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Breed and heterosis effects on reproduction and production traits of Girolando cows

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ABSTRACT - The objective of this study was to evaluate the breed and heterosis effects on reproductive traits, test-day milk yield, and 305-day milk yield in different lactations of crossbred Girolando cows. Data consisted of test-day milk yield records of first (118,831 records), second (63,227), and third lactation (44,512) and their relative productive (test-day milk yield, 305-day milk yield, and lactation length) and reproductive (age at first calving, calving interval, days open, and dry period) records of 35,582 Girolando cows from Brazil, collected from 1998 to 2014. The heterosis effect of the evaluated traits in Girolando cattle was estimated by MIXED procedure in SAS. Girolando cows showed a negative (favorable) and significant heterosis effect for reproductive traits. The dry periods between the first and second calving and between the second and third calving showed the greatest gains in heterosis (21.93 and 10.41%, respectively). All the evaluated productive traits showed a significant and similar heterosis effect between the three lactations. The use of crossbreeding strategies between the Holstein and Gyr breeds, instead of using the pure breed, is indeed a good alternative to increase the economic efficiency of the dairy activity in the different production systems in tropical environments.

Keywords: age at first calving, crossbreeding, dairy cattle, dairy milk yield, dry period

1. Introduction

The reproductive efficiency of a herd is one of the main components of the economic and productive performance of a dairy farm. One of the available strategies to make the production system more efficient is the use of crossbreeding, through which it is possible to introduce desirable genes in the target populations, changing the genotypic frequencies in the populations (Su et al., 2009), promoting improvements in the productive and reproductive efficiency of dairy herds (Clasen et al., 2018). The reason is that crossbreeding allows the exploration of complementarity and heterosis effects of the breeds (Canaza-Cayo et al., 2014).

It was through crossbreeding techniques that the formation of the Girolando breed began in 1989, aiming to produce milk in a sustainable way for the tropical and subtropical regions of Brazil (Silva et al., 2011). The benefits of this strategy on productivity and fertility were easily perceived by breeders, so the use of crossbreeding between animals of the Holstein and Gyr breeds spread quickly across the country (Silva et al., 2014), allowing the improvement of the performance of Brazilian herds. Currently, approximately 80% of the milk produced in Brazil comes from cows that have Holstein or Gyr genes in their genetic composition (Silva et al., 2015). In addition, Silva et al. (2020) reported an increase of 57% in milk production of Girolando cows between 2000 and 2018.

Therefore, it is important to know the heterosis effect resulting from crossbreeding, as there is a lack of studies on the Girolando breed, especially on reproductive traits. Some studies indicate that the heterosis effect can present an average gain of 11% for age at first calving (AFC) and of 9% for calving interval (CI) (Rege, 1998), but the effects are still vague and need further investigation.

Thus, the objective of the study was to evaluate the breed and heterosis effects on AFC, CI, days open, and dry period traits together with test-day milk yield (TDMY) and 305-day milk yield (MY305) of different lactations of Girolando cows.

2. Material and Methods

2.1. Data

Data consisted in reproductive and productive records collected by the Associação dos Criadores de Gado Holandês de Minas Gerais, Associação Brasileira dos Criadores de Gir Leiteiro, and Associação Brasileira dos Criadores de Girolando for the following breeds: Holstein (H), Gyr (G), and six genetic groups of Holstein × Gyr (1/4H, 3/4G (1/4H); 3/8H, 5/8G (3/8H); 1/2H, 1/2G (1/2H); 5/8H, 3/8G (5/8H); 3/4H, 1/4G (3/4H); 7/8H, 1/8G (7/8H)), officially called Girolando in Brazil, collected between 1998 and 2014 in 1,221 herds located in the state of Minas Gerais, Brazil.

When editing the data, only cows with records until the third lactation and that were milked twice a day were kept. A minimum of four and maximum of 10 test days, obtained from 5 to 305 days in milk, were considered for estimating lactation. Abnormal yield values or outliers were checked by graphical techniques such as normal probability plots and boxplots, as well as by median, mean, mode, skewness, and kurtosis. Cows with TDMY and MY305 or lactation period different from the mean (standard deviation higher than ±3.0) were not considered for the study. Then the MY305 records were removed if milk yield were out of the range from 900.00 to 13800.00 kg.

Regarding test-day milk yield (TDMY1, TDMY2, and TDMY3), productions lower than 3 kg/day and greater than 45 kg/day were discarded for the three lactations.

Following these criteria, the final data file consisted of 118,831 TDMY records of 17,004 first-lactation cows; 63,227 TDMY records of 9,570 second-lactation cows; and 44,512 TDMY records of 9,008 third-lactation cows.

The reproductive traits AFC, interval between first and second calving (CI12), interval between second and third calving (CI23), days open between the first and second calving (DO12), days open between the second and third calving (DO23), dry period between the first and second calving (DP12), and dry period between the second and third calving (DP23) were evaluated. Reproductive traits were obtained in the following ways:

Age at fist calving was calculated as the number of days from birth to the first calving of the cows, in which records of AFC lower than 545 and greater than 1280 days were excluded, as cows could have a previous lactation not properly registered (Eastham et al., 2018).

The CI12 and CI23 were calculated by the difference in days between the second and first calving and between the third and second calving, respectively. Cows with values less than 300 or more than 800 days were discarded because some type of failure could have occurred at the time of successive calving records.

The DO12 and DO23 were calculated through the number of days obtained individually for CI12 and CI23 subtracted from the average gestation period of cows (e.g., 284 days); cows with periods shorter than 50 or longer than 250 days were discarded.

Finally, DP12 and DP23 were calculated by subtracting the second calving date from the first lactation closure date and subtracting the third calving date from the second lactation closure date, respectively. Cows with periods shorter than 30 and longer than 300 days were excluded from the evaluation.

The productive traits MY305 in the first (MY3051), second (MY3052), and third (MY3053) lactation; test-day milk yield in the first (TDMY1), second (TDMY2), and third (TDMY3) lactation; and lactation length in the first (LL1), second (LL2), and third (LL3) lactation were evaluated. The descriptive analysis of the edited data, number of cows, and their genetic compositions are shown in Table 1. The population structure analysis was carried out through the program CFC (Sargolzaei et al., 2006), with 19 generations of ancestors being considered, with a description of the range of each ancestor up to the sixth generation. The number of sires and dams used in this study were 2.444 and 20.204, respectively.

2.2. Breed and heterosis effects

The databases made available by each of the three Association of Breeders presented the breed composition of each animal (with or without production or reproductive records), as well as their parents. According to the mating used by the breeders and previously registered by the technicians of these Associations, the genetic proportions are expected for each type of crossing present in the evaluated population (Table 1). The three dairy breeds (Hostein, Gyr, and Girolando) were considered with enough records to estimate breed and heterosis effects for all traits. The proportion of genes was calculated for each cow using the following equation (Dickerson, 1973; Penasa et al., 2010a, 2010b):

$$\alpha_i^p = \frac{\alpha_i^s + \alpha_i^d}{2},$$

in which α_i^p is the proportion of genes from breed i in the progeny, α_i^s is the proportion of breed i in the sire, and α_i^d is the proportion of breed i in the dam. Each proportion of the Holstein genes (1/4H, 3/8H, 1/2H, 5/8H, 3/4H, and 7/8H) plus the proportion of the Gyr genes was equal to 1.

Coefficients of specific heterosis were calculated between pairs of the dairy breeds, using the following equation (Dickerson, 1973):

$$\delta_{ij}^p = \alpha_i^s\alpha_j^d + \alpha_j^s\alpha_i^d,$$

in which δ_{ij}^p is the coefficient of expected heterosis between fractions of breeds i and j in the progeny; α_i^s and α_j^s are proportions of breeds i and j in the sire, respectively; α_i^d and α_j^d are proportions of breeds i and j in the dam. The specific heterosis effects were used in the six genetic groups of Girolando, as the distribution of cows across the classes of coefficients of expected heterosis was adequate for this purpose (Penasa et al., 2010b). The classes of coefficients of heterosis were defined as: 1 = 0%, 2 = 25%, 3 = 37.5%, 4 = 50%, 5 = 62.5%, 6 = 75%, 7 = 87.5%, and 8 = 100%.

2.3. Statistical analyses

The mean values of all traits of each genetic group were compared using analysis of variance (ANOVA). Estimation of Type III mean squares, F statistics, and least squares means were computed using PROC GLM of SAS.

The phenotypic values of all traits were analyzed using the MIXED procedure of SAS (Statistical Analysis System, version 9.1). Effects of breed and heterosis were estimated using the following mixed linear models:

Model 1:

$$y_{ijklmn} = \mu + S_i + YT_j + H_k + C_l + D_m + \sum_{q=1}^{2} \varphi_q a^q + \beta f + \lambda h + e_{ijklmn},$$

in which y_{ijklmn} is the n-th observation of TDMY of trait (TDMY1, TDMY2, or TDMY3) measured at the l-th cow, in m-th class of days in milk, in the i-th season of TDMY, in the j-th year of TDMY, in the k-th herd; μ is the constant; S_i is the fixed effect of the i-th season of TDMY; YT_j is the fixed effect of the j-th year of TDMY; H_k is the fixed effect of the k-th herd; C_l is the random effect of the l-th cow; D_m is the fixed effect of the m-th class of days in milk (equivalent to the lactation stage); φ_q are regression coefficients associated with the linear (q = 1) and quadratic (q = 2) effects of cow age; β is the regression coefficient associated with the linear effect of proportion of Holstein f; λ is the regression coefficient associated with the linear heterosis effect (h) between Holstein and Gyr; and e_{ijklmn} is the residual random error associated with observation y_{ijklmn}, assuming NID $(0, \sigma_e^2)$.

Table 1 - Number of cows and observations for each genetic group for traits

Genetic group	Percentage of Holstein gene	Trait															
		AFC (day)	CI12 (day)	CI23 (day)	DO12 (day)	DO23 (day)	DP12 (day)	DP23 (day)	MY3051 (kg)	MY3052 (kg)	MY3053 (kg)	TDMY1 (kg)	TDMY2 (kg)	TDMY3 (kg)	LL1 (day)	LL2 (day)	LL3 (day)
		Number of cows in each trait															
H	100	437	223	197	181	159	196	181	603	422	433	620	425	433	620	425	433
7/8H	87.50	1151	552	356	405	297	488	335	1359	778	744	1364	778	744	1364	778	744
3/4H	75.00	4483	2116	1252	1590	962	1879	1147	5278	2913	2599	5291	2915	2604	5291	2915	2604
5/8H	62.50	3663	1825	1077	1396	850	1613	951	4539	2645	2570	4561	2648	2573	4561	2648	2573
1/2H	50.00	3661	1658	995	1253	755	1458	897	4191	2211	2009	4219	2216	2017	4219	2216	2017
3/8H	37.50	320	147	104	120	82	132	94	420	227	256	423	228	256	423	228	256
1/4H	25.00	311	113	78	73	66	92	76	412	210	221	420	210	221	420	210	221
G	0	133	85	72	55	51	73	61	202	164	176	219	164	176	219	164	176
		Number of observations in each trait															
H	100	3180	4480	4115	3669	3410	4017	3806	4164	2970	2513	4264	3009	2639	4264	3009	2639
7/8H	87.50	8474	10927	7817	8207	6517	9763	7370	9710	5537	3999	9733	5564	4177	9733	5564	4177
3/4H	75.00	33464	40036	26458	30270	20343	35804	24433	38161	19888	12957	38241	20012	13524	38241	20012	13524
5/8H	62.50	25918	32075	21184	24699	16914	28645	18999	31360	16740	11715	31497	16871	12419	31497	16871	12419
1/2H	50.00	25330	30093	20875	23007	15920	26860	18982	28280	14270	10024	28458	14340	10315	28458	14340	10315
3/8H	37.50	2316	2623	2037	2146	1601	2377	1839	2945	1474	1413	2959	1476	1454	2959	1476	1454
1/4H	25.00	2139	1978	1437	1299	1203	1619	1394	2751	1319	1013	2803	1330	1084	2803	1330	1084
G	0	1006	1810	1574	1138	1116	1577	1377	1460	1029	878	1570	1129	1114	1570	1129	1114

H - Holstein breed; G - Gyr breed; 1/4H, 1/2H, 3/8H, 3/4H, 5/8H, 7/8H - crossbred (Holstein × Gyr) cows that are officially called Girolando in Brazil; AFC - age at first calving; CI12 - interval between the first and the second calving; CI23 - interval between the second and the third calving; DO12 - days open between the first and second calving; DO23 - days open between the second and third calving; DP12 - dry period between first and second calving; DP23 - dry period between second and third calving; MY3051 - 305-day milk yield in the first lactation; MY3052 - 305-day milk yield in the second lactation; MY3053 - 305-day milk yield in the third lactation; TDMY1 - test-day milk yield in the first lactation; TDMY2 - test-day milk yield in the second lactation; TDMY3 - test-day milk yield in the third lactation; LL1 - lactation length in the first lactation; LL2 - lactation length in the second lactation; LL3 - lactation length in the third lactation.

Model 2:

$$y_{ijklm} = \mu + S_i + CY_j + H_k + C_l + \sum_{q=1}^{2} \varphi_q a^q + \beta f + \lambda h + e_{ijklm},$$

in which y_{ijklm} is the m-th observation of trait (MY3051, MY3052, MY3053, CI12, CI23, DO12, DO23, DP12, or DP23) measured at the i-th calving season, in the j-th calving year, in the k-th herd, in the l-th cow; μ is a constant; S_i is the fixed effect of the i-th calving season; CY_j is the fixed effect of the j-th calving year; H_k is the fixed effect of the k-th herd; C_l is the random effect of the l-th cow; φ_q are regression coefficients associated with the linear (q = 1) and quadratic (q = 2) effects of cow age; β is the regression coefficient associated with the linear effect of proportion of Holstein f; λ is the regression coefficient associated with the linear effect of heterosis (h) between Holstein and Gyr breeds; and e_{ijklm} is the residual random error associated with observation y_{ijklm}, assuming NID (0, σ_e^2).

Model 3:

$$y_{ijkl} = \mu + S_i + BY_j + H_k + C_l + \beta f + \lambda h + e_{ijkl},$$

in which y_{ijkl} is the observation of the AFC trait measured at the i-th birth season, in the j-th birth year, in the k-th herd, in the l-th cow; μ is the general mean; S_i is the fixed effect of the i-th birth season; BY_j is the fixed effect of the j-th birth year; H_k is the fixed effect of the k-th herd; C_l is the random effect of the l-th cow; β is the regression coefficient associated with the linear effect of proportion of Holstein f; λ is the regression coefficient associated with the linear effect of heterosis (h) between Holstein and Gyr breeds; and e_{ijkl} is the residual random error associated with observation y_{ijkl}, assuming NID (0, σ_e^2).

Test-day milk yield and calving and cow birth seasons in the models were equally defined as dry season (April to September) and rainy season (October to March). The TDMY of each cow during lactation were grouped into ten classes of days in milk (class 1: 5 to 30 days; class 2: 31 to 60 days; class 3: 61 to 90 days; and up to class 10: 270 to 305 days).

3. Results

3.1. Average estimates of different traits

The least square means of reproductive (AFC, CI12, CI23, DO12, DO23, DP12, and DP23) and productive (MY3051, MY3052, MY3053, TDMY1, TDMY2, TDMY3, LL1, LL2, and LL3) traits differed among the eight genetic groups evaluated (P>0.05) (Table 2).

The mean AFC estimates ranged from 965.62 to 1084.91 days for the different genetic groups. It was observed that cows with the smallest ages at the first calving were those belonging to the genetic groups 1/2H (965.62 days), 3/4H (983.38 days), and H (992.08 days), that is, around 32 months for the first calving. In contrast, cows that had the highest mean AFC estimates were those belonging to groups 3/8H (1084.91 days) and G (1082.44 days), that is, around four months more to have the first calving.

The CI12 ranged from 427.37 to 485.31 days in the different genetic groups, with group 1/2H, followed by groups H and 3/4H, presenting the lowest mean CI. The means of CI23 ranged from 414.14 to 471.33 days in the different genetic groups, in which group 1/2H, followed by 3/4H and 1/4H, had the second lowest CI. It is possible to observe that there was a reduction in CI with the advancement of parities (except for H).

Regardless of the genetic group, mean DO12 values ranged from 120.41 (H) to 149.53 (1/4H) days, and DO23 ranged from 118.81 (1/2H) to 147.49 (G) days, allowing to observe that, on average, the days open decreased in the second period of CI. On average, DP12 ranged from 93.96 (3/4H) to 169.86 (G) days among the different genetic groups. Together with the genetic group 7/8H (94.37 days), groups 3/4H (93.96) and 1/2H (101.53 days) had the shortest dry periods, while the longest periods were observed for groups 1/4H (158.71 days) and G (169.86 days).

Table 2 - Least square means and standard errors for reproductive (AFC, CI12, CI23, DP12, and DP23) and productive (MY3051, MY3052, MY3053, TDMY1, TDMY2, and TDMY3) traits of Holstein (H), Gyr (G), and Girolando (1/4H, 1/2H, 3/4H, 3/8H, 5/8H, and 7/8H) cows

Trait	Genetic group							
	H	7/8H	3/4H	5/8H	1/2H	3/8H	1/4H	G
AFC (day)	992.08bc±157.92	995.27c±161.82	983.38b±160.21	1016.07d±155.76	965.62a±151.99	1084.91f±126.58	1050.67e±144.14	1082.44f±124.19
CI12 (day)	427.37ab±96.27	438.26bc±101.35	432.00ac±98.75	440.01b±99.05	427.24a±95.19	434.81ab±93.06	485.31d±114.08	484.98d±103.85
CI23 (day)	432.60b±93.82	424.10ab±87.41	418.91a±88.64	432.19b±93.82	414.14a±90.81	422.50ab±85.66	420.87ab±76.11	471.33c±91.58
DO12 (day)	120.41a±51.95	126.97a±53.02	125.20a±51.70	127.40a±53.17	124.47a±52.86	128.57ab±54.34	149.53c±60.51	142.45bc±50.00
DO23 (day)	125.40ab±50.50	121.47ab±48.77	121.11a±51.15	126.54b±53.52	118.81a±51.95	121.64ab±51.10	122.56ab±48.52	147.49c±51.10
DP12 (day)	102.53ab±64.02	94.37a±55.71	93.96a±58.48	120.99c±68.00	101.53b±63.49	117.57c±62.61	158.71d±75.38	169.86d±59.07
DP23 (day)	100.22a±51.89	102.67a±55.89	104.99a±63.71	122.34c±66.89	112.12b±63.49	133.71cd±71.48	147.40de±59.64	161.90e±51.49
MY3051 (kg)	4686.11a±2084.27	4619.28a±2021.42	4636.56a±2045.78	3918.18b±1956.56	4631.96a±2232.63	3856.34b±2035.95	4084.76b±2375.30	2913.74c±1150.62
MY3052 (kg)	5104.85ab±2167.55	5129.88a±2209.03	4903.13b±2198.81	4162.26d±2110.32	4725.95c±2271.17	3838.08e±2041.62	3706.08e±2078.40	2619.21f±1099.79
MY3053 (kg)	5261.48a±2255.66	5137.54a±2240.49	4937.29b±2175.87	4166.85c±2079.65	4933.68b±2411.06	4139.14c±2379.59	3634.40d±2219.92	2855.75e±1506.34
TDMY1 (kg)	13.91c±7.70	14.91b±7.09	15.05b±7.15	12.85d±7.17	15.51a±8.15	12.13d±6.40	13.65c±8.25	8.94e±4.70
TDMY2 (kg)	15.70a±8.19	15.52a±7.59	15.60a±7.27	13.86b±7.24	15.24a±8.00	13.19bc±7.16	12.13c±6.73	8.85d±4.44
TDMY3 (kg)	15.07ab±7.94	14.43b±7.44	15.19a±7.72	13.22c±7.42	15.02ab±8.46	12.35cd±7.90	11.72d±6.66	8.47e±5.03
LL1 (day)	296.24a±94.92	304.31a±108.73	301.54a±105.67	285.39bc±101.03	287.24b±100.79	272.62d±95.80	276.14cd±96.97	278.26bd±81.51
LL2 (day)	294.18ab±97.95	299.09a±96.58	286.43b±100.30	273.61c±97.75	269.63cd±92.24	258.82d±86.74	269.06cd±90.36	266.08cd±79.38
LL3 (day)	299.44a±101.23	295.17a±99.28	282.96b±100.79	273.70c±99.59	270.84ce±94.94	260.43de±85.16	257.96e±89.05	277.84bcd±76.80

AFC - age at first calving; CI12 - interval between first and second calving; CI23 - interval between second and third calving; DO12 - days open between the first and second calving; DO23 - days open between the second and third calving; DP12 - dry period between first and second calving; DP23 - dry period between second and third calving; MY3051 - 305-day milk yield in the first lactation; MY3052 - 305-day milk yield in the second lactation; MY3053 - 305-day milk yield in the third lactation; TDMY1 - test-day milk yield in the first lactation; TDMY2 - test-day milk yield in the second lactation; TDMY3 - test-day milk yield in the third lactation; LL1 - lactation length in the first lactation; LL2 - lactation length in the second lactation; LL3 - lactation length in the third lactation.
a-f - Estimates of least square means with different letters in rows are significantly different (P<0.05).

In the following dry period (DP23) the genetic group H (100.22 days) had the shortest period, followed by groups 7/8H (102.67 days) and 3/4H (104.99 days) and the longest, as occurred in DP23, were also for groups 1/4H (147.40 days) and G (161.90 days).

For reproductive traits, it is possible to observe a superiority of genetic groups 3/4H and 1/2H, which have 0.75 and 0.50 Holstein genetic proportion, respectively.

In general, there was an increase in the mean MY305 during lactations (MY3051, MY3052, and MY3053) for most genetic groups, except for groups 1/4H, 3/8H, and G. The means for MY3051, MY3052, and MY3053 of the different genetic groups ranged from 2913.74 to 4686.11 kg, 2619.21 to 5129.88 kg, and 2855.75 to 5261.48 kg, respectively. It was observed that cows of genetic groups H, 7/8H, 3/4H, and 1/2H presented MY3051 similar and with the highest yields. For the second-lactation cows, the highest milk yields were observed for groups 7/8H, followed by H. Again, in the third lactation, the highest yields of milk production were observed for cows in groups 7/8H and H. Thus, regardless of the lactation order, there is a superiority in milk production at 305 days for cows of genetic groups H and 7/8H, followed by groups 3/4H and 1/2H.

In contrast, TDMY showed the highest mean values in the second lactation for most genetic groups, except for G, 1/4H, and 1/2H. Following the same behavior of MY305, the highest TDMY means were observed for genetic groups H, 7/8H, 3/4H, and 1/2H, regardless of the lactation.

The mean estimate for TDMY at each lactation in the different genetic groups studied ranged from 8.94 to 13.91 kg for TDMY1, from 8.85 to 15.70 kg for TDMY2, and from 8.47 to 15.19 kg of milk for TDMY3.

The length of lactation of first- (LL1), second- (LL2), and third-lactation (LL3) cows of different genetic groups showed significant differences that varied regardless of the lactation order ($P<0.05$). In this sense, the LL1 mean varied from 272.62 to 304.31 days in the different genetic groups, being higher in groups 7/8H and 3/4H. The mean LL2 ranged from 258.82 to 299.09 days in different genetic groups, being higher in groups 7/8H and H. However, it was observed that cows in group 3/8H had the shortest duration of lactation in both lactations. In contrast, for the third lactation the shortest duration was observed for cows in group 1/4H. Thus, the LL3 means ranged from 257.96 to 299.44 days in the different genetic groups, being higher in groups H and 7/8H. The superiority in the lactation duration of cows of the genetic group 7/8H was observed regardless of the lactation order.

3.2. Breed and heterosis effects on the traits

The breed effect was significant for most evaluated traits (Table 3). In the reproductive traits, there was a negative and favorable breed effect. Age at first calving (−159.80±10.01 days), DP12 (−75.52±7.27 days), and DP23 (−62.14±8.34 days) had the greatest breed effect ($P<0.001$), followed by CI12 (−35.61±10.62 days) and DO12 (−23.37±6.70 days). The productive traits (TDMY and MY305) also showed a positive and favorable significant breed effect ($P<0.001$) for all lactations. The highest means were observed in the second lactation, MY3052 of 2697.23±156.23 kg and TDMY2 of 9.50±0.56 kg.

A negative (favorable) and significant heterosis effect was also verified for the evaluated reproductive traits. However, the greatest heterosis effect observed was on AFC (−104.04±5.97 days), CI12 (−29.80±6.14 days), and DP12 (−29.87±4.18 days). All production traits showed a highly significant ($P<0.001$) and positive (favorable) for the heterosis effect. The greatest mean positive heterosis effects were for MY3053 (976.57±96.28 kg) and TDMY3 (3.98±0.15 kg) of the crossbred cows compared with the average daily milk yield of purebred animals.

In general, we observed that the heterosis effect promoted a favorable reduction in AFC in the intervals or periods evaluated of the reproductive traits, as well as a favorable and expected increase in milk yield in different lactations. When we analyzed heterosis in percentage (%), it was possible to observe the greatest gains when using a crossbred cow instead of purebred, this mainly in productive traits, TDMY3 (33.81%) and MY3053 (24.06%). In reproductive traits, the greatest impacts could be observed in DP (10.41% and 21.93%) and AFC (10.03%).

Table 3 - Breed and heterosis effects and their standard errors and heterosis percentage for reproductive (AFC, CI12, CI23, DP12, and DP23) and productive (MY3051, MY3052, MY3053, TDMY1, TDMY2, and TDMY3) traits

Trait	Breed effect[1]	Heterosis effect	Heterosis (%)
AFC (day)	−159.80±10.01***	−104.04±5.97***	10.03
CI12 (day)	−35.61±10.62**	−29.80±6.14***	6.53
CI23 (day)	−1.45±11.30	−20.60±6.64**	4.55
DO12 (day)	−23.37±6.70**	−12.57±3.86**	9.56
DO23 (day)	−4.68±7.19	−9.05±4.28*	6.63
DP12 (day)	−75.52±7.27***	−29.87±4.18***	21.93
DP23 (day)	−62.14±8.34***	−13.65±4.87**	10.41
MY3051 (kg)	2165.24±114.09***	799.03±69.56***	21.02
MY3052 (kg)	2697.23±156.23***	897.46±93.95***	23.23
MY3053 (kg)	2500.68±157.12***	976.57±96.28***	24.06
TDMY1 (kg)	7.69±0.38***	3.04±0.23***	26.62
TDMY2 (kg)	9.50±0.56***	2.05±0.34***	16.71
TDMY3 (kg)	8.81±0.24***	3.98±0.15***	33.81
LL1 (day)	42.31±6.14***	0.15±3.76	0.05
LL2 (day)	49.73±7.71***	−4.19±4.64	−1.49
LL3 (day)	37.98±7.70***	−7.44±4.72	−2.57

AFC - age at first calving; CI12 - interval between first and second calving; CI23 - interval between second and third calving; DO12 - days open between the first and second calving; DO23 - days open between the second and third calving; DP12 - dry period between first and second calving; DP23 - dry period between second and third calving; MY3051 - 305-day milk yield in the first lactation; MY3052 - 305-day milk yield in the second lactation; MY3053 - 305-day milk yield in the third lactation; TDMY1 - test-day milk yield in the first lactation; TDMY2 - test-day milk yield in the second lactation; TDMY3 - test-day milk yield in the third lactation; LL1 - lactation length in the first lactation; LL2 - lactation length in the second lactation; LL3 - lactation length in the third lactation.
[1] Breed effect was calculated based on Holstein breed.
* P<0.05; ** P<0.01; *** P<0.001.

4. Discussion

4.1. Estimation of reproductive and productive traits

According to Facó et al. (2005), the first calving marks the beginning of a cow productive life and greatly influences its productive and reproductive life. The heritability of the trait is generally low, indicating that it is highly influenced by the management system (Häggman et al., 2019). The lowest AFC means (best performance) were found in cows with a greater presence of the proportion of the Holstein breed, such as those belonging to genetic groups H, 7/8H, 3/4H, and 1/2H (Table 2). Cows in these genetic groups also showed higher milk yield, both for MY305 and TDMY, in the evaluated lactations. Facó et al. (2005) also observed lower AFC for 1/2H, 3/4H, and 7/8H animals. The results obtained also meet those found by Lemos et al. (1992), in which the 1/2H (F1) had the first calving at younger age than other genetic groups, revealing that there may be a greater intensity of selection and use of best sires in the formation (matings) of these genetic groups. The advantage of using precocious animals, according to a study performed by Eastham et al. (2018), is that younger cows at first calving showed higher mean daily milk yield over the productive life and a higher probability of reaching the second lactation, in addition to expressing good fertility associated with shorter CI.

Calvin interval is an important reproductive index, as it affects the total milk yield during lactation and the number of calves born from cows; thus, the dairy systems should aim at an ideal 12-13-month CI (Ibrahim and Seid, 2017). In addition to a reproductive problem and with direct consequences in the number of lactating cows, CI often reflects problems associated with management (Dono et al., 2013). In our study, we found a mean of 446.24 days (14.6 months) for CI12 (Table 2). Although close to the recommended ideal range, this longer period for CI12 may be associated with the genetic variation existing among the evaluated genetic groups, since groups G and 1/4H presented means well above the general population mean for this trait. McManus et al. (2008) reported that the later cows among several genetic groups were those that have a higher proportion of the Gyr breed, such as G, 3/8H, and 1/4H; in addition, they also reported that animals with more proportion of the Holstein breed were reproductively more efficient and probably had higher calf production in the same interval than crossbred animals. The results obtained so far corroborate. Titterington et al. (2017), who stated that as the age of the animal increases, CI tends to decrease. Our results also reveal the superiority of adaptation of crossbred animals with a higher proportion of Holstein to Brazilian production systems, corroborating McManus et al. (2008). Thus, we observed that the second CI (CI23) decreased in relation to the first (CI12) for the evaluated genetic groups, except for group H (Table 2). The decrease in CI is directly related to the days open, which include anestrus and service periods of cows and should vary from 80 to 85 days to allow the production of one calf per year (Ibrahim and Seid, 2017). We observed in our study that for DO12, cows that had the longest period of days open were those of genetic groups 1/4H (149.53 days) and G (142.45 days) (Table 2). The observed values indicate an increase in CI and, consequently, cause a smaller number of calves born, interfering in the livestock profitability (Krpálková et al., 2020).

Differently from what was observed for DO12, in DO23, cows with longer periods were those of the genetic groups G (147.49 days) and 5/8H (126.54 days). Singh et al. (2016) reported that reproductive efficiency varies not only between breeds but also among the animals within the same breed. However, these observed variations can also be explained by regional seasonal changes, such as calving season or other environmental factors (Boni et al., 2014), i.e., the reproductive efficiency of animals does not depend only on the genetic merit of the animals (genetic composition), but also on other factors such as nutrition, management, health, and the environment (Ibrahim and Seid, 2017). In addition to CI and days open, the efficiency of the milk production chain can be assessed through the duration of animals' dry period, recommended as 60 days as ideal (Schaeffer and Henderson, 1972; Capuco et al., 1997). Days open also affect farm profitability; they are expensive for the production system, as they cause a reduction in yield per unit of time. According to Louca and Legates (1968), more days open are identified with an extended late lactation period in which daily production is low, whereas fewer days open are similarly identified with a shorter period of low daily production. The first and second dry periods (DP12 and DP23) differed ($P<0.05$) for the genetic groups, with the evaluated population presenting a higher mean than that recommended as ideal (Schaeffer and Henderson, 1972; Capuco et al., 1997), in particular, some of the genetic groups that have a greater presence of the proportion of the Gyr breed (Table 2).

According to Rangel et al. (2009), the shorter the lactation duration, the longer the dry period and the lower the percentage of lactating cows in the herd, causing lower daily milk yield, directly compromising the economic efficiency of the dairy activity, because the longer the lactation duration, the greater the milk yield of cows. It is known that Holstein cows have a longer mean lactation duration, which varies from 279 (McManus et al., 2008) to 303 (Mellado et al., 2011) days, followed by Holstein × Gyr crossbred cows with 283 days (Ribeiro et al., 2017); Gyr cows have the lowest mean lactation duration with 275 days (Ruas et al., 2014).

Similarly, we identified in our study that cows showed differences in the lactation duration according to the genetic group (values not shown), which may have generated the variation of the dry period of cows with a tendency to prolong this period for the genetic groups of animals with greater presence of the proportion of the Gyr breed. However, it is noteworthy that a short dry period will not provide adequate rest and time for mammary regeneration, while long dry periods will result in higher feed costs with no income from milk production and can also result in fat cows that are more prone to health and reproductive performance problems (Annen et al., 2004).

In the productive traits, there was a significant difference ($P<0.05$) in milk yield in different lactations for the evaluated genetic groups, with a higher mean for MY3051 verified in cows belonging to genetic groups H, 7/8H, 3/4H, and 1/2H. The same sequence in the classification for greater productivity was observed by Daltro et al. (2020), while Balancin Júnior et al. (2014) observed greater milk yield in the first lactation for genetic groups 7/8H, 3/4H, and 1/2H. For MY3052, the same genetic groups had the highest milk yields, while the lowest productive means were observed in cows of genetic groups 1/4H, 3/8H, and G, similarly to those identified by McManus et al. (2008), that is, animals with a higher genetic proportion of Gyr. It is believed that this result is linked to improvement in environmental conditions, frequent use of genetically superior bulls selected by existing breeding programs, and the greater intensity of selection applied to the Holstein breed, which is specialized in milk production (Freitas et al., 2004).

The mean MY305 increased with the progress of the lactation order, in a way that genetic groups H, 7/8H, 3/4H, and 1/2H remained the most productive. In relation to 1/2H animals (F1 generation), Ruas et al. (2014) found that F1 animals (Holstein × Gyr) showed a greater amount of milk produced with advancing lactations, with peak production in the seventh lactation. Similarly, according to Cerdótes et al. (2004), cow age strongly influences milk production, constituting an important source of variation. Cobuci et al. (2000) claimed that the variations that occur with advancing cow age are mainly caused by physiological factors and provide maximum performance with animal maturity. On the other hand, genetic groups 1/4H and G had a higher mean MY305 in the first lactation order. Choudhary et al. (2017) verified that the performance of F1 (Tharparkar × Holstein Friesian) crosses was found to be superior during the first lactation when compared with the third one. The same authors reported this could be due to larger number of gene pairs interacting to express a certain trait, for the Holstein Friesian is a well-known productive breed in the world, and when crosses are done between a low-productive breed like Tharparkar, there is an improvement of the performance of the first generation by the increase in heterotic responses of the animals. However, this advantage of increased productivity is lost with the effects of gene recombinations that lead to the breaking of this heterotic superiority in subsequent generations. This effect of gene recombination may be a likely factor that explains the fact that animals from genetic group 1/4H (F2 generation) have a higher milk production in the first lactation when compared with the others.

Although an increase in milk production during cows' lactations is also expected by the recurrent effect of selection, our results can be attributed to a possible inferior genetic merit of this group of cows, indicating that selection for milk production appears not to have been as effective in these animals as subsequent lactations performed poorly. In this sense, Freitas et al. (2004) found that only 1/4H presented an estimate of predicted transmitting ability (PTA) lower than the population mean when compared to the other groups of the Girolando breed. However, it is noteworthy that this superiority in milk production in the first lactation was only 9.28 and 11.03%, respectively, when compared with the second and third lactation. Another factor that could explain this superiority in the first lactation is its duration when compared with the following lactations. It was also found that the mean TDMY was higher in the second-lactation cows (TDMY2), except for those belonging to genetic group G. These results may be related to a longer lactation time in the second order (H, 7/8H, and 3/4H) or the small difference in the duration of the second lactation in relation to the third one, which can be observed in genetic groups 5/8H and 1/2H. These results show that both mean MY305 and TDMY were affected by lactation duration. Choudhary et al. (2017) found that total milk production in lactation was significantly affected by lactation duration during the second lactation. Thus, to maintain milk production in subsequent lactations, several factors such as body condition, season of the year, and feed management can be responsible for maintaining a good level of milk production in the subsequent lactations of crossbred cows (Choudhary et al., 2017).

4.2. Breed and heterosis effects

As expected, AFC showed a significant heterosis effect ($P<0.001$), with gains of −104.04 days, which represents a 10.03% reduction in AFC in relation to the mean of their purebred parents (Table 3). Vergara et al. (2009) observed a negative and not significant effect for AFC in animals from different crosses (Angus × Blanco Orejinegro × Zebu); the authors attributed the influence of management, nutrition, and climate to this result. Meanwhile, Penasa et al. (2010a) found very low, but significant, heterosis effects for AFC in F1 cows (Pure Holstein × British Holstein). However, it is important to keep in mind that this trait depends more on the decisions of the breeders than on the physiological aspects of the animal itself.

Knowing that AFC is benefited by the heterosis effect, it is possible to make use of crosses aiming at the economic benefits for this trait and to improve the reproductive performance of the herd. In addition, the use of selection for AFC can generate moderate genetic advancement for sexual precocity in Girolando herds (Canaza-Cayo et al., 2018). On the other hand, significant and favorable heterosis effects for the first and second CI were expected, as traits of low heritability (0.00-0.08), according to Canaza-Cayo et al. (2018), tend to be more benefited from the heterosis effect (Facó et al., 2008).

In our study, crossbred cows showed a reduction of 6.53 and 4.55%, respectively, for CI12 and CI23, in relation to the mean of their purebred parents (Table 3), indicating that the use of crossbreeding can be a strategy to improve reproductive efficiency, since heterozygosity and, consequently heterosis, can reduce CI (Facó et al., 2008). The magnitude of this heterosis in these first two CI, obtained through the value of the coefficient of heterosis effect (Table 3), allows for an indirect increase in profitability since the increase in income for the dairy activity is based on shorter CI and the use of crossbred animals, since these animals tend to be less dependent on inputs (nutrition, health, and technology) (Gazzarin et al., 2018). In reducing CI, it is important to assess the behavior of the days open of the cows; in this way, there was a significant and favorable effect for DO12 (−12.57±3.86 days) and DO23 (−9.05±4.28 days), with reductions that represented 9.56 and 6.63% of heterosis, respectively, in these traits (Table 3).

The DP12 and DP23 were the reproductive traits most benefited by the heterosis effect, with reductions of −29.87 and −13.65 days, respectively. Thus, the possible gains in performance verified in crossbred animals, when compared with the means of their purebred parents, represented 21.93 and 10.41%, respectively, for DP12 and DP23 (Table 3). These results are important as the reduction in dry periods contributes to the increase in lactation duration, in addition to having shorter CI (Canaza-Cayo et al., 2018). Therefore, the reductions in AFC, days open, CI, and dry periods of animals verified by the heterosis effect are advantageous indications for the different production systems that use crossbred animals, because the increase in these traits represent economic losses in properties and affect, in general, the reproductive activity of dairy cows.

Heterosis can be beneficial for several economically important traits, including reproductive ones. These traits are particularly important in low-management production systems common in the tropics, with significant potential to improve profitability (Bunning et al., 2019). Thus, one way of improving the performance of dairy production systems in regions of warm climate, by means of genetic improvement, is the utilization of crosses between zebu breeds, which exhibit excellent adaptation to tropical environment (Perotto et al., 2010).

Regarding the productive traits, we found that there is an effect of positive heterosis on TDMY and MY305 for Girolando cows, regardless of the lactation order. Some of the few studies carried out in Brazil with Girolando cattle also reported a significant heterosis effect on milk yield in the first lactation (Daltro et al., 2019, 2020; Facó et al., 2008).

Heterosis caused an increase in the milk yield of Girolando cows of 21.02, 23.23, and 24.06%, respectively, for MY3051, MY3052, and MY3053, when compared with the means of their purebred parents (H and G) (Table 3). Bunning et al. (2019) found gains in heterosis of 35.15% for milk yield in crossbred cows (*Bos taurus taurus* × *Bos taurus indicus*), while Lembeye et al. (2016) found a heterosis variation of 3.3-5.9% for crossbred animals (Holstein - Friesian × Jersey) and concluded that lactation and production level (evaluated as low, medium, and high) of cows affect the expression of heterosis.

Higher milk yield implies better economic parameters, as they represent lower costs per kilogram of milk produced and, thus, greater profitability (Němečková et al., 2015). For this reason, the heterosis effect is an important benefit of the use of crosses in the adaptation of dairy cattle in tropical conditions as in Brazil, because in addition to being animals more adapted to the environment, they are more competitive due to their low maintenance costs (Lopez-Villalobos et al., 2000), resulting in greater production efficiency. Although crossbreeding is tacitly associated with the exploitation of heterosis, it cannot be neglected that, in conditions where management and other environmental factors are also being improved, the use of additive breed differences via increases in the proportion of *Bos taurus* genes in the breed composition of animals can result in great benefits to dairy production (Facó et al., 2002).

For TDMY, a greater heterosis effect was observed for TDMY1 (3.04±0.23 kg), which represented an increase of 26.61% when compared with the mean of their purebred parents (Table 3). Similar to what was observed for the first lactation, we verified that the heterosis effect for TDMY2 (3.01±0.32 kg) and TDMY3 (2.91±0.34 kg) represented potential gains of 24.53 and 24.72%, respectively, in the daily milk yield when compared with the means of their purebred parents (H and G). We also observed that the heterosis effect was more significant in the third lactation for MY305, while for TDMY, it was greater in the first lactation. Lembeye et al. (2016) observed lower heterosis effect in the first lactation (105±11 kg) for Holstein-Friesian × Jersey animals and greater heterosis effect in the fourth lactation (219±12 kg). The magnitude of the heterosis effect values generated gains of 4.7 and 6.6% in milk production, respectively, in the first and fourth lactations.

According to Buckley et al. (2003), a higher milk yield from crossbred cows (Holstein × Friesian) is associated with better reproductive performance. In contrast, Němečková et al. (2015) observed a worse reproductive performance for Holstein cows with high milk yield when compared with cows of the same breed, but with lower milk yield.

In our study, we observed that cows with a higher genetic fraction of the Holstein breed (7/8), in general, had higher productive indexes, whereas crossbred cows showed better reproductive performance (3/4H and 1/2H). This indicates a significant influence of the heterosis effects on these two groups of traits and, when used appropriately for the interest of the breeders, they contribute to the improvement of the herds. In general, it is possible to improve the efficiency of the animals for the main reproductive and productive traits of economic interest for the breeders through the exploration of the heterosis effect, by the careful choice of the genetic material of the parents to be used in the crosses.

5. Conclusions

This type of crossing, widespread in Brazil, allows an expressive heterosis effect to occur (between 4.5 and 26.5%), benefiting the cows of the different genetic groups of the Girolando breed, with the reduction in age at first calving, calving intervals, days open, and dry periods, in addition to the expressive increase in test-day milk yield and 305-day milk yield, regardless of the calving order. The results indicated that the cows of the genetic groups 7/8H, 3/4H and 1/2 were superior for the productive and reproductive traits, allowing the producer to choose the most suitable type of genetic composition according to the production system adopted in his property. Thus, expanding the use Girolando in Brazil tends to be very advantageous, in terms of performance and economic efficiency, for the national dairy industry.

Conflict of Interest

The authors declare no conflict of interest.

Author Contributions

Conceptualization: M.T. Vieira and J.A. Cobuci. Data curation: M.T. Vieira and D.S. Daltro. Formal analysis: M.T. Vieira and D.S. Daltro. Methodology: M.T. Vieira and J.A. Cobuci. Project administration: J.A. Cobuci. Resources: M.T. Vieira and J.A. Cobuci. Supervision: J.A. Cobuci. Visualization: M.T. Vieira and D.S. Daltro. Writing-review & editing: M.T. Vieira, D.S. Daltro and J.A. Cobuci.

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