SARCOGLYCAN COMPLEX IN HUMAN TRACHEAL TISSUE: AN IMMUNOFLUORESCENCE STUDY

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The sarcoglycan complex (SGC) in made up by transmembrane glycoproteins, α-, β-, γ-, δ-sarcoglicans, and as a part, by dystrophin-glycoprotein complex (DGC). The DGC also includes dystrophin, dystroglycan (α- and β-) and syntrophins; the interaction of the DGC with components of the extracellular matrix may have an important role in force transmission and sarcolemma protection. Moreover, α-dystroglycan appears to have a core protein consisting of two roughly globular domains connected by a segment, which most likely corresponds to a mucin-like central region.[1] Several studies also demonstrated that sarcoglycans, which are involved in stabilization of sarcolemma during muscle contraction, are expressed in some epithelial tissues as gingival, breast and prostatic one.[2] The aim of our study is to demonstrate the expression of sarcoglycans in tracheobronchial epithelial tissue, related with the presence of mucins. Mucins, which are a superfamily of highly glycosylated proteins, are the main constituent of mucus. In the conducting airways mucus is produced by the tracheobronchial glands and it plays a key role in maintaining the health of epithelial tissues. Biochemical studies shows that human tracheobronchial gland mucous (HTMG) cells secreted typical airway mucins, and immunohistochemical studies showed that these cells expressed different isoforms of MUC, especially those of our interest: MUC4 and MUC16.[3] We performed an immunofluorescence reaction, using antibodies against β-sarcoglycan and against MUC4. To avoid any possible fluorescence interference between the primary antibody and the mucus gel the sample was pre-treated following a mucolytic protocol (N-acetylcystein 10% in Phosphate buffer 0.2 M).

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MESENCHYMAL STEM CELLS IN HIDRADENITIS SUPPURATIVA: A NEW PIECE OF THE DISEASE MOSAIC

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Hidradenitis suppurativa (HS) is a chronic inflammatory skin disease. It generally involves the apocrine gland-bearing area of the body and induces deep painful lesions, often associated with other physical and psychosocial comorbidities. Onset of HS is caused by an early occlusion of follicle, followed by dilatation of pilosebaceous unit, rupture and release of follicular contents into the dermis. Then, an inflammatory state is established, as demonstrated by the various immunological abnormalities found in skin lesions. Mesenchymal stem cells (MSCs) were successfully isolated from different tissues, included skin. Researches support the hypothesis of their early involvement in various skin diseases, such as psoriasis and atopic dermatitis. However, the role of MSCs in HS has never been evaluated. In this study, MSCs were isolated from lesions of patients affected by HS (HS-MSCs) and characterized. Their immunological profile was examined by assessing the levels of twelve cytokines related to Th1, Th2 and Th17 pathways on conditioned medium by ELISA array. Inflammatory activity of HS-MSCs was also studied by immunocytochemistry. MSCs isolated from healthy subjects were used as controls. HS-MSCs revealed an over-expression of most of the molecules analyzed, demonstrating the activation of MSCs in the disease towards a pro-inflammatory state. Even more, the fact that these same cytokines play a role in HS, as demonstrated by previous studies, strengthens the hypothesis of MSCs contribution in disease onset and progression.

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PROGESTERONE RECEPTOR MEMBRANE COMPONENT 1 (PGRMC1) EXPRESSION IN CANINE MAMMARY TUMORS

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Canine mammary tumors (CMT) are the most common neoplasms in female dogs and histopathological examination is the gold standard for their diagnosis and classification.[1] Recently, PGRMC1 has been considered a putative biomarker for diagnosis and prognosis in human breast cancer, which shares some common features with canine counterpart.[2] This study represents the first description of PGRMC1 expression in CMT.
First, we evaluated PGRMC1 expression by immunohistochemistry in three major histopathological types: normal/hyperplastic tissue, simple adenomas and simple adenocarcinomas. Epithelial cells showed positivity to PGRMC1 and we applied a scoring system considering the percentage of positive cells and the intensity of expression defined as A (weak), B (moderate) and C (strong). Normal/hyperplastic sample showed almost 100% of positive cells and a strong intensity of PGRMC1 expression. The same features were present in adenomas but with a more variable intensity. In adenocarcinomas, the percentage of positive cells was lower (30-60%) and the intensity was weak in tubular parts, while both features became progressively negative in the solid parts of the tumor. Western blot analysis in healthy and neoplastic mammary tissue biopsies of dogs undergoing surgery revealed the presence of the 25 kD PGRMC1 band in both types of tissue. Further investigations are in progress to determine differences in protein level according to the 3 different major types of tumors. Moreover, PGRMC1 will be assessed in blood samples of dogs affected by different types

of CMT to evaluate if it could represent a prognostic serum biomarker as previously demonstrated in human lung and kidney cancer.[3]

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CONTRIBUTION OF miR-145-5p/Ago2 COMPLEX TO THE REGULATION OF EPITHELIAL-MESENCHYMAL TRANSITION

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The epithelial-mesenchymal transition (EMT) is essential for cell fate determination during development but it is involved in pathological processes like cancer as well, being one of the first steps in the mechanisms leading to metastasis. miR-145-5p is one of the most widely recognized tumor-suppressor miRNAs, able to regulate cell migration and EMT through the contribution of the RISC complex in which Argonaute (Ago) proteins are required for target recognition and gene silencing.[1] Ago2 is an important member of the Ago family and its overexpression correlates with a transformed phenotype in breast cancer cells.[2] With the aim to unravel miR-145-5p/Ago2 contribution to the suppression of cancer progression in epithelial tumors, here we show that: i) miR-145-5p and Ago2 are down-regulated in breast tumor *vs* normal tissues; ii) the restored expression of miR-145-5p in breast cancer cell lines results in the reduction of tumor phenotype; iii) Ago2 expression is positively and specifically regulated by miR-145-5p; iv) miR-145-5p-dependent Ago2 induction is necessary for the inhibition of cell migration; v) when Ago2 is depleted, the formation of an alternative miR-145-5p/Ago1 active complex redirects miR-145-5p tumor suppressor function and correlates with a more invasive phenotype in breast cancer cells. These results open to the identification of miR-145-5p/Ago2-dependent molecular networks involved in the maintenance and progression of cancer phenotype.

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SARCOGLYCAN SUB-COMPLEX IN CARDIAC MUSCLE OF PATIENTS DECEASED FOR SEPSI

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The sarcoglycan sub-complex (SGC), member of the DGC, is made up of six subunits of transmembrane glycoproteins named α-, β-, γ-, δ-, ε-, and ζ-sarcoglycan (SG). This complex is localized in the sarcolemma of skeletal muscle fibers and its function is to connect the extracellular matrix to the cytoskeleton playing a key role in stabilizing muscles and their sarcolemma during contraction and relaxation. Clinical studies have shown that myocardial contractility is reduced in severe sepsis which is a life-threatening organ dysfunction which prevalence has increased significantly in recent decades. It has been shown that in 40% of patients with sepsis and cardiac dysfunction the range of mortality is of 70-90% if compared to the 20% mortality in septic patients without cardiovascular involvement. Recent studies support that an increased plasma membrane permeability in cardiac fibers, could be responsible for sepsis-induced myocardial dysfunction; they have also shown a decreased expression of some DGC proteins, suggesting a role of DGC in the pathogenesis of sepsis induced cardiomiophaty. The critical importance of the SGC in maintaining sarcolemmal stability led us to hypothesize that even these proteins could be involved in sepsi-induced cardiomyopathy. The aim of the present work was to analyze the expression of sarcoglycans in human cardiac muscle samples from deceased patients because of sepsis, with cardiovascular involvement. By histological and immunofluorescence techniques we obtained results which show critical structural alteration of cardiac fibers sarcolemma and that the expression of β-, γ- and ε-sarcoglycans, but not the α-sarcoglycans, is reduced. We hypothesize that members of the sarcoglycans sub-complex could be involved in structural alteration of sarcolemma, leading to sepsis induced cardiomiophaty, even if the pathogenetic mechanism are still unclear.

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