# Secondary Sex Ratio in Gir Cattle and Assessment of Risk Factors

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### ABSTRACT

Present study was conducted to assess the influence of calf, dam and sire factors on female calves born in Gir cattle. Records pertaining to calf sex (n=1307), spreaded over 10 years (2010-2019) and various calf factors viz. period, month, season and sex of calf; dam factors viz. age, parity and gestation period and sire factors were used for the study. Female calf born in Gir cattle was 49.96%. Calf sex was not affected by period of birth (p=0.26), but significantly higher proportion of female birth than male (60.22% vs. 39.78%) occurred in 2013 (p<0.05). Odds of female calf being born was 1.75 times higher in 2013 as compared to 2010 (OR=1.75, p=0.04). Month (p=0.31) and season (p=0.41) of birth did not affect calf sex. There was significantly higher chances of female birth during February (OR=1.66, p=0.05) and March (OR=1.67, p=0.04) as compared to January. Calf sex was not affected by age (p=0.44) and parity (p=0.85) of dam. With increase in gestation length the birth frequency of female calf decreased significantly (p<0.001). Compared to gestation period of >290 days, the odds of female calf born increased by 2.25 (OR=2.25, p=0.001) and 2.27 (OR=2.27, p=0.001) times with gestation period of <275 and 275-290 days, respectively. Sire wise data showed that proportion of female birth ranged from 22 to 90%, but female calf born from different sires did not differ statistically (p=0.13). Chances of female birth was significantly higher (OR=3.79-31.5, p<0.05) in 12 sires. The results indicated that gestational length had significant effect on secondary calf sex ratio in Gir cattle and dam with shorter gestation period delivered more number of female calves.

#### HIGHLIGHTS

- Gestational length had significant effect on proportion of female calf born in Gir cattle.
- There was significantly higher probabilities of female birth during February and March as compared to January.

Keywords: Secondary sex ratio, Gir cattle, Female calf, Risk factors

Gir cattle is one of the important milch breeds of India spotted in South Saurashtra Agro-climatic zone of Gujarat having 2810 and 2573 kg lactation and standard lactation milk yield, respectively (Patbandha et al., 2020). This region housed different types of domestic animals as well as wild animals. This region has 0.98 and 1.70 million breedable cattle and buffalo population, respectively (20<sup>th</sup> livestock census, 2020). Female birth is more essential for dairy herd as compared to the male counterpart as the former is kept for herd replacement (Yilmaz et al., 2010; Sawa et al., 2014). Furthermore, financial attraction of dairy farmers is more when the replacement rates are better (Berry and Cromie, 2007). In current scenario,

determination of secondary sex ratio i.e. either male to female or female to male live births of calves is having keen interest of many researchers. Various factors have been reported by the earlier workers with variation in expected 50:50 sex ratio in dairy animals (Kaygisiz and Vanli, 2008; Yilmaz et al., 2010; Berry et al., 2011; Sawa et al., 2014; Khan et al., 2013). Sex ratio can be manipulated by enhancing the selection and genetic improvement programmes (Seidel, 2003). As per the sex ratio allocation

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theory of Trivers and Willard (1973), mothers in better physiological conditions would give birth to male calves. Reproductive success varies more among one sex than the other. Factors responsible for difference in the secondary sex ratio might be due to the classification of breed, age of dam and sire, season, parity, year, herd size, body condition score and the levels of sex hormones (Singh *et al.*, 2004; Roche *et al.*, 2006; Yilmaz *et al.*, 2010). There is paucity of reports on secondary sex ratio in indigenous breed like Gir. Hence, this study was planned to identify the factors and determine the association of relationship between the odds of female births in zebu cattle under tropical to subtropical climate of Gujarat.

## MATERIALS AND METHODS

#### Study area and data collection

Data pertaining to the present experiment were collected from records of Gir cattle (Bos indicus) maintained at Cattle Breeding Farm, Junagadh (Gujarat), India. The farm is located at 70.5° west longitude, 21.4° north latitude and an altitude of 60 meters above mean sea level. Tropical to sub-tropical climate prevails in the study area. Animals were maintained under loose housing system with pucca floor, fed seasonal green fodder (maize, jowar and berseem) (a)10 kg/ cow, ad-lib dry fodder and measured amount of concentrate mixture to meet the nutritional requirement as per Indian Council of Agricultural Research (ICAR, 2013) feeding standards. Health monitoring protocols were followed as per routine farm standards. Artificial insemination programme was followed using semen from proven bulls evaluated under All India Co-ordinate Research Project (AICRP). Information of calves (date of birth, sex and weight), dam (age, parity, gestation period) and sire (sire number/ name) were collected from the records of 10 years period (from 2010 to 2019). In the study, records of normal full term birth (n=1307) comprised of insemination from 28 sires were included.

#### Classification of data and statistical analysis

Factors affecting secondary sex ratio (female to male calf) in Gir cattle were first grouped into different classes using certain criteria. Total 10 year study period was first divided into 10 classes based on date of birth ( $P_1$ :2010,

data were categorized into 12 classes (January through December) and seasons into 3 classes (Winter: November-February, Summer: March-June and Rainy: July-October). Considering previous calf sex, data were classified into 2 groups such as female and male calves. Data were further classified based on dams' age into 5 classes ( $\leq 4$  years, 5-6 years, 7-8 years, 9-10 years and >10 years); dams parity into 8 classes  $(1^{st}, 2^{nd}, 3^{rd}, 4^{th}, 5^{th}, 6^{th}, 7^{th} and \ge 8^{th})$ ; dams? gestation period into 3 classes (<275 days, 275-290 days and>290 days) and service sire into 28 classes (1-28). Effect of period of birth, month of birth, season of birth, previous calf sex, dams' age, dams' parity, dams' gestation period and service sire on proportion of female calves was analysed by chi-square test. Considering the probability of 0.5 (or 50%) a calf being born either female or male, the observed and expected frequencies were also compared by chi-square test. Further the association of risk factors with odds of female calf being born was estimated by binary logistic regression where the birth of a female calf was coded as '1' and birth of male calf as '0'. Results were considered as significant when  $p' \leq 0.05$  and a 'trend' if  $p' \leq 0.1$  but >0.05. All the statistical analyses were carried out using SPSS software version 16.

## **RESULTS AND DISCUSSION**

#### Secondary sex ratio

The overall female calf born during 10 years of study period (from 2010 to 2019) in Gir cattle was 49.96%, which did not deviate from the hypothesized probability of calf sex as 0.5 or 50% (Table 1). The secondary sex ratio in Gir cattle is in agreement with Hossein-Zadeh (2012) who found the overall ratio of females to males was 50.4: 49.6 in Holstein cattle. Delesa *et al.* (2014) studied the calves' sex ratio in naturally and artificially bred cattle and stated that AI did not alter the female to male ratio in dairy cattle. Contrary to the present study, Sawa *et al.* (2014) and Del Rio *et al.* (2007) observed female to male ratio as 47.21: 52.79 and 53.3: 46.7, respectively in HF dairy cows. The differed expected ratio was also observed by Goshu and Singh (2013) in HF cattle and Banik and Naskar (2006) in Sahiwal cattle. Several factors *viz.* vaginal and uterine pH,

maturity of the oocyte, timing of AI and climatic factors at AI also affect secondary sex ratio (Pursley *et al.*, 1998).

#### Effect of calf factors on female calf born

### Period of calving

Period of birth did not affect calf sex ( $\chi^2 = 11.18$ , df=9, p=0.26), but significantly higher proportion of female birth (60.22% vs. 39.78%) occurred during 2013 as compared to male calf ( $\chi^2$  = 3.88, df=1, p<0.05; Table 1). Proportion of female calves born increased from 2010 to 2013, from 2014 to 2016 and from 2017 to 2019. Similarly, Sawa et al. (2014) found number of female calves increased every year during the analyzed period (45.53% in 2000-2001 to 52.12% in 2012). Roche et al. (2006) and Kaygisiz and Vanli (2008) found non-significant effect of calving year on secondary sex ratio. A skewed sex ratio from year to year and its dependent on year also observed by Yilmaz et al. (2010). Contrary to Gir cattle, earlier researchers found a significant relationship between the year of calving and sex ratio of calves (Lathwal and Kumar 1994; Berry and Cromie, 2007 and Goshu and Singh, 2013). Roche et al. (2006) and Kaygisiz and Vanli (2008) found nonsignificant effect of calving year on secondary sex ratio. The variation in female calves born in different season might be due to the large differences in climate within season.

 Table 1: Period wise proportion of female calves (%) born in
 Gir cattle (n=1307)

Total	No. of females	Female (%)
123	57	46.34
107	54	50.47
117	63	53.85
93	56	60.22*
156	69	44.23
132	62	46.97
178	99	55.62
155	71	45.81
110	54	49.09
136	68	50.00
1307	653	49.96
	<b>Total</b> 123 107 117 93 156 132 178 155 110 136 <b>1307</b>	No. of females           123         57           107         54           117         63           93         56           156         69           132         62           178         99           155         71           110         54           130         68

Considering calving year 2010 as reference, the odds of female calf being born was 75% higher in 2013 (OR=1.75, p=0.04; Table 7). Hossein-Zadeh (2012) studied the period effect on odds of male births in Iranian Holsteins and observed higher male to female ratio (52.5:47.5) during 1996-1999 which decreased during 2004-2007 *i.e.* 48.5: 51.5. The odds of male birth was 1.18 in 2004-2007 as compared to 1996-1999 and 1.01 in 2004-2007 as compared to 2000-2003, which reflected decrease in male to female sex ratio in recent years (Hossein-Zadeh, 2012). Although there was significant effect of year of birth on probability of female calf but there is no clear and conclusive reason (Berry and Cromie, 2007).

### Month and season of calving

Month ( $\chi^2 = 12.73$ , df=11, p=0.31) and season ( $\chi^2 = 1.76$ , df=2, p=0.41) of birth did not affect the calf sex in Gir cattle (Table 2 & 3). Calf sex also did not differ from the expected probability of 50% in any month or season. Proportion of female calves born was higher in rainy season (52.39%) followed by summer (49.77%) and winter season (47.91%), respectively. Singh *et al.* (2004) stated that calving season has slight effect on sex ratio of the calves born. Uniform female calves born in individual calving season was observed by Sawa *et al.* (2014), whereas, other researchers did not observe any relationship between season and sex ratio of calve (Mukherjee *et al.*, 2000; Kaygisiz and Vanli, 2008; Goshu and Singh, 2013).

 Table 2: Month wise proportion of female calves (%) born in
 Gir cattle (n=1307)

Month of birth	Total	No. of females	Female (%)		
Jan	133	58	43.61		
Feb	103	58	56.31		
Mar	117	66	56.41		
Apr	113	55	48.67		
May	113	56	49.56		
Jun	91	39	42.86		
Jul	78	34	43.59		
Aug	110	61	55.45		
Sep	109	59	54.13		
Oct	121	65	53.72		
Nov	87	41	47.13		
Dec	132	61	46.21		
$\chi^2 = 12.73$ , df = 11, p = 0.311					



**Table 3:** Season and previous calf wise proportion of femalecalves (%) born in Gir cattle

Parameters	Total	No. of females	Female (%)	
Season of birth (	(n=1307)			
Winter	455	218	47.91	
Summer	434	216	49.77	
Rainy	418	219	52.39	
$\chi 2 = 1.76$ , df =2,	p = 0.415			
Previous calf sex	x(n=860)			
Female calf	435	215	49.43	
Male calf	425	207	48.71	
$\chi 2 = 0.04$ , df =1,	p = 0.833			

Although proportion of male calves born increases with increasing air temperature and humidity around the time of conception (Roche *et al.*, 2006), such association was not observed in Gir cattle.

Logistic regression analysis reflected that there was significantly higher chances of female birth during February (OR=1.66, p=0.05) and March (OR=1.67, p=0.04; Table 7) as compared to January. Moreover, a trend of higher female calf being born in the month of August (OR=1.61, p=0.06) was observed. In Holstein cows, higher proportion of male calves born in spring (OR=1.02; P< 0.0001), and the lowest male calves born in summer or fall calvings as reported by Hossein-Zadeh (2012). Further, Berry and Cromie (2007) reported higher proportion of male birth during warm months (February-June) as compared to colder months (December-January).

### **Previous calf sex**

Sex of previous calf did not affect secondary sex ratio in Gir cattle ( $\chi^2 = 0.04$ , df=1, *p*=0.833; Table 3). Further, calf sex also did not deviate from the probability of a female calf being born with a chance of 50%. Although previous studies reported that the sex of the previous calf to the same cow had significant (p<0.05) effect on the sex of the subsequent calf (Roche *et al.*, 2006; Berry and Cromie, 2007; Hossein-Zadeh, 2012), such effect was not observed in Gir cows. The repeatability of calf sex of a particular dam is primarily influenced by the permanent environmental factor of the dam (Hossein-Zadeh, 2012).

#### Effect of dam factors on female calf born

### Age and parity of dam

Calf sex was not affected by age ( $\chi^2 = 3.73$ , df=4, p=0.44; Table 4) and parity ( $\chi^2 = 3.38$ , df=7, p=0.85; Table 5) of the dam. The sex ratio also did not deviate from the probability of 50% in any age or parity group. Highest proportion of female calves born in 5th parity (56.18%) followed by 1st (51.24%) and 2<sup>nd</sup> (49.85%) parity cows. The proportion of female calves born at 1st calving was 48% followed by 44% and 51% in second and fifth-seventh calving, respectively (Goshu and Singh, 2013). The present study favored that the proportion of female calves was higher to primiparous (51.24%) compared to multiparous (48.65%) cows. Sawa et al. (2014) and Singh et al. (2004) observed similar results. The probability of male calves born was significantly higher in aged cows as compared to younger cows (Berry and Cromie, 2007). Earlier researchers did not find any significant effect of number of calving on the sex ratio of the calves born (Kaygisiz et al., 2003, Roche et al., 2006) which is supported by present study.

 Table 4: Age wise proportion of female calves (%) born in Gir cattle (n=1307)

Age of dam	Total	No. of females	Female (%)
≤4 yr	337	171	50.74
5-6 yr	426	211	49.53
7-8 yr	266	139	52.26
9-10 yr	142	61	42.96
>10 yr	136	71	52.21
$\chi 2 = 3.73$ , df =	=4, <i>p</i> = 0.443		

 Table 5: Parity wise proportion of female calves (%) born in Gir cattle (n=1307)

Parity of dam	Total	No. of females	Female (%)
1	404	207	51.24
2	331	165	49.85
3	216	103	47.69
4	137	67	48.91
5	89	50	56.18
6	57	28	49.12
7	30	12	40.00
≥8	43	21	48.84
$\gamma 2 = 3.38$ , df =7	p = 0.848		

#### Gestation period of dam

With increase in gestation length the frequency of female birth decreased significantly ( $\chi^2 = 21.13$ , df=2, p<0.001; Table 6). Frequency of female birth was significantly lower (32.69% vs. 67.31%) as compared to male counter parts in Gir cows with gestation length beyond 290 days ( $\chi^2 = 18.69$ , df=1, p<0.001). The chances of female calf birth increased by 2.25 (OR=2.25, p=0.001; Table 7) and 2.27 (OR=2.27, p=0.001; Table 7) times in case dams having gestation period of <275 days and 275-290 days, respectively as compared to those with gestation period of >290 days.

 Table 6: Gestation period wise proportion of female calves (%)

 born in Gir cattle

Gestation period	Total	No. of females	Female (%)	
<275 days	139	73	52.52	
275-290 days	1012	529	52.27	
>290 days	156	51	32.69*	
$\chi^2 = 21.13$ , df =2, $p < 0.001$ ;* = Significant at 0.1% ( $p < 0.001$ ).				

 Table 7: Associated risk factors (period of birth, month of birth and gestation period) for female calf in Gir cattle

Parameters	Estimates	SEM	OR	95% CI	P value
Period of birth					
2013 vs. 2010	0.561	0.279	1.752	1.015-3.025	0.044
Month of birth					
February vs. January	0.511	0.265	1.667	0.992-2.800	0.054
March vs. January	0.515	0.256	1.673	1.014-2.762	0.044
August vs. January	0.476	0.260	1.610	0.968-2.677	0.067
Gestation period					
<275 days vs. >290 days	0.823	0.241	2.277	1.420-3.651	0.001
275-290 days vs. >290 days	0.813	0.182	2.255	1.579-3.221	< 0.001

There was a scanty report regarding effect of gestation period on calf sex but some information available on calf sex effect on gestation period. Gestation period is affected by several factors *viz*. calf sex, single or twin pregnancy, age of dam, calving season and genetic factors (Silva *et*  *al.*, 1992). Higher birth weight in male calves might be due to the higher androgen hormone intensity of male fetus serum (Manzi *et al.*, 2012). A significant amount of production of testosterone hormone starts from 45 days of gestation which reaches to peak up to 70 days. During this fetal life, presence of androgen receptors within muscle cells enhances the muscle tissue growth results into weight differences between the sexes (Holland and Odde, 1992). Additional fetal growth in male calves prolongs the gestation period (Rezende *et al.*, 2020). A positive relationship exists between the gestational length and birth weight of calf. Gestation length prolonged by 1.1 (Silva *et al.*, 1992) to 2.0 days (Hayr *et al.*, 2015) in case of male calves.

#### Effect of sire on female calf born

#### Service sire

Sire wise data showed, proportion of female birth ranged from 22.22 to 90.00% (Fig. 1 & Table 8). However, the sex of calf born from different sires did not differ statistically ( $\chi^2 = 35.24$ , df=27, p=0.13). Moreover, out of 28 sires, the proportion of female birth significantly (p<0.05) deviated from probability of 50% in 3 sires and the frequency of female calf was 22.22% ( $\chi^2 = 5.56$ , df=1, p<0.05), 70.37% ( $\chi^2 = 4.48$ , df=1, p<0.05) and 90.00% ( $\chi^2 = 6.40$ , df=1, p<0.05). Variations of secondary sex ratio in HF dairy sires have been documented in earlier studies (Berry *et al.*, 2011; Hossein-Zadeh, 2012).



Fig. 1: Histogram of female Gir calves (%) and service sires (%)

The variation might be due to the relatively small number of sires represented in most of the studies. The ejaculation



Percent of female calves	Estimates ± SEM	OR	95% CI	P value
Reference sire- 22.22% female calves	-1.253±0.567	0.286	_	0.027
44.04	1.013±0.599	2.754	0.852-8.907	0.091
45.16	1.059±0.622	2.882	0.852-9.749	0.089
45.45	$1.070 \pm 0.628$	2.917	0.851-9.993	0.088
45.78	$1.084 \pm 0.608$	2.956	0.897-9.736	0.075
46.15	1.099±0.632	3.000	0.870-10.343	0.082
47.06	1.135±0.663	3.111	0.848-11.408	0.087
47.62	1.157±0.608	3.182	0.967-10.468	0.057
50.00	1.253±0.737	3.500	0.825-14.848	0.089
50.00	1.253±0.737	3.500	0.825-14.848	0.089
50.00	1.253±0.642	3.500	0.994-12.321	0.051
52.00	1.333±0.634	3.792	1.095-13.129	0.035
52.22	1.342±0.605	3.826	1.169-12.521	0.027
52.38	1.348±0.646	3.850	1.086-13.647	0.037
52.63	1.358±0.653	3.889	1.080-13.997	0.038
52.94	1.371±0.633	3.938	1.140-13.604	0.030
56.02	$1.495 \pm 0.588$	4.459	1.408-14.120	0.011
56.67	1.521±0.676	4.577	1.216-17.223	0.024
57.78	$1.566 \pm 0.642$	4.789	1.360-16.865	0.015
59.09	1.620±0.714	5.056	1.248-20.480	0.023
70.00	2.100±0.748	8.167	1.885-35.380	0.005
70.37	2.118±0.706	8.313	2.082-33.194	0.003
90.00	3.450±1.197	31.500	3.017-328.930	0.004

Table 8: Associated sire factor for female calf in Gir cattle

of an unequal ratio of X and Y-bearing sperm, differences in capacitation and fertilization rate, differences among sexes in the rate of intrauterine attrition at different stages of development from zygote to the fetus might be the possible reasons of variable sex ratio (Berry *et al.*, 2011). Logistic regression analysis of sire data showed wide variation of female calf being born from different sires considering the lower frequency of female calf born from the sire (22.22% female calf) as reference. The chances of female calf birth increased significantly (OR=3.79-31.5, p<0.05) in 12 sires and there was a trend of higher odds of female birth in 10 sires (OR=2.7-3.5, p<0.1).

## CONCLUSION

The proportion of female Gir calves born was not affected by period, month, season of birth and age or parity of dam. In Gir cows, there was no significant effect of previous calf sex on secondary sex ratio. Gestational length had significant effect on proportion of female calf born in Gir cattle. The frequency of female birth was found significantly lower as compared to male counter parts in Gir cows with gestation length beyond 290 days. Sire wise data suggested the proportion of female birth ranged from 22.22 to 90.00%.

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