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Effect of clinical mastitis on lactation curves of Murrah buffaloes

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ABSTRACT

India accounts 18.5% of the world milk production and ranked first in milk production. Buffaloes (*Bubalus bubalis*) produce 54% of total milk production of India. However, mastitis remains the most expensive production disease of the buffaloes. The present study was based on the lactation records of Murrah buffaloes maintained at Cattle and Buffalo Breeding Farm of LPM Section, ICAR-Indian veterinary research institute, Izatnagar over a period of 10 years (2005–2014). The aim of present study was to find best fitted lactation model explaining the lactation behaviour of Murrah buffaloes in healthy and mastitis condition. The data consisted of 80068 daily test day milk yield records of 296 Murrah buffaloes. Different standard lactation curve models such as Ali and Schaeffer (1987), Cobby and Le Du (1978), Sikka (1950), Mitscherlich × Exponential (Rook *et al.* 1993), Mixed log (Guo and Swalve 1995), Wilmink (1987) and Wood (1967) models were fitted by Proc NLIN Procedure of SAS 9.3. The goodness of fit was judged by the high value of R^2_{adj} and low value of MSPE, AIC and BIC. Durbin-Watson test was used to test autocorrelation and Shapiro-Wilk's test and Kolmogorov Smirnov test was used to test the normality of the residuals. Based on the analysis of the data Ali and Schaeffer model was the best fitted model to explain the lactation behaviour of the healthy as well as mastitic Murrah buffaloes.

Key words: Daily test day milk yield, Goodness of fit, Lactation curve, Mastitis

Agriculture and allied activities remain the major source of livelihood for nearly half of the Indian population. The share of agriculture in employment was 48.9% of the workforce (National Sample Survey Office 2011–12) while its share in the Gross Domestic Product (GDP) was 17.32% in 2016–17 at current prices. Animal husbandry sector contributes approximately one-fourth of the Agricultural GDP, whereas the dairy sector contributes two-third of GDP from the animal husbandry sector.

India ranks first in milk production, accounting 18.5% of world milk production, achieving an estimated annual output of 155.49 million tonnes during 2015–16.Whereas per capita availability of milk in India has increased from 112 grams per day (1970–71) to about 337 grams per day (2015–16) which is higher than the world average (285 grams per day) and minimum nutritional requirement of 280 grams recommended by the Indian Council of Medical Research (ICMR). However, there are wide inter-state and inter-regional differences in terms of per capita availability of milk. The average per capita availability varies from as low as 57 g/day in Mizoram to 1,032 g/day in Punjab.

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The milk production is a continuous process but the rate of secretion of milk is not constant and follows a curvilinear pattern. There is a marked increase in milk yield in dairy animals which reaches to peak at 2 to 8 weeks and then gradually declines. However, the nutrition, disease conditions, genetic makeup of the animals and managemental practices affect the milk production. Macciotta *et al.* (2006) studied and concluded that the age, parity and calving season have the significant effect on shape and scale of lactation curves.

Mastitis is known to be most economically devastating disease hampering desired progress in the dairy industry. The presence of bacteria and other infectious agents in animals suffering from mastitis are harmful to human beings. The presence of antibiotic residues in milk due to mastitis treatment renders the milk quality and milk is unsuitable for human consumption and further processing (Costa *et al.* 1997). However, mastitis remains the most expensive production disease of buffaloes. Thirunavukkarasu *et al*. (1999) reported that total losses due to mastitis in affected buffaloes were ₹ 404.73 per lactation. Sasidhar (2002) had reported from an organized dairy farm in Hyderabad, there were losses of ₹ 326 per infected cow due to mastitis while the overall national economic loss in India due to mastitis was to the tune of ₹ 16,072 million and average decrease in milk yield due to clinical mastitis was estimated to be 50% (Joshi 2006).

Bardhan (2013) had reported that the average loss due to mastitis per animal per month to be ₹ 1708.89 in an optimistic scenario and ₹ 1934.78 in the pessimistic scenario in case of buffaloes. Singh *et al.* (2014) estimated the total losses due to mastitis per lactation in buffalo as ₹ 1272.36 and the annual economic loss due to udder infections in India has been estimated to 908 million US Dollar (Srivastava *et al.* 2015).

Profitable milk production in India has become highly dependent on intensive methods of husbandry, large herd sizes, group handling of dairy animals and better management practices. Diseases are very important production-limiting factors, of which mastitis is very important and excellent example. Losses caused by mastitis have been attributed mainly to decreased milk production from mastitis. Costs associated with mastitis include lower production, discarded milk because of antibiotic therapy, treatments costs and culling or death. The prediction of mastitis is nonlinear complex phenomenon hence its prediction with classical statistical methods is not appropriate (Tasdemir *et al.* 2011). Due to nonlinearity in the milk production data Panchal *et al.* (2016) tried to classify healthy and mastitis Murrah buffaloes by using neural network models using yield and milk quality parameters.

Lactation curve is defined as a graphic representation of milk production and lactation time starting at calving (Papajcsik and Bodero 1988). Lactation curve can be very useful in genetic breeding programs, herd nutritional management, decision taking on the culling of animals and milk production systems. The lactation curve is also important because of its wide characterization of the animal production throughout lactation and allows estimating the peak yield, lactation persistency and days in milk (Ferreira and Bearzoti 2003). Several studies related to lactation curves of dairy animals are available however scant literature are available related to modelling the effect of lactation curve of Murrah buffaloes in disease conditions. Keeping in view the above facts, the objective of the present study was to select best lactation curves model in Murrah buffaloes under healthy and mastitis condition.

MATERIALS AND METHODS

Source of data: For the present study, the milk production, lactation order, season of calving, season of disease occurrence, data related to lactation record of Murrah buffaloes were compiled from livestock production record maintained at Cattle and Buffalo farm of LPM Section, ICAR-Indian Veterinary Research Institute, Izatnagar, Bareilly, Uttar Pradesh over a period of 10 years (2005–2014). In the present study, the data related to lactation record of Murrah buffaloes were divided into two groups, viz. (1) Healthy and (2) Mastitic. We have considered those Murrah buffaloes as mastitic in which clinical mastitis occurs during the 2–8 week of the milk production, i.e. when Murrah buffaloes were in peak production stage. The data consisted of 80068 test day milk yield records of 296 Murrah buffaloes (222-Healthy and 74 Mastitic).

Lactation curve model: To model the lactation behaviour of Murrah Buffaloes in disease and healthy condition seven standard lactation curve models were selected. The Parabolic exponential model, Incomplete Gamma function, Linear Decline Model, Wilmink Model and Mixed Log Model are based on three parameters whereas, Mitscherlich x Exponential model and Ali and Schaeffer Lactation Curve Model are based on four and five parameters respectively.

Parabolic exponential model (Sikka *et al.* 1950)

$$Y_t = a \exp(bt\text{-}ct^2)$$

Incomplete Gamma function (Wood *et al.* 1967)

$$Y_t = at^b \exp(\text{-}ct)$$

Linear Decline Model (Cobby and Le Du 1978)

$$Y_t = a\text{-}bt\text{-}a \exp(\text{-}ct)$$

Wilmink Lactation Curve Model (Wilmink *et al.* 1987)

$$Y_t = a + b \exp(\text{-}kt) + ct$$

Mixed Log Model (Guo and Swalve 1995)

$$Y_t = a + bt^{1/2} + c \ln(t)$$

where, Y_t is the milk production at day ‘t’, ‘a’ is the initial milk production, ‘b’ is the increasing rate from initial milk production to peak production, ‘c’ is the decreasing rate after peak production and the factor ‘k’ was related to the time of peak of lactation and usually assumes a fixed value, derived from preliminary analysis made on average production it is equal to 0.61 (Olori *et al.* 1999); 0.10 (Brotherstone *et al.* 2000) and 0.065 (Silvestre *et al.* 2009).

Mitscherlich × Exponential (Rook *et al.* 1993)

$$Y_t = a(1–b \exp(–ct))–dt$$

where, Y_t is the milk production at day t, ‘a’ is the scale factor or milk yield at the beginning of lactation, ‘b’ is the rate of change from initial production to maximum yield, ‘c’ is the rate of change from maximum yield to the end of lactation and ‘d’ is a parameter related to maximum milk yield

Ali and Schaeffer Lactation Curve Model (Ali and Schaffer 1987)

$$Y_t = a + b\delta + c\delta^2 + d\,\theta_i + e\,\theta_i^2$$

where, Y_t is the milk production at day t, ‘a’ is the scale factor or milk yield at the beginning of lactation, ‘b’ and ‘c’ is the rate of change from initial production to peak yield at increasing rate and decreasing rate,‘d’ and ‘e’ is the rate of change from maximum yield to the end of lactation at increasing rate and decreasing rate, $\delta = t/305$ and $\theta i = \ln(305/t)$

Statistical analysis: Each standard lactation curve models were fitted to DTDMY of Murrah buffaloes in healthy and disease condition separately by using PROC NLIN statement of the statistical package SAS 9.3 version (SAS Institute Inc. 2011. USA). Levenberg-Marquardt algorithm (Marquardt’s iteration) was used to estimates the parameter of standard lactation curves and all the parameters were estimated separately. The best fitted models were selected and evaluated by using different goodness of fit criterion.

Goodness of fit: The goodness of fit explains that “How well some specified model fits the data”. The goodness of fit was accessed by

Mean square prediction error (MSPE)

$$MSPE=\sum_{i=1}^{n}\frac{(o_i-p_i)^2}{n}$$

Adjusted coefficient of determination (R^2_{adj})

$$R^2_{adj} = 1-(1-R^2) \times \frac{n-1}{n-p-1}$$

where, 'R^2' coefficient of determination, 'n' is the number of experimental observation, 'p' is the number of parameters in the model. R^2_{adj} is used to compare the model that involves different number of parameter.

Akaike's Information Criteria (AIC)

$$AIC= n \log_e MSE+2p$$

Bayesian information criterion (BIC)

$$BIC= n \log_e (MSE) + p \log_e (n)$$

where, 'MSE' is the mean square error. The preferred model is the one which has minimum MSPE, AIC, BIC value.

Examination of residuals: Residuals or errors are defined as the difference between the observed and predicted value of the response. For modeling purpose, there are two assumptions, viz. 1. The errors are independently and identically distributed; 2. The errors have constant variance $\varepsilon \sim N(0, \sigma^2_e)$

Test for autocorrelation: The Durbin–Watson (1951) statistic is a test statistic used to test the presence of autocorrelation.

The Durbin–Watson test statistic (d) is;

$$d=\frac{\sum_{i=2}^{n}(e_i-e_{i-1})^2}{\sum_{i=1}^{n}e_i^2}$$

Test for normality: Shapiro and Wilk (1965) test was used to test the normality of the residuals. The Shapiro-Wilk Test statistics (W) is:

$$W = S^2/b$$

$$S^2 = \Sigma\, a_{(p)} \{X_{n+1-p}-X_{(p)}\};\; b = \Sigma\, (X_i-\overline{X})^2$$

where, 'e_i' is the difference between observed and predicted value, if $W_{cal} < W_{tab}$: H_0 (Errors are normally distributed) should be rejected. $X_{(p)}$ is the p^{th} order statistics of the set of Residuals. The value of coefficient $a_{(p)}$ for different value of n and p are mentioned in Shapiro-Wilk's table.

Effects of mastitis on milk production: To study the effect of disease (mastitis), season, time (week) and interactions of mastitis and season, disease and time, season and time and disease, season and time the following general linear model was used

$$Y_{ijk}=\mu+D_i+S_j+(DS)_{ij}+ID(D,S)\ Rand + W_k+(DW)_{ik} + (SW)_{jk} + (DSW)_{ijk}+e_{ijk}$$

where, y_{ijk} = Observed value of the response variable for i^{th} disease at j^{th} season and at k^{th} week; μ = General mean effect; D_i = Effect of i^{th} disease; S_j = Effect of j^{th} season; ID(D,S) Rand = Random term; W_k = Effect of k^{th} week; e_{ijk} = Error term, and the comparison between healthy and Murrah buffaloes was done by t-test.

Table 1. Summary of animals for the study, 305 day milk production, average lactation length and average peak production of Murrah buffaloes

Type of Murrah buffaloes	No. of animals	Total milk production in 305 days (kg)		Lactation length		Peak production (kg)	
		Mean	SE	Mean	SE	Mean	SE
Healthy	222	1835.21	0.12	278.59	2.66	11.53	0.16
Mastitis	74	1372.75	0.10	242.18	14.78	9.63	0.48

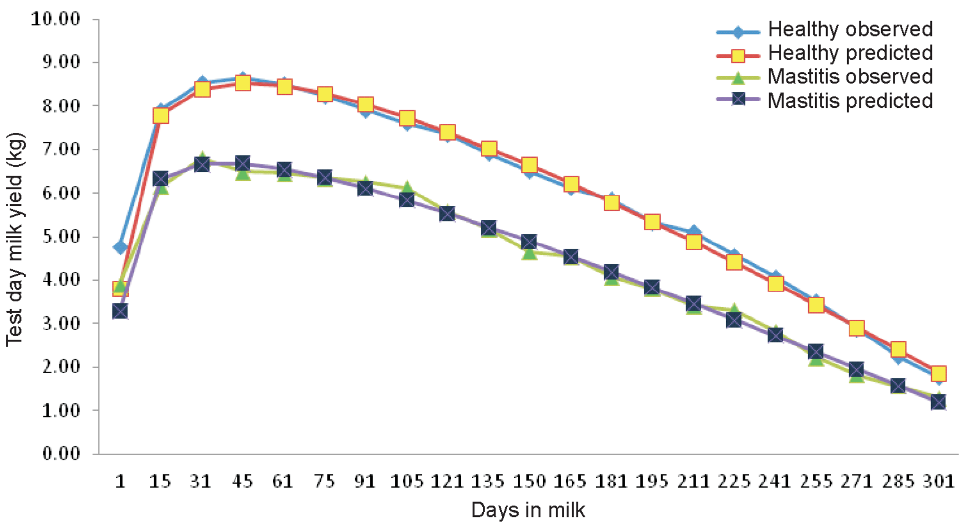

Fig. 1. Comparison of the observed and predicted test day milk yield of healthy and mastitic Murrah buffaloes

RESULTS AND DISCUSSION

The descriptive statistics observed from 222 healthy and 74 mastitic Murrah buffaloes are mentioned (Table 1). The average milk yield production, peak production and average lactation length during 305-days of healthy Murrah buffalo were 1835±0.12 kg, 278±2.66 days and 11.53±0.16 kg respectively. In case of the Murrah buffaloes suffering from clinical mastitis, the average milk yield production, peak production and average lactation length during 305-days were 1372±0.10 kg, 9.63±0.48 kg and 242±14.75 days respectively.

Fitting of lactation curves: Ali and Schaeffer (AE), Cobby and Le Du (CL), Sikka (SK), Mitscherlich x Exponential (ME), Mixed log (ML), Wilmink (WL) and Wood (WD) lactation models were fitted to the lactation records of Murrah buffaloes. The lactation curves were fitted to daily test day milk yield records of both healthy and Mastitic Murrah buffaloes.

Lactation curves of healthy Murrah buffalo: The AS model was best fitted model to the DTDMY (Daily test day milk yield) data of Murrah buffalo (healthy condition) ($R^2_{adj.}$=0.9949; MSPE=0.022; AIC=–1150.8 and BIC=–1132). ME model was 2nd best fitted model ($R^2_{adj.}$=0.9889) followed by WL ($R^2_{adj.}$= 0.9869), ML ($R^2_{adj.}$=0.9707), WD ($R^2_{adj.}$=0.9687) and SK ($R^2_{adj.}$=0.9576).CL model was least fitted to the DTDMY data of healthy Murrah Buffalo ($R^2_{adj.}$=0.9546). CL under predicted and WD over predicted the total milk production. CL over predicted (0.51 kg) and SK under predicted (0.52 kg) the peak production. ML and CL model under predicted the 1st week milk production whereas SK model over predicted the 1st week milk production. All model under predicted the milk production from 41st week to 44th week. The value of parameter estimates of different lactation curves models along with the various measures of goodness of fit are mentioned. (Table 2) and the overall observed and predicted DTDMY of healthy Murrah buffaloes from best fitted lactation curve function was plotted graphically (Figure. 2). Similar to present finding, Catillo *et al.* (2002) observed that AS model was the best fitted model to describe the lactation behaviour of Italian water buffaloes. Catillo *et al.* (2002) also suggested that the possible reason of closeness of the AS Model, may be due to the fact that the AS model account rising and declining segments of the model. Singh *et al.* (2015) reported that the AS model has highest accuracy to predict the milk production of Murrah buffaloes. Sahoo *et al.* (2016) observed that the AS model (R^2 value =99.30%; RMSE = 0.02 kg) was best fitted model for prediction of weekly test day milk yield in Murrah buffaloes. However, Dimauro *et al.* (2005) reported comparatively lower R^2 value (0.967) of AS model to describe the lactation behaviour of Italian water buffaloes.

Testing of autocorrelation and normality of residuals for healthy Murrah buffaloes: Durbin-Watson test was used to test autocorrelation among the residuals of different fitted models. Shapiro-Wilk's test and Kolmogorov Smirnov test were used to test normality present in residuals. After the fitting of different model residuals were estimated for all the models. After applying the Durbin Watson test to the

Table 2. Estimated value of parameter of different lactation curve models of healthy Murrah buffalo along with different measure of goodness of fit

Model	Parameter	Estimate	SE	MSPE	R^2 (adj)	AIC	BIC
AS	a	13.734	0.408	0.022	0.9949	–1150.8	–1132
	b	–11.333	0.723				
	c	–0.658	0.343				
	d	–1.888	0.210				
	e	0.028	0.027				
CL	a	10.178	0.062	0.198	0.9546	–485.1	–473.9
	b	0.026	0.000				
	c	0.133	0.005				
ME	a	10.837	0.048	0.048	0.9889	–916.5	–901.6
	b	0.546	0.009				
	c	0.046	0.002				
	d	0.029	0.000				
ML	a	2.693	0.162	0.127	0.9707	–620.9	–609.7
	b	–1.298	0.017				
	c	3.904	0.071				
SK	a	7.352	0.077	0.184	0.9576	–506.7	–495.5
	b	0.003	0.000				
	c	0.000	0.000				
WL	a	10.521	0.035	0.059	0.9869	–856.3	–845.1
	b	–6.410	0.112				
	c	–0.027	0.000				
WD	a	3.467	0.098	0.136	0.9687	–599.6	–588.4
	b	0.329	0.009				
	c	0.007	0.000				

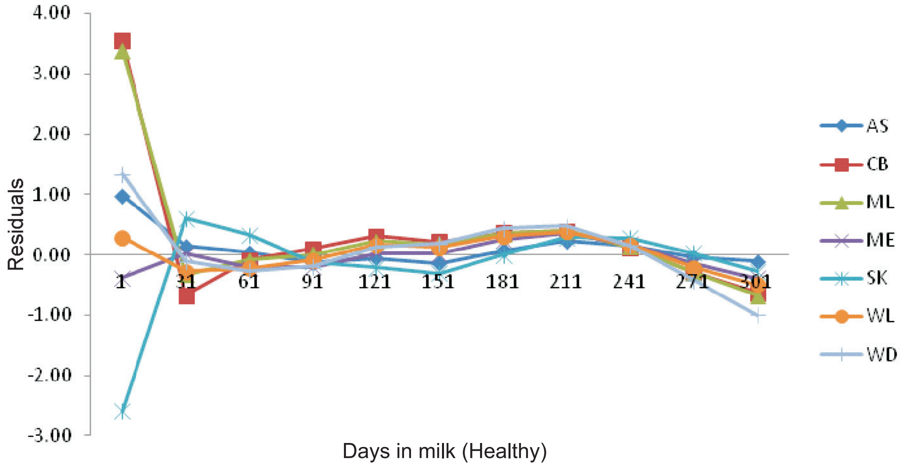

Fig. 2. Plots of residuals against day in milk for healthy Murrah buffalo.

residuals of different models we have observed that DW statistic was close to zero, which indicated that in all the models residuals were positively auto correlated. The value of the autocorrelation was highest (0.57) for AS model and lowest value (0.053) for the SK model. (Table 3) Shapiro-Wilk's test indicated that residuals of all the fitted models were not normally distributed (Fig. 2). The values of the Kolmogorov-Smirnov test also indicated that residuals of most of the fitted models were not normally distributed. However, Kolmogorov-Smirnov test indicated that residuals of ME model were normally distributed.

Lactation curves of Murrah buffaloes suffered from clinical mastitis: The AS model was best fitted model to the DTDMY (Daily test day milk yield) data of Murrah buffalo having clinical mastitis ($R^2_{(adj)}$ =0.9868; MSPE=0.041; AIC= –964.24 and BIC=–945.64). ME model was 2nd best fitted ($R^2_{(adj)}$=0.9818) followed by WL ($R^2_{(adj)}$=0.9817), ML ($R^2_{(adj)}$ =0.9727), SK ($R^2_{(adj)}$=0.9616) and WD ($R^2_{(adj)}$=0.9626). CL

Table 3. Test for the presence of autocorrelation and normality of residual in healthy Murrah buffalo by different models

Healthy Murrah	DW Statistic	Kolmogorov-Smirnov		Shapiro-Wilk	
		Statistic	Sig.	Statistic	Sig.
AS	0.576	0.051	0.006	0.938	<0.001
CL	0.076	0.178	<0.001	0.803	<0.001
ML	0.146	0.131	<0.001	0.793	<0.001
ME	0.167	0.049	0.075	0.984	0.002
SK	0.053	0.142	<0.001	0.771	<0.001
WL	0.136	0.137	<0.001	0.941	<0.001
WD	0.079	0.110	<0.001	0.924	<0.001

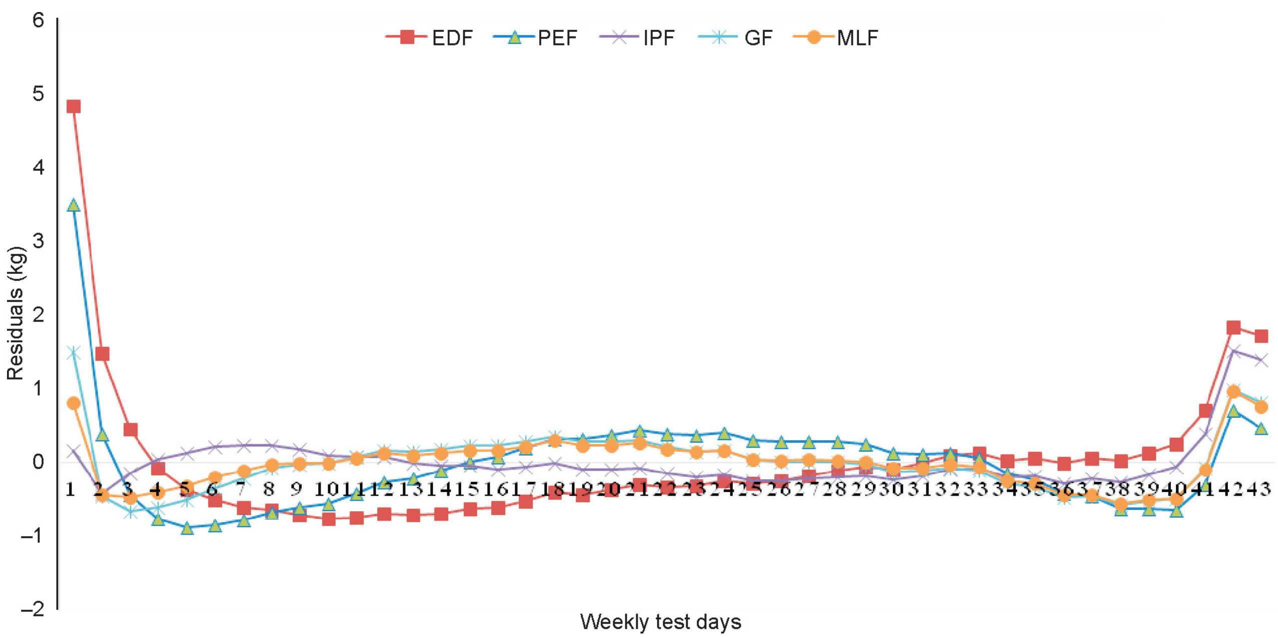

Fig. 3. Plots of residuals against day in milk for Mastitic Murrah buffalo.

Table 4. Estimated value of parameter of different lactation curve models of Mastitic Murrah buffalo along with different measures of goodness of fit

Model	Parameter	Estimate	SE	MSPE	$R^2_{(adj)}$	AIC	BIC
AS	a	10.233	0.553	0.04	0.9868	–964.2	–945.6
	b	–9.240	0.981				
	c	0.092	0.466				
	d	–1.109	0.286				
	e	–0.017	0.037				
CL	a	7.822	0.047	0.128	0.9586	–618.4	–607.3
	b	0.021	0.001				
	c	0.185	0.008				
ME	a	8.274	0.048	0.055	0.9818	–869.9	–855
	b	0.490	0.014				
	c	0.053	0.003				
	d	0.023	0.001				
ML	a	2.770	0.131	0.083	0.9727	–750.2	–739
	b	–1.015	0.013				
	c	2.868	0.057				
SK	a	5.944	0.063	0.119	0.9616	–651.2	–639.8
	b	0.003	0.001				
	c	0.000	0.000				
WL	a	8.155	0.034	0.057	0.9817	–864.6	–853.5
	b	–4.295	0.111				
	c	–0.023	0.000				
WD	a	3.049	0.097	0.114	0.9626	–653.4	–642.3
	b	0.303	0.010				
	c	0.007	0.000				

model was least fitted to the DTDMY data of Murrah buffalo suffered from mastitis ($R^2_{(adj)}$=0.9586) (Table 4). All model equally predict the total milk production but, CL under predicted (3.58 kg) and WD over predicted (5.26 kg) the total milk production. However, CL accurately predicted the peak production and SK model under predicted the peak production. ML and CL model under predicted the 1st week milk production whereas all model over predicted the milk production from 37th week to 44th week. The comparison between lactation curves of observed vs. predicted DTDMY (by best fitted model) in mastitis conditions are represented in form of the graph. (Fig. 1)

Testing of autocorrelation and normality of residuals for Murrah buffaloes suffered from clinical mastitis: After applying the Durbin Watson test to the residuals of different models in case of mastitic buffaloes we have observed that in all the models residuals were positively auto correlated. The value of the autocorrelation was highest (0.73) for AS model and lowest value (0.17) for the ML model (Table 5). Shapiro-Wilk's test indicated that residuals of AS model were normally distributed whereas residuals of all other fitted model were not normally distributed (Fig. 3). The values of the Kolmogorov-Smirnov test also indicated that residuals of most of the fitted models were not normally distributed. However, Kolmogorov-Smirnov test indicated that residuals of AS, ML, ME and WL model

Table 5. Test for the presence of autocorrelation and normality of residual in Mastitic Murrah buffalo by different models.

Mastitis Murrah	DW Statistic	Kolmogorov-Smirnov		Shapiro-Wilk	
		Statistic	Sig.	Statistic	Sig.
AS	0.735	0.035	0.200	0.995	0.390
CL	0.252	0.060	0.009	0.925	<0.001
ML	0.189	0.047	0.099	0.924	<0.001
ME	0.483	0.048	0.088	0.986	0.005
SK	0.247	0.110	<0.001	0.824	<0.001
WL	0.468	0.043	0.200	0.989	0.022
WD	0.247	0.106	<0.001	0.951	<0.001

Table 6. The effect of disease, season and week on the milk production in Murrah buffalo

Source	DF	SS	MSS	F Ratio	Prob>F
Disease	1	1382.04	1382.04	15.6463	0.0001*
Season	2	160.074	80.037	0.9066	0.4061
Disease× Season	2	0.64167	0.32084	0.0036	0.9964
ID[Disease, Season]&Random	148	19670.9	132.911	36.8562	<.0001*
WK	43	14963	347.977	96.4938	<.0001*
Disease× WK	43	202.686	4.71364	1.3071	0.0859
Season× WK	86	397.748	4.62498	1.2825	0.0407*
Disease× Season×WK	86	142.231	1.65385	0.4586	1.0000
Error	10456	37706.58	3.606		
Corrected total	10867	106711.55			

were normally *distributed.*

Effects of mastitis on the milk production of Murrah buffaloes: The results of the general linear model (Table 6) indicated that the effects of the disease was significant on milk production and the reduction in milk production was about 463 kg per lactation. The effects of disease and season-period interaction were significant but the effect of season, disease-season interaction and period-disease-season interaction were non-significant. The loss in milk production due to mastitis with compared to healthy Murrah buffalo was 28.81%, 24.37% and 18.06%, when mastitis occurred in rainy, summer and winter season respectively.

Similar to present finding, Lucey *et al.* (1986) concluded that the mean reduction in recorded 305-day yield of 540 kg when mastitis occurred before the week of peak yield. Houben *et al.* (1993) estimated the production loss due to clinical mastitis was 527 kg of milk (8.1%) in the second lactation. Seegers *et al.* (2004) stated that the Mastitis losses of milk production were at 375 kg for a clinical case. Wilson *et al.* (2004) estimated that production loss from clinical mastitis during the whole lactation was 598 kg. Bar *et al.* (2008) estimate the milk losses due to mastitis was 164 kg and 253 kg in primiparous and multiparous dairy animal respectively.

It has been possible to find a single well fitted model to represent the shape of the lactation curve in Murrah buffalo. In the present study, the value of R^2_{adj} varied between 0.955–0.995 and it showed that lactation curve models predicted the milk yield with great accuracy in healthy as well as mastitis condition of Murrah buffaloes. The lactation curve based on Ali and Schaeffer model was the best fitted model based on the different goodness of criteria for both healthy and the disease condition. The loss of milk production due to mastitis with compared to healthy Murrah buffalo was more when mastitis occurred in rainy season as compared to mastitis occurred in summer and winter season.

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