Correlation of Human Mesenchymal Stem Cell Characteristics with Donor Age
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Introduction
New strategies in tissue engineering favour mesenchymal stem cells because they are multipotent, self-renewing cells which can be easily isolated from various tissues. A differentiation into chondrocytes and osteoblasts is well established and attractive for regeneration of damaged cartilage and bone. Loss of cartilage is a result of osteoarthritis, a disease which mostly occurs with advanced age. Impaired bone fracture healing is frequently observed in patients with age-related osteoporosis. Therefore the questions arise whether MSC properties change with the age of their donor and should MSC therapies be restricted to a special age group.

Materials and Methods
MSC were isolated from bone marrow of 28 donors, 14 male and 14 female, and classified into three age groups: A young group, aged from 5 to 15 years, a middle group, aged from 24 to 49 years and an elderly group, aged from 64 to 80 years. Colony forming unit fibroblasts (CFU-Fs) were measured by counting colonies consisting of more than 30 cells after 7 days of culture. For single cell cloning efficiency one cell per well was seeded and cultured for 14 days. Differentiation of cells into osteogenic and adipogenic lineage was induced in monolayer culture for 3 weeks with differentiation medium. Osteogenesis was quantified by an alkaline phosphatase (ALP) assay, adipogenesis by Oil Red O measurement. Chondrogenic differentiation was induced in high density pellet culture with 0.5x10⁶ MSC for 6 weeks in chondrogenic medium supplemented with TGF-β1. Pellets were analyzed for proteoglycan- and collagen type II content via Alcian Blue Assay and ELISA. Data were statistically evaluated by Mann-Whitney-U Test and p ≤0.05 was considered significant.

Results
The number of MSC obtained per total number of mononuclear cells of bone marrow was independent of donor age (Fig.1A). Single cell cloning efficiency of MSC was lower in the elderly group compared to the young one, p=0.025 (Fig.1B). Osteogenic and adipogenic differentiation potential was maintained during aging. Histological staining as well as proteoglycan and collagen type II quantification for chondrogenic differentiation yielded no significant difference between the three age groups. Although there is a tendency towards less differentiation capacity in the elderly group in all three lineages this trend did not reach significance due to donor variability.

Discussion and Conclusions
While fewer cells can be expanded from old MSC donors the multilineage differentiation capacity of these MSC is maintained during aging. The high donor variability can therefore not be explained by donor age alone and possible further factors affecting the stem cell characteristics will be discussed. We conclude that age is no factor excluding patients from stem cell therapy.

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Disclosures
The authors have nothing to disclose.