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- 2 The role of birth weight on litter size and mortality
- 3 within24 h of lifein purebred dogs: What aspects are
- 4 involved?

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Abstract

In humans, scientific evidence emphasizes the role of birth weight on 16 17 neonatal welfare, morbidity and mortality. In canine species, defining 18 normal ranges of birth weight is a harder issue due to a great morphological 19 variability in size, body weight and breed. The aim of this study was to 20 correlate birth weight with litter size and mortality within24 h of life 21 in 789 pups from 140 litters of purebred dogs and to investigate the aspects 22 that might affect these factors. Birth weight was influenced by maternal 23 size, weight and age (P < 0.001). The lightest pups were from toy sized or 24 weighing up to 10 kg bitches. Conversely, bitches aged 2-8years whelped 25 heavier pups than younger and older mothers. Birth weight was also related 26 both to litter size, with heavier pups in smaller rather than in larger litters from medium sized bitches, and breed (P < 0.05). Unexpectedly, 27 28 birth weight did not differ between liveborn and stillborn pups. However, birth weight was lower in pups dying within 24 h of life (P < 0.05). High 29 30 mortality of pups was related both to short pregnancies (P < 0.05), also showing lighter litters (P < 0.001), and to dystocic parturitions (P <31 32 0.001). Litter size was associated with parity, type and number of mating, 33 and length of pregnancy (P < 0.001). Low birth weight appears to predispose 34 to early neonatal mortality suggesting a predominant role of the breed 35 rather than size and weight in determining birth weight in pups.

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Keywords

38 Birth weight, Litter size, Neonatal mortality, Purebred dog.

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Introduction

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born).

42 Physical characteristics, which consist of large surface area-to-volume ratio, little body fat, poor vasomotor control, and inability to shiver, 43 44 make neonates extremely susceptible to hypothermia with underweight infant 45 seven more prone to its potential complications (Manani et al., 2013). 46 Moreover, since babies are born with limited capacities for gluconeogenesis and glycogen stores, they are at great risk for developing hypoglycaemia 47 48 (Maromet al., 2010; Fawcett, 2014). In human perinatal clinics and 49 research, low birth weight is regarded as a reflection of the immaturity 50 of the organism and used as a predictor of mortality and morbidity risk 51 (Melve and Skjaerven, 2003). Similar to humans, it can be assumed that 52 underweight newborn pups are more exposed to hypothermia than littermates on account of their reduced adipose tissue as well as to hypoglycaemia due 53 54 to sibling competition for maternal resources (nipples and milk) with important implications for neonatal survival. Stillbirth and neonatal 55 mortality are quite common in canine species and involve many factors 56 57 including the quality of labour, congenital and acquired disorders, and 58 neonatal environment (Davidson, 2003). While some factors are controllable, the easily evaluation of birthweight could optimize neonatal 59 60 attendance focusing it on needy underweight pups. 61 Fetal development and the resulting birth weight is also considered a 62 predisposing factor for elective Caesarean-section in canine reproduction 63 as the size of the pups in case of singleton pregnancy may cause dystocia (Smith, 2007; Lopate, 2008). Various factors can influence body weight at 64 65 birth: environmental, nutritional and genetic components, besides fetal 66 uterine position in polytocous species (Bautista et al., 2015). In Boxer, 67 also the genetic role in birth weight was reported (Nielen et al., 2001). Due to different birth weight distributions in different ethnic groups, a 68 population-specific standard for birthweight has been proposed in human 69 70 medicine (Wilcox and Russell, 1990). Domestic dog (Canis familiaris) shows the greatest morphological variability of any mammal (Sutteret al., 2008). 71 72 In fact, adult canine size and body weight may vary from 15 to 120 cm 73 height at the withers and from 500 g to 120 kg, respectively, depending on 74 the breed (Fiszdon and Kowalczyk, 2009). This heterogeneity makes it 75 challenging to determine the physiological ranges of birthweight in this 76 species. We firstly investigated which maternal feature (size, weight, age 77 and breed) can affect birth weight in pure-bred dogs. Then, birth weight 78 was correlated to litter size and neonatal mortality within 24 h of life. 79 Moreover, some reproductive aspects that might influence birth weight, 80 litter size and early neonatal mortality were explored (parity, type and number of mating, length of pregnancy, type of parturition, sex of new-81

2. Materials and methods

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2.1. Clinical records This study is based on data collected by a 84 questionnaire administered to breeders/owners from January 2012to September 85 2014. One hundred and forty litters from 119 bitches belonging to 31 86 87 different breeds were enrolled (Table 1). Bodyweight, age and the 88 reproductive history of bitches were carefully recorded. The height at the 89 withers for each breed was taken from Federation Cynologique Internationale (F.C.I.), Ente Nazionale Cinofilia Italiana (E.N.C.I.) and breed clubs. 90 91 The length of pregnancy was calculated both from the estimated LH surge by 92 blood progesterone measurement, when available (Concannon et al., 1977; 93 Kutzler et al., 2003; Michel et al., 2011), and from the day of 94 mating/artificial insemination (AI) with fresh semen when hormonal 95 monitoring of the reproductive cycle was not performed. In the latter case, 96 the first day of pregnancy was considered the same of the mating/AI when 97 single, or the first mating/AI when repeated within 24 h interval, or the 98 intermediate day when multiple mating/AIs were performed in an interval of 99 at least 48 h. Type of parturition (vaginal eutocic, vaginal dystocic, 100 elective and emergency C-section) was also recorded. Body weight and sex of the pups were noted at birth. Mortality of pups (regardless of the 101 cause) was verified at birth and after 24 h. 102

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2.2. Statistical analysis 104 Data were analyzed using a commercial statistical pro-gramme (SPSSTM 22.0 105 for Windows, IBM). Dogs were stratified in groups according to maternal 106 size, i.e. height at the withers (toy: <20 cm; small: 20-40 cm; medium: 107 40-65 cm; large: >65 cm) (Bonetti, 1995); maternal weight(≤10 kg; 10-20 108 kg; 20-30 kg; 30-40 kg; >40 kg); age (≤2years; 2-4 years; 4-8 years; >8 years); litter size (small:≤3 pups; medium: 4-8 pups; large: ≥9 pups); 109 birth weight (≤200 g; 200-400 g; 400-600 g; >600 g); parity (primiparous; 110 multiparous); type of mating (natural; AI); numbers of mating/AI (1; 2-3; 111 112 >3); length of pregnancy (\leq 60days; 61-63 days; 64-66 days; >67 days) and 113 type of parturition (vaginal eutocic; vaginal dystocic; elective C-section; 114 emergency C-section). Breeds represented by at least eleven litters 115 (Bernese Mountain dog, Chihuahua, German Shepherd dog, Labrador retriever) 116 were considered a significant sample as determined using the internet-117 based sample size calculator and statistically analyzed. These breeds were 118 compared against birth weight and litter size using a generalized estimating equation (GEE) where the dependent variables had an inverse 119 120 Gaussian distribution, and Log link function was used. Goodness of fit was 121 assessed using a quasi likelihood under independence model criterion (QIC). Birth weight, litter size and mortality of pups (at birth and within 24 h 122 123 of life) were related to clinical variables (maternal size, weight and age,

breed, parity, type and number of mating/AI, length of pregnancy, type of

- 125 parturition, sex) and to each other by GEE, Kruskal-Wallis and Mann-Whitney
- 126 U tests. Values of P < 0.05 and P < 0.001 were considered statistically
- 127 significant and highly significant, respectively.
- 128 3. Results
- 129 Distribution of the bitches according to their size, weight, age, breed
- and litter size is shown in Tables 1 and 2.
- 131 3.1. Birth weight
- 132 Weight of pups at birth was between 55 and 900 g. The lightest pup, born
- 133 alive but died within 24 h of life, was from a litter of seven French
- 134 bulldog pups weighing 121.8 \pm 37 g. The heaviest pups were one from a
- 135 litter of four New-foundland pups and one from eight German Shepherd pups
- weighing 765 \pm 106.6 g and 674.3 \pm 163.9 g, respectively. Neither of these
- 137 two litters recorded neonatal mortality. Birth weight was related to
- maternal size, weight and age (P < 0.001; Table 2). Toy sized bitches, or
- up to 10 kg in weight, whelped pups lighter than 200 g, and medium-large
- sized bitches, or more than 20 kg in weight, whelped pups heavier than 400
- q. Bitches aged from 2 to 8 years delivered heavier pups than younger and
- older mothers. Birth weight differed significantly among the four breeds
- 143 considered (German Shepherd with P < 0.05; Bernese Mountain dog, Chihuahua
- and Labrador retriever with P < 0.001; Table 2). Birth weight was related
- to litter size with heavier pups in smaller rather than in larger litters
- from medium sized bitches (P < 0.05; Table 2). The same occurred according
- 147 to maternal weight (P < 0.05) and in Bernese Mountain dog (P < 0.001) and
- 148 Labrador retriever (P < 0.05; Table 2), but not in bitches weighing 30-40
- 149 kg (Table 2).
- 150 3.2. Litter size
- 151 A total of 789 pups were delivered (Table 2) with litters ranging from 1
- to 13 pups. The lowest prolific breed was Chihuahua with an average of 3.1
- 153 ± 0.8 pups per litter, while the largest litter was from Rhodesian ridgeback
- with an average of 10.8 \pm 2.9 pups per litter. Litter size was related to
- 155 maternal size, weight and age (P < 0.001). Toy and small sized bitches,
- 156 weighing up to 20 kg or older than 8 years, never whelped more than 8 pups
- 157 (Table 2). Litter size was different among the four breeds considered (P <
- 158 0.05), except between Bernese Mountain and German Shepherd dog.
- 3.3. Mortality within 24 h of life
- 160 Mortality of pups registered at birth and within 24 h of life was 3.5% and
- 161 5.8%, respectively (Table 3) and was not related to maternal size, weight,
- age and breed. Birthweight did not affect mortality at birth, while living
- pups at birth were heavier than pups dying within 24 h of life(P < 0.05;
- 164 Table 3). Mortality of pups was not influenced by litter size, except in
- the German Shepherd breed with larger litters showing a lower mortality
- than smaller ones (P < 0.05).

167 3.4. Reproductive aspects

Natural mating was performed in 97 bitches whose reproductive cycle was 168 169 not monitored (n = 53) or monitored (n = 44). Artificial insemination with 170 fresh semen was performed in the remaining 43 bitches, without (n = 22) or 171 with reproductive monitoring (n = 21). Number of mating/AI per bitch is shown 172 Table 4. Litter size was smaller in bitches artificially 173 inseminated rather than naturally mated (P < 0.001; Table 4). Litter size was also smaller in bitches mated/inseminated more than three times, while 174 no differences were found in number of pups born by single mating/AI or 175 176 repeated 2-3 times (P < 0.001; Table 4). Birth weight, adjusted for litter 177 size, was statistically related to the length of pregnancy calculated both from mating/AI (n = 75) and from estimated LH surge (n = 65; P < 0.001). 178 Birth weight was higher in pregnancies lasting at least 64 days than in 179 180 shorter pregnancies (Table 5). Litter size did not affect the length of 181 pregnancy calculated from mating/AI, while it was inversely proportional when calculated from LH surge (P < 0.001). The average length of small, 182 medium and large litter pregnancies was $66.2\pm~1$, $65.8\pm~0.9$ and $64.9\pm~0.8$ 183 184 days from LH surge, respectively. Litter size was influenced by parity (P 185 < 0.001). Multiparous bitches aged 4-8 years whelped smaller litters(6.6 \pm 186 2.5 pups) than primiparous peer bitches (10.1±3.8 pups). Parity influenced 187 also the length of pregnancy, calculated both by mating/AI (P < 0.05) and 188 LH surge (P < 0.001), with the longest pregnancies in multiparous bitches. 189 Mortality of pups, excluding pups born by elective C-section, was higher 190 in shorter pregnancies (60.4±1.1 and 64.8±1 days, calculated from mating/AI 191 and LH surge, respectively) compared to longer ones (61.3± 1.5 and 65.4±1 days, calculated from mating/AI and LH surge, respectively) (P < 0.05). 192 193 Type of parturition was related to neonatal mortality, which was high in pups born by vaginal dystocic delivery (P < 0.001; Table 6), but no 194 195 difference in birth weight was recorded between eutocic and dystocic pups. 196 The sex of pups did not affect their birth weight: the average weight was 411.1 ± 170.2 g and 410.2 ± 184.4 g in 407 male and 382 female pups, 197 198 respectively.

4. Discussion

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To date, despite the growing interest in breeding and selecting purebred dogs, no studies are available on the effect of birth weight affecting litter size and neonatal mortality. Although long since accepted in canine reproductive practice, some aspects have not been scientifically verified yet. Our results showed that maternal size, weight and age as well as breed, even if proved in four breeds only could strongly impact on birth weight. Interestingly, a significant breed-dependent difference in birth weight among breeds of the same size and weight (German Shepherd dog and Labrador retriever) was recorded. These data suggest a predominant role of

the breed rather than size and weight in determining birth weight likely based on standardized morphological phenotypes defining distinct dog breeds. Birth weight is an important survival determinant inmost mammalian species (Gatel et al., 2011). Low birthweight is accompanied by immature development and adaptive postnatal failure that can predispose to neonatal mortality (Tonnessen et al., 2006). Unexpectedly, in this study, birth weight was not related to stillbirth; however, it was significantly lower in pups dying within 24 h than in living pups (P < 0.05). It is reported a greater predisposition of purebred pups to congenital defects leading to neonatal mortality (Rickard, 2011). Contribution of stillborn pupssuffering from malformations involving an increase rather than a decreasein birth weight (i.e. anasarca) can justify our findings. We speculated that low birth weight should be considered as prognostic of survival within 24 h from birth, but not as discriminating between live born and stillborn pups. Intrauterine growth retardation, a condition affecting polytocous species, including dogs, is characterized by low weighing and distressed pups compared to littermates (Wootton et al., 1983). Assistance attempts should be focused on these underweight live born pups (2.3% in our study) in order to reduce the percentage of perinatal mortality. In previous studies, neonatal mortality was associated with maternal age, litter size and breed (Tonnessen et al., 2012). We did not record any significant relation even comparing neonatal mortality within 24 h of life to maternal size and weight and to sex of the pups. A different constitution of the canine study population may explain this divergence. Dystocia is among the principal causes of neonatal mortality in dogs, mainly due to uterine inertia and fetal malpresentation (Tonnessen et al., 2012). We recorded the highest percentage of neonatal mortality (33%) in cases of dystocic parturition (P < 0.001). Although elective C-section is recommended in singleton pregnancies as the size of the pups may predispose to dystocia (Smith, 2007), in our sample birth weight did not differ between dystocic and eutocic pups. It should be noted that neonatal mortality in German Shepherd dog was higher in smaller litters than in larger ones (P < 0.05). Moreover, the birth weight in German Shepherd dog was remarkably high in relation to the breed size and weight. We can assume that breed-specific characteristic such as mesocephalic mesomorphous conformation makes German Shepherd dog able to whelp oversized pups by eutocic parturition but resulting in distressed pups and higher neonatal mortality as in the case of small litters. Mortality of pups born both by emergency C-section and eutocic parturition was lower than previously reported (Moon et al., 1998). However, our data refer to an observation period limited to 24 h after birth. Moreover, advances in breeding management and, mainly, in protocol

and anesthetic techniques could be responsible for this improvement in

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neonatal survival. Similar to humans (Reddy et al., 2011), neonatal mortality was higher in shorter pregnancies (about one day less) than in longer ones (P < 0.05), even when both occurred at physiological time. These data are consistent with a lower birth weight recorded in pregnancies shorter than 64 days when compared to longer ones in medium sized bitches (P < 0.001). Likewise in cats (Gatel et al., 2011), birth weight of pups was higher in smaller than in larger litters although it was not statistically significant in all categories of size, weight and breed. Since no data on the Body Condition Score of the dogs included in this study are available, we cannot exclude that the nutritional status ofpatients may affect the birthweight of pups as described in humans (Vasudevan et al., 2011). Litter size is reported inversely proportional tomaternal age (Borge et al., 2011; Mir et al., 2011), with the highest prolificacy reached between 2 and 4 years of age (Munnich and Kuchenmeister, 2009). In our sample, litter size was significantly lower in dogs over 8 years than in younger bitches (P < 0.001). Moreover, bitches aged 2-8yearswhelped heavier pups than younger and older mothers (P < 0.001). These aspects are in agreement with fertility reduction due to immature development and to degenerative age-related processes affecting the reproductive organs (Gavrilovic et al., 2008), and support the recent guidelines for breeder ethical code from an Italian Kennel Club (E.N.C.I.) that strongly advise against breeding too young (before the second heat) or too old dogs (over 7 years). In contrast with previous reports (Borge et al., 2011), parity significantly affected litter size, with primiparousbitches (2-8-year-old) being more prolific than multiparous peer bitches (P < 0.001). Ethically discouraged, reproductive exploitation is not evenjustified in terms of efficiency. Litter size was significantly correlated also to the number of mating/AI and showed lower in bitches that underwentmore than 3 mating/AI (P < 0.001), while there were no differences in prolificacy between single mating/AI or repeated 2-3 times. Therefore, once the period of optimal fertility is accurately identified, avoiding repeatedmating/AI seems to be more rational and convenient, even in terms of economic management. In agreement with literature (Borge et al., 2011), naturally mated bitches had larger litters than those undergoing AI (P < 0.001). This result may depend on different mating management by differentoperators and on the quality of semen from stud dogs. In conclusion, maternal size, weight and age as well as breed features can

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In conclusion, maternal size, weight and age as well as breed features can affect birth weight in dogs. The huge heterogeneity of breed morphometry and the lack of a standard reference for birth weight in purebred dogs make it challenging to determine the physiological ranges of birth weight in this species. Increased knowledge about the impact of birth weight on litter size and mortality of pups will make the identification of pups in

- 293 need of any breeder and/or veterinarian intervention easier as well as
- 294 their attendance to support birth and neonatal survival better.

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- 296 Conflict of interest
- 297 We disclose all possible conflict of interest from all the authors.

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 Table 1

 Distribution of the bitches based on their breed and size and the corresponding number of litters and pups.

Breed	Size	Number of bitch	Number of litters	Number of pups
Appenzeller Mountain dog	Medium	2	2	11
Beagle	Small	1	1	7
Bernese Mountain dog	Medium	14	16	88
Border collie	Medium	3	4	30
Boston terrier	Small	3	5	10
Bullmastiff	Large	1	1	10
Cane Corso	Medium	1	1	6
Cavalier King Charles spaniel	Medium	1	1	8
Chihuahua	Toy	9	12	34
Chinese crested dog	Small	1	2	5
Dobermann	Large	1	1	9
Drahthaar	Medium	1	1	9
English bulldog	Small	1	1	6
Epagneul Breton	Medium	3	4	26
French bulldog	Small	5	5	30
German shepherd	Medium	16	18	114
Golden retriever	Medium	2	3	25
Great Dane	Large	1	1	11
Hovawart	Medium	3	7	59
Jack russel terrier	Small	3	3	14
Labrador retriever	Medium	19	21	117
Little Lion dog	Small	1	1	4
Miniature dachshund	Toy	2	2	11
Newfoundland	Large	3	3	13
Pug	Small	5	7	28
Rhodesian ridgeback	Large	4	4	40
Rottweiler	Medium	1	1	4
Segugio dell'Appennino	Medium	1	1	6
Staffordshire bull terrier	Small	8	8	42
Yorkshire terrier	Toy	1	1	1
Zwergpinscher	Small	2	2	11
Total		119	140	789

Bold indicates a breed showing significant numbers of litters/pups.

Table 2Birth weight and number of pups based on maternal size, weight, age, breed and litter size.

	bBW (gr)	Litter size						Number of pups		Number of bitches
		≤3		4-8		≥9		100		
		n	bBW (g)	n	bBW (g)	n	bBW (g)	n	mean (±SD)	
Maternal size										
Toy	147.1 ± 30	23	153.1 ± 37.8	23	141 ± 17.3	nd	nd	46	3.6 ± 1.3	15
Small	244 ± 144.3	30	247.2 ± 143.5	135	243.5 ± 145	nd	nd	165	5.4 ± 1.7	36
Medium	471.8 ± 134.9	34	544.4 ± 155.9	273	450.6 ± 143.2	188	489.5 ± 109.6	495	7.5 ± 2.5	79
Large	523.32 ± 139	1	760	27	547 ± 157	55	507.4 ± 126.1	83	9.7 ± 2.5	10
Maternal BW (kg)										
≤10	161.9 ± 43.4	50	180 ± 50	99	152.8 ± 36.3	nd	nd	149	4.7 ± 1.9	40
10-20	399.1 ± 127.7	3	673.3 ± 20.5	106	326.7 ± 122.7	9	382.2 ± 34.9	118	6.5 ± 1.2	19
20-30	409.1 ± 84.6	12	458.7 ± 132.5	130	398.6 ± 58.4	67	420.6 ± 110.3	209	7.1 ± 2.3	34
30-40	571 ± 120.6	7	506.4 ± 205.7	65	599.5 ± 137.4	80	553.2 ± 88.5	151	8 ± 2.6	22
≥40	544.2 ± 118.3	16	638.7 ± 103.7	58	574.8 ± 121.4	88	506.9 ± 103.5	162	8.7 ± 3	25
Maternal age (years)										
≤2	327.2 ± 182.2	16	248.2 ± 207.9	109	260.8 ± 144.2	40	539.8 ± 64.3	165	6.9 ± 2.4	28
2-4	449.9 ± 165.5	51	341.4 ± 211.2	217	440.8 ± 171.4	169	494.4 ± 119.8	437	7.2 ± 2.7	76
4-8	408 ± 171.7	17	377.5 ± 195.5	106	404.4 ± 185.3	34	434.7 ± 101.8	157	6.9 ± 3	30
>8	311.9 ± 143.4	4	602.5 ± 235.1	26	267.2 ± 40.4	nd	nd	30	6 ± 1.8	6
Total		88	343.3 ± 216.8	458	379.7 ± 181.1	243	493.5 ± 133.5	789		140
Breed										
Bernese Mountain dog	558.3 ± 86.9	15	630.7 ± 102	31	599 ± 73	42	502.4 ± 46.7	88	7.8 ± 3.2	14
Chihuahua	147.3 ± 32.8	22	151.9 ± 38.3	12	138.9 ± 17.5	nd	nd	34	3.1 ± 0.8	9
German Shepherd	539.3 ± 149	9	522.8 ± 220	55	577.3 ± 163.5	50	500.6 ± 103.3	114	7.8 ± 2.7	16
Labrador retriever	397.2 ± 57.2	4	462.5 ± 75	95	405.1 ± 47.2	18	340.9 ± 65.8	117	6.1 ± 1.6	19
Total		50		193		110		353		58

^{&#}x27;n' means number of pups; 'bBW' means weight of pups at birth; 'nd' means not detected

Table 3
Neonatal and perinatal mortality based on maternal size, weight, age and breed.

	Live born pups		Stillborn pups		Alive p	ups within 24 h of life	Pups dying within 24 h of life	
	n	bBW (g)	n	bBW (g)	n	bBW (g)	n	bBW (g)
Maternal size	151501	1-12-18-18-18-18-18-18-18-18-18-18-18-18-18-	100	-2009 A	re-Norma	to describe and control to the	C 200 D	
Toy	45	146.8 ± 29.9	1	160	45	146.8 ± 29.9	nd	nd
Small	160	243.2 ± 144.6	5	275 ± 146.7	156	246.9 ± 144.5	4	98.2 ± 40
Medium	476	471.6 ± 136	19	475.6 ± 105.4	462	475.2 ± 135.4	14	352.8 ± 100.6
Large	80	523.4 ± 137.3	3	469 ± 207.1	80	523.4 ± 137.3	nd	nd
Maternal body weight (kg)								
<10	145	161.4 ± 41.6	4	180 ± 96.3	141	163.2 ± 40.4	4	98.2 ± 40
10-20	116	337.9 ± 128.5	2	407.5 ± 17.7	112	341.4 ± 129.4	4	240.7 ± 21.8
20-30	199	407.7 ± 85.6	10	436.3 ± 58.3	194	409.2 ± 85.4	5	371.2 ± 79.8
30-40	148	573.7 ± 117.9	3	433.3 ± 195.5	144	577.7 ± 116.9	4	432.5 ± 53.8
≥40	153	545 ± 118.1	9	531.2 ± 127.6	152	545.4 ± 118.4	1	490
Maternal age (years)								
≤2	159	326.6 ± 182	6	356.3 ± 203.3	155	330.5 ± 181.5	4	153 ± 108.6
2-4	422	449.2 ± 166.2	15	469.4 ± 149.5	413	451.1 ± 166.6	9	363.1 ± 127
4-8	150	408.4 ± 175.2	7	400 ± 67.8	149	407.8 ± 175.6	1	490
>8	30	311.9 ± 143.4	nd	nd	26	322.9 ± 151.2	4	240.7 ± 21.8
Total	761	410 ± 178	28	427.8 ± 150.2	743	412.8 ± 178	18	296.3 ± 141
Breed								
Bernese Mountain dog	82	558 ± 88.1	6	562.3 ± 73.6	81	588.9 ± 88.4	1	490
Chihuahua	33	146.9 ± 33.2	1	160	33	146.9 ± 33.2	nd	nd
German Sheperd	108	542.8 ± 151.1	6	476.7 ± 93.3	104	548 ± 151	4	407.5 ± 72.3
Labrador retriever	112	396.4 ± 56.6	5	414 ± 75.7	112	396.4 ± 56.6	nd	nd
Total	335		18		23		5	

^{&#}x27;n' means number of pups; 'bBW' means weight of pups at birth; 'nd' means not detected.

Table 4
Number of bitches and pups based on the type and number of mating.

	Type of mating		Number of mating/AI			
	Natural mating	AI	1	2-3	>3	
Number of bitches	97	43	61	73	6	
Number of pups	$566(7.4\pm2.9)$	$223(6.2 \pm 2.1)$	$353(6.8 \pm 2.3)$	$415(7.4\pm3)$	$21(4.2\pm2)$	

Table 5Birth weight and length of pregnancy according to litter size.

Litter size												
Pregnancy length (days)	≤3			2.0	4–8				≥ 9			
	Mating/AI		LH surge		Mating/Al		LH surge		Mating/AI		LH surge	
	n	bBW(g)	n	bBW (g)	n	bBW(g)	n	bBW(g)	n	bBW(g)	n	bBW(g)
≤60	23	300.9 ± 197.5	nd	nd	153	333.2 ± 178.3	nd	nd	73	503.8 ± 79.1	nd	nd
61-63	56	335.6 ± 206.9	1	160	286	395.5 ± 176.1	7	121.8 ± 37.4	161	480.4 ± 121.5	9	511.1 ± 48.6
64-66	9	499.2 ± 276.4	34	404.4 ± 209	19	515.3 ± 177.7	125	428.7 ± 204.9	nd	nd	132	483.7 ± 101.5
≥67	nd	nd	12	520.6 ± 246.2	nd	nd	39	552.9 ± 204.4	9	644.4 ± 91.7	nd	nd

^{&#}x27;n' means number of pups; 'bBW' means weight of pups at birth; 'nd' means not detected.

Table 6Mortality of pups at birth and within 24 h of life based on the type of parturition.

Type of parturition	n	Mortality at birth		Mortality within 24	h
		Live born pups (%)	Stillborn pups (%)	Alive pups (%)	Dead pups (%)
Vaginal eutocic	614	598 (97.4)	16(2.6)	585 (95.3)	29(4.7)
Vaginal dystocic	9	6(66.7)	3(33.3)	6(66.7)	3(33.3)
Elective C-section	99	97 (98)	2(2)	92(92.9)	7(7.1)
Emergency C-section	67	60(89.6)	7 (10.4)	60(89.6)	7(10.4)
Total	789	761 (96.4)	28(3.5)	743 (94.2)	46(5.8)

Numbers indicate the number of pups; numbers in parentheses indicate percentages.