



Standardization of pulsation ratios in crossbred cows milked in automated herringbone milking parlour*

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Pulsation has a fundamental role in milking and is used to regulate the different phases of milking when milk flow is established. It causes cyclic pressure change which regulates the opening and closing of the cluster liners in the proximity of teat-ends, providing massage to the lower part of the teat followed by the milking phase. The pulsation ratio of a milking machine plays a significant role in optimizing the milking performance and ensuring good teat-ends and udder health (Strobel *et al.* 2016). Evidences from earlier report show that pulsation ratio affects milk flow and milking time, and influence the milk somatic cell count (SCC) (Gleeson *et al.* 2004, Bade *et al.* 2009). Incorrect pulsation setting is found to cause over-milking, especially in front teats in herds where conventional cluster milking (whole-udder level) is practiced in comparison to farms where each udder quarter is milked separately as in automatic milking system (AMS farms) (Svennersten-Sjaunja and Pettersson 2008). Despite the milking machine being introduced in dairy production since long in India, there are very few studies concerning technical improvement in mechanized milking system that could meet the demand for efficient milking in native cows.

The present study was conducted at Livestock Research Center of the institute on the crossbred cattle herd maintained in loose housing system under group management practice. Crossbred dairy cows (56) in lactation 1st to 4th were selected for the study. The animals were categorized into 3 groups based on the level of production i.e. low yielder (cows yielding less than 12 kg

of milk, 16), medium yielder (cows yielding milk between 12–18 kg, 32), high yielder (cows yielding more than 18 kg of milk/day, 8). The animals were monitored through automatic animal identification system consisting of neck transponder, portal identification antenna, system controller and software based herd management programme on personal computer (PC). Milking of animals was done separately for each group thrice a day i.e. in morning between 0500 to 0700 h, in afternoon (between 1230 to 1330 h) and in evening (between 1730 to 1830 h) in 2 × 8 De Laval low-line automated herringbone milking parlour. The pulsation ratio tested during the study were 65:35 and 60:40 for each cow keeping the vacuum level constant at 42 kPa, and automatic cluster removal (ACR) at 0.2 kg/min. The pulsation system was alternating type (2×2) and the duration of different phases of pulsation ratio (a, b, c and d) are summarized in Table 1. The cows were milked at the 2 ratios for 3 days, each treatment given 1 day adjustment time before taking the observations. The same treatment was repeated thrice to balance results for variation in milk yield on daily basis. Statistical analysis of data generated was analyzed using general linear model procedure in SAS 9.3 version software.

The milkability traits in crossbred dairy cows at 65:35 and 60:40 pulsation ratios showed no effect on mean daily milk yield and milk yield/milking session (morning) (Table 2). The yield first 2 min of milking, machine-on time and average milk flow rate were similar at the 2 ratios irrespective of the level of production in crossbred cows. The peak milk flow rate also did not vary and was similar at 2 ratios and levels of production. Ferneborg and Sevennersten-Sjaunja (2015) also reported non-significant effect on milk yield comparing the 2 ratios in addition to other higher ratios (70:30 and 75:25). However, contrary to our results, they observed decrease in average and peak milk flow at 60:40 as compared to standard 65:35. This may be due to difference in milking machine and the mechanization, as in their case, milking was quarter based in which each udder quarter is milked separately in contrast to our conventional udder based milking system. Another possible reason could be due to difference in vacuum as in

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Table 1. Duration of the four phases (a-d) of pulsation

Pulsation ratio	Pulsation phase length (ms)			
	a	b	c	d
65:35	146	500	106	245
60:40	146	450	106	287

Pulsation rate, 60 cycles/min; System vacuum, 42 kPa.

Table 2. Milkability (Mean±SE) of crossbred cows at 65:35 and 60:40 pulsation ratios

Parameter	Level of production	Pulsation ratio		
		65:35	60:40	Overall
Total milk yield (kg)	<12 kg	10.92±0.26	11.35±0.26	11.14±0.19 ^A
	12–18 kg	15.00±0.32	14.50±0.32	14.75±0.23 ^B
	>18 kg	20.85±0.50	20.26±0.50	20.55±0.33 ^C
Milk yield first session (kg)	<12 kg	5.10±0.13	5.10±0.13	5.10±0.09 ^A
	12–18 kg	6.88±0.17	6.64±0.17	6.76±0.12 ^B
	>18 kg	9.75±0.20	9.65±0.20	9.70±0.14 ^C
Yield first 2 min (kg)	<12 kg	2.14±0.07	2.10±0.07	2.12±0.05 ^A
	12–18 kg	2.77±0.07	2.75±0.07	2.76±0.05 ^B
	>18 kg	3.79±0.11	3.74±0.11	3.76±0.08 ^C
Machine-on time (min)	<12 kg	4.34±0.09	4.11±0.09	4.23±0.06 ^A
	12–18 kg	4.74±0.12	4.58±0.12	4.66±0.09 ^B
	>18 kg	5.24±0.14	5.14±0.14	5.19±0.10 ^C
Average milk flow (kg/min)	<12 kg	1.12±0.06	1.14±0.06	1.13±0.04 ^A
	12–18 kg	1.39±0.07	1.28±0.07	1.34±0.05 ^B
	>18 kg	1.66±0.08	1.47±0.08	1.56±0.06 ^C
Peak milk flow (kg/min)	<12 kg	2.49±0.10	2.51±0.10	2.50±0.07
	12–18 kg	2.61±0.14	2.51±0.14	2.56±0.10
	>18 kg	2.78±0.16	2.66±0.16	2.72±0.11
<i>Milk flow rate (0–120 sec)</i>				
Flow 0–15 sec (kg/min)	<12 kg	0.21±0.03 ^a	0.10±0.03 ^b	0.15±0.02
	12–18 kg	0.10±0.02	0.11±0.02	0.11±0.01
	>18 kg	0.12±0.04	0.21±0.04	0.16±0.03
Flow 15–30 sec (kg/min)	<12 kg	1.29±0.10 ^a	1.00±0.10 ^b	1.10±0.07
	12–18 kg	0.92±0.07	0.95±0.07	0.93±0.05
	>18 kg	1.01±0.15	1.30±0.15	1.15±0.10
Flow 30–60 sec (kg/min)	<12 kg	1.07±0.10	1.09±0.10	1.08±0.07 ^a
	12–18 kg	1.22±0.06	1.19±0.06	1.21±0.04 ^{ab}
	>18 kg	1.43±0.18	1.33±0.18	1.38±0.13 ^b
Flow 60–120 sec (kg/min)	<12 kg	1.41±0.08	1.45±0.08	1.43±0.06
	12–18 kg	1.64±0.09	1.54±0.09	1.59±0.06
	>18 kg	1.59±0.15	1.57±0.15	1.58±0.11

Means bearing different superscript within a row in small letter and within a column in capital letter differ significantly at 0.01 level of significance.

our case the vacuum level was kept constant at 42 kPa, which was lower as compared to above study. Other researchers compared wider ratios and found no difference in milk production on individual cow basis (Thomas *et al.* 1993). The results were in agreement to these researchers as milk production did not differ at two ratios. For conventional milking systems, it was reported that higher pulsation ratio (70:30 or 75:25) could be associated with alteration in milking time and milk flow (Gleeson *et al.* 2004) which was not within the scope of present study. The results of present study on milkability suggest that either

Table 3. Milk EC, SCC and milking irregularities (Mean±SE) in crossbred cows at different pulsation ratio

Milkability parameters	Level of production	Pulsation ratio		
		65:35	60:40	Overall
Electrical conductivity (ms/cm)	<12 kg	6.66±0.14	6.71±0.14	6.68±0.10
	12–18 kg	6.58±0.19	6.52±0.19	6.55±0.14
	>18 kg	7.03±0.12	6.90±0.12	6.96±0.08
Somatic cell count (X10 ⁵)	<12 kg	1.52±0.17	1.60±0.17	1.56±0.12
	12–18 kg	1.64±0.16	1.65±0.16	1.64±0.11
	>18 kg	1.36±0.26	1.37±0.26	1.37±0.19
Cluster slips (%)	<12 kg	—	—	—
	12–18 kg	1.00±0.50	1.00±0.50	1.00±0.40 ^A
	>18 kg	3.90±2.20	4.70±2.20	4.30±1.60 ^B
Cluster re-attach (%)	<12 kg	—	—	—
	12–18 kg	1.30±0.70	1.40±0.70	1.40±0.50 ^A
	>18 kg	6.40±2.70	6.40±2.70	6.40±1.90 ^B
Post milking strip yield	<12 kg	124.88±14.93	128.44±14.93	126.66±10.56
	12–18 kg	165.16±16.90	172.91±16.90	169.03±11.95
	>18 kg	152.13±54.26	173.75±54.26	162.94±38.37

Means bearing different superscript within a row in small letter and within a column in capital letter differ significantly at 0.01

of the two ratios i.e. 65:35 and 60:40 were equally suitable for milking as far as Indian dairy crossbred cows are concerned.

The mean milk flow rate in 0–15, 15–30, 30–60 and 60–120 sec of milking did not differ on changing the pulsation ratio (65:35 and 60:40) for medium and high yielders. However, for low yielder cows producing on an average less than 12 kg daily milk production, showed significant ($P<0.05$) effect of pulsation ratio at the start of milking. At 65:35 pulsation ratio, the initial flow at 0–15 and 15–30 sec of milking was significantly higher as compared to 60:40, which may be due to faster let down of milk. This was responsible for the bimodality seen in the milk flow rate curve at 65:35 ratio in crossbred cows producing lower milk in a session. Such milk flow rate curve is characterized by steep decrease in milk flow in time less than 60 sec after the beginning of milking in cows and reported to have significant ($P<0.001$) negative correlation with milk production traits (Strapak *et al.* 2011, Fahim *et al.* 2017). The results of present study were in agreement with Strapak *et al.* (2011) for the low yielder cows as such cow produced 0.43 kg, although the difference was statistically non-significant. Similar findings were reported by Fahim *et al.* (2017) on studying the effect of parity, stage of lactation and udder type on milkability of crossbred cows.

The milk EC and SCC were not affected by the change in pulsation ratios due to which it may be suggested that the milk was of similar quality and both the ratios were equally suitable for milking of crossbred cows. The results were in agreement with the findings of Thomas *et al.* (1993) comparing even higher ratios at large commercial dairy herds without any significant differences for the udder health measures of bulk tank SCC, clinical mastitis cases, or number of cows culled for mastitis. Similarly, Gleeson *et*

al. (2004) reported that increasing the pulsation ratio from 60:40 to 67:33 with a pulsation rate of 60 cpm during three consecutive days had no effect on teat tissue and milk SCC. Ferneborg and Svennersten-Sjaunja (2015) also compared 3 pulsation ratios (60:40, 70:30 and 75:25) with the standard pulsation ratio (65:35) during 6 weeks and found no negative effects on teat condition or milk SCC with any of the pulsation ratios applied during the study. The results of our investigations were in complete agreement with the results of above researchers. No negative effects on milk EC or SCC were observed with any of the pulsation ratios applied during the study.

The milking irregularities observed at the two pulsation ratio studied were within the prescribed standards of milking procedure given in ISO and ASABE standards (Mein and Reid 1996) and did not differ significantly. However, there were fewer cluster slips, almost same cluster reattachments and slightly better milking in terms of post-milking strip yield at 65:35 as compared to 60:40. No other detrimental effects on milking performance were seen for these two ratios. In a report submitted for milking-time test and guidelines for milking unit, a goal of <5–10/slips/100 cow milkings has been suggested as a thumb rule to efficiently handle the milking operations (Mein and Reid 1996). A wide range of variation (0–25%) in unit reattachment rate was reported in a farm survey data of commercial dairy which makes milking efficiency poorer (Ruegg *et al.* 2005). The results of present investigation were in line with the prescribed standards mentioned above for an efficient milking in mechanized system. Milking irregularities being common problems encountered during milking operations at most of the dairy farms, still there are very few reports available on this aspect. It reduces the efficiency of milking parlour by increasing time consumed per batch of cows milked (Fahim *et al.* 2017). The occurrence of milking irregularities with respect to machine settings, with an aim to minimize them may help in improving the performance of milking herd.

SUMMARY

It was concluded from the study that both the pulsation ratios (65:35 and 60:40) performed equally well in crossbred cows and did not affect the milk yield. There was no significant effect on milkability traits, although in low yielder cows, significant effect on milk flow rate was observed at the start of milking. The initial milk flow was significantly higher at 65:35 as compared to 60:40, responsible for the bimodality in the milk flow rate curve in low producers.

There were no negative effects on milk EC and SCC at the two ratios. The milking irregularities were also within the prescribed standards at both the ratios. Thus, both the ratios are equally suitable for milking of crossbred cows under Indian conditions.

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Effectiveness of various sire evaluation methods in ranking Mehsana bulls based on FL305MY and TMY under field progeny testing programme

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Buffaloes in India have a place of pride in all respects, viz. production, productivity and population in the world, diversity, service to the underprivileged, clean draft power, sure-footed transport, rich food/nutrition, support to a host of industries and livelihoods etc. Mehsana is one of the world's best dairy type buffalo with superior genetic potential for milk production. Dudhsagar Research and Development Association (DURDA), Dudhsagar Dairy, Mehsana, is involved in breeding and improvement programme for Mehsana buffaloes in Gujarat by undertaking field progeny testing programme since 1985. The accurate measurement of the milk production of individual buffalo is of major interest in dairy genetic improvement programs (Parmar *et al.* 2018). It is expected that demand for milk in India is likely to increase to besides 200 million tonnes by 2021–22. To meet the growing demand, it is necessary to improve the native breeds through proper breeding system besides identification of high genetic merit bull for healthy semen production in field. Keeping the above facts in view, the present study was undertaken to assess the breeding values of sires by least squares (LS) technique, BLUP Sire model and BLUP Animal model based on first lactation milk yield under field progeny testing programme as well as effectiveness of different sire evaluation methods by studying their efficiency to discriminate amongst Mehsana buffalo bulls.

The first lactation monthly test-day milk yield data of Mehsana buffaloes that calved between 1989 and 2013 at the farms located in the field progeny testing areas of Dudhsagar Research and Development Association were used. Villages (74) were clustered into 3 groups based on the geographical location. The duration of 25 years was

classified in to 5 periods each of 5 years. Each year of calving was further classified into 3 seasons, viz. winter (November to February), summer (March to June), rainy (July to October). Only the sires having records on at least 10 daughters were included in the present study.

First lactation milk yield was calculated using two approaches. First, test-day recordings were used to generate FL305MY using the formula as recommended by Department of Animal Husbandry and Dairying (DAHD 2009), Government of India. Second approach was to calculate total first lactation milk yield (TMY) as per the procedure suggested by the International Committee for Animal Recording (ICAR 2014).

Three different sire evaluation methods, viz. least-squares analysis of variance (Harvey 1979) to estimate the breeding values for bulls; best linear unbiased prediction sire model (BLUP-SM), where (co)variance components were estimated by best linear unbiased prediction animal model (BLUP) in WOMBAT genetic analysis tool (Meyer 2007) and best linear unbiased prediction animal model (BLUP-AM) were employed. The single trait animal model was considered for estimation of breeding values.

The effectiveness of sire evaluation methods was judged by using various criteria, viz. within sire variance (or error variance) and rank correlations. The method with the lowest error variance was considered as the most efficient method. Spearman's rank correlation between the ranks based on breeding values of sires derived by 2 methods was estimated. Higher rank correlation between the sire evaluation methods indicated higher degree of similarity of ranking by 2 methods.

Sire evaluation involves the estimation of breeding value of the bulls on the basis of first lactation 305-days milk yield of their daughters with a rationale to minimise the generation interval. The average expected breeding value of Mehsana buffalo bulls by LS method for FL305MY was 2,284.02 litres (Table 1), which ranged from 2,767.52 litres 1,987.10 litres. The breeding values of as many as 81 sires (44.51%) were more than the average expected breeding value. Similarly, the expected breeding value by best linear

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Table 1. Average breeding value (BV) estimates of sires for first lactation 305-day milk yield and total milk yield by different methods

Trait	Sire evaluation method	Average BV (Litre)	Number of sires above average	Number of sires below average	Maximum BV (Litre)	Minimum BV (Litre)	Range of BV (Litre)
FL305MY	LS	2284.03	81 (44.51%)	101 (55.49%)	2767.52	1987.10	780.42
	BLUP-SM	2242.11	81 (44.51%)	101 (55.49%)	2713.44	1946.15	767.29
	BLUP-AM	2256.81	81 (44.51%)	101 (55.49%)	3072.86	1737.13	1335.73
TMY	LS	1913.74	83 (45.61%)	99 (54.39%)	2345.03	1598.74	746.29
	BLUP-SM	1870.57	85 (46.71%)	97 (53.29%)	2283.84	1553.61	730.23
	BLUP-AM	1891.24	87 (47.81%)	95 (52.19%)	2590.17	1347.00	1243.17

Table 2. Relative efficiencies of different sire evaluation methods for different first lactation traits in Mehsana buffaloes

Trait	Method	Error variance	Relative efficiency (%)
FL305MY	LSM	1591471963.00	67
	BLUP-SM	1577000000.00	68
	BLUP-AM	1071570000.00	100
TMY	LSM	1642950151.00	53
	BLUP-SM	1361890000.00	64
	BLUP-AM	875461000.00	100

unbiased prediction sire model (BLUP-SM) was 2,242.11 litres, which ranged from 2,713.44 to 1,946.15 litres. The average expected breeding value of Mehsana buffalo bulls obtained by both LSM and BLUP-SM was higher than those previously reported by Singh *et al.* (2014) and Chaudhari *et al.* (2015) in Murrah buffaloes. The average expected breeding value obtained by BLUP-AM for FL305MY was 2,256.81 litres, which ranged from 3,072.86 to 1,737.13 litres. The obtained breeding value was higher than those reported by Singh *et al.* (2014) and Yadav and Singh (2015) in Murrah buffaloes. Number of sires above and below the average expected breeding value was similar with all the methods used and it was same as estimated by the least squares method (Table 1).

The breeding values of the Mehsana bulls under study for TMY by LS, BLUP-SM and BLUP-AM were 1,913.74, 1,870.57 and 1,891.24 litres, respectively (Table 1). Similarly, range of these estimates was 2,345.03 to 1,598.74, 2,283.84 to 1,553.61 and 2,590.17 to 1,347.00 litres, respectively, by LS, BLUP-SM and BLUP-AM methods. The number of bulls having breeding values more than the average breeding value was highest by BLUP-AM followed by BLUP-SM and LSM.

The sire evaluation method/model which gave the lowest error variance was considered to be most efficient and taken as the base for efficiency estimation of other methods. BLUP-AM method had lowest error variance both for FL305MY and TMY as compared to other methods of sire evaluation, indicating it as most efficient methods (Table 2). The relative efficiency of LSM and BLUP-SM method for FL305MY were 67 and 68%, respectively. Similarly, the relative efficiency of LSM and BLUP-SM

method for TMY was 53 and 64%, respectively. Singh *et al.* (2014) and Dash *et al.* (2014) also found BLUP-AM method to be more efficient than other methods in Murrah buffaloes and in Holstein Friesians Crossbred respectively.

The values of rank correlation coefficients of LSM and BLUP-SM methods with most efficient BLUP-AM method were 0.954 and 0.953, respectively. Similarly, for TMY, the values of rank correlation coefficients of LSM and BLUP-SM with BLUP-AM method were 0.920 and 0.922, respectively. These estimates of rank correlation coefficients for both the traits studied were highly significant ($P \leq 0.01$). As such these two methods are expected to rank sires with similar accuracy as compared to the most efficient method. However, the magnitude of the coefficients of rank correlation indicated that in comparison to BLUP-AM, relative accuracy was highest for LSM followed by BLUP-SM methods (Table 3). The present findings regarding the ranking of different sire evaluation methods was in agreement with the reports of Singh *et al.* (2014), Chaudhari *et al.* (2015) and Kumar *et al.* (2015) in Murrah buffaloes.

Based on these results, it was concluded that the breeding values of Mehsana buffalo bulls for first lactation milk yield by all the 3 methods were higher than those documented for Murrah buffaloes by various workers. This suggests that Mehsana buffaloes have higher genetic potential for milk production. BLUP-AM method was relatively more efficient, accurate and stable with lowest genetic variation than other methods of sire evaluation. Hence this model is recommended for genetic evaluation of Mehsana buffalo

Table 3. Rank correlations between breeding values of Mehsana buffalo bulls for FL305MY and TMY by different methods

Method	LSM	BLUP-SM	BLUP-AM
FL305MY			
LSM	1	1.000**	0.954**
BLUP-SM		1	0.953**
BLUP-AM			1
TMY			
LSM	1	0.984**	0.920**
BLUP-SM		1	0.922**
BLUP-AM			1

**Significant ($P \leq 0.01$).

bulls. The rank correlation coefficients among breeding value of sires by all the methods are expected to rank sires with fairly high accuracy. The results appear to promise further improvement in milk production through the use of progeny tested bulls in the years to come and use of top ranking sires under field conditions appears to promise substantial improvement in the milk yield of Mehsana buffaloes.

SUMMARY

First lactation records of 8,072 Mehsana buffaloes, progeny of 182 bulls, spread over a period of 25 years (1989–2013) maintained under field progeny testing programme of DURDA, Dudhsagar Dairy, Mehsana, Gujarat, India were used to estimate the breeding values of Mehsana buffalo bulls for first lactation milk yield (FL305MY and TMY) by least squares, BLUP-SM and BLUP-AM methods. The effectiveness of different sire evaluation methods was compared using error variance and rank correlations among the estimated breeding values of sires. The average breeding values for FL305MY by LS, BLUP-SM and BLUP-AM were 2284.02, 2242.11 and 2256.81 litres, respectively. The corresponding values for TMY were 1913.74, 1870.57 and 1891.24 litres, respectively. BLUP-AM method had lowest error variance for both FL305MY and TMY as compared to LS and BLUP-SM methods making it as the most efficient method. The rank correlation coefficients indicated that all the methods could rank the sires with fairly high accuracy with highest relative accuracy for BLUP-AM.

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