however, it is now impossible to propose the interval between the injection of the DNA and mating, the exact site and the amount of exogenous DNA incorporated into spermatozoa and the expression and stability of exogenous DNA in testes.
Objective: In present study, we investigated the stability of expression of pIRES-EGFP liposome complex in rat testis after sperm mediated gene transfer.
Materials & Methods: A chimeric pIRES-EGFP plasmid (50 ng) labeled with EGFP reporter gene was mixed with cationic liposome (DOTAP) in HSB solution and injected into rat testis and 2 , 5 , 10 , 30 and 60 days post injection the testes were removed and the expression level of EGFP gene was examined by RT-PCR, Realtime PCR and fluorescent microscopy. Animals which were injected with only DOTAP or plasmid without DOTAP were assigned as control groups.
Result: A fluorescent microscopic observation showed that EGFP gene was expressed in testes from 2 to 60 days post injection. Similar results were observed when we analyzed EGFP expression by RT-PCR. Quantitative analysis of EGFP expression by Realtime PCR showed that its expression level was the highest in 10 days post injection ($p < 0.05$). EGFP expression level was the lowest in 2 and 60 days after TMGT.
Conclusions: these results demonstrated that, in the case of our testis-mediated gene transfer, exogenous DNA injected with liposomes is permanently expressed 60 days after TMGT, however the 10 day is the best time for mating because the highest expression of foreign DNA was detected.
Keywords: SMGT, rat, pIRES-EGFP, gene expression

O-1-44721-Binding of p53 mutant and wild type to BRCA1 and Rad51 protein in vitro and in vivo
Tayebeh Azimi, Mozhgan Rasti
1. Recombinant Protein Lab, Department of Biochemistry, School of Medicine, Shiraz University of Medical Sciences, Shiraz, Iran

Background: P53 is one of the key tumour suppressor genes that is targeted for inactivation during human tumour genesis. Point mutations or deletions in the p53 gene are found in approximately 40% of all cance .In normal cells p53 plays a crucial rolein the negative regulation of cell growth in response to DNA-damage and other stress-activated signalling pathways.
Objective: In this study, we investigated the binding capacity of p53 mutant at codon 273 and wild type p53 to BRCA1and Rad51 in vivo and in vitro.
Materials & Methods: The human breast cancer cell lines, MDA-MB-468 cells (ER- negative; hemizygous p53 mutant) and MCF-7 cells (ER-positive; wild type p53) were used. By in vivo studied, the Immunoprecipitation of mutant p53 from MDA-MB-468 cells were compared to wild type p53 in MCF7 cells. Western blotting for bound p53 revealed that mutant p53 could bind BRCA1, similar wild type p53 in vivo. However, it could not bind to Rad51 in MDA-MB-468 cells.
In vitro studies were performed by using GST-p53 fusion protein which does not posttranslational modification. We determined the affinity of the wild type p53 fusion protein for partner proteins by GST- pull down assays.
Results: The results from GST- pull down experiments indicated that the GST-p53 fusion protein retained its binding capacity for Rad51in MDA-MB-468 and MCF7 cell lines. GST-p53 only binds to BRCA1 in MDA-MB-468 cells.
Conclusions: In conclusion, In vivo binding studies suggested that mutation at codon 273 of p53 did not effect on its binding capacity to BRCA1 protein but not Rad51.Since pervious reports showed that BRCA1 bound physically to GST-p53, therefore the low levels of these proteins in our study may be the reason for our results.
Keywords: p53, MDA-MB-468 cells, MCF7 cells, Rad51, BRCA1

O-1-49687-Characterisation of the expression of GABAA receptor subtypes in the mouse enteric nervous system
Mohsen Seifi[1], Jeremy Mills[1], James Brown[1], Pradeep Bhandari[2], Jerome Swinny[1]
1. School of Pharmacy and Biomedical Sciences, University of Portsmouth, PO1 2DT
2. Portsmouth Hospitals NHS Trust, Queen Alexandra Hospital, Cosham, Portsmouth, PO6 3LY

Background: The enteric nervous system (ENS) is a collection of neurons located in the muscular wall of the gastrointestinal tract (GIT) which provide the intrinsic neural control of vital GI functions such as motility, endocrine, microcirculation and inflammatory processes. Importantly, numerous GI disorders arise due to changes in the activity of the neurons which compose the ENS. Understanding the underlying pathophysiology of such GI disorders is hindered by our lack of insight into the molecules such as neurotransmitters which regulate ENS neuronal function.
Objective: Here, we investigated the expression of GABAA receptor ($GABA_AR$) subunits in the ENS of the mouse colon using immunohistochemistry and confocal microscopy
Materials & Methods: In our ongoing research, we demonstrate the expression of $GABA_A$ α1-5, β1-3, γ1-3 and δ subunits mRNA using reverse transcription PCR. We also show that different $GABA_A$ alpha subunits are localised to neurochemically distinct populations of ENS neurons including those expressing Nitric oxide synthase (NOS), Choline acetyltransferase (ChAT), Serotonin (5-HT) and corticotropin-releasing factor (CRF).
Results: Our data show the expression of $GABA_AR$ subunits, at the mRNA and protein levels, in the mouse enteric nervous system which suggests that these receptors may be involved in the control of enteric neuronal activity.
Conclusions: The functional implications are that different $GABA_AR$ subtypes are responsible for regulating the activity of neurochemically distinct cell-types within the ENS.
Keywords: GABAA Receptor, Mouse Enteric Nervous System

O-1-73638-Co-culture of human bone marrow derived-mesenchymal stem cells with k562 cells conferred resistance to k562 cells against cytarabine treatment
Vadoud Malikzadeh [1], Ahmad Gharehbaghian [1, 3], Ahmad Mehdipour [2], Mehryar Habibi Roudkenar [3*]
1. Hematology Department, Paramedical Faculty, Shahid Beheshti University of Medical Sciences
2. Tissue Engineering and Cell Therapy Department, Faculty of Advanced Medical Technologies, Tehran University of Medical Sciences
3. Blood Transfusion Research Center, High Institute for Research and Education in Transfusion Medicine, *Corresponding Author:* P.O. Box 14665-1157 Tehran, Iran e-mail: roudkenar@ibto.ir

Background: Human mesenchymal stem cells (H-MSCs) are population of non-hematopoietic cells that have many applications in medicine most notably in cell therapy. Despite the many advantages, there are still concerns regarding the use of these cells such as induction of drug resistance.
Objective: This study was performed to evaluate the effects of MSCs on k562 cells (human erythroleukemia cell line) in terms of cytotoxicyty or apoptosis following treatment with a well-known chemotherapy agent i.e. cytarabine
Materials & Methods: H-MSCs were isolated from health volunteers and characterized by flowcytometery. MSCs were co-cultured with k562 cell line in presence of serial concentrations of cytarabine. As a control, K562 cells were also treated with cytarabine without cultivation with H-MSCs. The cell viability was determined by trypan blue exclusion test and MTT assay. Apoptosis was studied by ELISA kit.

Results: Cultivation of MSCs with k562 in the presence of cytarabine conferred resistance to k562.In other words, higher concentration of cytarabine was required to induce cytotoxicity when k562 cells cultivated with MSCs. Cytarabine also induces apoptosis cells in k562 in the concentration of (0.1-0.4μg) however and interestingly apoptosis cells were lower when they co-cultured with MSCs.
Conclusions: This study indicated that MSCs confer resistance to erythroleukemia cell line following treatment with chemotherapy agent and prevent leukemic cells apoptosis. Our finding may open new window why erythroleukemia cell become resistance to chemotherapy and may suggest new strategy for treatment of them.
Keywords: H-MSC, K562 Cells, Cytarabine, Apoptosis

O-1-9103-Evaluation of DNA strand breaks associated with apoptosis in lymphocytes of Phototherapy-treated icteric newborns

Maryam Shahidi[1], Seyed Alireza Mesbah-Namin[2]
1. Department of Biochemistry and Biophysics, Mazandaran University of Medical Sciences, Sari, Iran
2. Department of Clinical Biochemistry, Tarbiat Modares University, Tehran, Iran

Background: The aim of this study was to evaluate the DNA double strand breaks associated with apoptosis in hyperbilirubinemic newborns during continues phototherapy.
Materials & Methods: Neutral comet assay was used to evaluate DNA damage levels and apoptosis value in peripheral blood lymphocytes from 15 healthy full term newborns with physiological jaundice (bilirubin levels < 13 mg/dl) as the control group (not receiving phototherapy) and 30 healthy newborns with non-physiological jaundice (bilirubin levels >13 mg/dl) undergoing 16 hours continues phototherapy (wavelength range was about 480-520nm) as the phototherapy-treated group.
Results: Neutral comet assay results showed that DNA damage significantly decreased in peripheral blood lymphocytes from phototherapy-treated newborns during 24-hours incubation, while apoptosis value significantly increased at the same time.
Discussion : These finding indicated that DNA double strand breaks associated with apoptosis significantly increased after phototherapy in hyperbilirubinemic newborn's lymphocytes compared with the control group. Further studies are needed to evaluate of the phototherapy effects observed in this study on the long-term health in jaundiced newborns.
Keywords: DNA breaks; Apoptosis; Neutral Comet assay; Phototherapy; icteric newborns

O-1-5576472-ICG-001 influence on chemo-therapy resishtance factors in MXT-resistance HepG2.

Ghaffari S
1. Institute of Biochemistry and Biophysics, University of Tehran ,Enghelab Ave., Tehran , Iran P.O. Box: 13145-1384

Background: Hepatocellular carcinoma (HCC) is one the most common cause of death in the world and it treatment failure is still a major problem in scientific circles. Methotrexate has been used for decades to treat psoriasis and some cancers but chemotherapy resistance is a major obstacle in the treatment of liver cancer. ICG-001 is a selective Wnt/ß-catenin signaling inhibitor that binds to the amino terminus of CREB, thus preventing ß-catenin/TCF signaling in the nucleus
Objctive: In this study, we intend to determine the important markers in related to chemo-therapy resistance in HepG2 as a MTX-resistance cell line after it treatment with ICG-001.
Materials & Methods : Cell culture and MTT assay of HepG2 for MXT resistance and ICG-001 treatment, RT-PCR for b-catenine, lgr-5, MDR-1, Hif-1(alpha) and NF-kB factors expression, and western blot analysis of caspase 3, 9, bcl-2 and Livin.
Conclusion: These data indicate targeting Wnt/ß-catenin Pathway with ICG-001 could inhibit apoptosis and also somewhat reduce chemotherapy resistance.
Keywords: ICG-00, chemo-therapy resistance factors

O-1-14903-Differentialexpression of two novel variants of SOX2OT in different types of brain tumors

Youssef Fouani[1], Mahshid Malakootian[1], Alireza Shahryari[1], Hooshang Saberi[2], SeyedJavad Mowla[1*]
1. Department of Molecular Genetics, Faculty of Biological Sciences, TarbiatModares University, Tehran, Iran
2. Department of Neurosurgery, Tehran University of Medical Sciences, Tehran, Iran
* Corresponding author's email: sjmowla@modares.ac.ir

Background: Numerous reports have beenrecently published on the misregulatedexpression of long non-coding RNAs (LncRNAs) invarious cancer types, suggesting that abnormal expression of LncRNAs is a major contributor to tumorigenesis. A broad spectrum of LncRNAs is expressed in central nervous system (CNS), where these transcripts have been shown to play indispensable roles in brain development and function.
Objective: So X2OT isaLncRNA gene with a proposed role in the regulation of neural development. Interestingly, it also embeds SOX2, a master regulator of pluripotency, in its 3rdintron.Based on our previous studies, alternative splicing of SOX2OT generates several spliced variants.
Materials & Methods: In the present study, we investigated the expression pattern of two SOX2OT variants (SOX2OT-S1 and SOX2OT-S2) in three types of brain tumors (Glioma, Meningioma, and Adenoma). Total RNA was extracted from 35 brain tumor samples, and the expression of both S1 and S2 variants was measured using real-time RT-PCR.
Results: Our data revealed a differential expression ofS1 and S2 variants in different brain tumors, in a way that its expression level could discriminate glioma, meningioma and adenoma tumors from each others ($P < 0.05$). In conclusion, the differential expression and splicing of SOX2OT in different types of brain tumors indicates a potential involvement of SOX2OT variants in brain tumorigenesis.
Conclusions: Our data also highlights a potential usefulness of SOX2OT variants in molecular diagnosis and classification of different types of brain tumors.
Keywords: Long non-coding RNAs, SOX2OT, Brain Tumors, Tumor Marker

O-1-46693-Estrogen receptor-(gers2228480polymorphism among Iranian women with breast cancer

Sakineh Abbasi[1], Samira Kalbasi[2], Cyrus Azimi[3],Fariba Nabatchian[4]
1. Department of Medical Biotechnology, School of Allied Medicine, Tehran University of Medical Sciences, Tehran, Iran
2. Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran
3. Department of Genetics, Cancer Institute, Imam Khomeini Hospital Complex, School of Medicine, Medical Sciences /University of Tehran, Iran
4. Department of Medical laboratory Sciences, School of Allied Medicine, Tehran University of Medical Sciences, Tehran, Iran

Background: Receptor-mediated estrogen activation participates in the development and progression of breast cancer. Evidence suggests that alterations in estrogen signaling pathways, including estrogen receptor-a (ESR1) occur during breast cancer development. ESR1 gene polymorphism has been found to be as-