human Adipose Stem Cells and their Secretome contrast painful symptoms induced by peripheral diabetic neuropathy in the mouse experimental model.

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Diabetic neuropathy is one of the most common forms of neuropathic pain. Even today, the treatment of these pain forms is very difficult; it is therefore necessary to explore new approaches. A new possibility might be the use of stem cells. In this study we evaluated the effect of Mesenchymal Stem Cells isolated from human adipose tissue (hASC) and of their secretome/conditioned medium (CM-hASC) on the neuropathic symptomatology, in a preclinical experimental mouse model of diabetic neuropathy induced with low dose of Streptozotocin (STZ, 80 mg/Kg once a day for 3 consecutive days, intraperitoneal injection). The development of mechanical and thermal allodynia after STZ was monitored by using respectively a Dynamic Plantar Aesthesiometer (Von Frey test, ranging up to 10 grams in 10 seconds) and Acetone test (50μL of acetone, cold-allodynia).
When allodynia was well established (2 weeks after STZ), mice were treated by intravenous administration with either $1*10^6$ hASC or CM-hASC obtained from $2x10^6$ serum-free cultured cells. As control we evaluated the effect on neuropathic pain of the CM obtained from a human fibroblasts (CM-hF). The effect of treatments on allodynia was monitored over time (14 weeks from STZ). The effect of hASC was compared to CM: both hASC and CM-hASC were able to significantly reduce mechanical allodynia, although the efficacy of hASC was always higher than that of CM-hASC; on the contrary CM-hF was unable to evoke any antiallodynic effect in diabetic neuropathic mice. In STZ-mice treated with hASC or CM-hASC the anti-allodynic effect is very rapid in fact, after only three hours from therapeutic treatment, we observed a slight but significant pain relief, moreover the effect on pain is long lasting, it is still evident 12 weeks from therapeutic treatment. Moreover, treatments were effective also when performed in a more advanced stage of the disease (6 week after STZ), in fact the data obtained were fully comparable with those observed after 1st treatment. Besides, we evaluated the effects of treatments with hASC and their CM also on cold-allodynia at different times: 3h, 24h, 3 and 7 days after therapeutic treatments. We observed that both hASC and CM-hASC are able to significantly modulate the sensorial alteration typical of STZ-mice at all observed times. Moreover our data suggest that both hASC and their CM were able to contrast the typical body weight reduction of STZ treated mice. 14 weeks after STZ, that is 12 or 8 weeks after therapeutic treatment, six animals of each group were sacrificed for the biochemical evaluation. In order to understand the mechanisms at the basis of the observed effects on pain, we studied the involvement of neuroinflammation evaluating the levels of the cytokines IL-1 and IL-10 in the spinal cord. We confirmed that since neuropathic mice are characterized by a proinflammatory profile, high IL-1 and low IL-10 levels. 14 weeks from STZ both hASC and CM treatments were able to restore a correct pro/antinflammatory cytokine balance in this nervous tissue. Furthermore in this model of diabetes, peripheral immunity is altered and in particular it is present an alteration of lymphocytes toward a T-helper 1 (Th) profile. We confirmed, in STZ-animals, the presence of Th1 profile in splenocytes, characterized by high IFN levels and low IL-10 levels. Also in this case all treatments were able to restore the Th1/Th2 balance by decreasing IFN and increasing IL-10.
The data obtained in this study confirm that hASC treatment may be a favorable approach for neuropathic pain treatment and indicate that cells may eventually be substituted with their CM, moving toward a cell-free therapy.