

Abstrakt 6

Mesenchymal stem cells differentiate into striated muscle with connection to motor end plates - a new therapeutic option for urinary incontinence

Autor(en):

S. Baumann¹), G. Feil¹), R. Schäfer²), J. Schäfer¹), R. Möhle³), A. Stenzl¹), K.-D. Sievert¹)

1)Department of Urology, 2)Insitute of Clinical and Experimental Transfusion Medicine, 3)Department of Medicine II, Eberhard Karls University, Hoppe-Seyler-Strasse 3, 72076 Tübingen, Germany

Einleitung:

Human mesenchymal stem cells (MSCs) differentiate into both striated and smooth muscle cells. Urinary incontinence is associated with damaged sphincteric myofibers or with an age-dependent decrease of myofibers by apoptosis. Therefore, autologous MSCs might be an option for a functional treatment of urinary incontinence. The aim of the study was to investigate survival and myogenic differentiation of transplanted human MSCs in a rat model and to demonstrate a linkage to the host's nerve system.

Material und Methodik:

Human MSCs were isolated from bone marrow aspirates by plastic adherence and propagated in vitro. To induce myogenic differentiation in vitro, human MSCs were exposed to 5-azacytidine (AZA) in passage (P) 1. Native MSCs in P1 and P3 as well as AZA-exposed MSCs of subsequent P2 or P3 were directly injected into the rectus abdominis muscle of athymic nude rats. For in vivo tracking MSCs were labeled with red fluorescent PKH26 cell linker. Integration and myogenic differentiation of MSCs in rat muscle tissue was monitored histologically from 4 days up to 16 weeks after cell injection. The muscle markers desmin (clone D33) and skeletal muscle myosin heavy chain (clone NOQ7.54D) were immunologically detected. Innervation of newly formed skeletal muscle was investigated by staining cryosections with alpha-bungarotoxin conjugate (α -BTX-AF 488) that binds to the acetylcholine receptors of motor end plates.

Ergebnisse:

Both native and AZA-exposed MSCs of all passages could be demonstrated in all animals investigated. Histology of animals in the short-term experiments up to 8 days revealed well-defined clusters and beginning migration of transplanted MSCs (red PKH26 fluorescence) in the rectus abdominis muscle. After 4 and 8 weeks of cell injection, a continuous dissemination of transplanted MSCs was detected. Histology of animals in the long-term study revealed PKH26-positive myofibers that were in parallel with the native skeletal muscle fibers. Immunohistochemistry for myogenic desmin demonstrated striated myofibers. Skeletal muscle myosin was expressed in PKH26-positive myofibers. Staining for acetylcholine receptors showed motor end plates adjacent to newly formed PKH26-positive myofibers.

Schluss:

The experimental athymic rat model revealed a development of human MSCs into myotubes and subsequently the formation of myofibers that were well integrated into the host tissue. Connection to the nerve system of the newly formed muscle indicates a functional integration of transplanted and myogenically differentiated MSCs. This is promising for a regeneration of damaged rhabdosphincter muscle based on autologous adult stem cells.