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acterized by Electrochemical Impedance Spectroscopy (EIS) and CV.

Results: The hybridization event between the DNA probe and DNA oligomers was determined by changes in Differential Pulse Voltammetry (DPV) signal of the accumulated indicator. Effective discrimination against non-complementary DNA and the point mutation was obtained.

Conclusions: The new biosensor can be detected complementary and non-complementary of amyloenzyme ssDNA.

Keyword DNA biosensor, DNA hybridization, Amyloenzyme gene, Differential pulse voltammetry, Electrochemical impedance spectroscopy, Au nanoparticles

P-2-37039-Optimization of two Iranian RNA extraction kits for extraction of RNA from Gram-negative bacteria

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Background: A good RNA extraction is the base of good cDNA synthesis, RT-PCR, Real-time PCR and cDNA Libraries. Conventional RNA extraction methods cost much time and money, so nowadays a variety of kits which at least reduce time had been developed. Some of these kits have global purposes and are not optimized to special purposes and cause RNA degradation. Gram-negative bacteria produce much quantities of RNA and need special approaches.

Materials & Methods: Here we worked on two Iranian kits, Sinaclon RNXplus and DenaZist RNA extraction kit, which are very common in Iranian researches. Through modification of Cell density, Cell lysis methods, centrifuge duration and chloroform quantity, instructions optimized and tested on *Pseudomonas aeruginosa*. Here is the optimized instruction: 5 ml of bacteria culture in OD₆₀₀ 1.2 centrifuged for 7 minutes in 7000 rpm in 15 ml falcons. The supernatant discarded and 5 ml distilled water added to bacteria cells. After 15 sec vortex, another centrifuge in 7000 rpm for 7 minutes performed. The supernatant discarded and 1 ml kit buffer added to falcon and mixed. The Bottom half inserted in liquid nitrogen for 1 minute. After melt, falcon was in environment temperature for 5 minutes. Procedure followed by Manufacturers instructions.

Results: Quality of extracted RNA is much better than common instruction.

Keywords: RNA extraction, Gram Negative bacteria

P-2-55405-Study on the binding affinity of chromium oxide (VI) to DNA and chromatin: Equilibrium dialysis

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Background: Chromium hexavalent compounds are well known as human carcinogens, but their molecular mechanism is still unclear.

Objectives: The objective of the study was to investigate the binding affinity of chromium oxide (Cr (VI)) to soluble-chromatin and DNA.

Materials & Methods: Soluble-chromatin and DNA were prepared in 10 mM Tris-HCl (pH 7.2) and dialyzed against the same buffer containing various concentrations of chromium oxide using dialysis tubing at room temperature. The equilibrium was achieved within 72 h at 23 °C. The total metal concentration (C_t) and the concentration of free metal (C_f) in the dialysate were measured directly from the absorbance at 350 nm before and after dialysis. The amount of bound metal (C_b) was obtained from $C_b = C_t - C_f$. Binding parameters were determined from the plot of r/C_f versus r according to Scatchard method.

Results: The binding of chromium oxide to chromatin ex-

hibited a cooperative binding pattern as illustrated by the positive slope observed in the lower regions of the binding isotherm. The curve reached a maximum at a value of $r = 0.5$ and decreased in the slope was observed at higher r values. The binding of chromium oxide to chromatin represents a binding constant of $K = 2.4 \times 10^2 \text{ M}^{-1}$ and drawing $\ln(r/n-r)$ against $\ln C_f$ gives a straight line with a slope of the n_H which was 1.8, confirming the positive cooperative binding of the metal to chromatin. The binding isotherm of DNA also exhibits cooperative binding pattern with a binding constant of $K = 0.9 \times 10^2 \text{ M}^{-1}$ and n_H value of 1.7.

Conclusions: The result demonstrates that chromium oxide has almost the same binding affinity to DNA and chromatin which weakens the participation of histone proteins in this processes.

Keywords: Chromium oxide; Equilibrium dialysis; binding affinity; cooperative binding

P-2-56033-Effects of hyaluronic acid on the behaviors of adipose-derived mesenchymal stem cells cultured on decellularized gingival matrix in vitro

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Background: Biological scaffolds derived from decellularized tissues and organs are used progressively in tissue engineering and regenerative medicine. Hyaluronic acid (HA) is a major component of extracellular matrix that regulates cellular behaviors during the morphogenesis, development and regenerative processes.

Objectives: The effects of soaking decellularized gingival matrix in HA on the behaviors of adipose-derived mesenchymal stem cells (AD-MSCs) aimed to be investigated by histological methods in the present research.

Materials & Methods: After decellularization of human gingival tissue pieces using physical methods (slow and rapid freezing) and chemical agents (sodium dodecyl sulfate (SDS) and Triton X-100), washing and sterilization procedures were performed. Scaffolds were then divided into two groups including soaked scaffolds in HA (0.3% solution) and scaffolds without HA as controls. In the next step, scaffolds were seeded by a density of 6×10^3 AD-MSCs/cm² in both groups. The scaffolds histological studies were performed after 1, 2, 3 and 4 weeks.

Results: Histological studies revealed no changing in the cell density but increase in cell migration one and 2 weeks of culture, while a decrease in cell density and migration was observed at 3rd and 4th weeks after culture in both groups but there were no significant differences in cell density and migration between the two groups.

Conclusions: The evaluated concentration of HA in the present study did not show a significant impact on increasing the survival and migration of AD-MSCs.

Keywords: Hyaluronic acid, Mesenchymal stem cells, Gingival tissue, Decellularization, Tissue engineering.

P-2-56457-Improvements of thermal stability in chondroitinase ABC I by site-directed mutagenesis

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