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Breed, parity, and cycle season effects on life-time reproduction in bitches: a retrospective study

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Abstract: The aim of this retrospective study was to evaluate the effects of breed, parity, and season on the reproductive parameters in bitches reared under a temperate climate. Breeding and pregnancy statuses, proestrus, estrus, and pregnancy durations, litter size, and cycle interval were recorded from German shepherds (GS, n = 34), Labrador retrievers (LA, n = 23), Belgian Malinois dogs (BM, n = 13), and Pointers (PO, n = 9) up to 10th parities. The mean age at first breeding (464.8 ± 26.2 days, mean ± SD) and the pregnancy rate (74.6%) were not different among breeds. The pregnancy duration was shortened as the number of puppies born increased in each whelping ($P < 0.008$). The cycle interval varied by breed (208.2, 215.1, 208.6, and 237.0 days for GS, LA, BM, and PO, respectively, $P < 0.01$) and decreased linearly from 241.1 to 202.0 days as parity increased from 1 to 10 ($P < 0.04$). The season did not affect the cycle interval, proestrus, and estrus length ($P > 0.05$). Reproductive parameters varied among the GS, LA, BM, and PO bitches reared in Turkey. The effect of seasonal variation was negligible.

Key words: Bitches, litter size, estrus cycle, pregnancy duration, season

1. Introduction

Bitches are monoestrous, spontaneous ovulatory and traditionally nonseasonal mammalians, except for some breeds such as Basenji and Chow-Chow (1). The estrous cycle of the bitch is considerably longer than that of other domestic species and the estrous period is followed by a longer interestrous interval (diestrus and anestrus) (2). The interval is highly variable (2–10 months) among the breeds (3,4). Various factors that affect the interestrous period and pregnancy duration, such as breed, age, parity, and litter size have been described in previous studies (3,5,6).

Although bitches are considered nonseasonal, the seasonal effect on the reproductive parameters are still contradictory. Studies confirming (1,7,8) and denying (3,9–11) seasonal effects on bitch reproduction are available. To our knowledge, this was the first comprehensive retrospective study in Turkey investigating reproduction in bitches to reveal: 1) the distribution of whelping among breeds; 2) the breed and parity effect on pregnancy rates; 3) the breed, parity, and estrus onset season effects on the cycle interval, the proestrus, and estrus length; and 4) the breed and parity effects on the pregnancy duration and litter size.

2. Materials and methods

2.1. Regional characteristics

Seasonal characteristics of the study area (40°11′N, 29°04′E) are presented in Table 1. These categorizations were based on statistics released by the Turkish State Meteorological Service.

2.2. Section of animal and management

Nine years of reproductive records of 465 estrous cycles from 79 bitches [German shepherds (GS; 196 / 34), Labrador retrievers (LA; 148 / 23), Belgian Malinois dogs (BM; 78 / 13), and Pointers (PO; 43 / 9)] were recorded. The bitches were living in the same environment and with the same husbandry. They were housed in individual kennels and were fed with commercial dry food. Vaginal cytology and behavioral signs were evaluated in all bitches. Proestrus bleeding was monitored every morning and estrus was detected by daily vaginal cytology, which was started when proestrus bleeding was first detected. Neutrophil disappearance, a decrease of erythrocytes, an increase of superficial cells in a microscopic area, and the acceptance of mating were regarded as the estrus onset (12). Pubertal age data were not available. When vaginal cytology revealed >80% cornified superficial cells, the bitches were brought to the mating area. The bitches were

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Table 1. Seasonal characteristics of the study environment.*

Variable	Season			
	Winter (Dec–Feb)	Spring (Mar–May)	Summer (Jun–Aug)	Fall (Sep–Nov)
Min. temp. (°C)	–6.09 ± 3.34	0.83 ± 4.13	12.42 ± 2.28	3.37 ± 4.91
Max. temp. (°C)	19.54 ± 2.38	28.39 ± 4.11	36.85 ± 2.27	29.73 ± 4.47
Av. temp. (°C)	6.01 ± 1.85	13.19 ± 4.42	24.29 ± 1.63	15.28±4.24
Humidity (%)	68.53 ± 3.33	64.26 ± 5.43	58.22 ± 4.89	68.28 ± 4.93
Daylight (h)	10:10 ± 00:42	13:28 ± 01:13	14:44 ± 00:38	11:27 ± 01:15
	Day-light duration (h) **			
	10:43 ± 00:59 (Oct–Mar)		14:11 ± 00:56 (Apr–Sep)	
Min. temp. (°C)	–3.37 ± 4.35		8.63 ± 5.07	
Max. temp. (°C)	23.01 ± 4.73		34.24 ± 3.74	
Av. temp. (°C)	8.71 ± 3.92		20.67 ± 4.45	
Humidity (%)	68.44 ± 4.27		61.20 ± 5.88	

*Data are mean ± SD.
**The mean daylight duration was 12:27 ± 02:01 h. Short and long days were categorized based on the mean daylight duration, shorter and longer than 12:27 h.

mated naturally every other day starting from the first voluntary mating until the last day of estrus (refusal to mate) under observation. The total number of male dogs was 41 [(GS, 12), (LA, 10), (BM, 5) and (PO, 4)] in the kennel. The bitches were not tested for LH or progesterone levels before mating for the calculation of gestation, and the last mating date was accepted as day 1 (8). Ultrasonographic examination for pregnancy diagnosis was performed 25 days after the last mating date.

2.3. Data handling and statistical analysis

The climatological characteristics of the study environment were subjected to the PROC MEAN procedure (Table 1). The whelping season by breed was subjected to cross-tabulation and chi-square analysis. For continuous variables (pregnancy duration, litter size, cycle interval, proestrus length, and estrus length), the linear model included the effects of breed, parity, and estrus onset as well as 2-way interactions using the PROC MIXED procedure. In significant model terms, the data were subjected to hierarchical backward elimination to avoid overparameterization. The dog within breed × parity was a random term in the models. Due to missing values (dogs), the 10th parity P value was generated using the Satterthwaite approximation option. The mean differences were attained using the LSD option. Daylight was replaced with season in another linear model when the daylight duration effect was obtained (10:43 ± 00:59 vs. 14:11 ± 00:56 h, occurring in October–March vs. April–September). Moreover, interrelationships among reproductive parameters were determined using the PROC CORR procedure. Finally, the mathematical relationship between the pregnancy duration and the litter size was established using the PROC REG procedure. In all statistical analyses (13), $P < 0.05$ was considered significant.

3. Results

The distribution of bitches for the whelping season was not different among breeds ($\chi^2 = 10.38$, $P < 0.32$). There were no main effects of the breed ($P < 0.32$) and whelping season ($P < 0.63$) and the effect of the breed by whelping season interaction was $P < 0.26$ on the first breeding age (Figure 1). The mean first breeding age was 467 days. The mean pregnancy rate was 71.21% (n = 253) among the breeds (Figure 2).

Overall, the mating ($\chi^2 = 12.59$, $P < 0.19$, in total of 465 cycles) and pregnancy ($\chi^2 = 7.08$, $P < 0.63$, in total of 339 mated bitches) rates were independent from the breed and parity. As the parity advanced, the pregnancy rates did not differ among the breeds (Table 2).

The pregnancy duration varied by breed ($P < 0.02$), but not by parity ($P < 0.33$) (Table 3). PO had the longest pregnancy duration (65.19 days), whereas BM had the shortest pregnancy duration (62.24 days). The mean litter size varied by both breed ($P < 0.001$) and parity ($P < 0.002$)

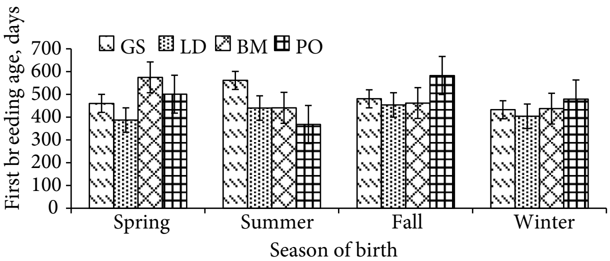

Figure 1. The interaction between breed, season of parturition, and first breeding age.

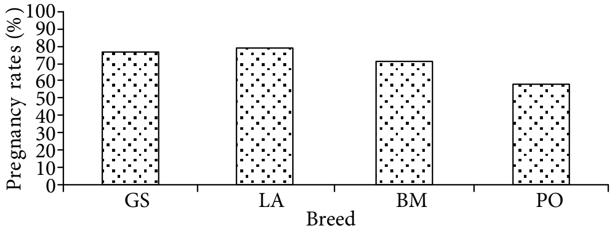

Figure 2. The effect of breed on pregnancy rate.

Table 2. The effect of parity on pregnancy rates (%).*

Parity	Pregnancy rate				
	GS (n = 34)	LA (n = 23)	BM (n = 13)	PO (n = 9)	Statistics
1 (n = 79)	78.6 (34)	86.7 (23)	72.7 (13)	71.4 (9)	χ^2 = 1,02, P < 0.80
2 (n = 77)	76.7 (34)	73.3 (23)	42.9 (11)	42.9 (9)	χ^2 = 5.28, P < 0.15
3 (n = 67)	66.7 (27)	80.0 (22)	62.5 (9)	42.9 (9)	χ^2 = 4.06, P < 0.38
4 (n = 61)	81.0 (26)	64.3 (19)	85.7 (9)	60.0 (7)	χ^2 = 2.24, P < 0.52
5 (n = 51)	86.7 (18)	81.8 (19)	71.4 (9)	66.7 (5)	χ^2 = 1.11, P < 0.77
6 (n = 41)	83.3 (15)	100.0 (15)	60.0 (9)	100.0 (2)	χ^2 = 4.28, P < 0.23
7 (n = 33)	66.7 (14)	75.0 (11)	100.0 (7)	100.0 (1)	χ^2 = 1.38, P < 0.50
8 (n = 25)	100.0 (12)	66.7 (7)	75.0 (5)	100.0 (1)	χ^2 = 2.43, P < 0.30
9 (n = 17)	75.0 (9)	50.0 (5)	100.0 (3)	-	χ^2 = 1.33, P < 0.51
10 (n = 14)	50.0 (7)	100.0 (4)	100.0 (3)	-	χ^2 = 2.86, P < 0.24

*GS: German shepherds; LA: Labrador retrievers; BM: Belgian Malinois; PO: Pointers. Numbers in parentheses indicate the number of bitches.

(Table 3). The mean litter size was largest in BM (n=6.70) and lowest in GS (n = 5.33). The number of puppies per whelping decreased as the parity advanced (P < 0.002), especially after the 2nd whelping (inflection point: 2.4).

There was no difference in the proestrus length among the breeds (8.74 ± 0.37 days; Table 4). However, the cycle interval (P < 0.01) and the estrus length (P < 0.001) differed by breed (Table 4). The longest and the shortest cycle intervals were observed in PO (237.0 days) and GS (208.2 days), respectively. The estrus length was the longest in GS (9.54 days) and shortest in BM (7.98 days). The estrus season did not affect the cycle interval, proestrus length, and estrus length (Table 4). As the parity advanced, the cycle interval shortened by 16%, whereas the proestrus and estrus lengths remained unchanged (Table 4). Furthermore, the pregnancy duration became 0.24 days shorter as the number of puppies born in each whelping increased (P < 0.008) (Figure 3). However, precaution is needed, because the duration of pregnancy was counted from the last day of mating, which could be slightly erroneous as compared with LH surge determination.

The seasons varied not only by the environmental temperature, but also by the photoperiod length (Table 1). However, none of the variables was affected by the daylight duration when the statistical model included the daylight duration instead of the season. The lack of season and daylight duration effects could be related to the simultaneous changes in temperature and daylight duration within the category of "season" and "daylight".

The reproductive parameters were autocorrelated (Table 5). Parity was negatively correlated with the cycle interval (r = –0.18), proestrus length (r = –0.09), and litter size (r = –0.60). As the cycle interval became longer, the litter size increased (r = 0.21). The proestrus length was

Table 3. The effects of breed and parity on pregnancy duration and litter size.

	Variable	
	Pregnancy (days)	Litter size (n)
Breed		
GS	63.72 ± 0.33^{ab}	5.33 ± 0.23^{b}
LA	63.52 ± 0.44^{bc}	6.38 ± 0.30^{ab}
BM	62.24 ± 0.55^{c}	6.70 ± 0.37^{a}
PO	65.19 ± 0.83^{a}	5.47 ± 0.56^{b}
Parity		
1 (n = 48)	62.80 ± 0.51	7.03 ± 0.34
2 (n = 40)	64.00 ± 0.57	6.96 ± 0.38
3 (n = 36)	64.65 ± 0.59	5.84 ± 0.39
4 (n = 35)	63.58 ± 0.59	5.87 ± 0.40
5 (n = 29)	63.34 ± 0.65	5.93 ± 0.44
6 (n = 23)	64.08 ± 0.72	4.48 ± 0.49
7 (n = 14)	63.81 ± 0.93	6.26 ± 0.63
8 (n = 12)	64.93 ± 1.00	5.79 ± 0.67
9 (n = 9)	63.19 ± 1.15	6.00 ± 0.78
10 (n = 7)	62.28 ± 1.29	5.52 ± 0.87
Effect	--------------------------- P <	---------------------------
Breed	0.02	0.001
Parity	0.33	0.002
Breed × Parity	0.82	0.23

GS: German shepherds; LA: Labrador retrievers; BM: Belgian Malinois, PO: Pointers.
^{a,b,c} Superscripts represent statistical significance.

inversely correlated with the pregnancy duration (r = −0.24). As the number of puppies per whelping increased, the pregnancy duration (r = −0.17) was shortened.

4. Discussion

The season influences reproduction in many species. High ambient temperatures can cause infertility (10) by compromising the ovarian functions (14), estrus and ovulation (15), embryonic implantation, survival, and sperm production (16) in pigs. Similarly adverse consequences are known in cows (17). The adverse effects of heat stress on bitch reproduction have not been documented sufficiently.

Although bitches are considered nonseasonal or monoestrous breeders, showing estrus, mating, and whelping throughout the year (9), there are several aspects of the seasonal effect on estrus in bitches. The season affects the estrous cycle (8) and the estrus incidences (1,7) in bitches. For instance, the highest number of estrous cycles were reported in November in Kenya (7), between February and May in the United Kingdom, and between November and March in Sweden. These were also attributed to variations of daylight or photoperiod, as shown in tropical and polar regions (8,10). Studies reporting a lack of a seasonal effect on estrus in bitches (3), especially those living in temperate climates (4), are also available. Ortega-Pacheco et al. (11) reported that estrous cycles could be observed in each season of the year. This might explain the lack of a seasonal effect on reproductive variables in the present study (Table 1), which was

Table 4. The effects of breed, parity, proestrus season, and estrus onset (EO) season on the cycle interval, proestrus, and estrus duration.*

	Variable		
	Cycle interval (days)	Proestrus duration (days)	Estrus duration (days)
Breed			
GS	208.21 ± 4.08[b]	8.84 ± 0.26[ab]	9.54 ± 0.25[a]
LA	215.14 ± 4.68[b]	8.86 ± 0.29[ab]	8.53 ± 0.34[ab]
BM	208.58 ± 6.20[b]	8.03 ± 0.39[b]	7.98 ± 0.41[b]
PO	237.00 ± 8.58[a]	9.21 ± 0.55[a]	8.22 ± 0.54[b]
Proestrus/EO season			
Spring	225.09 ± 5.26[a]	8.52 ± 0.33	8.62 ± 0.32
Summer	211.46 ± 5.42[b]	8.64 ± 0.34	8.31 ± 0.34
Fall	213.01 ± 6.04[ab]	9.09 ± 0.38	8.89 ± 0.42
Winter	219.37 ± 5.48[ab]	8.69 ± 0.35	8.46 ± 0.38
Parity			
1	241.12 ± 6.17	9.67 ± 0.39	9.07 ± 0.39
2	225.38 ± 6.31	8.78 ± 0.40	8.75 ± 0.41
3	221.02 ± 6.68	8.48 ± 0.42	8.65 ± 0.41
4	216.25 ± 7.07	9.22 ± 0.45	8.89 ± 0.44
5	219.65 ± 7.65	8.55 ± 0.48	8.61 ± 0.49
6	216.19 ± 8.55	8.35 ± 0.54	8.04 ± 0.57
7	208.81 ± 9.57	8.63 ± 0.61	8.25 ± 0.69
8	208.19 ± 10.84	9.48 ± 0.68	7.17 ± 0.76
9	213.71 ± 13.19	7.67 ± 0.83	8.80 ± 0.87
10	202.01 ± 14.44	8.51 ± 0.94	9.47 ± 0.93
Effect	----------------------------------	P <	-------------------------------
Breed	0.01	0.20	0.001
Proestrus/EO season	0.18	0.64	0.67
Parity	0.04	0.29	0.54

*There were no 2- or 3-way interactions of main effects. GS: German shepherds; LA: Labrador retrievers; BM: Belgian Malinois; PO: Pointers.

[a,b,c] Superscripts represent statistical significance.

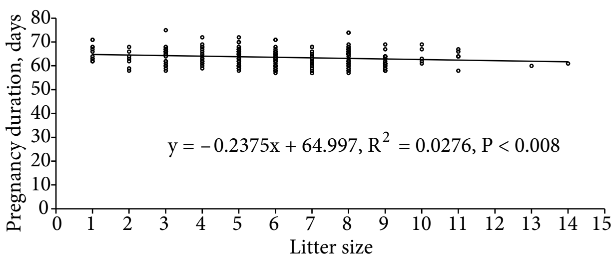

Figure 3. The relation between pregnancy duration and litter size.

Table 5. Correlations among reproductive parameters in all dogs.

	(1)	(2)	(3)	(4)	(5)	(6)
Parity (1) (n = 465)	1	–0.18**	–0.09*	–0.07	0.003	–0.20**
Cycle interval (2) (n = 465)		1	–0.08	0.05	0.03	0.21**
Proestrus duration (3) (n = 462)			1	0.02	–0.24**	0.008
Estrus duration (4) (n = 343)				1	0.06	–0.04
Pregnancy duration (5) (n = 253)					1	–0.17**
Litter size (6) (n = 253)						1

**P < 0.01; *0.01 < P < 0.05.

performed in a temperate climate area. There was no effect of daylight duration in the present study when the model included daylight duration, even though seasonal. The daylight duration is different from that reported in Sweden (min = 6 h vs. max = 18 h) (8).

Studies from different regions of the world reported various pregnancy rates, ranging from 64% to 95% (7,10). It was reported to be 64.4%, 85.0%, and 78.6% in a tropical region (10), in England (18), and in Sweden (8), respectively. In the current study, the pregnancy rate was 71.21% after mating (Table 2). This result was higher than the study performed in the tropical region (10). Although not well documented, these differences could partially result from heat stress. Moreover, the dog breeds in these countries also differ, suggesting that the effect may not totally be attributed to seasonal variations in the literature.

The cycle interval and estrus length were varied among breeds. The effect of breed on the cycle interval was also reported in previous articles (10,19). In agreement with previous studies (19), GS had the shortest cycle interval in the current study. The age was another factor that affected the cycle interval; as the age advanced, the cycle interval shortened by 16% in the current study. Conversely, previous studies reported a positive correlation between age and cycle interval (7,8,20,21).

The pregnancy duration in the bitches was about 65 days after the LH peak, about 57 days after cytological diestrus, and about 62 days after the last mating (8). Thus, breed differences for pregnancy duration in the present study (Table 3) could be artifactual, because differences between breeds were less than 3 days. In addition to parity and litter size, the breed was reported to affect pregnancy duration (22–24). It was reported that Golden retrievers have a longer pregnancy duration than other breeds such as large hounds, GS, and LA (6). Okkens et al. (5,22) reported that West Highland white terriers had a longer pregnancy duration than GS, LA, and Dobermans. In agreement with the literature, the pregnancy duration varied among breeds in this study too. The pregnancy duration was the shortest in BM (62.24 days) and longest in PO (65.19 days).

As the litter size per whelping increased, the pregnancy duration shortened (Table 3). Furthermore, each puppy was associated with a 0.24 days decrease in pregnancy duration. The negative correlation between litter size and pregnancy duration is known from previous work (6,8,22). Eilts et al. (2005) reported that bitches that whelped up to 5 puppies had a 1-day shorter pregnancy duration than bitches that whelped up to 4 puppies. A shorter pregnancy and a larger litter could be the result of a better planned mating time in relation to ovulation. Conversely, some authors stated that the number of puppies did not affect the pregnancy duration (23,24). Moreover, in the present study, because the LH surge was not determined, the actual mating time was not known (8), suggesting that the actual mating time appears to influence the duration of pregnancy, and hence the litter size (Table 3).

The litter size depends on age as reported previously (8,25,26). The parity did not influence pregnancy duration, but the litter size decreased with advanced age in this study, especially after parity 2.4. Borge et al. (26) suggested that productivity decreased after the second parity. Some authors declared a decrease in litter size after 5–6 years of age (8,20,26,27).

Bitches living in a temperate climate showed estrus throughout the year. Besides, the pregnancy duration, as assessed by the last mating, could vary by breed. The pregnancy duration shortened as the number of puppies increased, especially after the second parity. Moreover, the number of puppies affected pregnancy duration. In conclusion, the reproductive parameters are affected by breed and age characteristics, rather than by seasonal variations under a temperate climate.

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