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Factors affecting reproductive efficiency in German Shepherd bitches producing litters for Police of the Czech Republic

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ABSTRACT: The present study on German Shepherd bitches bred in the same environment of a private kennel and producing offspring for working purposes aimed to ascertain if any of the parameters: litter size, number of live puppies, stillbirth and mortality of puppies is determined by the month of whelping, parity, number of whelpings for one bitch over the course of one year, pregnancy length or bitch age at the time of whelping. Reproductive records of 73 bitches of German Shepherd breed (GSB) documented since 2001 till 2013 were processed. A total of 298 whelpings resulted in 2075 puppies; mortality of puppies was evaluated within the first 3 weeks of life. A significant seasonal influence on litter size and number of live born puppies, but not on stillbirth or mortality, was found. The parity of GSB significantly influenced litter size, live born puppies, and the incidence of stillbirths. The number of whelpings in one bitch over the course of one year significantly influenced litter size, while puppy losses were the same when bitches were bred once or twice a year. Litter size and number of live born puppies had a significant negative effect on pregnancy length. Furthermore, the number of stillbirths significantly increased when pregnancy was prolonged. Bitch age did not influence litter size, number of live born puppies and stillbirths, however it significantly affected mortality.

Keywords: dog; reproduction; whelping; litter size; puppy losses

INTRODUCTION

The German Shepherd (GS) is one of the dog breeds most frequently utilized for working purposes (Ozcan et al. 2009; Parr and Otto 2013). Working dogs are indispensable part of military and police organs in many countries and are still more needed e.g. due to increasing terroristic attacks (detection, guard, and tracking dogs) and natural disasters (rescue dogs). In the Police forces of the Czech Republic, GS bitches producing suitable offspring for subsequent working purposes are bred intensively (i.e. as much as possible upon maintaining health and welfare conditions). To meet the increased demands for recruitment of new dogs, successful and highly effective working dog breeding is essential.

Successful dog breeding could be defined as the number of puppies produced from one pregnancy, i.e. litter size (Borge et al. 2011). To evaluate reproductive efficiency and increase the effectivity

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of reproductive management of intensively bred bitches in military or police kennels, more information about "litter size" is necessary (e.g. live puppies born, stillbirths).

Factors influencing litter size are for example season of the year, number of previous litters (Gavrilovic et al. 2008), age of parents (Borge et al. 2011), and breed (Okkens et al. 2001; Borge et al. 2011). However, results published in various studies are inconsistent (Gavrilovic et al. 2008; Borge et al. 2011), possibly because they are based on reproductive efficiency data sets of bitches pertaining to various breeds (Bouchard et al. 1991; Okkens et al. 2001; Eilts et al. 2005; Chatdarong et al. 2007; Ortega-Pacheeo et al. 2007; Borge et al. 2011) and only very few studies deal with reproductive efficiency of only one dog breed, more specifically Drever (Gavrilovic et al. 2008), Irish hounds (Urfer 2009), German Shepherds (Mutembei et al. 2002), Beagles (Shimatsu et al. 2007), Boxers (Forsberg and Persson 2007), and English bulldog breed (Wydooghe et al. 2013). Moreover, results of some studies are only based on data from a questionnaire (Mutembei et al. 2002; Forsberg and Persson 2007; Wydooghe et al. 2013) and there is no information (or no rigorous information) about reproductive management of the breeders concerned. Other possible factors influencing the reproductive pattern of bitches are said to be different housing, nutritional conditions, and probably management (e.g. proper timing) of mating or insemination (Gavrilovic et al. 2008), i.e. specific environmental influence (Mutembei et al. 2002). Borge et al. (2011) mention that even though mean litter size is breed-specific, information on the expected number of puppies born for a particular breed is lacking and this knowledge is useful both for breeders and veterinarians.

The number of puppies surviving until weaning is greatly influenced by puppy losses (Indrebo et al. 2007), which are relatively high and breed-specific, and information on this phenomenon is still lacking for the majority of dog breeds (Tonnessen et al. 2012). Indrebo et al. (2007) studied prevalence rates and risk factors concerning stillbirth and mortality during the first three weeks of life in four large breeds. Gill (2001) described the incidence of neonatal mortality (the first six weeks after birth) for toy breeds. These studies used records from Norwegian Kennel Club (Tonnessen et al. 2012) and from a survey among breeders (Gill 2001). Information on the incidence of puppy losses in postpartum period in particular breeds would be highly valuable for breeders and especially veterinarians (Tonnessen et al. 2012) and might contribute to reducing the rate of stillbirths and deaths of puppies (Indrebo et al. 2007).

The present study on German Shepherd bitches producing offspring for working purposes and bred in the same environment aimed to determine, if any of the parameters: litter size, number of live puppies, stillbirth and mortality of puppies is determined by month of whelping, parity, number of whelpings for one bitch over the course of one year, pregnancy length or age of bitch at the time of whelping.

MATERIAL AND METHODS

Animals. Reproductive records for a period 2001–2013 were obtained for 73 bitches of German Shepherd breed (GSB) aged 2–8.5 years with a total of 300 whelpings and 2067 puppies. The parity of bitches used in this study ranged from 1 to 8. The data was collected from a breeding kennel belonging to Police of the Czech Republic (Domažlice). The bitches were individually housed in separate pens (2.2 × 3.5 ×2 m), fed twice daily (Royal Canin, Sensible; Royal Canin, Gard, France) with *ad libitum* access to water. All the bitches had the same daily exercise program.

Reproductive management and data. All the bitches were treated and subjected to natural mating by the same veterinarian for the whole period (2001–2013). Natural mating was scheduled according to vaginal cytology and blood serum progesterone (P4) levels. Generally, bitches were mated for the first time when vaginal cytology revealed >90% of cornified cells and P4 was >10–15 ng/ml in two consecutive blood samplings. Despite of the general pattern of mating described above, dynamics of the vaginal cytology and P4 levels were monitored for each bitch individually. It means that bitches were mated in various time intervals from the first blood collection. Levels of P4 were estimated by the chemiluminescence immunoassay (Immulite 2000 Immunoassay System; Siemens, Erlangen, Germany). Only proven sires (litters after natural mating were registered in the kennel book under the sire) were used. Pregnancy was diagnosed by ultrasonography 30 days after the last mating.

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The reproductive data for each bitch included: name, date of birth, date of whelping, length of gestation, age of bitch at whelping, parity and number of whelpings in one bitch during one year, litter size, number of live puppies born, and number of stillbirths and mortality of puppies.

Definitions. The litter size is defined as the number of all puppies born (including born live and dead). Live puppies born means the number of puppies alive at the time of whelping. Losses of puppies incorporate stillbirths and mortality and these are defined as the percentage proportion of puppies reported as dead during parturition or at birth and dead within three weeks after parturition, respectively. Losses of puppies from whelping until three weeks of age were calculated at litter level as a proportion of litters which contains stillbirths and dead puppies (Dohoo 2009).

When the effect of month of whelping was calculated, the litters were grouped by month of birth (1 = January, 2 = February, etc.). Parity means the number of whelpings for each bitch during her life. The number of whelpings for one bitch during one particular year is one or two (this parameter served for comparison between groups of bitches that whelped twice or once within one year). Gestation length is calculated as the number of days from the last mating to whelping. Age of bitch represents the period between the birth of a particular bitch and whelping. Percentages of stillbirths and mortality were calculated on the litter level by dividing the number of stillbirths and puppies that died within three weeks of life by litter size, respectively.

Statistical analyses. Fixed effects influencing all traits were analyzed by a mixed linear model with repeated measurements using MIXED procedure of the SAS software (Statistical Analysis System, Version 9.1, 2006). In the model, random (co)variances were summarized by residual **R** matrix, which was assumed to be a block diagonal with identical submatrices, each corresponding to an individual bitch. Statistical models for the traits analyzed (litter size, number of live puppies born, stillbirths and mortality of the puppies, or pregnancy length) were as follows:

Model 1:

$$y_{1ijklmn} = \mu + Month_i + AgeB_j + Parity_k + NumberW_l + pe_n + e_{ijklmn}$$

Model 2:

$$y_{2ijklmn} = \mu + Month_i + AgeB_j + Parity_k + NumberW_l + b_1NumberP + pe_n + e_{ijklmn}$$

where:

$y_{1ijklmn}$ = traits analyzed (litter size, number of live puppies born, stillbirths and mortality of the puppies)
$y_{2ijklmn}$ = traits analyzed (pregnancy length)
μ = overall mean
$Month_i$ = month of whelping (i = 1, …, 12)
$AgeB_j$ = age of bitch at whelping (j = 1.5, …, 8.5)
$Parity_k$ = parity (k = 1, …, 8)
$NumberW_l$ = number of whelpings in one bitch during one year (l = 1, 2)
$b_1NumberP$ = linear regression on (a) litter size or (b) number of live puppies born
pe_n = permanent effect of bitch (n = 1, …, 73)
e_{ijklmn} = residual error

For the analysis of the trait pregnancy length, two variants of Model 2 were used with the effect (*NumberP*) being (a) litter size, (b) number of live puppies born.

RESULTS AND DISCUSSION

Reproductive data on bitches bred in the kennel producing working dogs of GSB for Police of the Czech Republic are given in Table 1.

Month of whelping. Litter size in the German Shepherd (GS) bitches retrospectively evaluated in our study was significantly influenced by the month of whelping ($P < 0.001$) (Table 2). The number of puppies delivered varied during the year and the smallest litter was documented in April (4.5 ± 0.6 puppies) and the largest in November (8.8 ± 0.6 puppies) (Figure 1). Similarly, the month of year influenced the number of live puppies born ($P < 0.001$) (Table 2). The most viable litters were

Table 1. Descriptive statistics for reproductive data for German Shepherd bitches bred in the evaluated police kennel during 2001–2013

Police kennel data	Mean ± SD (range)
Number of whelpings	300
Total number of puppies born	2067
Parity	3.5 ± 2.3
Age of the bitch at whelping (years)	4.6 ± 1.8
Pregnancy length (days)	60.4 ± 3.2 (53–79)
Number of puppies per litter	6.9 ± 3.1 (1–14)
Number of live born puppies per litter	6.1 ± 3.1 (0–14)
Stillbirth per litter	0.8 ± 1.1 (15%) (0–6)
Mortality per litter	0.8 ± 1.2 (12%) (0–6)

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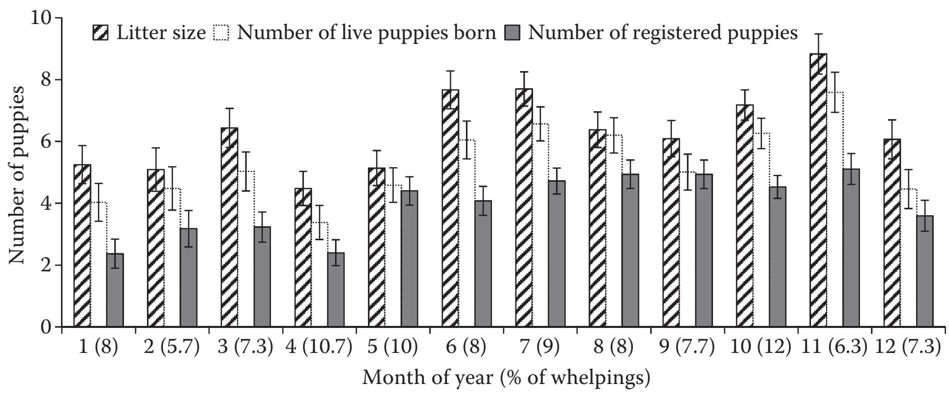

Figure 1. Seasonal influence on litter size and live puppies born in German Shepherd bitches in the evaluated police kennel

born in November (7.6 ± 0.7 live puppies) and the least viable in April (3.4 ± 0.6 live puppies) (Figure 1). The percentage of stillbirth (15.9 ± 4.6%) and mortality (15.8 ± 4.5%) was not influenced by the month of year (Table 2).

A seasonal (4 seasons) effect was found in the study by Gavrilovic et al. (2008), who reported the largest litters in a private kennel (Drever breed) in spring months, which is characteristic for the majority of breeds in Sweden. Hare and Leighton (2006) also reported a significant influence of the season (GBS), contrarily Mutembei et al. (2002) did not note any variations during the year in GSB bred in Africa. The reason for such conflicting results (non-seasonality) could be the effect of the comparison of reproductive data enrolling several breeds instead of evaluating sufficient records from each breed separately (Eilts et al. 2005). Another reason could be different housing conditions of animals which are kept as a colony. In bitches kept as family pets Lindeforsberg and Wallen (1992) observed evenly distributed heat periods, while those in bitches kept as a colony and housed outdoors without heating or supplementary light differed over the year (Lindeforsberg and Wallen 1992; Elmaz et al. 2008). The results of our study support this conclusion and it seems that natural conditions in pens represented by normal day length (no supplementary sources of light) and temperature (no supplementary heating) provoke seasonal variations in litter size in GSB kept as a colony in the Czech Republic.

There is a lack of studies investigating the influence of the month of year on live born puppies for one breed, but the study of Chatdarong et al. (2007) pointed to a seasonal influence on the number of live born puppies in the tropics in four large breeds. In our study, the results of seasonal influence on live puppies born indicate that breeding GS in the second half of the year can result in higher numbers of live puppies for the conditions of Central Europe, particularly the Czech Republic.

It is known that mortality of puppies is breed-specific (Tonnessen et al. 2012), highly variable (Sturgess 1998; Lawler 2008), and can be quite high (16%) in large breeds (Indrebo et al. 2007). In a two-year retrospective study by Tonnessen et al. (2012), no seasonal influence on perinatal mortality was found, which is in line with our results. Conversely, a seven-year Australian study reports on the seasonal influence (Gill 2001). The inconsistency of results might be explained by different altitude, however this is rather a hypothetical conclusion and the reason for different mortality in various parts of the world remains to be elucidated. Our study is the first to describe the influence of the month of year on mortality of puppies, specifically in GS bred in a colony, and our results indicate that puppy losses in the

Table 2. Statistical significance of evaluated effects (individual values stand for *P*-value)

Trait	Effect				
	month of whelping	parity	number of whelpings per year	pregnancy length	age of bitch
Litter size	0.0001	0.0410	0.0323	< 0.0001	0.0985
Live puppies born	0.0006	0.0054	0.0530	< 0.0001	0.2975
Stillbirth	0.0967	0.0009	0.7717	0.0007	0.4014
Mortality	0.1066	0.2578	0.3591	0.1015	0.0121

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Table 3. Relationship of parity and litter size, number of live puppies born, stillbirths, and mortality in German Shepherd bitches (mean ± Least Squares Means)

Parity	Bitches belonging to parity (%)	Litter size	Number of live puppies born	Stillbirth (%)	Mortality (%)
1st	24.3	5.2 ± 0.5[a]	4.9 ± 0.5[a]	15.8 ± 4.2[a]	16.2 ± 4.1
2nd	19.3	6.5 ± 0.5[b]	6.4 ± 0.5[bd]	14.2 ± 3.2[a]	14.1 ± 3.9
3rd	14.3.	7.1 ± 0.6[c]	6.2 ± 0.5[bc]	8.6 ± 4.0[ac]	15.2 ± 4.3
4th	11.0	6.4 ± 0.6[d]	6.3 ± 0.5[bd]	2.6 ± 4.3[bc]	13.0 ± 4.5
5th	9.7	7.0 ± 0.6[e]	6.5 ± 0.6[b]	6.2 ± 4.6[abc]	18.4 ± 4.6
6th	8.0	5.2 ± 0.7	4.7 ± 0.7[ad]	17.9 ± 5.2[a]	28.1 ± 5.9
7th	6.7	6.1 ± 0.8	4.8 ± 0.8[ad]	39.1 ± 7.0[d]	11.2 ± 6.8
8th	6.7	5.6 ± 0.8	4.4 ± 0.8[acd]	21.7 ± 6.3[a]	10.4 ± 7.0

[a–e]different superscripts in a column denote significant differences ($P < 0.05$)

kennel recorded for more than 12 years are within the same range throughout the year.

Parity. In our study, the parity (Table 2) of GSB significantly affected litter size, number of live puppies born and stillbirth (Table 3). Mortality of puppies was not influenced by parity of the bitch ($P > 0.05$).

Reproductive efficiency of a bitch is influenced by many factors (Tonnessen et al. 2012). A combination of these (Mandigers et al. 1994) stand behind the final numbers of puppies registered in kennel clubs, and parity of the bitch can be classified among the major factors (Borge et al. 2011). Studies evaluating the effect of parity on litter size in different breeds revealed a significant influence of parity when only parity (not parity and breed) was incorporated to a statistical model (Borge et al. 2011; Polat et al. 2015). In Drever bitches kept as a colony in a private kennel, the litter size at the time of birth increased slightly from the 1st to the 3rd parity (Gavrilovic et al. 2008). Mutembei et al. (2002) found litter size in GSD bitches was affected only by the 5th and higher parities. In the private kennel evaluated litter size was the largest in bitches belonging to the 2nd–5th parity, thus the significant influence of parity on litter size found in our study corresponds to the above mentioned studies. The highest number of live puppies was born in the period between the 2nd and 5th parity, which agrees with the largest litters at these parities. Surprisingly, there is a lack of studies investigating the influence of parity on the number of live puppies born in dogs, although knowledge about this relationship could help the breeders, especially those producing working dogs, decide about how many times they should mate the bitches for whelping the highest numbers of live puppies.

From the literature it is known that risk of stillbirth increases with breed size and can be as high as 21.6% in large breeds (Tonnessen et al. 2012). The same study documented that the rate of stillbirths declined with increasing parity (up to the 6th) and then sharply increased during the 7th parity. This trend is also traceable in our study, moreover litters at the 3rd–5th parity had the lowest incidence of stillbirth. Also Gill (2001) found parity of bitch was a significant predictor of stillborn puppies. Losses due to stillbirth were constant up to the 4th parity, after that the percentage of stillbirths increased with the increasing parity (Gill 2001). According to our results concerning the parity, it seems reasonable to anticipate the largest litters, highest numbers of puppies born, and lowest incidence of stillbirths from bitches bred on the 3rd to 5th parity. Breeding GS at 6th or higher parities should be carefully and individually considered.

Number of whelpings for one bitch over the course of one year. The number of whelpings for one bitch over the course of one year significantly influenced the litter size (Table 2). When the bitches whelped once ($n = 233$) or twice ($n = 67$) a year, the breeding resulted in 6.1 ± 0.3 or 5.3 ± 0.4 puppies in litter, respectively ($P = 0.03$). The numbers of live puppies born ($P = 0.05$) and losses of puppies ($P > 0.05$) were not dependent on the frequency of breeding of one particular bitch during one year (Table 2).

The long period of anestrus allows most bitches to breed maximally twice a year (Okkens and Kooistra 2006) and the number of breedings is also

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regulated by the Federation Cynologique Internationale (www.fci.be) and/or by local federations. According to the above-mentioned specifications, GS may be bred twice a year as maximum. Nevertheless, this is not a common breeding practice and it is recommended to breed only during alternate oestrous cycles (von Heimendahl and England 2010). In our study, we found larger litters when the bitches whelped once than twice a year. However, this difference represented approximately only one puppy. A similar trend was also apparent in more live puppies born when the bitches whelped only once compared to twice a year (difference of 1.1 ± 0.5 live puppies, $P = 0.05$). Based on our results we may state that even though there was a difference in litter size, it was by only one puppy, and therefore responsibility how frequently the bitches will be bred lies on individual breeders. Interestingly, we did not find disturbed survival of puppies when bitches were bred once or twice a year.

Pregnancy length. The mean pregnancy length in our study was 60.4 ± 3.2 days. We found a significant negative effect of the litter size (b = –0.24) and number of live puppies born (b = –0.24) on pregnancy length. When the litter size and number of live puppies born increased by one puppy, pregnancy length shortened by 0.24 ± 0.06 and 0.24 ± 0.06 days, respectively ($P < 0.0001$). In addition to all evaluated traits, the regression of pregnancy length on stillbirth was calculated by adding this effect to the model we used, and a significant negative effect of pregnancy length on the incidence of stillbirth (b = 1.4) was found. When the pregnancy was prolonged by one day, the stillbirth incidence was by 1.5 ± 0.5% higher ($P < 0.0007$).

The results of our study are in line with Eilts et al. (2005), Mir et al. (2011), and Polat et al. (2015) who found a negative relationship between litter size and pregnancy length in large breeds as well. Our study documented for the first time the negative effect of gestation length on the number of live puppies born. Prolonged pregnancies often lead to parturition problems and can cause foetal death (Irons et al. 1997; Gill 2001). Some studies detected a negative influence of prolonged parturition on stillbirth (Indrebo et al. 2007), however the so far published information did not concern the GSB. We found a higher frequency of stillbirths in prolonged pregnancies, and based on our novel results it is advisable to manage the timing (e.g. induction) of whelping in GSB if prolonged duration is observed.

Age of bitch at whelping. In this study, bitch age did not influence the litter size, number of live puppies born and stillbirth (Table 2). When the relationship between puppy losses and bitch age was evaluated, a significant effect ($P = 0.01$) on the mortality of puppies was found (Table 4).

Table 4. Effect of bitch age on litter size, number of live puppies born, and incidence of stillbirth and mortality in puppies of German Shepherd bitches (Least Squares Means ± standard errors of the means)

Age of bitch at whelping (years)	Bitches involved in age group (%)	Litter size	Number of live puppies born	Stillbirth (%)	Mortality (%)
2	2.7	6.5 ± 1.1	6.1 ± 1.1	9.4 ± 8.2	4.1 ± 8.0$^{a–c}$
2.5	9.7	8.9 ± 0.7	7.1 ± 0.7	18.1± 5.7	17.3 ± 5.8$^{a–c}$
3	7.7	6.7 ± 0.7	6.2 ± 0.7	13.2 ± 5.0	6.1 ± 5.1$^{a–c}$
3.5	11.0	7.1 ± 0.6	6.0 ± 0.6	21.3 ± 4.1	11.4 ± 4.3$^{a–c}$
4	12.7	7.0 ± 0.6	5.8 ± 0.6	18.6 ± 4.8	10.6 ± 4.7$^{a–c}$
4.5	8.7	7.0 ± 0.6	6.0 ± 0.6	15.6 ± 4.9	15.6 ± 4.6$^{a–c}$
5	9.7	7.4 ± 0.6	5.8 ± 0.6	20.2 ± 4.2	15.9 ± 4.1$^{a–c}$
5.5	8.0	6.1 ± 0.6	5.9 ± 0.6	15.2 ± 4.7	7.1 ± 4.9^{b}
6	6.3	6.3 ± 0.7	6.1 ± 0.7	13.3 ± 5.1	5.4 ± 5.9^{b}
6.5	6.0	6.8 ±0.7	5.8 ± 0.7	16.5 ± 5.7	17.9 ± 5.0$^{a–c}$
7	5.7	6.2 ± 0.8	7.0 ± 0.8	4.7 ± 6.2	19.4 ± 6.2$^{a–c}$
7.5	4.7	5.9 ± 0.8	5.0 ± 0.8	6.0 ± 6.6	24.1 ± 6.6^{c}
8	4.0	4.4 ± 0.9	3.3 ± 0.9	28.3 ± 7.8	45.6 ± 7.6^{d}
8.5	3.3	3.9 ± 1.0	3.5 ± 1.0	20.9 ± 8.1	19.5 ± 8.2$^{a–c}$

$^{a–d}$different superscripts in a column denote significant differences ($P < 0.05$)

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The influence of bitch age on litter size is frequently discussed in literature, although what this relationship really means is not clear. When many different breeds were evaluated to test for a relationship between age and litter size using unconditional analysis, no relationship was found, while multivariable analyses showed a curvilinear relationship and a significant interaction between age and breed size (Borge et al. 2011). Similarly to our results, in a private kennel, where only the Drever breed was kept, no influence of age on litter size and registered puppies was found (Gavrilovic et al. 2008). Mandigers et al. (1994) found a slightly negative influence of age on litter size in a small breed (Dutch Koiker dog).

The relationship between the age of bitch and mortality has not been studied yet in a closed population of one breed. When data from different breeders and breeds were evaluated (Gill 2001), a low bitch age was associated with low mortality, and this trend was also found in our study covering only GSB (Table 4). Similarly to our study, Gill (2001) reported mortality significantly increasing with age, and our results are also in line with a study evaluating the relationship between age and perinatal losses of 224 breeds (Tonnessen et al. 2012).

Based on our results we may state that in kennels with good reproductive and health management GS can be intensively bred between 2–6 years of bitch age. In the case of breeding older GSB, the breeders are strongly advised to take into account bitches' health and reproductive status, and slightly smaller litters can be expected.

CONCLUSION

In intensively bred GSB producing puppies for working purposes the litter size was influenced by the month of year in which the puppies were born, however there were no losses of puppies. The largest litters with the lowest puppy losses were produced by bitches at the age of 2–6 years and on the 3rd–5th parity. The litter size in bitches whelping once a year was by only one puppy larger if compared to those whelping twice a year; puppy losses were similar. We found a significant negative effect of the litter size and number of live puppies born on pregnancy length, moreover prolonged pregnancy significantly increased stillbirths.

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