002). Interviews were carried out to develop a broad understanding of informal milk chains and markets, the details of which are as follows:

- Twenty seven milk producers of varied land and livestock holdings, producing and selling at least some milk, from four villages in each of the two districts. These producers were identified and the interviewer introduced by the ongoing ACIAR project and government of Punjab's dairy extension staff.
- Twenty four middlemen of various scales were interviewed. At a rural level, interviews were carried out with immediate buyers or large milk collectors. At the urban level, those delivering milk at retail shops were interviewed.
- Twenty two specialized milk retailers were interviewed in different areas of the cities of Lahore, Bhakkar, Sahiwal and Dera Ismail Khan.
- Eleven milk consumers were interviewed at the retail shops in the cities mentioned above.

After initial analysis the study focused on one traditional ruraly based chain that was studied as a specific case (Yin, 2009). This value chain is characterized by the presence of many intermediaries in the flow of product to the marketplace. This provided an example of what we describe as a complex rural-urban informal value chain model. This chain connected farmers in Okara with consumers in Lahore. To identify the participant participants, a snowball sampling method was used where one or more identified individuals provided the information on other parts of the chain (Patton, 2002; Robson, 2002). Thus producers identified their small buyers, which led to medium and large buyers, and then the milk retailer. It took three days to elucidate the structure and study this chain. The point of origin was a village in rural Okara, 135 km southeast of the final consumption market in metropolitan Lahore city.

Results:

A number of milk value chain models existed in both the districts and are depicted in Figure 1. The results presented here focus on one specific model from rural Okara district to metropolitan Lahore city (depicted by the

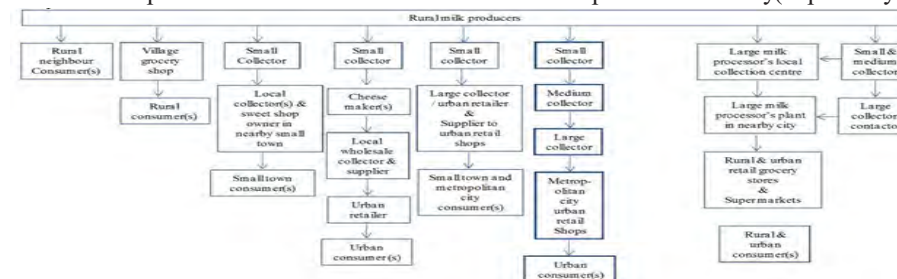


Figure 1: Milk value chain models from rural producer to fine consumer

bolded path). In this chain, milk was collected and transported with minimal use of current milk transport technology and the chain had a complex structure with many tiers of collectors.

As illustrated in Figure 2, the rural-urban informal fresh unpackaged milk chain linked hundreds of rural producers with thousands of consumers (consumption estimates based on household income economics survey 2011 per capita milk consumption estimates) in the final urban market destination and created more than one hundred employment opportunities.

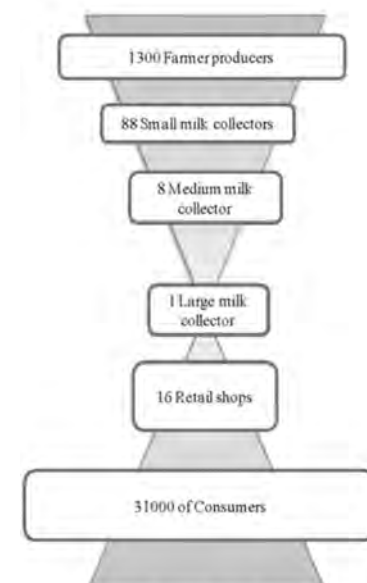


Figure 2: Pyramid of Okara-Lahore fresh unpackaged milk value chain

1. Technical subsystem and actors of the value chain: This section introduces specific actors and their functions, geographical location as well as providing a time course of movement of product down the chain to the consumer (Table 2).
2. The typical producer was located in a village in rural Okara. The small producer owned 5 acres of land and 5 dairy animals of which 1 cow and 1 buffalo were lactating. He and his wife spent three hours per day performing husbandry practices and hand milking. The farmer was selling almost half of his mixed cow and buffalo milk production, with the remainder being used domestically. He was selling part of both his morning and evening milking. The evening milking was kept in a refrigerator or ice was added when electricity was not available and sold the next day.

The small collector collected small volumes of milk from ten to fifteen farmer's doorstep and delivered it to a central collection point once every morning, a process that took 4 hours. He used his motor cycle and steel pot to collect milk and travelled 40 kilometres daily. He even milked the animal himself, if there was some doubt about the farmer's integrity in maintaining the purity of the product. In addition, he delivered groceries or agricultural inputs on demand to farmer's household.

The medium collector's operation was located at a bus stand, a midpoint between a few villages. His 3 hour morning operation did not require travel as milk was delivered at his centre by eleven small collectors. A big open top steel tank was available but not used, as milk was measured directly by the large collector and transferred to his truck. The medium collector's job was to record milk volume supplied by each small collector, to measure and record fat content.

The large collector was from a powerful Gujjar clan that dominates the Lahore milk market. He was collecting milk from 8 middle medium collectors and supplying to 16 different shops in Lahore including his own shop. He made a 300 km return trip between Lahore and Okara on a daily basis. This was a 18 hour operation seven days a week. This was a very basic operation as plastic containers having a holding capacity of 138 litres were used to transport milk. Five kg of ice was added to every 4 litres of milk (8.2 milk: 1 ice) to carry it to the final destination.

The milk retailer was related to the large collector and was being supplied milk only by this chain. He was selling milk directly to the final consumers in an impoverished area of Lahore city, around 150 km from its origin. His operation required little investment in infrastructure to store and sell milk, and two refrigerators for milk storage. Ice was added in the case of electricity disruption, which was a common occurrence. The only value adding activity undertaken was 1300 Farmer producers 8 Medium milk collector 88 Small milk collectors 1 Large milk collector 16 Retail shops 31000 of Consumers Figure 2: Pyramid of Okara-Lahore fresh unpackaged milk value chain making yogurt the milk was boiled. Milk and yogurt were both sold in transparent polythene bags.

The consumers at retail shops in Lahore milk market preferred fresh unpackaged milk. The main choice criterion was taste, which was defined by sweetness and thickness. That is, the more cream found in the purchased product after boiling, the happier the consumer was. The consumers were not sure of the unit to purchase, but the quantity consumed by each purchaser was based on income and family size.

Table 2: Functions of actors, geographical location, time input and technology used to handle the product and technology and infrastructure along the chain

Actors	Producer(s)	Small collector(s)	Medium collector (s)	Large Collector	Retail shop(s)
Functions, geographical location and time along the chain	Milk production from both cows and buffalo at village	Milk collection from local villages and delivery at central rural collection point	Milk exchange facilitation at a central rural collection point	Milk collection from rural central collection points and distribution to urban retailers	Milk sale to final urban consumers i.e. Lahore city
	3 hours that includes 0.5 hours for hand milking two animals twice and 2 hours for husbandry practices	4 hours from 6:00 to 10:00am for the collection and delivery of morning milking only	3 hours from 10:00 am to 1:00pm for delivery, recording and transfer onwards	18 hours as starts 5:00am and last delivery at 11:00pm	17 hours as shop opens at 6:00am and closes at 10:00pm
Technology and Infrastructure (Storage Transport Cooling Processing)	Hand milk and storage of evening milk in refrigerator. Ice added when electricity breakdown	Motor cycle used for milk collection (40 km travelled) in steel pots. Ice used to store evening milking.	No travel required. Open top steel tank available but not used. Milk directly transferred to large collector's containers / truck	Plastic containers of 138 litre capacity used to collect milk. 5 kg of ice added to every 41 litre. 300 to 400 km return trip on truck	Freezers available at the shop for storage of milk. Ice also added in case of electricity breakdown. Milk is sold in polythene bags. Yogurt is the only processing done

2. Economicsubsystemofthevaluechain: Thissectionintroducesgrossmarginsperactor,excludingthe owneroperator'sopportunitycostoflabourandgivesanestimateofthecapitalinvestedbyeachparticipant (Table3).Themeasuringunitsusedfomilksalesareexplainedlaterinthegovernancesection.Thefiguresgiven werecollectedonthedayofsurveyandarerepresentativeofransactionsthatoccurduringthisseasonoftheyear.

Theproducersold4.5gadviomixedcowandbuffalomilktosmallthecollectorandkeptthesameamountfor domesticuse.Hisgrossmarginfrommilksoldwas13Rsthatday(Godfrey,etal.,2011).Hiscapitalinvestment toproducemilk,consideringlandandlivestocktobethemajorassets,was2.56millionRs.

Thesmallcollector'smarginbasedonactualvolumecollectedwouldhaveactuallybeenalossof-18Rsbutat thisstepofthechain,therewasaconversionfromgadvitolitresandareward/penaltysysteminplaceforfat content(tobedescribedinthegovernancesection).Theseadjustmentsincreasedthecollectionvolumeby8litres andthushismarginwasRs238thatday.Hiscapitalinvestmenttoenterthisbusinesswas0.1millionRsand comprisedofRs50,000foramotorcycleandmilkcollectionpots,withasimilaramountbeingextendedas advancetofarmers.

Themediumcollector'smarginwas1,580Rsbasedonacommissionof3Rs/litreandatotalcollectionof660 litresatday.Hiscapitalcosttoenterthisbusinesswas1.1millionRsextendedasinterestfreeloanstoelven localsmallcollectors,collectingmilktoservicetheneedsofhislargecollectorcustomerfromLahore.

Thelargecollector'sgrossmarginwas46,600Rsbasedonacollection6700litres(8.2milk:1ice).Hiscapital investmentwasinatruckusedfortransportationofmilkworth1.4millionRsandinloansestimatedtobeworth1 millionRsfomilksuppliedoncredittoretailcustomers.

Themilkretailer'sgrossmarginwas500Rs/day,assumingaverage salesof465 litresateachshop.Asmall proportionofthisiscomprisedoftheadjustmenttothesaleofalitreofmilkto900mlincommercialoutlets.This pointwillbeexplainedfurtherintheinformationsection.Thiswasbasedonamilkretailpriceof42Rs/litre whereasyogurtwasactuallysoldat55Rs/kg,whichisignoredinthiscalculation.Hiscapitalinvestmentof 50,000Rspaidfortworefrigeratorsusedfomilkstorage.

3. Information flow on pricing of product: There was no evidence of how farm gate milk price was fixed. In the Lahore city market, the city district government annually fixes milk price/litre, whereas there is no set mechanism to fix the milk price at the farm gate. The producer was given the price of milk by the small collector, which was negotiable. The producer did however also consult with neighbouring farmers on prevailing prices.

The small collector was given a price by the medium collector at the central collection point that changed every few weeks based on supply and demand. The medium collector still got the same commission irrespective of price change. The large collector was of the opinion that the major processors such as Nestle, Haleeb and Engro actually control the milk farm gate price. A difference of 2Rs/litre was noted between this chain and big processor farm gate prices, with the latter using accurate litre scales when buying milk and offering stable prices across seasons. The producer still preferred to sell to the informal milk chains due to the fact that they were getting advance payment for the milk.

The retail shops in Lahore city claimed to change price only once per year as set by the city district government. At the time of this study the fresh un-packaged milk price was set at Rs50/litre. The retail shops were selling milk in litres, kilograms and gadi with different units being sold at different prices. The retail sector in the chain were selling milk between 42 to 60Rs per litre (though units varied) and market prices were observed to vary from 42 to 70Rs, and the different units of sale that included gadi, litre and kilogram. This difference in units is assumed to be away to get around the price per litre set by the government. The shops are assumed to be selling a lesser quantity (litre) for a given price keeping the margin so high that the price for a bigger unit (such as gadi). It was evident that the price was not strictly being enforced across the commercial sector. The ultra heat treated (UHT) packaged milk processed by large processors was being sold at Rs85/litre.

4. Governance in the value chain: There are no formal statutory regulations for this chain and no written contracts among chain actors. However there is a set of unwritten rules to exercise control. Thus business practices and their impact on the quality of milk products reaching the consumer can be adjusted to advantage lead actors of the chain, that are large collectors and retailers, without recourse to any authority. Rules currently operating within the chain investigated are detailed in Table 4. This chain operated seven days a week and the only holiday was the tenth day of every Islamic month when milk is given as charity to the poor and deserving.

Rule 1): Cash Advance, Loans and Payment Schedules: The cash advance, loans and payment schedules varied at each step of the chain: these arrangements made the chain cohesive. Upstream, at the rural end, the medium collector extended a loan of 0.1 million Rs to each of the small collectors, which enabled them to provide a monetary payment to their farmers suppliers who demanded a two week cash advance for the milk sold to the small collector. This cash advance was based on the milk volume sold. The term “economichostages” has been coined (Williamson, 1991) to describe the interdependency of the relationship between operators within these 3 phases of the marketing chain.

The large collector made cash payments to medium and small collector every eighth day of the business cycle but downstream at the retail end he was supplying milk to some of his customers on credit. It is assumed that he was locked in with his customer too, due to this credit arrangement.

Rule 2): Inconsistent units, fat premium/quality and dilution: Inconsistent units of volume were found all along the chain. It is hypothesized that these have developed historically to generate incomes for operators within the chain, part of which is to distort the volume of milk sold as a litre to the consumer to the advantage of the retail outlet. Given that there is no statutory regulatory authority to protect the consumer this is not entirely unexpected. Similarly retail prices can be adjusted beyond the price fixed by the city government.

In this chain the most important attribute of the raw product was its fat content as it was sought after by the urban consumer, thereby raising the importance of high fat buffalo milk which can therefore be diluted more prior to sale. There was little understanding of the nutritional value of milk protein and microbial quality. The temperature of the milk was however, maintained by adding gicet to the milk directly, which apart from acting as a diluent, may have potentially introduced microbial agents.

The small collector's gadv contained 100 grams more milk than it should (1130 grams instead of 1030 grams for milk having a higher density). This is a deliberate ploy to gain illicitly 100 grams of milk per gadv collected from the producer. These additional grams were taken into account at the central collection point, and were converted to litres at the point of exchange of product. Thus his milk collection increased from 4 l gadv to 4.5 litres that is a gain of 4 litres because of this deception.

He preferred buffalo milk due to its higher fat content, but was generally buying mixed cow and buffalo milk. He assessed quality of milk collected by visual appearance and smell as cultural norms did not allow any further checks. The large collector had placed a reward/penalty system in place at the medium collector's central collection point where the milk exchanged hands. The medium collector measured and recorded fat content on behalf of the large collector against the 6% standard. As the small collector's milk had 6.5% fat, he gained another 4 litres as it was diluted to provide the standard 6% fat content. There was a centrifugal machine present at the collection point to test fat, but it was rarely used as estimation was at the discretion of the medium collector. The following formula was used as a tool to encourage the supply of unadulterated high fat content buffalo milk (Table 4).

The large collector diluted the milk with ice at this stage (8.2 litres of milk: 1 litre equivalent of ice which varied with season) to ensure that it does not get spoiled on its way to the Lahore market which was an hour trip. The retail milk shops in the city, not owned by the large collector, had their own formula to test fat based on which the price was fixed. On arrival of the truck, 2.5 litres of milk was boiled to get a standard 400 gram soft khoya (milk thickened by heating in an open iron pan). The price was fixed based on grams of khoya obtained from this process. This was to ensure collection of higher fat content milk preferred by the final consumers.

The milk was sold by the retail shop to the consumer in gadv, litres and kilograms. It is assumed that the litre was 900 grams, which meant a lower quantity for the price paid. A maund (a local unit) was actual 140 kg at the farm gate and 46 litres in the Lahore market due to the ready acceptance of dilution by the vendors.

Table 3: Physical and financial flows along the chain

	Farmer(s)	Small collector	Medium collector	Large Collector	Retail shop(s)
Volumes	4.5 gadv (5.1 litres based on 130 grams excess collection pot)	41 gadv but 49 litres after unit conversion and fat standardization	660 litres based on fat content premium	6700 litre that includes 5970 litres of milk and 820 kg ice (8.2 milk:1 ice)	465 litres/shop (sells 0.9ml for every litre sold and thus makes 45 litres extra)
Price at each step	30 Rs/gadv	32 Rs/litre	35 Rs/litre	40 Rs/litre	42/litre
Estimated Revenue / day (P*Q)	135 Rs = 30*4.5	338 Rs = 41*2 + 32*8	1,980Rs = 3*660	59,050Rs = milk 5,970*5 + ice 730*40	840Rs = 465*2
Estimated Variable costs / day		100 Rs/day motor cycle fuel	400 Rs/day based on -8000Rs/month to record keeper -2000Rs shop rent & utility bills -2000Rs for miscellaneous such as entertainment of shop guests	12,500 Rs/day based on -8000Rs/day truck fuel -8000/month truck driver's salary -270 to 400 Rs for 6 loaders -2460 Rs of ice for 830kg ice @ 3Rs/kg	430 Rs/day based on -8000Rs/month shop and utility bills -4000Rs/month shop helper's salary -1000Rs/month polythene bags for selling milk
Gross margins per actor	Rs 12.58/day	238 Rs/day	1,580Rs/day	46,600Rs/day	503 Rs/day
Capital Investment	2.56 million Rs (0.31 million for livestock and 2.25 million for land)	0.1 million Rs (50,000 Rs for motor cycle and pots, 50,000Rs approx. advances to farmers)	1 million Rs as loans to small collectors	Truck worth 1.4 million Rs and 1 million Rs as milk on loan to some retail customers	50,000 Rs as refrigerator(s) for milk storage

Note:

- The unit conversions are gadv = 1130 grams at farmer's door step, litre = 1030 gram or ml during transport and exchange in the chain, litre = 900 grams at the retail shop in the city when sold to final consumer
- OANDA <http://www.oanda.com/currency/converter>; exchange rate of 15th Sept. 2011 where 1AUD=89.3Rs

Discussion and conclusion:

The research has given an insight into how a rural-urban informal fresh un-packaged milk value chain operates in Pakistan, which has not been described previously. It has quantified the number of participants and employment opportunities along the chain.

There is no formal contract at any stage in this chain, yet producers obtain advances for their milk, regular cash flows and additional services such as agricultural inputs as a part of the arrangement. Small collectors obtain interest-free loans to operate and earn their livelihood while the medium collector generates a viable return on his

capital. The small collector is in part trapped, by both the producer to whom he extends advances, and the medium collector from whom he borrows the money. The large collector holds the most power at the rural end of the chain yet he has to keep the demands and expectations of his retail customers. This chain collects, transports and sells fresh un-packaged milk with a fat content of 4 to 5%, which is higher or equivalent to the packaged milk usually containing 3.5% fat, but which is marketed at almost half the price of the packaged product.

The risks associated with this chain and the raised from the study include:

- a) The chains operate with minimal use of technology and the answer partly is that the consumer pays a lower price but is this practice sustainable in the long run? What about the microbial quality and protein content?
- b) The margins, in particular those earned by the producer and small collectors are negligible. The actors downstream are gaining competitive advantage only by lowering prices and distorting the quantity and quality of milk on sale.
- c) There is no formal mechanism for setting a farm gate price while the price of fresh milk in urban centres is set by the government that should be reconsidered.
- d) The milk unit conversions deprive both the farmer and final consumer of quantities for which they receive no payment/milk. Having fat content as the only determinant of milk quality is a matter for concern where the living standards and expectations of consumers is becoming more discerning.

This chain operates in an industry environment which is complex and competitive. There are a large number of small producers and final consumers catered for by a large number of collectors and distributors at different tiers of the chain and retailers. More chains and different models will have to be studied before making any concrete recommendations for the more effective marketing of such an important component of the Pakistan diet.

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PLANNING DAIRY DEVELOPMENT PROGRAMS IN ASIA



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Effect of improved extension services on adoption rates and farm economics of smallholder dairy farmers in Pakistan

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Abstract

The objective of the study is to demonstrate the effect of improved extension services on adoption rates and farm economics of smallholder dairy farmers in Pakistan. During the first two years of the project a simple approach of extension targeting male farmers was used. Whereas in the second phase of the project a whole family approach targeting all the family members was utilized. The effect of improved extension services on adoption rates and farm economics was collected on a monthly basis from the farmers. These farmers were working in the districts of Kasur, Okara, Pakpattan, Jhelum and Bhakkar within the Punjab province. Preliminary results indicated significantly higher ($P < 0.05$) adoption rates when extension services were provided using the whole family approach (50%) compared to the simple approach (14%) after one year of both phases. Improved extension services have significantly increased ($P < 0.05$) the farm income of smallholder dairy farmers. Overall there was about \$US 100/month increase in the income of smallholder dairy farmer. In conclusion, this study indicates that improved extension services have a significant impact on adoption rates resulting in an increase in farm economics of smallholder farmers. The data generated from this study will be helpful to devise better strategies for improved extension services in order to optimize the dairy production of smallholder farmers and will have a ripple effect on the others to follow.

Keywords: Extension services, adoption rate, farm economics, smallholder farmer, dairy

Introduction

Pakistan, like many developing countries, has an agrarian rural based economy. Livestock is a major contributor to the national (12%) and agricultural (50%) economy (Pakistan Economic Survey, 2006). The livestock sector has been recently declared as one of the fastest growing sectors and provides improved livelihoods for more than 35 million people with farmers/households deriving 30 to 40% of their income from livestock.

Milk remains the major contributor to income derived from livestock. The value of milk alone exceeds the combined value of wheat, rice, maize and sugarcane in the country. Milk is produced under different production systems namely, rural subsistence small holding, rural market oriented small holding, rural commercial farms and peri-urban dairying. It is estimated that around 70% of the dairy households in Pakistan still operate under conditions of subsistence by maintaining herds of three or four animals (Burki et al., 2005). The productivity of livestock is still lagging behind its potential level. In order to meet the requirements of a rapidly growing population, dairy production needs to be increased. This can be done by adopting modern techniques of dairy farming. New technologies developed by researchers are disseminated among the farmer through an effective extension program.

The role of extension has been to provide research-based education and information to the production sector. The most important management areas on a dairy farm are feeding and forages, udder health, reproduction, calf raising, and herd health (Dahle et al., 1991a). Problems solving in these areas requires a broad base of knowledge and expertise, and often the implementing agency must organize a multidisciplinary team of extension specialists or other professionals to assist producers (Dahle et al., 1991b). Services to the dairy sector are being provided by government agencies and a range of NGOs, and virtually all services providers who interact with the farmers are veterinarians or para-veterinarians who perform vaccination, treatment and A.I. Limitations in the extension

service and the research/extension interface are considered to be bottlenecks in the development of the dairy sector. In particular, the style of communication between farmers and extension staff, the information available to extension staff, the number of skilled extension staff and a failure to consider problems and solutions in a whole-of-farms systems context are important limitations. Thus the major objective of this study is to demonstrate the effect of improved extension services on adoption rates and farm economics of small holder dairy farmers in Pakistan.

Materials and Methods

In 2007, an Australian Centre for International Agricultural Research (ACIAR) research project LPS/2005/132 was commenced with the aim of increasing dairy production through improved extension services. Small dairy farmers having 4-10 (buffalo and/or cattle) for production were the main target group for this project. During the first phase of the project a simple approach of extension targeting male farmers was used. In 2011, ACIAR extended this research project for an additional five years. The project is currently working in five districts of Punjab (Okara, Pakpattan, Kasur, Jhelum, Bhakkar) and two districts (Thatta and Badin) of Sindh province. During this second phase of the project a whole family approach targeting all family members was utilized. A number of innovative ways of extension have been adopted including the use of video practices, demonstration plots, problem based learning, staged drama, radio and TV shows to improve the effectiveness of the program. Benchmark data were collected on whole farming systems from 228 farmers during the first phase and 292 farmers during the second phase of the project. Subsequently, at the end of every year data have been collected to monitor the impact of these initiatives on the rate of extension message adoption. In order to analyze the farm economics, data was collected from 10 primary leading farmers, two from each of four project districts of Punjab annually.

The project placed emphasis on a comprehensive interdisciplinary educational program of meetings, workshops and training of both farmers and extension workers. Basic husbandry, nutrition, and calf management were the initial subjects addressed during both phases of the project. Adoption rates between the whole family approach and simple approach after one year of both phases were analyzed using a Chi-square test. Similarly, adoption rates of various modules at the start and after one year of project phase-II were analyzed using a Chi-square test. Comparisons of various average monthly incomes of small holder farmers were analyzed using a t-test. Statistical analysis was carried out using SPSS (Version 10.0) with $P < 0.05$ regarded as significant.

Results

Adoption rates (50%) were significantly higher ($P < 0.05$) by more than three-fold when a whole family approach was implemented compared to the simple approach (14%) after one year of both phases (Figure 1). Comparison of adoption rates of various modules (animal husbandry, basic concept of nutrition and calf nutrition) at the start and after one year of the second phase is shown in Figure 2. Untying animals and giving free access to water together with twice daily cleaning of sheds were readily adopted, however investing in infrastructure in the form of shed construction was not as easy for farmers (Figure 2a). Offering fodder ad libitum, feeding concentrate and improved extension services have significantly increased ($P < 0.05$) the farm income of small and mineral mixes were perceived as being easy and of direct benefit to productivity: in contrast feeding cotton seed cake was not (Figure 2b). Irrespective of the message for calf rearing feeding colostrum, offering concentrate and sufficient milk and water were perceived as being readily adoptable messages (Figure 2c). Overall there was about US\$ 100/month increase in the income of small holder dairy farmer (Table 1).

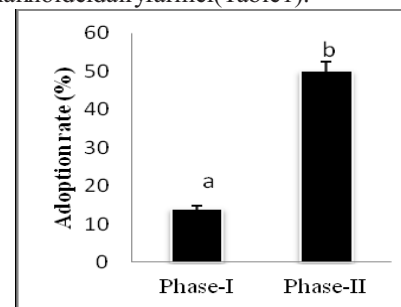


Figure 1. The difference in adoption rates (%) of extension messages following the use of a traditional "male only" extension approach (phase I) as compared with a whole family approach (phase II).

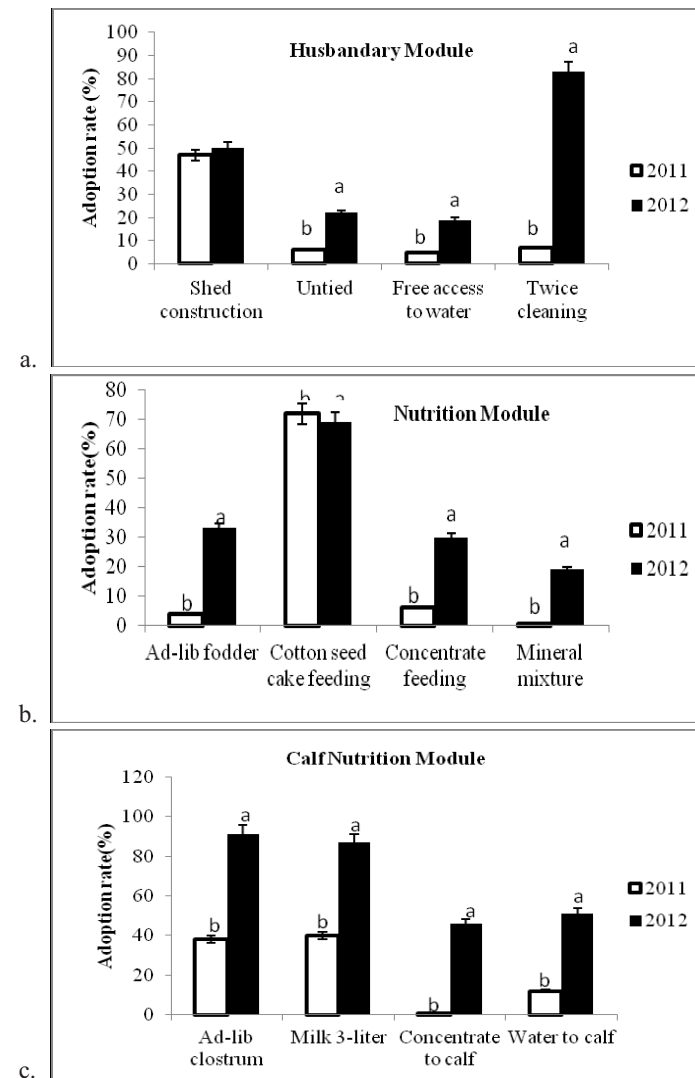


Figure 2. The difference in adoption rates (%) of specific extension messages following the use of a traditional “male only” extension approach (phase 1, 2011) as compared with a whole family approach (phase 2, 2012). 2a: watering, cleaning and shed construction; 2b: aspects of animal nutrition; 2c: aspects of calf feeding

Table 1. Comparison of various average monthly incomes of small holder farmers

Survey	No. of milking animals	Land (Acers)	Dairy income/ month (Rs)	Crops income/ month (Rs)	Income from other sources/ month (Rs)	Total income/ month (Rs)
2011	2	6.7	7161a	5172a	4710a	17043a
2012	2.2	6.7	12758b	9358b	5400b	27516b

a, b Means with different superscripts within a column are significantly different ($P < 0.05$)

Discussion

To our knowledge this is the first report which clearly describes the effect of improved extension services on adoption rates and farm economics of small holder farmers in Pakistan. Higher adoption rates (50%) were achieved when we introduced a whole family (male, female and children) approach compared to simple approach (14%) relative to the traditional male only approach to extension. An effective extension program with participation by the whole family is highly desirable to enhance farm productivity. Many organizations aiming at improving small holder dairying fail to appreciate this fact and ignore women and children in their training and

skills development programmes. Women normally cannot leave their home and families for a few days to participate in training programmes and usually require female trainers for effective communication. Thus during the second phase of the project we arranged parallel sessions for women and children co-ordinated by women trainers. This approach, along with training of the male farmers resulted in higher adoption rates.

In order to demonstrate the role of children in rural communities this project initiated a buffalo calf raising competition among the children of farmers (10-16 years) in Pakpattan district. These results demonstrated that under field conditions the average daily buffalo calf live weight gain (431 gm/day) was comparable to many controlled studies in various leading research institutes of Pakistan (Bhatti et al., 2009; Iqbal and Iqbal, 1992). These findings clearly indicated that we can effectively enhance animal productivity through the active involvement of children in our extension program.

In the present study, adoption rates of various modules at the start and after one year of project phase-II are significantly higher. Possible reasons of high adoption rates other than the whole family approach are the implementation of the innovative ways of extension like video practices, demonstration plots, problem based learning, staged drama, radio and TV shows. Innovative methods of extension played a significant role in order to achieve higher adoption rates. While devising training programmes, one should keep in mind that "Seeing is believing" and "Farmers do not have ears, they only have eyes."

The present study clearly demonstrated that improved extension services resulted in significantly increased farm income for small holder dairy farmers. There was an average increase of about \$US 100 in monthly income after one year of effective extension. Farmers were shown to have adopted basic husbandry and improved nutrition practices. Providing ad-libitum access to water and feeding resulted in the increase of approximately 1 lit/animal/day (Warriach et al., unpublished data). However, there is a need to investigate further the effects of various adoption rates on the productivity and farm economics of small holder farmers. In conclusion, these

preliminary results indicate that improved extension services have a significant impact on higher adoption rates resulting in an increase in farm incomes of small holder farmers.

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Sustainability of milk production from small-holder farmers: a Pakistan perspective

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Abstract

Maintaining the milk supply for a population of 180 million people who depend heavily on this commodity for their animal protein needs remains a challenge given the poor level of training provided for the 8.5 million small holder dairy farmer to whom this task falls. The key objective of any extension program needs to focus on boosting the production per animal beyond the mean of 3.15 litres per day. In striving to attain this goal farmers must be able to achieve a profit margin sufficient to justify their investment in improving productivity. One of the key objectives of the dairy industry in the future must be to rationalise milk marketing chains so that a significant proportion of the profits derived from selling high quality milk to consumers is passed directly to the producer. With the provision of a clear economic signal the farmer will strive to improve feeding, disease control, reproductive management and the environment in which animals are kept. Changes to the industry are not necessarily going to come from direct government investment, but are more likely to be achieved by the implementation of government policies designed to encourage private and corporate investment in the industry.

Keywords: Milk production, small holder dairies, marketing, extension

Introduction

Pakistani Minister for Livestock and Dairy Development, Hamayun Aziz Kurd proudly informed the National Assembly early in 2014 that Pakistan stands as fourth in the ranking of the largest milk producing countries with an annual production of 36.2 million tonnes from eight million farming households. Pakistan is the fourth largest producer of milk in the world behind India, China and the United States. He further stated that the annual milk production across the country is worth as much as Rs 177 billion and is easily the largest product in the entire agriculture sector (Anon 2014).

The contribution of the dairy to the national economy is to the tune of Rs 540 billion with 97 percent of it being informal non-documented economic activity. Approximately two thirds of this is consumed by the domestic householders sold in local markets, leaving one third to be sold as fresh un-packaged milk through informal chains (Zia et al. 2011). The formal production and processing sector comprises only 3% of national production.

While the Minister has good reasons to be proud of these statistics, they belie the fact that total milk production is derived from 34.6 million buffalo and 39.7 million cattle (Ahmad 2014) each producing a little less than 3 litres per day. Furthermore the estimated 5% increase in production annually is offset by a 15% increase in consumer demand which is currently at 43.2 million tonnes, some 5 million tonnes above annual total production.

The government's response to this issue involved funding the national Livestock and Dairy Development Board to install 150 milk cooling tanks, train 168 milk cooling tank operators, 176 village livestock workers and establish 3871 farmers in "milk producing groups" similar to the concept of farmer co-operatives. Further to this the Board had already registered and trained 566 quality breeders and registered 1004 Progeny Testing program Farmers. Are these the answers that the increasingly urbanised population of 188 million people of Pakistan are looking for?

It is important to remember that 60% of the population lives on less than \$2 a day (World Bank 2013) and milk products provide 10.6% and 18.7% of the total calories and protein intake per capita per day (Pakistan Bureau of Statistics 2011).

Some of the answers lie within the problem areas identified within the industry by the Pakistan Dairy Development Company, an independent government organisation established under the Ministry of Industries, Production and Special Initiatives in 2006 (PDDC 2006).

The key issues raised at that time were:

- The low profitability of small-holder dairy operations; These separation of farmers from the milk marketing sector and market forces operational in regional urban centres;
- Inadequate infrastructure for both milk storage and transport: this includes refrigeration to preserve milk quality;
- Poor educational facilities available for farming communities;
- Little knowledge of how to feed a lactating animal; Inadequate training for the government veterinary extension officers who advise farmers on animal productivity;
- Lack of awareness of the consumers of the quality attributes of milk; Adulteration of the product along the marketing chains to maximise profit margins for milk chain operatives;
- The poor range of dairy products available to consumers suggesting that the industry lacked innovative entrepreneurial thinking; Inadequate research facilities across dairy production systems to provide the innovative thinking required to sustain a vibrant and profitable industry.

The nature of small-holder dairy operations

Close analysis of small-holder dairy production units most often are not profitable. It is important to note that the rationale for owning buffalo and cattle does not often relate to boosting family income. A dairy animal most often provides the sole source of animal protein for the family for an average lactation of more than 200 days. The amino balance, vitamins and minerals provided by milk protects children and women from nutritional deficiencies. Where production exceeds family need then the attraction of making a small income for the same duration helps a family pay for the essential foods, clothing and pharmaceuticals.

As most of these farming operations also rely on cash cropping from their small areas and irrigation water allocation, income from livestock provides an insurance against crop failure as well as providing dung which enables nutrients to be recycled to the soil profile thus avoiding the use of expensive inorganic fertilisers. Animals also act as an insurance policy to pay for weddings, funerals, medical expenses and education fees (Afzal 2006). Both cropping and forage growing operations are seasonal and it is important that the farmer manages his limited arable area to maximise production from both of these operations.

Given the need to milk animals twice daily and feed animals constantly, dairy operations generate employment opportunities. Most of these tasks are the domain of the women in families as their domestic duties keep them close to the family home where animals are most often kept for reasons of security and ease of management.

The pricing of milk

Even with all of these factors, milk price received is very important to the farmer and his family. Yet the very nature of traditional milk marketing chains dictates that the relatively uneducated dairy farmer is isolated from the price setting principles of supply and demand in operation in the urban market place. Factors influencing price received on farm are dependent on species, location of farms relative to markets, season, the educational status of the farmer as well as the pricing policies of statutory organisations affected the price received for milk on farm (Ishaq et al. 2015).

Given the convoluted structure of informal marketing chains the fixing of milk prices in the urban market place leaves little room for profit sharing for the milk collectors or the herdists that form the chain and even less scope for a

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Given the convoluted structure of informal marketing chains the fixing of milk prices in the urban marketplace leaves little room for profit sharing for the milk collectors or the distributor that form the chain and even less scope for a realistic price being offered to the farmer. The dilution of milk with water and the addition of other adulterants to boost perceived protein and fat contents by these operators is essential for profit sharing along the chain.

We have surveyed marketing chains in 3 districts of Punjab and found that the addition of water added up to 25% of the volume of milk sold in the urban marketplace (Table 1).

Districts	Fat%	Protein%	SNF%	Added water%	Lactose%
Okara	4.527	2.570	6.818	19.65	3.598
Pakpattan	4.175	2.568	6.850	18.51	3.618
Kasur	4.231	2.367	6.303	24.61	3.327

Table 1: Composition of milk collected in the urban marketplaces of Okara, Pakpattan and Kasur in Central Punjab Province Pakistan

It is clear that shop owners are able to balance the fat content of milk to remain above the 4% benchmark by strategically blending the higher fat buffalo milk (<6%) with cow milk together with water to maintain fat at a level acceptable by the consumer who places a high emphasis on the yield of cream in the raw product and ghee upon boiling. It is the low protein level that should be of concern to the consumer with concentrations of less than 2% often being found in the marketplace, suggesting that the dilution rate can be as high as 1:2 along some marketing chains. The containers used for measuring milk volume along the chain also favour the trader, while in many commercial outlets the consumer is provided with 900ml when in fact he has paid for one litre.

Pakistan, however, is no different to other developing countries where informal marketing systems facilitate the supply of cheap calories for the consumer (FAO 2013). Policies and development strategies in many countries often fail to recognize and provide adequate support for smallholder production systems and associated marketing chains, focusing instead on higher profile industrial production (Echeverría et al. 2011).

The importance of milk quality

While the dilution of milk with water is well known, give the amount of feces that is added to minimize the spoilage of product, other more covert contaminants can be found in milk. Concentrate feeds based on cottonseed and wheat gluten are highly susceptible to fungal contamination with the result that mycotoxins are often transferred from feed constituent to milk. Our studies have shown that up to 5% of aflatoxin B₁ in feed will be transferred to the milk of buffalo as the aflatoxin M₁ metabolite resulting in a concentration that exceeds international standard up to 100-fold. Given the highly carcinogenic nature of these toxins, this represents a "sleeping" hazard for all consumers of milk in Pakistan (Aslam et al. 2015).

The future of marketing dairy products

The traditional informal marketing chains described above are very reminiscent of mechanisms for the distribution and sale of agricultural produce in the western world up to the start of the industrial revolution of the 19th century.

This involved producing grain, milk, meat and fibre at the local level which was sold to traders and then through small scale merchants in the raw state or perhaps with a single step of processing such as churning cream to form butter or grinding grain to flour. Governments often set the price but there was little regulation of product quality. International trade seldom played a role, although sadly the dumping of heavily subsidized products like milk powder in the marketplace of developing countries has the potential to distort traditional marketing systems. This unethical practice has compromised small-holder farming systems in both Africa and Asia often destroying the sole source of income for marginal populations such as nomadic graziers. This has compromised the price of milk on offer to small-holder producers on some occasions while this product can also be beneficial by filling seasonal milk shortages.

Then with the advent of the industrial revolution the food production and marketing systems of Europe and North America underwent profound change. The establishment of large scale first stage processing companies for example grind grain (e.g. Cargill) or process milk led to the development of secondary processing companies producing refined food products (e.g. Nestle).

The first U.S. milk processing plant to install pasteurization equipment was the Sheffield Farms Dairy in Bloomfield, New Jersey in 1891, while Chicago became the first major American city to pass a law requiring commercial milk to be pasteurized. Mass distribution of milk was not possible until the advent of the milk bottle sealed with wax in 1884 and then the plastic coated cardboard milk carton in 1932 (Bellis 2014).

These changes lead inevitably to large scale chains of stores and supermarkets with associated large wholesale and logistics companies. Inevitably this leads to pressure on farmers to produce more with lower profit margins per litre. If farmers were not able to increase their herd they went out of business, which has led to commercial herds in California of more than 2000 cows. Similar trends have been seen in Australia. While these sceptics said this would not happen in developing countries, this has not proved to be the case. Food systems in developing countries in Latin America, Asia, Eastern Europe, and some of Africa have undergone a revolution since the 1980's resulting in the demise of small-holder farming systems (Reardon and Timmer 2012).

Will Pakistan follow this trend? Perhaps we will learn more from the experience of India which has recently allowed multinational supermarket companies to trade in large population centres.

Increasing the resilience of small-holder dairy farmers

Given the poor education available to most small-holder farmers, practices used for animal husbandry are most often of a traditional nature which transcend the generations. The low productivity of cattle and buffalo herds is in part due to the poor extension services on offer from the government veterinary advisory services. It is also important that only around 40% of small holder farmers are accorded any government services at all. The veterinary curriculum of most universities places a strong emphasis on curative medicine and management of reproduction. Seldom is nutrition included as a major component of any curriculum. Similarly veterinary

extension staff know little about calf rearing and its importance for farm profitability. Thus the training of extension staff is an important component of any assistance program.

The traditional mechanism for farmer extension is to provide formal presentations on site to male farmers in their villages. Our experience was that even with the most straightforward messages, success with this approach was limited to adoption rates of no better than 30%. This was even the case with our initial approach to training to untie animals and provide free access to water and feed. The identification of sentinel early adopting farmers in villages helped in encouraging others to join our program. It was important to note that Punjab farmers have an innate scepticism to work together co-operatively within a village to achieve a common goal.

However much greater success was achieved when a "whole family" approach was taken to the provision of extension services. This involved offering simultaneous extension services to men and women, with the village school female teacher attending the female extension session. The school teacher then displayed the extension messages in the school room for the children to read. The material provided needed to be largely pictorial to ensure illiterate adults and poorly educated children understood the message. The content of the women's sessions was broadened to discuss informal matters relating to female and child health and for this reason the village health worker was often invited to attend also. Some sessions were also designed to broaden recreational activities such as training with henna, stitching and even making ice-cream. The end result of this approach was that many families then discussed the content of the meetings around the family meal that evening and then the adoption of extension messages became a family decision rather than that of the male farmer by himself. Often it was the ever perceptive female who foresaw business opportunities for the family.

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Assessing the impact of training programs.

Inevitably funding bodies like to be able to quantify the success of the programs they have funded. Yet this is often the most difficult aspect of any project.

In order to achieve this goal we developed a simple questionnaire which sought to gain an understanding of the level of adoption of the simple messages we had provided to some 900 farmers for up to 5 years.

The simplicity of questions is shown here:

Un-tying of animals: Un-aware Aware

In developing this program we categorized farmers as either

1. Farmers registered with our program and most likely to adopt our messages;
2. Farmers not registered but whom we maintain contact with registered farmers or who attend our meetings without formally registering.
3. Traditional farmers who have not shown significant interest in our programs.

Table2: Rates of adoption of extension messages delivered to farmers on specific topics. The categories of farmers are given above and they were randomly selected from 5 districts in Punjab and 2 in Sindh. The extension messages are detailed as follows:
 Untying: Farmers were advised to untie animals and build a small yard to allow animals access to water ad libitum.

Farmer Group	Untying	TwoMonthsDry	Vaccination	BreedingBull	Ad Lib Colostrum	BalFeed	ImpSeed
1 (n=179)	98.3	95.5	Awareness Level(%)	Recommendation	97.2	92.7	89.8
2 (n=116)	88.7	82.8	93.1	80.2	67.0	77.6	75.7
3 (n=104)	75.0	78.9	80.8	71.8	42.2	68.6	73.5

Two months dry: Farmers were advised to dry off animals for 2 months prior to calving.

- Vaccination: Farmers were advised to use effective vaccines for Foot and Mouth disease and Hemorrhagic Septicemia
- Breed Bull: Farmers were advised to use semen from proven sires in breeding animals.
- Ad libitum colostrum: Farmers were advised to provide for calves access to colostrum ad libitum in the first 24 hours after calving.
- Bal Feed: Farmers were advised to provide sufficient quantities of a balanced ration including forage, concentrate and trace mineral to lactating animals to maximise production. Imp seed: Farmers were advised to source improved forage seed to increase yields

The surprising result from these data was the poor understanding of the importance of colostrum to the neonatal calf. This highlights the effectiveness of impact assessments in determining future emphases for extension programs. In this case, more effective extension messages need to be delivered on the importance of colostrum: it is possible also that the message needs to be delivered in a different way.

Conclusion

Of the factors that were listed above as limiting for the development of the dairy industry, perhaps the most important is the lack of appreciation of the quality attributes of milk. Every year we learn more about the health-giving properties of this wonderful biological human food.

We have known for some time about the virtues of the major macronutrients in milk such as protein, lactose and fat along with vitamins and minerals. We understand now that the caseins are highly complex proteins capable of yielding peptides that control a range of biological activities including behaviour, insulin sensitivity, blood pressure, gut absorption, immunity, gut motility and absorption and cholesterol status. Furthermore, fatty acid composition and complex carbohydrates are now seen as key metabolic regulators for the consumer of milk. Yet most milk in Pakistan is boiled because of problems with food safety thereby destroying many labile milk components as well as denaturing the key proteins. Of course, alternative methods to boiling such as heat treatment through pasteurization are needed to minimise the incidence of major milk-borne illnesses like tuberculosis, diphtheria and typhoid. The consumption of raw milk through commercial outlets is strictly limited worldwide for this reason. An education program developed with a joint government private enterprise consortium would go a long way to change consumer knowledge and demand for quality products at the right price. Part of this may be a school milk program where the same consortium funds the distribution of high quality milk to primary school children, while at the same time providing an education program for the whole community.

So where does this leave the small-holder dairy farmer who is losing money for every litre of milk he markets through the traditional chains? As is seen in most countries it is not the government extension services that are

destined to change the profitability of milk production: public sector organisations are typically ineffective because of their structural inertia. It is the private sector that needs to step forward to develop a much wider range of high quality dairy products and to educate the consumer on their health giving virtues. Extension services for farmers must be run using our "whole family approach" to achieve the best results. Inevitably the role of the traditional marketing chains will pass to innovative private entrepreneurs, both large and small as suggested by Reardon and Timmer above, to provide much more direct links between the farm gate and the consumer. Payment to farmers must be based on strict quality attributes including the elimination of heavy metals, insecticides, herbicides and mycotoxins from the milk. These regulations need to be implemented by private industry and not government organisations.

There are other contributing issues that will lead to the restructuring of the industry, however once the pricing of milk is linked to consumer demand for quality product, other changes in the production system will follow. However it is unlikely that 8.5 million dairy farmers will still be well managed viable production systems in 5-10 year time. The Pakistan dairy industry has the potential to be one of the most efficient in the world given the rich natural resources available to these farmers. We await the partnership of government and private industry to deliver for the dairy farmer of the future.

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The impact of extension programs to increase the productivity of the small-holder dairy farming industry of Pakistan

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Summary text for the table of contents

The 8.5 million small-holder dairy farmers of Pakistan produce over 90% of the nation's milk from 74 million animals averaging little more than 3 litres per day. Current marketing practices determine that they produce milk at a loss. This paper reviews our strategies to improve efficiencies in the industry to ensure that high quality milk will be available for the consumer into the future.

Abstract

Dairy farming operations with small animal numbers producing low volumes of around three litres per animal per day predominate in Pakistan's dairy industry. Although much of this is consumed domestically, many farmers sell small volumes into traditional milk marketing chains which feed the product into urban retail outlets. Analysis of these marketing chains shows that these farmers make a loss on every litre sold, while at the other end milk available to the consumer is of poor quality and often diluted as much as 1:2 with water. Small incremental profit margins are achieved by dilution and the use of distorted volume measures as the product is passed from small dealers to larger distributors and then to retail outlets. It is important that farmers are able to improve the efficiency of production by boosting the productivity of animals. This can be achieved through the adoption of better nutrition and animal husbandry practices. At the same time small scale local marketing chains require refinement to ensure profits generated from milk production stay with local communities. This paper reports on the development of effective extension strategies involving the whole family including the farmer, his wife and children. They have led to significant improvements in the profitability of small-holder dairy farming and a growing awareness of farmers of the commercial potential for their household cows and buffalo. The sustainability of these small-holder production systems in the face of changing consumer demands for higher quality products and world dairy product trade remain to be seen.

Keywords: Small-holder dairy farming Pakistan

Introduction

The current status of the industry

Pakistan's economy is heavily reliant upon agricultural production which accounts for 21% of the country's GDP, with livestock production contributing 56% of agriculture's contribution (Anon, 2015c). The livestock sector is dominated by milk and meat production with 62% of milk coming from a buffalo population of 34.6 million and the remainder coming from 39.7 million cattle, not all of which are kept for milk production (Anon, 2015a). The combined value of milk and meat of \$16.7 million exceeds the economic value of all cash crops (Food and Agriculture Organization, 2013).

There are 8.5 million rural smallholder and peri-urban dairy producers milking two to three milking animals which produce about 95% of the total milk production in Pakistan (Afzal, 2010). Around 80% of this comes from rural areas, many of which are isolated from major urban communities, while 15-20% is derived from peri-urban production units (FAO, 2011). Larger herds comprising more than 30 milking animals constitute only 0.3% of the total holding (Afzal, 2010). Most of the milk is produced in Punjab (63%) and Sindh (23%) while KPK and Baluchistan provide only 12% and 2% of total milk production in Pakistan (FAO, 2011).

While the statistics for milk production are impressive, the productivity per cow is very low by world standards. Animals produce as little as 3.15 litres per day when in many cases they have the genetic capability to produce 4 or 5 times this volume. While annual production is increasing at the rate of 53.2% per annum (Anon, 2015a), consumer demand is increasing by 15% each year to an impressive 43.2 million tonnes, some 5 million tonnes more than is produced. While the average length of lactation for the international dairy herd is 305 days which in many cases can be extended to 600 days (Abdelsayed et al., 2015), Pakistani buffalo and cattle struggle to maintain productivity beyond 200 days.

Furthermore the estimated 3.2% increase in production annually is offset by a 15% increase in consumer demand which is currently at 43.2 million tonnes, some 5 million tonnes above annual total production. The shortfall is made up from imports of whole milk powder, of which there is up to 50,000 tonnes being traded globally every month (Anon, 2015b).

Sustainability of the industry.

Increasingly livestock rearing and the consumption of animal products are seen as being environmentally unsustainable. Yet global demand for meat and dairy products continues to rise with the increase in urbanization of four world populations (Stoll-Kleeman and O'Riordan, 2015). The contributions of methane, nitrous oxide and carbon dioxide from livestock to global warming are significant with 18% of total global emissions coming from this source (FAO, 2006). The use of poor quality roughages and overstocking of fragile environments leading to low productivity per animal in the developing world exacerbates this problem. Clearly improvements in the efficiency of production per animal is required to reduce averse environmental impacts while at the same time providing more high quality food for the world (Foley et al., 2011).

Given the structure of the dairy industry in Pakistan based on small-holder production and the reliance of both urban and rural communities on milk as the major source of animal protein, we have focused on ways of increasing milk production per animal on small-holder farms. Dairy animals are ideally suited to meeting these basic requirements since the family's daily needs can be provided in just a single milking of one or two animals for up to 250 days over the course of lactation. In contrast the lack of refrigeration prevents the storage of meat in village households and so slaughter of livestock is not viable unless the family is able to sell meat to the local community.

Challenges associated with the dissemination of information to small-holder farmers. Firstly it is important to understand the reasons that farmers own animals. Apart from providing milk the number of animals owned by the family conveys social status within their community, while at the same time acting as a bank or insurance to meet essential family expenses such as the financing of weddings, funerals and expensive medical care. So profitability of any small-holder dairy enterprise is not necessarily a prime concern for the farmer. Often the most profitable aspect of any farm is the cropping component, with the dairy enterprise very much a secondary concern. Very often there is no informal marketing chain for milk particularly in more isolated regions where up to 70% of farmers produce milk solely to feed their families (Raja, 2003).

In a recent survey reported by Godfrey (2015) of 115 small-holder dairy farming operations in the irrigated region of Okara, gross farm analysis showed that the predominance of these dairy operations were not profitable (Table 1). The financial viability of the whole operation was dependent on the amount of finance borrowed to remain operational. Given that the profitability of the farm was carried by the cropping component, farmers were less receptive to any advice offered to improve the profitability of their dairy operations. Interestingly the return on assets was similar to that observed in many Australian farming operations.

Milk production per year (kg)	<2,300	2,300-3,700	3,700-10,100	SED	P Value
Number of farms	39	38	38		
Ave production/animal (kg/yr)	780	990	1234		
Ave milk price (PKR/kg)	22.89	22.84	23.25	0.60	0.752
Cost of production (PKR/kg)	57.05	33.66	24.15	4.94	<0.001
Profit (PKR/kg)	-34.16	-10.82	-0.9		
Enterprise milk profit (PKR/kg)	-43,072	-32,064	-679	8,355	<0.001
Total crop GM (PKR)	271,487	353,346	442,862	68,443	0.047
Whole farm GM (PKR)	228,202	299,082	464,162	89,724	0.019
Whole farm op. profit (PKR)	134,651	170,029	303,281	78,667	0.082
Total finance costs (PKR)	401,817	489,926	594,840	77,836	0.049
Net profit	-267,166	-319,896	-291,559	70,390	0.755
Return on assets (%)	3.27	1.37	4.24	1.84	0.290

Table 1 Gross margin analysis of small-holder dairy operations in the Okara irrigated dairy region of Punjab state in 2011. The farms are categorized according to the volume of milk produced each year. Data are given as means and variance estimated as a mean SED for each variable (source: 2008-2009 Agriculture Sector Linkages Dairy Program farmers survey-unpublished)

The failure of farmers to be compensated adequately with a fair market price for their milk is due to the structure of marketing chains connecting them to consumers, most of whom are urban based. Factors influencing price received on farm are dependent on whether milk is derived from buffalo or cattle, location of farms relative to markets, season, the educational status of the farmer and the pricing policies of statutory organisations affected the price received for milk on farm. Milk processors have the potential to collude to control prices, which are based on the economic imperatives of the companies rather than the cost of production for small-holder farmers.

The structure of one marketing chain is described in detail by Godfrey (2015) is given in Figure 1.



Figure 1: Pyramid of the relationships between participants in a fresh un-packaged milk value chain servicing the needs of small-holder milk producers in the irrigated Okara-Lahore region (Godfrey, 2015)

Given the number of participants in these informal marketing chains relative to the number of farmers being serviced and the fixation of milk prices in the urban marketplace, there is little room for profit sharing for the milk collectors or the distributor that form the chain and even less scope for a realistic price being offered to the farmer. The dilution of milk with water and the addition of other adulterants to boost perceived protein and fat contents by these operators is essential for profit sharing along the chain.

In a survey of marketing chains in 3 districts of Punjab and found that the addition of water added up to 40% of the volume of milk sold in the urban marketplace (Table 2).

Districts	Fat%	Protein%	SNF%	Lactose%	Added water%
Kasur	2.2	1.7	4.5	2.4	30.5
Okara	2.7	1.9	5.1	2.7	41.2
Pakpattan	3.2	2.4	6.3	3.3	26.0

Table 2: Composition of milk collected in the urban marketplaces of Okara, Pakpattan and Kasur in Central Punjab Province Pakistan (Aslam, 2015).

Typically consumers are looking for milk containing 4% fat and this can be achieved by vendors strategically by blending the higher fat buffalo milk (<6%) with cow milk together with water. Yet in our survey of milk collected from milk outlets in Okara, Pakpattan and Kasur fat content was as low as 2.2% (Table 2). At this level the yield of ghee upon boiling is unacceptable for the consumer. It is also the low protein level that should be of concern to the consumer with concentrations of less than 2% (Table 2) often being found in the marketplace suggesting that the dilution rate can be as high as 1:2 along some marketing chains. The containers used for measuring milk volume along the chain also favour the trader, while in many commercial outlets the consumer is provided with 900ml when in fact she has paid for one litre. The alternative source of product from milk manufacturers is also problematic, since product labelling provides no fat concentration information at all, but rather an energy content; this provides no information on composition at all.

Pakistan, however, is no different to other developing countries where informal marketing systems facilitate the supply of cheap calories for the consumer (FAO, 2013). Yet the presence of 7 middlemen in a marketing chain (Figure 1) through which only small volumes of milk flow, each making a small margin on the product they handle, is unsustainable in the long run.

Such a finding is not unusual since policies and development strategies in many countries often fail to recognize and provide adequate support for small holder production systems and associated marketing chains. Instead the future is seen to lie with higher profile industrial production (Echeverría et al., 2011). However there is still a place for informal marketing chains in rural environments, particularly where there is no alternative for selling milk. These however must provide a more direct link between the producer and the consumer to ensure high quality product reaches the consumer while adequate profits generated from the sale pass back to the farmer.

The challenges of improving extension services to small-holder farmers

Given the poor scope for improved milk prices received on farm and the secondary importance of milk production in the whole farm budget for many farmers, it is little wonder that high rates of adoption for extension messages delivered by veterinary staff are rare. Of course it is important to note that no more than 40% of small-holder farmers in Pakistan receive any form of extension services from either government, NGO or private industry sources. The effectiveness of extension using the traditional format of didactic delivery of information to male farmers has also been found to be wanting.

This can easily be explained by the fact that males mostly have very little to do with the collection of fodder, feeding, shed management and value adding to the milk produced (Table 3).

Activity	Men	Women	Girls	Boys
Feeding	0.70 ^a	2.53	1.50	0.60
Collectingfodder	0.60	2.70	0.5	0.40
Shedmanagement	0.70	2.75	0.95	0.45
Marketingofproducts	2.00	1.00	1.00	1.00
Vaccinationandtreatment	2.10	1.30	0.80	0.30
Dairyproductpreparation	0	2.00	2.00	0
Average	1.01	2.04	1.13	0.45

Table 3. The extent of involvement of members of the small-holder farming family in livestock production practices on farm.

^a Ranking, 0-1, minimum participation; 1.1-2, Some participation; 2.1-3, Maximum participation (source: adapted from Zia, Mahmood, & Ali, 2011)

Given this information logic would dictate that an extension program focussing on improving animal productivity should focus on women and children and not the male members of the household. Yet the social structure of the traditional Pakistani family shows clearly that the male is the family leader and therefore the recipient of all information that is important for the sustainability of the family.

The Agriculture Sector Linkages program (ASLP) dairy program has focused on refining their extension approach to incorporate the farmer, his wife, his children and then those that provide ancillary services to the community. These included the village school teacher and community health worker. This has led to much higher adoption rates of key extension messages, which initially were as low as 15% using the traditional "male only" approach. Anecdotal evidence has suggested that this approach leads to significant discussion within the family unit on each extension message. In the case of calf rearing, the children were often active participants in the family debate, with their interest being engendered from the school environment in which extension messages are displayed prominently on bulletin boards.

Assessment of the impact of four extension programs

The success of any extension program can only be gauged by monitoring the increase in knowledge on the subject material among the farming communities. There was a requirement for a carefully constructed survey in which farmers and their families were assessed on their level of knowledge of the fundamental principles underlying the key messages. These messages related to major limitations to productivity that have been observed in small-holder dairy production systems.

They included:

- the need to untie animals to provide free access to water and feed;
- the requirement for a 2-month dry period between lactations;
- the need to vaccinate animals using high quality effective vaccines;
- the selection of provensires for mating;
- correctly balancing feed to provide adequate energy, protein and trace minerals;
- the use of high quality seed to maximise forage production.

The survey of 399 farmers showed that the knowledge of so-called traditional farmers who have not been trained in the program varied markedly over these subject areas (Figure 2). Remarkably their knowledge of the importance of colostrum to the survival of the newborn calf was very low, while at the other end of the spectrum, a very high proportion understood the concept of the importance of providing water and feed ad libitum.

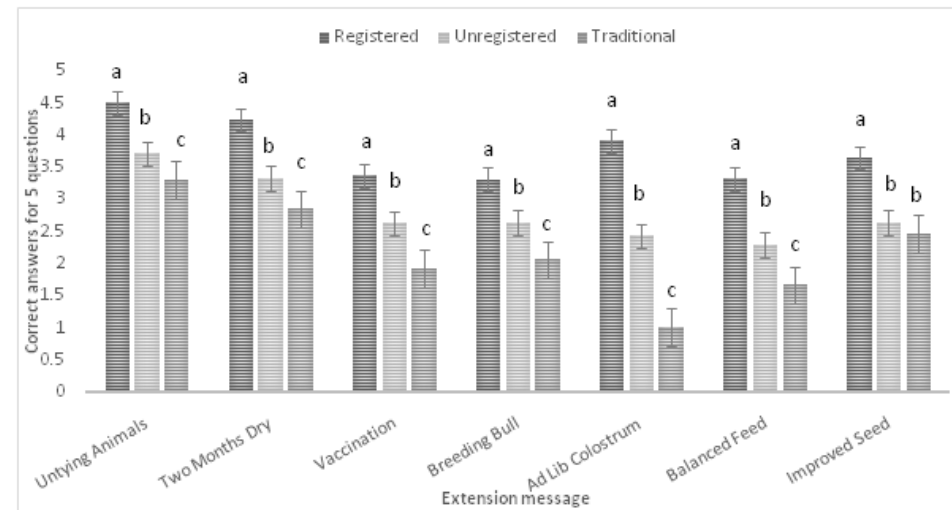


Figure 2: The impact of the provision of extension services to small-holder dairy farmers on their knowledge of 7 major parameters contributing to whole farm productivity. Farmers were asked 5 standard questions relating to each topic. Number of farmers from across 5 districts in Punjab interviewed in this survey, registered: 179; unregistered: 116; traditional: 104. Columns within production parameter with different superscripts (a, b, c) were significantly different ($p < 0.05$).

While the impact of the extension programme is clearly visible with these results, the interesting observation is that farmers who attend meetings but do not want to be engaged directly with our programme also benefit. However, it is not possible to assess whether these farmers implement the knowledge they have acquired as their production systems are not monitored.

It is fair to say that our programme refined over 7 years in the two major provinces, Punjab and Sindh, has generated significant advances in productivity in the farming communities we have worked with. The future of traditional marketing chains and small-holder dairy farming.

The future of traditional marketing chains and small-holder dairy farming.

History has shown that traditional informal marketing chains evolve over time. In North America and Europe in the 19th century, grain, milk, meat and fibre produced by small-holder farmers at the local level were traded to local small-scale merchants in the raw state or perhaps with a single step of processing such as churning cream to form butter or grinding grain to flour. Governments softened the price but there was little regulation of product quality.

Then with the advent of the industrial revolution, the food production and marketing systems of Europe and North America underwent profound change. The establishment of large-scale first-stage processing companies, for example grind grain (e.g. Cargill) or process milk, led to the development of secondary processing companies producing refined food products (e.g. Nestle). Mass distribution of milk was not possible until the advent of the milk bottle sealed with wax in 1884 and then the plastic-coated cardboard milk carton in 1932 (Bellis, 2014).

These changes lead inevitably to large-scale chain stores and supermarkets with associated large wholesale and logistics companies. Inevitably, this leads to pressure on farmers to produce more with lower profit margins per litre. If farmers were not able to increase their herd, they went out of business, which had led to commercial herds in California of more than 2000 cows. Similar trends have been seen in Australia. While the sceptics said this would not happen in developing countries, this has not proved to be the case. Food systems in developing countries in Latin America, Asia, Eastern Europe, and some of Africa have undergone a revolution since the 1980s, resulting in the demise of small-holder farming systems (Reardon and Timmer, 2012).

Inevitably Pakistan's 8.5 million small-holder dairy farmers will be subjected to the whims of the world's economy. The industry will be subject to change as the expanding middle class consumer demands higher quality dairy products that meet world food safety standards. However any expansion of the industry will have to involve improved efficiencies in production that also account for environmental sustainability. Whether small-holder farming communities can meet these challenges remain to be seen.

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