Genetic Recources

O- 53: Venom gland Transcriptomic analysis of Iranian yellow scorpion â€œOdontobuthus doria'revealed some new findings by medical purposes

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Introduction: Scorpion venom contains mixture of biologic molecules including selective toxins with medical capability. Odonthubuthus doriae (O.doriae) belonged to Buthidae family of scorpions and gained more interest among Iranian dangerous scorpion since 2005. The envenomation of this scorpion causes usually neurological signs because of existence of toxins affecting on ion channels.
Material & Methods: total RNA was isolated from yellow Iranian scorpion glands. A cDNA library was achieved by synthesize and insertion of dscDNA into special vectors and subsequent transformation to chemical competent E. coli as host. Library was screened by culturing of the liquid library on LB-agar plates. analysis of positive clones was performed by plasmid extraction and sequencing of inserts. Finally, sequences have been analyzed and characterized by bioinformatics software each.
Results: Analysis showed that toxins (42% of ESTs) had more venom transcripts than other venom components (antimicrobial peptide (10%), cell proteins (11%) and venom peptide (13%)) that may have capacity for medical used. Two EST didnâ€™t have any similarity by known scorpion peptides and may be new.
Conclusion: For the first time; we report a comprehensive study of an Iranian scorpion with interesting and novel findings and characterized a new putative sodium channel modifier and a new iron transporter in scorpions by some bioinformatics software, and then predicted their structures and functions.
Keywords: Transcriptome analysis; cDNA library; Venom gland; Iranian scorpion; Odontobuthus doriae

New Technologies and Technological Advances in Genetics

O-54: CRISPR/Cas9 targeting of SNHG15 as an oncogenic lncRNA inhibits cell proliferation capacity in colorectal cancer

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Recently, thousands of long noncoding RNAs (lncRNAs) have been found to be aberrantly expressed in various types of cancers. Since colorectal cancer (CRC) is the third most common human malignancy worldwide, we investigated lncRNAs misregulated in this type of cancer, and identified SNHG15 as a potentially oncogenic lncRNA. By analyzing expression data from CRC patients available on TCGA, we observed that SNHG15 upregulation is an early event in colorectal cancer promotion and its expression is maintained at high levels until later stages. As SNHG15 sequence has two E-box binding motifs for MYC, we analyzed colorectal adenocarcinoma RNA-seq data from TCGA and revealed SNHG15 is upregulated in the samples with high levels of MYC expression. After silencing MYC in a CRC cell line, the level of SNHG15 was decreased significantly. So SNHG15 transcription could be regulated by MYC. Using CRISPR-Cas9 system, we deleted the region containing exons 3 to 5 from SNHG15 and several clones were obtained with decreased expression of SNHG15. Results showed that these clones have low proliferation and colony formation capacity. However, we did not observe any significant changes in cell cycle and percentage of apoptotic cells. To investigate the effects of SNHG15 in vivo, we injected clones to immunodeficient mice and found that the tumors formed by knocked-out cells were obviously smaller and lighter than wild type cells. In summary, these results describe for the first time an important role of SNHG15 in promoting colon cancer and suggest a novel prognostic marker and target for RNA-based therapies.
Keywords: Long non-coding RNA, Colorectal Cancer, SNHG15, CRISPR-Cas9 system

O-55: Targeted next-generation sequencing revealed novel variants in Iranian families with hereditary loss of hearing

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Background: The syndromic and non-syndromic loss of hear-

ing is the most common sensory defect. This disorder is genetically heterogeneous. In recent years, efforts have been made to identify genetic variants associated with this disorder. Now, with the advent of next-generation sequencing (NGS) technology, provides a quick and low-cost approach to the genetic diagnosis of hearing impairments. Here, we use NGS to examine all of 127 known deaf-related genes in 12 Iranian families.

Methods: In this study, we examined 12 Iranian families with hereditary loss of hearing. Mutation screening was performed in 127 known deaf-related genes using NGS. The identified areas were also examined by direct sequencing. Finally, the identified variants were analyzed by SIFT and PolyPhen-2 to predict the effect of variants on protein function.

Results: By Using this approach, we were able to identify the causative gene variants in 7 of 12 families. The pathogenic role of these variants has already been reported. In addition, 7 novel variants were specifically identified in 6 deaf-related genes. These gene variants were included ADGRV1-c.12786C>G, GIPC3-c.265-266insAG, USH1C-c.1659T>A, LOXHD1-c.6463C>T, OTOF-Ex2-Ex47Dup, ALMS1-c.8665_8669delCAAAG and ALMS1-c.9457A>T. Direct sequencing co-confirmed the presence of these novel variants in patients. Analysis of these variants showed that GIPC3-c.265-266insAG and ALMS1-c.8665_8669delCAAAG are likely pathogenic.

Conclusions: In this study, we successfully used NGS technology to screen mutations in the deaf-related genes. Based on our findings, 7 novel variants were found in the deaf-related genes for the first time in Iranian population. These rare hereditary variants should be considered in genetic diagnosis and counseling.

Keywords: Hearing loss, Deafness, Novel variants, deaf-related genes, Next generation sequencing (NGS)

Stem Cell

O-56: Curcumin promote osteogenic differentiation of human bone marrow

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Mesenchymal Stem Cells (MSCs) are considered as therapeutic target for cell-mediated regenerative medicine to treat human metabolic bone diseases. Numerous efforts have been made to promote efficient differentiation of MSCs into osteoblast lineage. Accordingly, epigenetic signatures emerge as a key conductor of cell differentiation. Enhancer of Zeste Homolog 2 (EZH2), a histone methyltransferase appeared to suppress osteogenesis. Curcumin is an osteoinductive natural polyphenol compound supposedly modulates epigenetic mechanisms. The current study aims to address the role of EZH2 epigenetic factor in osteogenic activity of human bone marrow-derived MSCs (hBMSCs) after treatment with curcumin. We isolated MSCs from aspirated bone marrow and characterized for differentiation to mesodermal lineages and cell surface markers. The optimum concentration of curcumin was achieved by MTT assay. The effects of curcumin on cellular behavior of viability and osteogenic differentiation were evaluated at different time points under in vitro condition. Moreover, the expression level of EZH2 was assessed using quantitative real-time polymerase chain reaction (qRT-PCR) after 14 and 21 days. MTT results showed that curcumin at concentrations of 10 and 15 M had no cytotoxic effect and the cells were survived up to 70%. Quantitative-PCR results demonstrated that curcumin significantly enhanced the expression level of osteogenic markers including Runx2, osterix, collagen type I, osteopontin and osteocalcin at day 21. Interestingly, we observed that expression level of EZH2 gene down-regulated in the presence of curcumin compared to control group during osteogenesis. It is proposed that curcumin acts as an epigenetic switch to regulate osteoblast differentiation via decreasing histone methyltransferase EZH2 activity.

Keywords: Curcumin, Epigenetic, EZH2, Mesenchymal Stem Cell, Osteogenesis

O-57: Differential Expression of Long Non-Coding RNA SOX2OT in Gastric Adenocarcinoma.

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Gastric cancer (GC) is the third leading cause of cancer-related death in the world. Dysfunction of long noncoding RNAs (lncRNAs) in GC biogenesis are approved by increasing evidence. SOX2 overlapping transcript (SOX2OT) lncRNA, which harbors SOX2 transcription factor, is aberrantly expressed in different cancers. Materials and Methods: In this study, the expression of SOX2OT was evaluated in 33 matched pair tumor and non-tumor gastric samples and AGS and MKN45 gastric and NTERA2 embryonic carcinoma cell lines by real time PCR.

Results: Our finding revealed a significant decrease in the expression of SOX2OT in gastric tumor samples compared to their matched non-tumor samples (P=0.05). Also SOX2OT showed a lower expression in high grade compared to low grade of gastric malignancy. Furthermore, SOX2OT expres-