Quantitative Accuracy of SelexOn™ Canine CRP Device for C-Reactive Protein Determination in Dogs

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Abstract

C-reactive protein (CRP) is versatile and sensitive tools for diagnosis of various inflammatory conditions like infection, tissue damage, myocardial infarction, pulmonary disorders, panniculitis, acute pancreatitis, polyarthritis, hemangiosarcoma and metabolic syndrome both in human and veterinary practice. Accurate, easy and quick measurement way of this important parameter is the demand of time. The objective of this study was to evaluate the accuracy of SelexOn™ canine CRP device (Infopia, Korea) in comparison to the Life Assays canine CRP test (Life Assays, Sweden) point-of-care testing. Blood samples were collected from 200 dogs of various conditions and CRP was evaluated directly from blood by SelexOn™ canine CRP device while plasma was prepared and CRP was measured by Life Assays®. There was no statistically significant difference in the average value of CRP between two groups and Spearman’s correlation coefficient was r = 0.9385. The calculated association was significant (p < 0.0001) indicating its accuracy. Moreover, using SelexOn™ canine CRP is convenient and time savings as it is directly measured from the blood. Therefore, SelexOn™ canine CRP device is suitable to use for clinical test and alternatives to traditional laboratory devices for the virtue of its accuracy, time savings, and easiness.

Introduction

C-reactive protein (CRP) is a major acute phase protein and is synthesized by the liver in response to factors related to inflammation. The concentration of serum CRP is elevated up to several hundred folds within 24 ~ 48 hours during various inflammatory conditions like infection, tissue damage, myocardial infarction, pulmonary disorders, panniculitis, acute pancreatitis, polyarthritis, hemangiosarcoma and metabolic syndrome. Measurement of CRP levels is mainly used as a marker of inflammation diagnosis in human medicine[1-4]. In veterinary medicine, canine CRP is also known to rapidly increase in acute phase response similar to human beings. It has been reported that the level of canine CRP is increased in diverse diseases such as babesiosis, parvovirus infection, tumors, pyometra, and arthritis. So canine CRP test can be used for diagnosis, monitoring, and screening of canine patients, and is especially useful for prognosis of inflammation even when there is no alteration of white blood cell or band neutrophil count[5]. The detection of canine CRP has been developed various methods as enzyme-linked immunosorbent-assay (ELISA), electro immuno-assay, time-resolved immunofluorometric assay (TR-IFMA) and turbidimetric immunoassay (TIA)[6,7]. However, these methods require large amounts of blood and time for the assay and may have different results depending on the user’s proficiency. Point-of-care testing (POCT) is a diagnostic test that complements the disadvantages of these conventional methods and it enables real-time measurements at near the site of patients[8]. Many POCT technologies have been developed for decades and high-quality POCT devices with improved sensitivity and specificity are constantly being released in the market.

SelexOn™ (Infopia, Anyang, Korea) is POCT analyzer utilizing chromatographic immunoassay to quantitatively or qualitatively detect the concentration of more than 30 cardiac,
cancer, thyroid and infection markers in human blood such as cardiac troponin I (cTnI), alpha-fetoprotein (AFP), C-reactive protein (CRP), thyroid-stimulating hormone (TSH)\(^9\) and has been working authentically for a long time in the field. Recently SelexOn\(^T^M\) canine CRP is developed and it is a quantitative device capable of measuring CRP in dogs. We evaluated the clinical utility of SelexOn\(^T^M\) canine CRP in dogs and compared with Life Assays canine CRP test (Life Assays, Lund, Sweden).

Materials and Methods

**Animals and sample collection**

The dogs (n = 200) which were brought to Seoul local veterinary hospital by the client for different diseases or for the health assessments were used in this study. Primarily blood samples were collected in test tube containing EDTA for diagnosis or assessments purpose. Secondarily, the remaining blood was used for this study and the clients were informed about that. SelexOn\(^T^M\) canine CRP device (Infopia, Korea) was used to evaluate CRP by using blood. Then the remaining blood was centrifuged (3000 RPM for 5 minutes). Then prepared antibody specific to analyte, these produce visible red lines on analyte complexes pass through the test line that is immobilized antibody and then gold-nanoparticle labelled antibody-target Target analyte in plasma binds to gold-nanoparticle labeled antibody. EDTA-treated venous whole blood diluted quantitatively through Life Assays\(^\text{®}\) manual, the measuring range of canine CRP is 10 – 210 mg/L and the reference range is reported as less than 20 mg/L. SelexOn\(^T^M\) Canine CRP is a sandwich assay using two monoclonal antibodies specific to canine CRP and is quantified by the thickness of the test line depending on the color of gold nanoparticle. EDTA-treated venous whole blood diluted with buffer is injected into the blood inlet area of test strip and then the erythrocyte is separated and only plasma is transferred. Target analyte in plasma binds to gold-nanoparticle labeled antibody and then when gold-nanoparticle labelled antibody-target analyte complexes pass through the test line that is immobilized antibody specific to analyte, these produce visible red lines on nitrocellulose membrane. The optical system of the SelexOn\(^T^M\) meter measures the intensity of red lines and the integrated software converts the signal intensity to a quantitative result and shows it on the display. According to the SelexOn\(^T^M\) manual, the measuring range of canine CRP is 10 – 200 mg/L and the reference range is reported as less than 20 mg/L.

**Test devices and protocols**

The following two POCT analyzers were selected for this study: Life Assays Canine CRP\(^\text{®}\) test kit (IDEON Science Park, Lund, Sweden) and SelexOn\(^T^M\) Canine CRP (Infopia, Anyang, Korea). Life Assays Canine CRP\(^\text{®}\) test kit is a latex agglutination test for determination of canine CRP and is measured quantitatively through Life Assays\(^\text{®}\) Vet Reader instrument. According to the Life Assays\(^\text{®}\) manual, the measuring range of canine CRP is 10 – 210 mg/L and the reference range is reported as less than 20 mg/L. SelexOn\(^T^M\) Canine CRP is a sandwich assay using two monoclonal antibodies specific to canine CRP and is quantified by the thickness of the test line depending on the color of gold nanoparticle. EDTA-treated venous whole blood diluted with buffer is injected into the blood inlet area of test strip and then the erythrocyte is separated and only plasma is transferred. Target analyte in plasma binds to gold-nanoparticle labeled antibody and then when gold-nanoparticle labelled antibody-target analyte complexes pass through the test line that is immobilized antibody specific to analyte, these produce visible red lines on nitrocellulose membrane. The optical system of the SelexOn\(^T^M\) meter measures the intensity of red lines and the integrated software converts the signal intensity to a quantitative result and shows it on the display. According to the SelexOn\(^T^M\) manual, the measuring range of canine CRP is 10 – 200 mg/L and the reference range is reported as less than 20 mg/L.

**Statistical analysis**

Statistical analysis was done by using the Prism 5.03 software (GraphPad Software Inc., San Diego, CA, USA) for the of the data. Results are expressed as mean ± standard deviation of the mean (SD). The paired Student’s t-test was used and correlated with the Spearman’s rank correlation coefficient. The level of significance was set at p < 0.05.

**Results and Discussion**

A total of 200 dogs were included in this study after informing the owners, of these 137 dogs was purebred, including 29 different breeds, and 63 were mixed-breed. 91 of the dogs were male (37 castrated and 54 intact) and 109 of the dogs were female (38 spayed and 71 intact). 183 dogs had been diagnosed with various diseases while 17 of the dogs were healthy. All types of dogs were used for sample collection such as different breeds, sex, disease, or healthy were tested and the results were within expected levels because of the assessed CRP levels in 0-220 mg/L\(^7\).

All samples were analyzed by SelexOn\(^T^M\) Canine CRP (blood) and Life Assays Canine CRP\(^\text{®}\) (serum) device. The lowest CRP concentration measured by the Life Assays Canine CRP\(^\text{®}\) was 10 mg/L whereas at the same sample it was 11.21 mg/L. The height CRP concentration measured by the Life Assays Canine CRP\(^\text{®}\) was 86 mg/L whereas at the same sample it was 91.12 mg/L. The averages were 21.55 ± 32.53 (SD) mg/L and 22.46 ± 40.84 (SD) mg/L, respectively (Figure 1). Thus, it is clear that all the data either lower, upper or average values of CRP between these two groups were very close in these two devices.

The correlation coefficient (Spearman’s) between the SelexOn\(^T^M\) Canine CRP (blood) and Life Assays Canine CRP\(^\text{®}\) was 0.9380 for the 200 samples that were examined. The calculated association was significant (p < 0.0001) (Figure 2) indicating its accuracy and reliability. In general, the SelexOn\(^T^M\) Canine CRP (blood) assay tended to yield higher CRP concentrations than Life Assays Canine CRP. It’s might be due to sample type, as CRP was measured directly from blood SelexOn\(^T^M\) Canine CRP (blood) assay.

In addition, CRP was measured directly from blood by SelexOn\(^T^M\) Canine CRP, thus it can save extra 5 minutes which is needed for serum sample preparation for Life Assays Canine CRP. So, using SelexOn\(^T^M\) Canine CRP is convenient, easy and faster than Life Assays Canine CRP. Canine CRP measurement is performing to screen diseases and to monitor the efficacy of...
References


