



Veterinary clinical investigations: the use of heterologous mesenchymal stem cells in dogs with keratoconjunctivitis sicca

Michele A. de Barros^[a], Maura K. Bittencourt^[b], Cristiane V. Wenceslau^[a], João Flávio P. Martins^[c], Bruna P. de Morais^[d], Karine Evangelho^[e], José Paulo C. Vasconcelos^[b], Alexandre Kerkis^[c], Irina Kerkis^[f]

^[a] Federal University of São Paulo (UNIFESP), São Paulo, SP – Brazil

^[b] University of Campinas (UNICAMP), Campinas, SP – Brazil

^[c] Regenera Stem Cells, Campinas, SP – Brazil

^[d] University of São Paulo (USP), São Paulo, SP – Brazil

^[e] Universidad Nacional Rio Cuarto, Río Cuarto, Córdoba – Argentina

^[f] Butantan Institute, São Paulo, SP – Braz

Abstract

Keratoconjunctivitis sicca is a common disease in dogs which can be due to quantitative or qualitative deficiency of the tear film, or a combination of both. The current treatment is based on the use of topically applied artificial tear lubricants, stimulation of tear secretion and using anti-inflammatory drugs. Even advanced therapies, like punctual plugs, cyclosporine B administration, and salivary gland auto-transplantation have led to a limited success. Thus, an attractive alternative is the stem cell treatment, that needs to be explored in order to provide better and long term relief to these patients. This study aimed to evaluate the use of heterologous mesenchymal stem cells (MSC) derived from adipose tissue in dogs with keratoconjunctivitis sicca (KSC). MSC were obtained under good manufacturing practice conditions. KSC was defined by the quantitative Schirmer tear test (STT) below 15mm/min. Eleven dogs of different genders, ages and races were enrolled in present study. One eye of each dog was treated with 106 MSCs, which were administrated in a total dose of 0.5ml of physiologic solution. Each dose was applied into two sites: 0.3 ml of MSC solution was administered directly to the main lacrimal gland and 0.2 ml to the third eyelid gland. The eyes were evaluated weekly during 8 weeks using STT, fluorescein test and slit-lamp biomicroscopy. The severity of eye score (SES) was evaluated according to conjunctival hyperemia, ocular discharge, corneal opacity or irregularity and neovascularization. Dogs that did not show significant improvement five weeks after MSCs application were submitted to a second application following the same protocol and scheme of evaluation. After 3 weeks the dogs showed increased STT values when compared to baseline levels, and the values were statistically significant ($p=0.0023$). This increase of STT values remained significant until the fifth week of MSM application and 55% of the dogs showed improvement in tear production with STT measurements above 15 mm/min. The remaining animals, which needed to receive two applications of MSC, reached their peak of tear production at the seventh week, demonstrating statistically significant results as well ($p=0.003$). After 8 weeks the dogs showed increased STT values when compared to STT at the beginning of the treatment ($p=0.0228$). The clinical improvements of corneal opacity ($p=0.006$) and conjunctival secretion ($p=0.0376$) in eyes were also observed.

However, the data obtained regarding the degree of conjunctival hyperemia and corneal vascularization were not statistically significant. MSC used in this present study suggested its safety, since none of the animals demonstrated any type of rejection, allergic reaction or tumor formation. These cells demonstrated a clear clinical benefit in the treatment of KSC, thus improving the function of the lacrimal glands and of several other parameters. This study provided a basis for future clinical studies in humans with KSC.