1 **Title: Assessment of serotonin in serum, plasma and platelets of aggressive dogs**

2 Running title: Blood serotonin in aggressive dogs

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21 **Abstract**

22 Canine aggression is the most common reason for the referral of dogs to behavior practices. In

23 addition, dog bites represent an important problem for public health and animal welfare. The

24 serotonergic system is believed to play an important role in modulating aggression. The aim of

25 the present study was (1) to assess the suitability of different types of blood samples for

26 measuring circulating serotonin in canine clinical studies, and (2) to investigate the relationship

27 between the serotonergic system and canine aggression. The assessment of serotonin was

28 simultaneously carried out in serum, plasma and platelets of 28 aggressive and 10 non-

29 aggressive dogs with an enzymoimmunoassay technique. The mean serotonin concentration in

30 aggressive dogs was significantly lower than in non-aggressive dogs in all the assayed samples.

31 These findings suggest an inverse relationship between the activity of the serotonergic system

32 and canine aggression. Considering the simplicity of the methodology, the authors propose

33 sampling serum as the most suitable method for measuring circulating serotonin in dogs.

34 **Keywords:** serotonin, serum, plasma, platelet, dog, aggression

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Introduction

Canine aggression is the most common reason for the referral of dogs to animal behaviorists (Bamberger and Houpt, 2006; Fatjó et al., 2007). Dog bites represent an important problem for public health (Overall and Love, 2001; Palacio et al., 2005). They also pose a problem in the field of animal welfare since a significant proportion of aggressive dogs is euthanized or abandoned (Hunthausen, 1997; Mikkelsen and Lund, 2000). If these problems are to be prevented and effectively managed, it is necessary to deepen in the understanding of the biological basis of canine aggression.

Serotonergic neurotransmission is believed to play a critical role in the control of aggression in several species (Ferrari et al., 2005). Low concentrations of the main serotonin (5-HT) metabolite 5-hydroxyindoleacetic acid (5-HIAA) in cerebrospinal fluid (CSF) have been associated with low thresholds for aggression or impulsivity in human and non-human primates (Mehlman et al., 1994; Stanley et al., 2000; Soderstrom et al., 2001; Howell et al., 2007) as well as in dogs (Reisner et al., 1996).

Technical and ethical restrictions linked to the study of the serotonergic system in psychiatric disorders trough CSF and neurohistochemical studies have led scientists to search for more easily accessible biological samples. The platelet where blood 5-HT is mainly stored (Kema et al., 2000), has been proposed as a peripheral marker of the serotonergic neuron (Plein and Berk, 2001). Albeit blood 5-HT does not cross the hematoencephalic barrier, a correlation between blood and CSF serotonergic parameters have been found in humans (Sarrias et al., 1990). In the realm of human aggression, the measurement of blood 5-HT has offered inconclusive results since both low (Goveas et al., 2004) and high (Golubchik et al., 2009) concentrations have been found to be linked to aggressive behavior in different populations. In canine species, two recent studies have reported low serum serotonin concentrations in aggressive individuals (Çakiroglu et al., 2007; Rosado et al., 2010a).

The aim of the present study was two-fold. First, to assess the suitability of different types of blood samples for measuring circulating 5-HT in clinical studies in canine species. Second, to further investigate the relationship between the serotonergic system and canine aggression. To

64 this end, the assessment of 5-HT was simultaneously carried out in serum, plasma and platelets

65 of aggressive and non-aggressive dogs.

66 **Materials and methods**

67 *Aggressive animals*

68 Two Spanish veterinary teaching hospitals (Universidad de Zaragoza and Cardenal Herrera-

69 CEU, Valencia) contributed to the collection of cases. Dogs included in the present study were

70 referred to the Companion Animal Behavior Services within the respective hospitals owing to

71 problems of aggression towards people and/or other dogs. Displaying any other type of

72 behavioral problem did not constitute an exclusion criterion. Animals showing an underlying

73 causative medical condition to the problem of aggression were excluded from the study.

74 Twenty males (17 entire and 3 castrated) and 8 females (7 entire and one spayed) were included

75 in the aggressive group. The mean age was 4.2 years old (ranging from 7 months to 13 years).

76 The group consisted of 17 dogs of different breeds (four English cocker spaniels, two German

77 shepherds, two Rottweilers, one Alaskan malamute, one American pit bull terrier, one English

78 bull terrier, one Catalan sheepdog, one Lhasa Apso, one Poodle, one Teckel, one West Highland

79 white terrier and one Yorkshire terrier) and 11 cross-breeds and mongrels.

80 Diagnosis of aggression was carried out by means of a detailed standard questionnaire on the

81 dogs' behavior and daily routine. Clinical classification of aggression was established in

82 accordance with three main diagnostic criteria: target, context and dog's communicative signals

83 (based on Fatjó et al., 2007). In order to detect any underlying causative or contributory medical

84 condition to the aggression problem, all dogs were screened through physical examination,

85 complete blood count, serum biochemistry and thyroid hormone measurement at the time of

86 admission.

87 According to the main diagnosis, two animals showed intraspecific aggression whereas the rest

88 (n=26, 93%) showed aggression towards humans, mainly directed to the family members during

89 conflict or competitive situations (69.2%). Several forms of aggression were present in 39.3%

90 of the animals.

Control animals

The control group was made up of 10 beagles (5 entire males, 3 entire females and 2 spayed females) owned by the Universidad Cardenal Herrera-CEU for research purposes. The mean age was 3.1 years old (ranging from 1 to 6 years). All these dogs were healthy and lacked any history of aggression towards people and/or other dogs. The care and use of animals followed the European guidelines (European Union Directive 86/609/EEC, 1986).

Sample collection and preparation

Blood drawn (3 ml) into EDTA tubes was centrifuged at room temperature for 12 min at 152xg to obtain platelet-rich plasma (PRP). The PRP-supernatant was then transferred to another tube for platelet pellet preparation (according to the protocol by IBL, 1998; see below). The EDTA tubes were then centrifuged at 4500xg for 10 min at 4ºC to obtain plasma. Blood drawn (3 ml) into anticoagulant-free tubes was centrifuged at 4500xg for 10 min at 4ºC to obtain serum. Aliquots of plasma and serum were frozen and stored at –80ºC until the day of the analysis.

Platelet count was carried out in PRP (20 µL) using a Neubauer-counting chamber. To obtain the platelet pellet, an aliquot of 200 µL of PRP was added to 800 µL of physiological saline and centrifuged at 4500xg for 10 min at 4ºC. The supernatant was discarded and 200 µL of double-distilled water was then added to the pellet. Aliquots were stored at –80ºC. The day of the analysis the samples were centrifuged at 10,000xg for 2 min at room temperature after thawing of the frozen aliquots.

Serotonin analysis

Serotonin was measured in duplicate with a commercial EIA technique (Serotonin-ELISA, DLD Diagnostika GmbH, Hamburg, Germany). The intra and interassay coefficients of variations were 3.9-5.4% and 6%, respectively. Concentrations of 5-HT were expressed in ng/mL (serum and plasma) and $ng/10^8$ platelets (platelets).

Statistical analysis

A unifactorial multivariate analysis of variance was carried out to assess the effect of the factor "aggression" on the concentrations of 5-HT in the three types of samples, namely serum, plasma and platelets. Correlations were analyzed using the Pearson test.

119 Calculations were carried out using the statistical program SPSS 14.0. for Windows (SPSS, Inc,

120 Chicago, USA). *P* <0.05 denoted statistical significance.

Results

122 The unifactorial multivariate analysis of variance showed a significant effect of the factor

123 "aggression" on 5-HT concentrations (P<0.01). Aggressive animals showed lower 5-HT

124 concentrations in serum, plasma and platelets than non-aggressive animals. The higher mean 5-

125 HT concentration was detected in serum samples. Serotonin results according to the type of

126 sample are summarized in Table 1.

127 Serotonin concentration in platelets was positively correlated with that in serum (r=0.465;

128 P<0.01) and plasma (r=0.468; P<0.01). In addition, 5-HT concentrations in serum and plasma

129 were positively correlated (r=0.489; P<0.01).

Discussion

131 In the present study, the assessment of 5-HT in serum, plasma and platelets was carried out both

132 in aggressive and non-aggressive dogs. Regardless of the type of sample, aggressive dogs

133 showed significantly lower concentrations of circulating 5-HT than non-aggressive dogs. This

134 finding agrees with previous studies that found an association between high aggression or

135 impulsivity and low concentrations of CSF 5-HIAA and circulating 5-HT in primates (CSF:

136 Melhman et al., 1994; Stanley et al., 2000; Soderstrom et al., 2001; Howell et al., 2007;

137 platelets: Goveas et al., 2004) as well as in dogs (CSF: Reisner et al., 1996; serum: Çakiroglu et

138 al., 2007; Rosado et al., 2010a).

139 Several mechanisms regulate the functioning of the 5-HT system in the brain: (i) 5-HT synthesis

140 from tryptophan and degradation, (ii) 5-HT re-uptake from synaptic cleft, and (iii) the density

141 and sensitivity of 5-HT receptors (Popova, 2006). An alteration in any of these mechanisms

142 may be the underlying reason for the reduced activity of the serotonergic system in aggression.

143 Regarding canine aggression, an elevated plasma concentration of tryptophan was detected in a

144 group of aggressive dogs in comparison to a non-aggressive group (Juhr et al., 2005). In

145 addition, the administration of tryptophan-supplemented low-protein diets has shown to be

146 effective in reducing dominance aggression in dogs (DeNapoli et al., 2000). Moreover, a recent

147 study by the authors found a higher platelet 5-HT uptake in aggressive dogs than in non-
148 aggressive ones (Rosado et al., 2010b). Finally, a series of brain-imaging studies showed an
149 increase in the uptake of the 5-HT2A radioligand in all cortical areas of impulsive-aggressive
150 dogs compared with normal dogs (Peremans et al., 2002, 2003 and 2005). All of these findings
151 point towards an altered function of the serotonergic system in canine aggression.

152 In the present study, the mean concentration of 5-HT in the assayed samples reflected the
153 normal distribution of blood 5-HT, with the higher concentration in serum, followed by platelets
154 and finally, plasma. Circulating plasma 5-HT is taken up by platelets, and is accumulated during
155 the platelet's life cycle in dense granules. Thus, under normal conditions, 5-HT is almost
156 entirely confined in platelets whereas its concentration in plasma and other blood cells would be
157 very low (Ortiz et al., 1988; Kema et al., 2000). Mück-Seler et al. (2002) have suggested that
158 serum 5-HT concentration might indirectly represent a sum of platelet and plasma 5-HT
159 concentration. In the present study, the sum of the mean (estimated) platelet and plasma 5-HT
160 concentrations was similar, although not exact, to the mean serum 5-HT concentrations in each
161 group of dogs.

162 Platelet and plasma 5-HT concentrations in the control group were similar to those reported by
163 several studies on canine species (platelets: Meyers et al., 1982; Aranda et al., 1994; plasma:
164 Haga et al., 1996). Likewise, serum 5-HT concentration was also similar to previously
165 published data from whole blood in dogs (Ferrara et al., 1987; LaRosa et al., 1989; Chen et al.,
166 1993). It is worth mentioning, however, the striking difference in serum 5-HT concentrations in
167 the present study as compared with those in the study by Çakiroglu et al. (2007), where
168 relatively lower concentrations were detected both in non-aggressive and aggressive dogs. This
169 difference may be explained in terms of methodological differences in measuring serum 5-HT
170 (e.g., the use of different ELISA-kits).

171 A statistically significant difference in 5-HT concentration between aggressive and non-
172 aggressive dogs was noted both in serum and platelets as well as in plasma samples. This
173 indicates that any of them may represent a valid blood sample for measuring circulating 5-HT in
174 order to detect differences between both groups. The positive correlation found among the three

175   types of samples may also support this suggestion. The choice of either one of these different

176   types of blood samples for clinical applications, however, deserves some comments. Plasma is

177   easy to obtain but as previously mentioned 5-HT concentrations in this pool are low, and

178   therefore, more sensitive techniques are required to accurately detect the amine (e.g., high

179   performance liquid chromatography). In addition, plasma may be potentially contaminated with

180   platelet 5-HT (Anderson et al., 1987). Lastly, platelet preparation is an arduous task as an

181   additional platelet count in necessary to express the concentration of 5-HT apart from blood

182   centrifugation to obtain PRP. Since serum 5-HT concentration is considered an indirect measure

183   of platelet 5-HT content, it could be suggested that serum is the easiest sample type to assess

184   circulating 5-HT in dogs. Nevertheless, the assessment of plasma 5-HT may be of great interest

185   also in the field of research on the serotonergic system in canine aggression. It is suggested that

186   plasma 5-HT is a rapid turnover pool, affected by short-term changes in the 5-HT physiology

187   (e.g., synthesis, inactivation, uptake, etc.) while platelet 5-HT is a reserve, slow turnover pool

188   influenced by sustained changes. Thus, the measurement of plasma or platelet (serum) 5-HT

189   concentrations would be useful to study different aspects of 5-HT physiology (Ortiz et al.,

190   1988).

191   One limitation to the present work is the use of an experimental cohort of beagles as control

192   subjects for a group of aggressive pet dogs belonging to different breeds. In spite of their regular

193   contact with humans, the rearing and environmental conditions in the beagles group clearly

194   differed from those dogs in the pet group, and this may have affected the activity of the 5-HT

195   system even early in life. In addition, it is possible that breed-related differences may occur in

196   the 5-HT system functioning. It is important to note however that the use of experimental

197   beagles as a control group might provide some advantages such as low individual variability

198   (high group homogeneity) and, therefore, high reproducibility of the experiment in other

199   laboratory settings. Another limitation of the study is that the two groups of study were not

200   matched for sex and reproductive status, with proportionally more entire males in the aggressive

201   than the control group, and there is evidence that testosterone (but also estradiol) interacts with

202   the 5-HT system to influence the likelihood of aggression, probably by modulating 5-HT

203   receptors (Nelson and Chiavegatto, 2001; Soma et al., 2008). In another published study with a

204   larger sample of dogs, however, we did not find a significant effect of sex on serum 5-HT levels

205   (Rosado et al., 2010a). Finally, it is important to note that the above metioned limitations would

206   have not affected the first aim of the present study, that is: to assess and compare the suitability

207   of different types of blood samples for measuring circulating 5-HT in dogs. Anyway, future

208   studies on this field should consider breed, sex (and reproductive status) as well as age-matched

209   groups in order to assess these possible influencing factors.

210   In conclusion, the present results may suggest an inverse relationship between canine aggression

211   and the activity of the serotonergic system. To the authors' best of knowledge, this is the first

212   study that simultaneously assesses the concentration of 5-HT in serum, plasma and platelets in

213   aggressive and non-aggressive dogs. The three assayed blood samples showed differences

214   between both groups, but considering the simplicity of the methodology, the authors propose

215   serum as the more suitable sample for measuring circulating 5-HT in clinical studies in dogs.

216   The determination of circulating 5-HT may have important applications for behavior medicine

217   in the future. For example, it could be used for deciding which aggressive dogs might benefit

218   from drug therapy and for monitoring the response to these therapies. In addition, it could be

219   used for research purposes with the aim of creating new tools for diagnosis and treatment of

220   canine aggression.

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225   **References**

226   Anderson, G.M., Feibel, F.C., Cohen, D.J., 1987. Determination of serotonin in whole blood,

227   platelet-rich plasma, platelet-poor plasma and plasma ultrafiltrate. Life Sci. 40, 1063-1070.

228 Aranda, E., Pizarro, M., Pereira, J., Mezzano, D., 1994. Accumulation of 5-hydroxytryptamine

229 by aging platelets: studies in a model of suppressed thrombopoiesis in dogs. Thrombosis and

230 Haemostasis 71, 488-492.

231 Bamberger, M., Houpt, K.A., 2006. Signalment factors, comorbidity, and trends in behavior

232 diagnoses in dogs: 1,644 cases (1991-2001). J Am. Vet. Med. Assoc. 229, 1591-1601.

233 Çakiroglu, D., Meral, Y., Sancak, A.A., Cifti, G., 2007. Relationship between the serum

234 concentrations of serotonin and lipids and aggression in dogs. Vet. Rec. 161, 59-61.

235 Chen, Y.K., Richter, H.M. 3rd, Go, V.L., Tyce, G.M., 1993. Free and conjugated catecholamines

236 and serotonin in canine thoracic duct lymph: effects of feeding. Am. J. Physiol. 265, 84-89.

237 DeNapoli, J.S., Dodman, N.H., Shuster, L., Rand, W.M., Gross, K.L., 2000. Effect of dietary

238 protein content and tryptophan supplementation on dominance aggression, territorial aggression,

239 and hyperactivity in dogs. J. Am. Vet. Med. Assoc. 217, 504-508.

240 European Union Directive 86/609/EEC (1986) Council Directive of 24 November 1986 on the

241 approximation of laws, regulations and administrative provisions of the Member States

242 regarding the protection of animals used for experimental and other scientific purposes. Official

243 Journal of the European Communities Serie L358, 132.

244 Fatjó, J., Amat, M., Mariotti, V., Ruiz de la Torre, J.L., Manteca, X., 2007. Analysis of 1040

245 cases of canine aggression in a referral practice in Spain. JVB:CAR 2, 158-165.

246 Ferrara, A., Zinner, M.J., Jaffe, B.M., 1987. Intraluminal release of serotonin, substance P, and

247 gastrin in the canine small intestines. Dig. Dis. Sci. 32, 289-294.

248 Ferrari, P.F., Palanza, P., Parmigiani, S., de Almeida, R.M.M., Miczek, K.A., 2005. Serotonin

249 and aggressive behavior in rodents and nonhuman primates: Predispositions and plasticity. Eur.

250 J. Pharmacol. 526, 259-273.

251 Golubchik, P., Mozes, T., Vered, Y.; Weizman, A., 2009. Platelet poor plasma serotonin level

252 in delinquent adolescents diagnosed with conduct disorder. Prog Neuropsychopharmacol

253 Biol Psychiatry 33, 1223-1225

254   Goveas, J.S., Csernansky, J.G., Coccaro, E.F., 2004. Platelet serotonin content correlates

255   inversely with life history of aggression in personality-disordered subjects. Psychiatry Res. 126,

256   23-32.

257   Haga, N., Mizumoto, A., Satoh, M., Mochiki, E., Mizusawa, F., Ohshima, K., et al., 1996. Role

258   of endogenous 5-hydroxytryptamine in the regulation of gastric contractions by motilin in dogs.

259   Am. J. Physiol. 270, G20-28.

260   Howell, S., Westergaard, G., Hoos, B., Chavanne, T.J., Shoaf, S.E., Clevelan, A., et al., 2007.

261   Serotonergic influences on life-history outcomes in free-ranging mal rhesus macaques. Am. J.

262   Primatol. 69, 851-865.

263   Hunthausen, W., 1997. Effects of aggressive behavior on canine welfare. J. Am. Vet. Med.

264   Assoc. 210, 1134-1136.

265   IBL GmbH (hamburg, Germany), 1998. Instructions for use Serotonin-ELISA (RE59121).

266   Juhr, N.C., Brand, U., Riedel, E., 2005. Plasma amino acid concentrations in aggressive dogs.

267   Berl. Munch Tierarztl. Wochenschr. 118, 95-100.

268   Kema, I.P., de Vries, E.G., Muskiet, F.A., 2000. Clinical chemistry of serotonin and

269   metabolites. J. Chromatogr. B. Biomed. Sci. Appl. 747, 33-48.

270   LaRosa, C.A., Sherlock, D., Kimura, K., Pimpl, W., Money, S.R., Jaffe, B.M., 1989. The role of

271   serotonin in the canine secretory response to cholera toxin in vivo. J. Pharmacol. Exp. Ther.

272   251, 71-76.

273   Mehlman, P.T., Higley, J.D., Fauscher, I., Lilly, A.A., Taub, D.M., Vickers, J., et al., 1994. Low

274   CSF 5-HIAA concentrations and severe aggression and impaired impulse control in nonhuman

275   primates. Am. J. Psychiatry 151, 1485-1491.

276   Meyers, K.M., Homsen, H., Seachord, C.L., 1982. Comparative study of platelet dense granule

277   constituents. Am. J. Physiol. 243, R454-461.

278   Mikkelsen, J., Lund, J.D., 2000. Euthanasia of dogs due to behavioral problems: an

279   epidemiological study of euthanasia of dogs in Denmark, with a special focus on problems of

280   aggression. Eur. J. Comp. Anim. Pract. 10, 143-150.

281   Muck-Seler, D., Pivac, N., Sagud, M., Jakovljevic, M., Mihaljevic-Peles, A., 2002. The effects

282   of paroxetine and tianeptine on peripheral biochemical markers in major depression. Prog.

283   Neuropsychopharmacol. Biol. Psychiatry 26, 1235-1243.

284   Nelson, R.J., Chiavegatto, S., 2001. Molecular basis of aggression. Trends Neurosci. 24, 731-

285   739.

286   Ortiz, J., Artigas, F., Gelpí, E., 1988. Serotonergic status in human blood. Life Sci. 43, 983-990.

287   Overall, K.L., Love, M., 2001. Dog bites to humans–demography, epidemiology, injury, and

288   risk. J. Am. Vet. Med. Assoc. 218, 1923-1934.

289   Palacio, J., León, M., García-Belenguer, S., 2005. Aspectos epidemiológicos de las mordeduras

290   caninas. Gac. Sanit. 19, 50-58.

291   Peremans, K., Audenaert, K., Coopman, F., Hoybergs, Y., Slegers, G., Van Bree, H., et al.,

292   2002. Functional brain imaging of serotonin-2A receptors in impulsive dogs: a pilot study.

293   Vlaams Diergeneeskundig Tijdschrift. 71, 340-347.

294   Peremans, K., Audenaert, K., Coopman, F., Blanckaert, P., Jacobs, F., Otte, A., et al., 2003.

295   Estimates of regional cerebral blood flow and 5-HT2A receptor density in impulsive, aggressive

296   dogs with 99mTc-ECD and 123I-5-I-R91150. Eur. J. Nucl. Med. Mol. Imaging. 30, 1538-1546.

297   Peremans, K., Audenaert, K., Hoybergs, Y., Otte, A., Goethals, I., Gielen, I., et al., 2005. The

298   effect of citalopram hydrobromide on 5-HT2A receptors in the impulsive-aggressive dog, as

299   measured with 123I-5-I-R91150 SPECT. Eur. J. Nucl. Med. Mol. Imaging. 32, 708-716.

300   Plein, H., Berk, M., 2001. The platelet as a peripheral marker in psychiatric illness. Hum.

301   Psychopharmacol. 16, 229-236.

302   Popova, N.K., 2006. From genes to aggressive behavior: the role of serotonergic system.

303   Bioessasys 28, 495-503.

304 Reisner, I.R., Mann, J.J. Stanley, M., Huang, Y.Y., Houpt, K.A., 1996. Comparison of

305 cerebrospinal fluid monoamine metabolite levels in dominant-aggressive and non-aggressive

306 dogs. Brain Res. 714, 57-64.

307 Rosado, B., García-Belenguer, S., León, M., Chacón, G., Villegas, A., Palacio, J., 2010a. Blood

308 concentrations of serotonin, cortisol and dehydroepiandrosterone in aggressive dogs. Appl.

309 Anim. Behav. Sci. 123, 124-130.

310 Rosado, B., García-Belenguer, S., Palacio, J., Chacón, G., Villegas, A., Alcalde, A.I., 2010b.

311 Serotonin transporter activity in platelets and canine aggression. Vet. J. 186, 104-105.

312 Sarrias, M.J., Cabré, P., Martínez, E., Artigas, F., 1990. Relationship between serotonergic

313 measures in blood and cerebrospinal fluid simultaneously obtained in humans. J. Neurochem.

314 54, 783-786.

315 Soderstrom, H., Blennow, K., Manhem, A., Forsman, A., 2001. CSF studies in violent

316 offenders. I. 5-HIAA as a negative and HVA as a positive predictor of psychopathy. J. Neural

317 Transm. 108, 869-878.

318 Soma, K.K., Scotti, M.A., Newman, A.E., Charlier, T.D., Demas, G.E., 2008. Novel

319 mechanisms for neuroendocrine regulation of aggression. Front. Neuroendocrinol. 29, 476-489.

320 Stanley, B., Molcho, A., Stanley, M., Winchel, R., Gameroff, M.J., Parsons, B., Mann, J.J.,

321 2000. Association of aggressive behavior with altered serotonergic function in patients who are

322 not suicidal. Am. J. Psychiatry 157, 609-614.

323

324   **Table 1**. Mean (standard error) concentrations of serotonin in serum, plasma and platelets of

325   aggressive (n=28) and control non-aggressive dogs (n=10).