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Molecular Biology and Genetic Disorders

**Poster – [A-10-47-1]**

**Cellular death pattern in recurrent spontaneous abortion (RSA)**

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**Introduction:** Apoptosis or programmed cell death is normal process in which cells perish in an orderly, highly controlled manner so as to sculpt and control an organism's development. Abortion is the termination of a pregnancy associated with the death and expulsion of the embryo or fetus from the womb. In medicine, all terminations of pregnancy not resulting in live birth are defined as abortions. In common parlance, the terms miscarriage or stillbirth are applied to spontaneous (non-induced) abortions. The aim of this study was to investigate cell apoptosis in women with recurrent spontaneous abortion (RSA) of unknown etiology.

**Methods:** Twenty four women with a history of recurrent pregnancy losses with unknown etiology were included in this study. Women with anatomical, hormonal, infectious and genetic causes of RSA were excluded. We compared the percentage of mononuclear apoptotic cells by flow cytometry in these patients with controls. The pattern of cellular death (early apoptosis, late apoptosis and/or necrosis) was evaluated using annexinV-FITC and propidium iodide (PI) staining method by flowcytometry. Flowcytometric data were analyzed using FSC Express 3.0 software.

**Results:** The percentages of early apoptosis were 13% and 6.1% in RSA patients and controls, respectively. The percentages of late apoptosis were 6.4% and 1% in RSA patients and controls, respectively.

**Conclusion:** The results suggested that the higher percentage of early and late apoptosis in lymphocytes of RSA patients compared to control group might reflect uterine apoptosis cells. The percentage of apoptotic cell in lymphocyte may indicate the risk of recurrent pregnancy loss.

**Keywords:** Recurrent spontaneous abortion, Etiology, Apoptosis, Lymphocyte

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**Poster – [A-10-47-2]**

**Assessment of monoclonality in B cell non Hodgkin's lymphoma**

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**Introduction:** Most B-cell malignancies are diagnosed based on morphologic and immunohistochemical criteria. In some cases, however still present a challenge for the pathologist to discriminate between reactive hyperplasia and neoplastic disorders. In such cases, molecular techniques can be used as a helpful diagnostic tool. In this study, we assessed the value of polymerase chain reaction (PCR) technique in determination of the clonality of immunoglobulin heavy chain gene rearrangements for diagnosis of B-cell non Hodgkin’s Lymphoma in paraffin embedded tissue specimens.

**Methods:** DNA was extracted from paraffin embedded tissue of 31 diffuse B-cell lymphoma specimens. Framework 3 to joining regions (FR3/JH) of variable segment of immunoglobulin heavy chain genes were amplified using in house designed degenerate primers. PCR products were analyzed on 15% polyacrylamide gels following AgNo3 staining.

**Result:** Monoclonal rearrangements were identified in 22 of 31 cases (71%) of B-cell lymphoma specimens using FR3/JH primers.

**Conclusion:** PCR analysis, using degenerate primers could be used as sensitive, reliable and valuable diagnostic adjunct to conventional morphological and immunocytochemical evaluation of lymphoproliferative disorders particularly in cases with limitation in quantity and type of diagnostic material like needle aspirates and cellular fluids.

**Keywords:** Non Hodgkin’s lymphoma, Polymerase chain reaction (PCR), Immunocytochemical, (FR3/JH)
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**E.poster – [A-10-102-1]**

**An approach for the prevention of thalassemia in Mashhad**

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Introduction: Thalassemia is an inherited autosomal recessive blood disease that originated in the Mediterranean region. In thalassemia the genetic defect, which could be either mutation or deletion, results in reduced rate of synthesis or no synthesis of one of the globin chains that make up hemoglobin. This can cause the formation of abnormal hemoglobin molecules, thus causing anemia. The basic aim of this study was to identify a suitable molecular approach for prevention of thalassemia in Mashhad.

Material and methods: DNA was extracted from peripheral blood and the whole beta-globin and the entire alpha1 and 2 globin genes were amplified and DNA sequenced. The seven common deletion mutations for alpha-globin genes were investigated.

Results: Molecular basis of thalassemia was investigated in 88 mutant alleles. Eighty different mutations were found including one novel allele. Coincidental a-thalassaemia was found in 16% of cases of thalassemia. Consanguineous marriage and recessive disorders were studied. In 44 couples studied 70% were consanguineous and only 30% were completely unrelated.

Discussion: Over 98% of the diagnoses were done by direct mutation analysis. A multiplex polymerase chain reaction for mutation analysis was used and significantly reduced the total cost and time required for prenatal diagnosis. This pilot study appears to have identified a suitable approach for prevention of thalassemia in Mashhad.

Keywords: Thalassemia, Mashhad, Mutation

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Poster – [A-10-120-1]
Association between L55M paraoxonase-1 gene polymorphism and metabolic syndrome
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Introduction: Paraoxonase-1 (PON1) is a high-density lipoprotein (HDL) associated enzyme that exhibits antioxidant and anti-atherogenic activities. Three polymorphisms within the PON1 gene affect the enzyme activity. Two of them are located at coding region (L55M, Q192R) and the third one (−108C/T) is placed in promoter region. We looked for a possible link between L55M polymorphism and metabolic syndrome (MES) in samples collected from southeast of Iran.

Methods: DNA was extracted from whole blood of 119 patients with MES and 200 healthy controls. The allelic polymorphism at position 55 in the PON1 gene was studied by tetra amplification refractory mutation system-polymerase chain reaction (ARMS-PCR).

Results: We found the risk of metabolic syndrome in patients with MM and LM+MM genotypes of L55M gene is in marginal border (Odds ratio [OR], 1.33; 95% Confidence Interval [CI], 0.76–2.31, P = 0.34 and OR, 1.12; 95%CI, 0.68–1.85; P = 0.73 respectively ) with risk of MES.

Conclusion: The present study suggested that the L55M polymorphism is not a major risk factor for MES in the studied sample.

Keywords: Paraoxonase-1 (PON1), Metabolic syndrome, Gene, Polymorphism

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Poster – [A-10-125-1]
Nitric oxide synthase 3 VNTR (intron 4 a/b) polymorphism association with diabetic nephropathy
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Introduction: Nitric oxide synthase3 (NOS3) is involved in several functions playing important role in development of type 2 diabetes mellitus (T2DM), and insulin resistance (IR). It has been demonstrated that NOS3 modulates peripheral and hepatic glucose metabolism and insulin secretion, playing important role in the evolution of IR and T2DM. The aim of this study was to examine the association between NOS3 intron4 VNTR polymorphism and type 2 diabetes in an Iranian population. Patients with diabetes were diagnosed according to American Diabetes Association Criteria. Five CC of peripheral blood was collected in EDTA tubes from patients with type 2 diabetes attending diabetes clinic. Normal healthy controls were from same population. Then DNA was extracted from WBCs, using salting-out method and PCR was performed to determine allele and genotype frequencies for NOS3 gene VNTR polymorphism. A significant difference was found in genotype frequencies of NOS3 polymorphism between patients and controls (aa + ab vs bb p = 0.02, OR = 2.0, 95%CI; 1.05–3.96). Also allele a frequency was significantly increased in patients with diabetes compared with controls (p = 0.007, OR = 2.1, 95%CI; 1.19–4.08).

Conclusion: We found a significant difference in distribution of NOS3 polymorphic variants at both allele and genotype frequency level in diabetic patients. A allele is a risk factor of type 2 diabetes in an Iranian population.

Keywords: eNOS, Diabetic nephropathy, Polymorphism

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Poster – [A-10-125-2]
Nitric oxide synthase3 VNTR (intron 4 a/b) polymorphism association with diabetic retinopathy
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Introduction: Endothelial derived nitric oxide(NO), which is produced by NOS3 participates in several functions related to the vasodilation and mediates vascular action of insulin, and glucose and insulin delivery to the skeletal muscles. High NO levels and endothelial dysfunction have been observed in patients with type 2 diabetes and their first degree relatives as well as in patients with insulin resistance.