## Comparative study between double spin and the use of E-PET filter (equine platelet enhancement therapy) to obtain platelet rich plasma in horses – preliminary results

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## Abstract

The Platelet Rich Plasma (PRP) is a biotechnology technique that consists of a high concentration of autologous platelets in a small volume of plasma in order to obtain better results in the treatment of various diseases. The use of PRP is justified by growth factors present in the alpha granules of the platelet. There are several protocols to obtain PRP in horses described in the literature, among which stands out double spin, automated and the filters. This study aimed to compare the use of PRP obtained by double centrifugation protocol and by the E-PET (Equine Platelet Enhancement Therapy), taking into consideration the final platelet and leukocyte concentration and cost--effectivess among such methods. Four healthy horses were studied and they presented their complete blood count values within the normal range for the specie. It was used two samples of each animal to compare the two methods of achievement PRP. E-PET filter: each horse had 55mL of blood collected into a 60mL syringe containing 5mL of anticoagulant solution. The blood was transferred to the kit and filtered using a gravitation system in accordance with the manufacturer instructions. The manufacturer claims that the platelet concentration obtained by the filter is approximately seven times higher than the blood concentration. Double spin protocol: each horse had 20mL of blood collected in tubes containing sodium citrate (anticoagulant). Two centrifugations were done (300g for 5 minutes and 700g for 17 minutes) intercalated with rests of approximately 35 minutes in which the aim was to separate the plasma. After the second spin, it was discarded the upper 75%, being the remainder (lower 25%) the PRP. The platelet and the leukocyte were counted using the Neubauer's chamber. Double spin: A1: 936.775 platelets/µl of blood and 16.985 leukocyte/µl of blood; A2: 939.300 platelets/µl of blood e 17.587,5 leukocytes/µl of blood; A3: 1.121.000 platelets/µl of blood and 15.225 leukocytes/µl of blood; A4: 1.179.175 platelets/µl of blood and 13.440 leukocytes/µl of blood. Standard deviation: 124.713,7 platelets e 1.870,76 leukocytes. E-PET: A1: 1.262.500 platelets/µl of blood and 12.810 leukocytes/µl of blood; A2: 1.595.200 platelets/µl of blood and 22.257,50 leukocytes/µl of blood; A3: 808.000 platelets/µl of blood and 23.307,50 leukocytes/µl of blood; A4: 1.315.525 platelets/µl of blood and 19.897,50 leukocytes/µl of blood. Standard deviation: 326.031 platelets e 4.725,68 leukocytes. The amount of platelets and leukocytes obtained by the double spin and by the E-PET filter were similar, but they differed in relation to the use of a laboratory (not necessary on the filter), time (better at the

filter), to be a close system (filter) and in relation to cost (higher in the filter). However, it is necessary to study the differency beetween the concentration of growth factors in these two methods.

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