

In Vivo Effects of Bone Marrow Derived Mesenchymal Stem Cells on the Growth of Oral Squamous Cell Carcinoma: 1160

Gogilashvili, K.¹; Tsiklauri, M.¹; Zurmukhtashvili, M.¹; Menabde, G.¹; Amiranashvili, I.¹; Abiatari, I.¹

CONTENT NOT FOR REUSE

Cell Transplantation

604

Ex Vivo Generation of Glucose Sensitive Insulin Secreting Mesenchymal Stem Cells Derived from Human Adipose Tissue

Dave S.D.¹

¹Institute of Kidney Diseases and Research Centre and Institute of Transplantation Sciences, Department of Pathology, Laboratory Medicine, Transfusion Services and Immunohematology, Ahmedabad, India

Introduction: Diabetes is predicted to be the major killer of population all across the world with likelihood of 366 million people suffering from diabetes by the year 2030. Strategies to curb this problem are being established in the form of pancreatic transplantation or islet cell replacement from adult and embryonic stem cells producing insulin. We present prospective study of glucose-sensitive insulin-secreting mesenchymal stem cells (IS-MSC) generated from human adipose tissue (h-AD) sans xenogenic material.

Methods: After Institutional Review Board approval and informed consent forms from voluntary human donors ten grams anterior abdominal wall h-AD was collected in proliferation medium composed of α -Minimum Essential Media, albumin, Fibroblast-growth factor and antibiotics; minced, incubated in collagenase-I at 37°C with shaker and centrifuged. Supernatant and pellets were separately cultured in proliferation medium on cell+ plates at 37°C with 5% CO₂ for 10 days. Cells were harvested, checked for viability, sterility, counts, flow-cytometry (CD45-/90+/73+) and differentiated into insulin-expressing cells using differentiation medium composed of Dulbecco's modified eagle's medium, insulin gene expression up-regulating growth factors, hormones and antibiotics for 3 days. They were studied for transcriptional factors: paired box genes-6 (Pax-6), Islet-1 transcriptional factor (Isl-1) and pancreatic and duodenal homobox-6 (Pdx-6) (immunofluorescence). C-peptide and insulin were measured by chemiluminescence. In-vitro glucose sensitivity assay was carried out by measuring levels of insulin and C-peptide secretion in absence of glucose followed by 2 hours incubation after glucose addition. MSC cultured without use of differentiation medium was used as negative control.

Results: Total 50 IS-MSC cell lines were generated from h-AD derived MSC.

Hematoxylin and eosin stained cells showed large basophilic nuclei with distinct margins surrounded by eosinophilic cytoplasm.

The mean cell quantum was 3.16 ± 0.58 ml (range: 2-4 ml), mean cell count, $2.5 \pm 0.95 \times 10^4$ /ml (0.78 - 2.7×10^4 /ml), mean CD45-/90+ cells were 47.39 ± 15.77 % (range: 16.62-81.38%) and mean CD45-/73+ cells were 25.40 ± 13.03 % (range: 2.68-65.72%). All the IS-MSC showed presence of transcriptional factors Pax-6, Isl-1, pdx-1. Mean insulin level secreted by the cells themselves was 165.42 ± 665.85 mU/ml (0-3800 mU/ml) in absence of glucose and 2 hours after addition of glucose (following incubation at 37°C) the rise in insulin secretion level was observed with the mean of 510.41 ± 1502.24 mU/ml (range: 0.5-9500 mU/ml).

Mean C-peptide level secreted by the cells themselves was 0.32 ± 0.42 ng/ml (range: 0-2.26 ng/ml) in absence of glucose and after 2 hours after addition of glucose the rise in C-peptide secretion level was observed with the mean of 1 ± 1.67 ng/ml (range: 0.01- 9.35 ng/ml).

The mean rise in insulin and C-peptide secretion levels secreted by the cells themselves was 3.08 folds ($P=0.00001$) and 3.14 folds ($P<0.00001$) respectively after addition of glucose to the cells after 2 hours incubation. All the negative controls showed absence of insulin secretion and C-peptide in absence of glucose as well as 2-hours after addition of glucose.

Conclusion: Insulin-secreting h-AD-MSC can be generated safely and effectively showing in-vitro glucose responsive alteration in insulin and C-peptide secretion levels.

1091

Bone Marrow Mononuclear Cell Intracoronary Transplantation in Patients with Chronic Ischemic Cardiomyopathy

Shanava K.¹, Kakabadze Z.¹, Kipshidze N.², Berishvili E.³

¹Tbilisi State Medical University, Department of Clinical Anatomy, Tbilisi, Georgia, ²Cell Transplantation Unit, Central University Hospital, Tbilisi, Georgia, ³Division of Cell Transplantation, Georgian National Institute of Medical Research, Tbilisi, Georgia

Background: It has been shown that autologous bone marrow cells may contribute to myocardial repair after acute myocardial infarction. The aim of this study was to assess the beneficial effects of intracoronary transplantation of bone marrow cells in patients (n=9) with chronic ischemic cardiomyopathy.

Methods: Bone marrow was obtained and the cells were injected intracoronary after a brief balloon occlusion at a normal coronary segment. Dobutamine stress echo showed that all patients had left ventricular ejection fraction < 35%. Patients were followed up to 12 month. Clinical follow up was performed periodically and included electrocardiography, laboratory tests and echocardiography.

Results: Intracoronary bone marrow cell therapy improved ventricular performance, quality of life and survival in patients with chronic ischemic cardiomyopathy. No side effects were observed

Conclusions: Intracoronary transplantation of autologous bone marrow cells is safe and feasible in chronic ischemic cardiomyopathy.

1160

In Vivo Effects of Bone Marrow Derived Mesenchymal Stem Cells on the Growth of Oral Squamous Cell Carcinoma

Gogilashvili K.¹, Tsiklauri M.¹, Zurmukhtashvili M.¹, Menabde G.¹,

Amiranashvili I.¹, Abiatari I.¹

¹Institute of Medical Research, Iliia State University, Tbilisi Georgia, Tbilisi, Georgia

Background: Squamous cell carcinoma is one of the most common malignant tumors of the oral mucosa and associates with high morbidity and mortality in both Western and Asian countries. The purpose of the study was to detect the effect and possible mechanism of mouse bone marrow derived mesenchymal stem cells (MSCs) on the in vivo orthotopic growth of primary human oral squamous cancer cell line (OSCCs).

Methods and Results: Primary human OSCCs were injected in vivo in the submucosal layer of the nude mice oral cavity and development and growth of these tumors were monitored during the complete experimental period. MSCs were isolated from bone marrow of C57BL/6N mice and transplanted in the different sites of tumor model animals. Immunohistochemical and ELISA analysis revealed decreased local and systemic inflammation as well as reduced desmoplastic reaction of the tumor in animals after MSC transplantation. The role of MSCs in growth, spreading and neovascularisation of oral squamous cancer has also been characterized in this animal model.

Conclusions: Nude athymic mice are an useful in vivo model for studying oral squamous cancer growth. Transplanted MSCs are attracted by oral squamous cancer tissue and modulate their tumorigenicity in these animals.