5-azacytidine improves the osteogenic differentiation potential of aged human adipose-derived mesenchymal stem cells by DNA demethylation.

Abstract:
The therapeutic value of adipose-derived mesenchymal stem cells (Ad-MSCs) for bone regeneration is critically discussed. A possible reason for reduced osteogenic potential may be an age-related deterioration of the Ad-MSCs. In long term in vitro culture, epigenomic changes in DNA methylation are known to cause gene silencing, affecting stem cell growth as well as the differentiation potential. In this study, we observed an age-related decline in proliferation of primary human Ad-MSCs. Decreased Nanog, Oct4 and Lin28A and increased Sox2 gene-expression was accompanied by an impaired osteogenic differentiation potential of Ad-MSCs isolated from old donors (>60 a) as compared to Ad-MSCs isolated from younger donors (<45 a). 5-hydroxymethylcytosine (5 hmC) and 5-methylcytosine (5 mC) distribution as well as TET gene expression were evaluated to assess the evidence of active DNA demethylation. We observed a decrease of 5 hmC in Ad-MSCs from older donors. Incubation of these cells with 5-Azacytidine induced proliferation and improved the osteogenic differentiation potential in these cells. The increase in AP activity and matrix mineralization was associated with an
increased presence of 5 hmC as well as with an increased TET2 and TET3 gene expression. Our data show, for the first time, a decrease of DNA hydroxymethylation in Ad-MSCs which correlates with donor-age and that treatment with 5-Azacytidine provides an approach which could be used to rejuvenate Ad-MSCs from aged donors.