DETERMINATION OF Ca AND Fe IN BLOOD OF GRMD DOG SUBMITED TO A SYSTEMIC TRANSPLANTATION OF STROMAL CELLS (hASCs) USING NAA AND FRX TECHNIQUES

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Abstract

The use of alternative analytical techniques to investigate specific electrolytes in body fluids (mainly blood, serum and urine) has increased in past few years, presenting significant progress in clinical practices. Since 2004 Neutron Activation Analysis (NAA) and more recently X-ray Fluorescence (XRF) techniques have been applied to this clinical finality at IPEN/CNEN-SP, in collaboration with other research centers from Brazil. Several investigations in veterinary medicine, immunology and genetic fields were performed and also applications for medical diagnosis studies; particularly for Duchenne Muscular Dystrophy (DMD). The DMD is an illness of a hereditary character that affects approximately 1 in every 3,600 to 6,000 live male births in the world. Nowadays, many promising therapeutic strategies have been developed in animal models with DMD. An animal model which has a phenotype, which is similar to human patients with DMD, has been bred in Brazil: Golden Retriever Muscular Dystrophy dogs (GRMD). In these dogs, muscle degeneration and fibrosis are predominating, leading to a progressive loss of structure and muscle function, as in humans (once they have a comparable muscle mass to that of a human being).

Recently, the Human Genome Research Center (Biosciences Institute in Brazil) has shown that human adipose derived from stromal cells (hASCs) when injected systemically into GRMD dog cephalic vein paw are able to reach, engraft, and express human dystrophin in the host GRMD dystrophic muscle (up to 6 months after transplantation). This shows an improvement in the functional performance of injected animals without any immunosuppressant. In this study, Ca and Fe were investigated in whole blood during this period (before starting the transplantation process and after six months); due to functions they play in muscle to keep them healthy. Nonetheless, this disease is caused by a mutation of the dystrophin gene. The absence of dystrophin (a protein present in muscles) permits the excess of calcium to penetrate the sarcolemma (cell membrane). This protein is altered causing a critical muscular dysfunction in several body functions, such as: calcium homeostasis and dysfunction in the mechanisms of membrane permeability causing a degeneration of the membrane that involves the muscular cell, leading to its death. Another clinical point to be considered is related to Fe concentrations in blood. The hemochromathosis (overload of iron) can cause joint pain, abdominal pain, weakness, fatigue or significant organ damage, and that can eventually trigger another health problem for the DMD patients.

The present results have showed that this therapeutic procedure does not affect the Ca and Fe levels. Furthermore, some improvements in their physiological structure and mobility have been observed, suggesting that there is a need to continue this therapeutic strategy for DMD patients.

Keywords: steam cells, DMD, whole blood, Ca, Fe, NAA, FRX