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Evaluation of prothrombin time in canine hepatic dysfunction

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Abstract
The study was performed to calculate the prothrombin time in dogs affected with parenchymal, biliary and neoplastic hepatic disorders compared with dogs which are apparently healthy. Twenty apparently healthy dogs formed the control group. Hepatic dysfunction dogs where classified as parenchymal disorder (9), biliary disorder (6) and neoplastic disorder (5) formed the experimental group. Since liver functions as a site of production and removal of coagulation proteins, the present study was done to measure the thrombin time in healthy as well as dogs in experimental group. The prolonged prothrombin time was significantly noticed in dogs affected with parenchymal and neoplastic disorders of experimental group when compared with control dogs whereas the no significant alteration was observed in biliary disorder dogs. Thus the results of this study suggest that there is a significant change in the prothrombin time in dogs with hepatic dysfunction and it could be a prognostic indicator for the haemostatic disorders in dogs.

Keywords: canine, hepatic disease, prothrombin time, haemostatic disorders

Introduction
Liver is the largest endocrine gland in the body with high functional and regenerative capacity and to maintain its integrity even after extensive pathological damage. It acts as a center for various metabolic functions which includes haemostasis. It is the site of synthesis and clearance of most of the clotting factors including fibrinogen and the factors II, V, VII, IX, X, XI and XIII [1, 2]. The functioning of extrinsic and common pathway of coagulation cascade can be evaluated by measuring the prothrombin time In hepatic disorders, abnormalities of coagulation occurs due to impairment in the ability of the liver to synthesize these factors. Assessment of PT serves as a prognostic indicator in acute and chronic hepatocellular diseases. It is also a predictor of outcome in cases of acetoaminophen over dosage and acute alcoholic hepatitis. Evaluation of PT is also important in the management of patients with liver disease [3].

Materials and Methods
The study was conducted in dogs that were brought to Small Animal Clinical Medicine Outpatient Unit, Madras Veterinary College Teaching Hospital and at Department of Veterinary Physiology, Madras Veterinary College, Chennai. Twenty apparently healthy dogs formed the control group and twenty dogs with alteration in hepatic function formed the experimental group. As per WSAVA guidelines, the hepatic disorder dogs were further classified into parenchymal, neoplastic, biliary disorder group. About two millilitres of whole blood samples were collected in an aseptic manner by venipuncture of saphenous/cephalic vein from the selected dogs in vacutainers coated with sodium citrate as an anticoagulant in dilution rate of 1: 9 for haemostatic studies.
PT was estimated using commercial kits supplied by Agappe Diagnostics as per the standard protocol [4]. PT reagent was pre-warmed at 37 ˚C for 10 minutes prior to use. One hundred microliters of citrated plasma was pipetted into the test cuvette containing steel beads and incubated for 3 minutes at 37 ˚C. Two hundred microliters of pre-warmed PT reagent was added into the test cuvette and clotting time in seconds was recorded immediately using semi-automated coagulation instrument (Mispa Clog).

Result
The mean ± SE values of Prothrombin time in the control and experimental groups are compared and statistically interpreted.

The mean ± SE values of PT in control group was 10.98 ± 0.39seconds and the mean ± SE values of PT in parenchymal, neoplastic and biliary disorders of dogs were 25.31 ± 5.10seconds, 26.03 ± 7.47 seconds and 17.07 ± 2.41seconds, respectively. A significant increase ($P < 0.01$) was observed in the mean ± SE values of prothrombin time of dogs with parenchymal and neoplastic disorders when compared to the control group. No significant difference was observed in dogs with biliary disorder when compared to the control group.

Discussion

Tissue thromboplastin in the presence of Ca^{2+} activates extrinsic pathway of blood coagulation cascade. The activation time is proportional to the concentration of individual clotting factors taking part in the coagulation cascade. This assists in estimating cause and extent of haemorrhagic disorder. Clotting is the end result of a complex series of enzymatic reactions that involve at least thirteen clotting factors. The liver is the major site of synthesis of eleven blood coagulation proteins and prothrombin is one such protein synthesized in the liver.

The prothrombin time was significantly increased in dogs with parenchymal disorders. The present finding was similar to that of [5-7]. Prolonged PT is an indicative of measurable decrease in coagulation factor VII and a marked decrease in fibrinogen concentration. In the present study, the prothrombin time was significantly increased in the biliary disorder of dogs. Similar findings was reported [7, 8]. This increase in the prothrombin time in dogs may be due to deficiency of coagulation proteins involved in the extrinsic pathway of coagulation cascade as these coagulation proteins II, VII, IX and X are synthesized during vitamin K metabolism in the hepatocytes [9]. Due to cholestasis, bile flow by the contraction of gall bladder and relaxation of the sphincter of oddi into duodenum for the emulsification of fat soluble vitamin get reduced which in turn results in the decreased absorption of vitamin K. Hence, results in diminished synthesis of vitamin K dependent carboxylation of coagulation factors in the hepatocytes. Therefore, an increase in the prothrombin time was noticed in hepatic disorders of dogs [10].

Conclusion

The present study revealed that there was significant increase in dogs with parenchymal and neoplastic dysfunction and no significance was observed in dogs with neoplastic disorders. Thus alteration in hepatic function in dogs leads to alteration in the clotting mechanism due to impaired prothrombin synthesis.

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