Measurement of the Prothrombin Time, Activated partial thromboplastin time and Fibrinogen concentration in healthy dogs in Sudan

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ABSTRACT

The aims of this study was to determine the mean values of prothrombin time (PT), activated partial thromboplastin time(APTT), and fibrinogen concentration(FIB) for healthy police dogs (Labrador Retriever (20) and, German Shepherd (26) ) in Sudan ,and to evaluate the effects of the breed and sex on these parameters. 46 male (21) and female (25) dogs aged between (20-83 months) were enrolled in the study. Blood samples(2ml) were taken from the cephalic vein in a tube containing 3.2% trisodium citrate. The analysis was performed using semiautomatic BCA-2000-LED-light-coagulometer-with-Double-channel and Spinreac reagents kits for human medicine. The overall mean values were 7.35±0.91 second 15.12±1.32 second and 341.54±55.16 mg/dl for PT, APTT and fibrinogen concentration respectively. German Shepherd dogs showed significantly (P≤0.05) higher values than Labrador Retriever dogs for PT (7.58±1.01 vs 7.06±0.67) seconds respectively, and APTT (15.28±1.58 vs 14.9±0.85) seconds respectively. Males showed significantly (P≤0.05) higher values for FIB than the females (346.30±38.75 vs 337.73±65.99) mg/dl respectively. Effect of breed should be considered in clinical interpretation of dog PT and APTT, also effect of sex should be considered in clinical interpretation of dog FIB. The determined reference values may be useful information for an increasing clinical use.

Keywords: Prothrombin time (PT), Activated Partial Thromboplastine Time (APTT), Fibrinogen concentration (FIB), sex, breed.

INTRODUCTION:

Haemostasis is the mechanism to control blood loss (minor or major) arising from vascular or tissue trauma. Traditionally, the haemostatic system has been divided into 3 stages: primary haemostasis, secondary haemostasis and tertiary haemostasis, including fibrinolysis and natural anticoagulants (Hoffbrand, 2015). Secondary...
haemostasis can be assessed using PT, APTT, and FIB, which are specific tests, commonly used in laboratories to detect coagulation disorders in dogs. Disseminated intravascular coagulopathy (DIC) is one of those disorders which was detected using these specific tests (Vlasin et al., 2004, Machida et al., 2010, and Uhrikova et al., 2013). Liver diseases have been associated with abnormalities in haemostasis coagulation profile (PT, aPTT and FIB) (Prins et al., 2010).

The coagulation profile of healthy dogs was studied all over the world (Lopes et al., 2005, Ameri et al., 2011, Sumathi et al., 2012, Athanasiou 2013, Romao et al., 2013, Zapryanova 2013, Stockham 2013, and Walton 2014). To the best of our knowledge there are no published data on the coagulation profile of dogs in Sudan. So this work was undertaken to determine the normal values of PT, APTT and fibrinogen concentration of police dogs and to assess the effect of the breed and sex on these parameters under Sudan conditions.

MATERIALS and METHOD:

Study design and Animals: This was a prospective study conducted in 2017 in Khartoum State. The dogs belonging to the Police Directorate for Dogs at Khartoum State were 46 healthy police dogs (Labrador Retriever (20) and, German Shepherd (26), males (21) and females (25), their aged (20-83 months). They are individually housed in kennels and fed on imported dogs' food (Best Signor - Netherlands).

Blood collection and analysis: Blood samples (2ml) were withdrawn from the cephalic vein, while the animals were manually restrained, the blood was transferred into tube with anticoagulated (3.2% trisodium citrate). Each tube was gently inverted 3-5 times to guarantee mixing of the sample with the anticoagulant. The samples were immediately transported to the laboratory. The citrated plasma were separated by centrifuging citrated blood samples immediately for 15 min at 3000 RPM at room temperature. Using a semi automated-BCA-2000-LED-light-coagulometer-with-Double-channel (Biobase, China) and Spinreac reagents (Spinrea, Spain). The samples were analyzed for PT, APTT and FIB determination.

Statistical Analysis: The data of the studied parameters were presented as means ± standard deviation, range and median. The effects of the gender and breed were detected by student's t-test as described by Gomez and Gomez (1984). The statistical analysis of the data was performed using SPSS version 16 computer program.

RESULTS:

The mean values range and median were presented of PT, APTT and fibrinogen concentration for all the dogs, are displayed in Table (1). The effect of the breed is presented in Table (2). Prothrombin time (PT) and Activated partial thromboplastin time (APTT) were significantly higher (P≤0.05) in German Shepherd than in Labrador Retriever. Fibrinogen concentration (FIB) did not show any significant variation between the two breeds (P>0.05).

Table (1): APPT, PPT and fibrinogen values of police dogs.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Mean ± SD</th>
<th>Min-Max.</th>
<th>Median</th>
</tr>
</thead>
<tbody>
<tr>
<td>PT(second)</td>
<td>7.35±0.91</td>
<td>6-10</td>
<td>7.25</td>
</tr>
<tr>
<td>APTT(second)</td>
<td>15.12±1.32</td>
<td>13-18</td>
<td>14.80</td>
</tr>
<tr>
<td>FIB(mg/dl)</td>
<td>341.54±55.16</td>
<td>210-417</td>
<td>361</td>
</tr>
</tbody>
</table>
Table (2): The effect of the breed on some coagulation parameters of police dogs.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Labrador Retriever mean ± SD</th>
<th>German Shepherd mean ± SD</th>
<th>Overall mean ±SD</th>
<th>P. value</th>
</tr>
</thead>
<tbody>
<tr>
<td>PT (seconds)</td>
<td>7.06±0.67</td>
<td>7.58±1.01</td>
<td>7.35±0.91</td>
<td>0.019</td>
</tr>
<tr>
<td></td>
<td>6-8</td>
<td>6-10</td>
<td>6-10</td>
<td></td>
</tr>
<tr>
<td>APTT (seconds)</td>
<td>14.9±0.85</td>
<td>15.28±1.58</td>
<td>15.12±1.32</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>13-16</td>
<td>13-18</td>
<td>13-18</td>
<td></td>
</tr>
<tr>
<td>FIB (mg/dl)</td>
<td>344.41±50.63</td>
<td>339.44±59.15</td>
<td>341.54±55.16</td>
<td>0.764</td>
</tr>
<tr>
<td></td>
<td>254-417</td>
<td>210-416</td>
<td>210-417</td>
<td></td>
</tr>
</tbody>
</table>

Sex related differences in dogs coagulation profiles are presented in Table (3). Fibrinogen concentration (FIB) was significantly (P ≤ 0.05) higher in males than the females. Prothrombin time (PT) and Activated partial thromboplastin time (APTT) did not show any significant variation with the sex (P> 0.05).

Table (3): The effect of sex on some coagulation profile in police dogs.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Female mean ± SD</th>
<th>Male mean ± SD</th>
<th>Overall mean ±SD</th>
<th>P. value</th>
</tr>
</thead>
<tbody>
<tr>
<td>PT (second)</td>
<td>7.38±0.88</td>
<td>7.31±0.97</td>
<td>7.35±0.91</td>
<td>0.253</td>
</tr>
<tr>
<td></td>
<td>6-10</td>
<td>6-9</td>
<td>6-10</td>
<td></td>
</tr>
<tr>
<td>APTT (second)</td>
<td>15.02±1.27</td>
<td>15.23±1.39</td>
<td>15.12±1.32</td>
<td>0.736</td>
</tr>
<tr>
<td></td>
<td>13-18</td>
<td>13-18</td>
<td>13-18</td>
<td></td>
</tr>
<tr>
<td>FIB (mg/dl)</td>
<td>337.73±65.99</td>
<td>346.30±38.75</td>
<td>341.54±55.16</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>210-417</td>
<td>284-402</td>
<td>210-417</td>
<td></td>
</tr>
</tbody>
</table>

DISCUSSION:
The results obtained in this study showed higher values of PT and APTT in German Shepherd compared to values of Labrador Retriever (Table 2). However, sex did not show significant effect on PT and APTT value (Table 3). FIB result showed higher value in male compared to value of female (Table 3), however, breed did not show significant effect on FIB value (Table 2). There were lack of breed and sex effect on those indices in the previous reports on dogs (Vlasin et al., 2004, Uhrikova et al., 2013, Machida et al., 2010, Sumathi et al., 2012, Lopes et al., 2005, Prins et al., 2010, Athanasiou 2013, Romão et al., 2013, Zapryanova 2013). However, the effect of sex on FIB was reported by (Ameri et al., 2011).

The mean PT value in this study was 7.35±0.91 seconds with a minimum and maximum values of 6 and 10 seconds respectively. These values are more to or less similar to values reported by Prins et al., (2010), (7.4, range 6.7-9.5), Machida et al., (2010) and (7.9 ± 0.67, range 6.6-9.3), lower range values than of this work were reported by Lopes et al., (2005) (6.87 ± 1.4, range 4.07-9.67) and Vlasin et al., (2004), (8.12 ± 0.48, range 6.4-7.4 ), higher values of PT than of this study were reported by Uhrikova et al., (2013), (10.7, range 9.12-8.2), Romão et al., (2013), (11.80, range 11.03-13.05) and Sumathi et al., (2012), (12.12±0.57, range 6.7-16.6).

APTT mean value in this study was 15.12±1.32 seconds with a minimum and maximum of 13 and 18 seconds respectively. These values are more to or less similar to values reported by Machida et al., (2010), (15.8±3.0, range 9.8-21.8), and Lopes et al., (2005) (15.1± 1.6, range 11.9-18.3). Lower values of APTT than of this study were reported by Vlasin et al., (2004), (11.64±1.54, range 9.5-10.5) and Prins et al., (2010) (14.3 range, 10-17.2), higher values of APTT
were reported Uhrikova et al.,(2013),(17.5,range 16.4-19.6), Romão et al.,(2013),(18 ,range 15.75-19.21) and Sumathi et al., (2012),(43.17 ±2.39,range 22.6-56.6).
This differences in PT value and APTT among the different researchers could be due to differences in the studied animals like the number, sex or breed. There are many interlaboratory differences (Nagler et al., 2013) which may have caused this variation in the results obtained by the different researchers, such as the type of blood collection container, specimen transport, storage conditions, incubation time and temperature the used kits whether are for humans or specific for dogs .Trisodium citrate concentration was described by Adcock (1997) as an important factor which can affect the coagulation tests. Also the type of coagulometer, semi automatic or automatic may affect the results.

The FIB mean value in this study was 341.54±55.16 mg/dl with a minimum and maximum of 210 and 417mg/dl respectively which accord with the mean value (348), reported by Walton (2014) .Higher mean value (409.595± 3.554) and range (262.2-644.6) than of this work were reported by Athanasiou (2013).Lower FIB concentration than of this work was reported by Vlasin et al .,(2004),(257± 0.526) and Uhrikova et al.,(2013),(240). However the obtained results are near to the normal range (200-400) described by Stockholm (2013).

This variation could be due to variation in the studied animals as pregnancy and lactation are found to affect the FIB concentration (Antovic, 2013) or to interlaboratory differences (Van Blerk et al., 2015).

The present study shows a significant increase in FIB in males, this is on line with the results of Ameri et al, (2011). The total white blood cells count has a reverse effect on the concentration of fibrinogen Stockham (2013),so this may explain the higher concentration of FIB in the males than the females . The present results concluded that the commercially available kits for human medicine can be used to detect coagulation profile in dogs. The reference range values established in this study for PT in dogs was 7.35±0.91 seconds, for APTT it was 15.12±1.32 second, and for FIB it was 341.54±55.16 mg/dl. PT, APTT, and FIB mean value were affected by gender and breed, Effect of breed should be considered in clinical interpretation of dog PT, APTT and FIB, also effect of sex should be considered in clinical interpretation of dog FIB.

REFERENCES:


