

Genetic variation of antigen B2 among *Echinococcus granulosus* isolates in Tabriz, North West of Iran

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Cystic hydatid disease (CHD) is recognized as one of the most important zoonotic disease in the world. CHD is caused principally by the larval stage of the tapeworm *Echinococcus granulosus sensu lato*. Because of Antibody cross-reaction with antigens from other taeniid cestodes and lack of sensitivity and specificity in CE immunodiagnostic tests; this study designed to find the polymorphism of the Antigen B among animal isolates. The result of this genetic variability is necessary and important for evaluation, application, and standardization of diagnostic tests using AgB. 100 animal hydatid isolates (60 sheep cyst, 40 cattle cyst) collected from domestic animals at slaughterhouses of Tabriz. DNA extracted from protoscolices of sheep isolates and germinal layer of cattle isolates. All sheep isolates gave similar patterns of PCR-RFLP after digestion with AluI. The results of RFLP pattern showed high degree of genetic likeness in each strain. Amplification to the DNA samples extracted from germinal layer of cattle isolates not completed by using Eg AgB2 specific primer suggesting substantial level of inter-strain variation in AgB2 related genes. Therefore, In endemic regions and countries serologic evaluation and genetic variability of AgB prepared from different *E. granulosus* hosts must be considered.

Key words: *Echinococcus Granulosus*, Antigen B2, Genetic Variability.

The clinical report of cystic urolithiasis and surgical treatment in Caspian Miniature horse

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A 3-year-old, 300 kg body weight Caspian pony stallion has been referred to large animal clinic of faculty of Specialized Veterinary Sciences of Science and Research Branch of Islamic Azad University with urinary incontinence, tenesmus, hematuria, general weakness, dehydration and disquiet. After physical examination and transvaginal ultrasonography, a hyperechoic mass was observed in the bladder. The patient was prepared for cystotomy from ventral midline celiotomy approach. The bladder was incised and a relatively large (type II) and rigid urolith was found. It was removed gently from mucosa and sent it to laboratory. Based on laboratory analysis, the composition of urolith included 50% Calcium oxalate, 30% Calcium and 5% Ammonium was reported. After 10 days and appropriate postoperative care, signs of recovery were observed. The correct diagnosis of urolithiasis with early and standard surgical intervention may prevent urinary blockage and problems in Caspian Miniature horses.

Key words: Cystic urolithiasis, Calcium oxalate, Cystotomy, Caspian Miniature horse.

Cloning of *Mycoplasma agalactiae* P40 gene in prokaryotic system

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The P40 gene encodes adhesion protein that is expected to be involved in adhesion of *M. agalactiae* to eukaryotic cell and promote host colonization. Lipoprotein P40 is an immunodominant antigen and a vaccine candid. The target of this study is recombinant subunit vaccine, so P40 gene was cloned in prokaryotic system. In this study, the P40 gene of three vaccinal isolated *M. agalactiae* strains in Iran was amplified and sequenced. After changes, the selected sequence of P40 gene was synthesized and cloned in cloning plasmid, then transformed into *Escherichia coli* by heat shock protocol. After amplification, P40 gene was digested by two enzymes. Purified gene was cloned in expression plasmid for transforming into expression bacterial cell. The results showed that 920 bp fragment of P40 genes from three vaccinal strains was amplified, so three strains have P40 gene. Complete sequence of this gene was 1092 bp, was extracted from cloning plasmid and cloned in pET22b+ expression plasmid including T7 promoter and terminator. Finally, recombinant plasmid was transformed into bacterial cell. P40 gene cloning was first step in recombinant subunit vaccine production.

Key words: P40 Gene, *Mycoplasma Agalactiae*, Recombinant Subunit Vaccine, Cloning

Experimental study on inhibitory effects of Tomato (*Solanum lycopersicum*) pulp on oxidative stress in DMBA-induced skin carcinoma in the mouse

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Skin carcinogenesis is a malignant growth of the epidermis and the most common types of skin cancer are basal cell carcinoma and squamous cell carcinoma. Cancer chemoprevention is the use of natural, synthetic or biological substances to reverse or prevent the development of cancer. Tomato (*Solanum lycopersicum* L.) because of its lycopene and bioflavonoids contents has anti-carcinogenic effects. For this purpose, 50 mice were randomly divided into five equal groups including: Normal control, Carcinogen control and *Solanum lycopersicum* -treated Groups (SI 10, SI 20 and SI 40). Tomato treated groups and Carcinogen control mice received three topical applications of 7,12dimethylbenz[*a*]anthracene (DMBA) followed by croton oil on shaven dorsal skin for 12 weeks. Normal control mice received topical skin applications of the vehicle, acetone, only. Tomato pulp was simultaneously gavaged to three groups of mice at the doses of 10, 20 and 40 ml/kg. At the end of experiment, the activities of antioxidant enzymes superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and glutathione reductase (GR) in liver tissue from all groups were assessed. Rats treated with DMBA showed significant decline in antioxidants, and elevated lipid peroxidation index in liver ($p < 0.05$). Tomato pulp treatment significantly reduced lipid peroxidation product (MDA), and brought back the liver antioxidants in a dose dependent manner ($p < 0.05$). The results obtained showed, tomato inhibits DMBA-induced skin carcinoma in mice through the induction of cellular antioxidant defense systems.

Key words: *Tomato, Oxidative Stress, Skin Carcinoma, DMBA, Mouse*

Immunohistochemical study and survey on P53 gene alternations in ovine pulmonary adenomatosis

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P53 is a protein associated with the DNA and is a tumor suppressor gene. TP53 is encoded in human. This is a critical regulator of cellular proteins in organisms with cell cycle and tumor suppressor function as it is meant to prevent cancer. As protector of the P53 gene in the genome or arrowhead or the protection seen above is real dodgy genome, all of which suggest a protective role of P53 in preventing DNA mutations. P53 gene is a best place for mutation in all type of human cancer like Lung cancer in this study in a period of 90 Days we visited 5300 sheep lung in Kurdistan slaughterhouses. The Lungs that have lesions (5.6% of the total lung) like hydatid cyst, white fibrouse and adenomatous nodules choose for sampling. The sample are taken from superficial, deep and marginal edge of each lung with lesions and stained with hematoxylin and eosin, 15 samples had adenomatous, for determining mutation in P53 we block the pulmonary adenomatous positive with Immuno-histochemistry method, nine samples have this mutations in our study. The frequency of this mutation was 60% in positive cases. This study showed that sheep lung adenomatous in P53 gene mutation in 25 – 40% with human lung adenocarcinoma cases are similar.

Key words: P53, Mutation, Adenomatous Lung, Sheep

Histopathological study on sub-acute toxicity of paraquat on liver of benny fish fingerling (*Barbus sharpeyi*)

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Agricultural pesticides and specially herbicides are one of the most important water ecosystems pollutants. Aim of this research is histopathological study of sublethal effects of Paraquat in liver of *Barbus sharpeyi* fingerlink. 60 fishes were divide into 3 aquarium had the following concentrations 25%, 50% and 75% of LC₅₀ 96h and one other was contained control fishes and exposed to paraquat for 96 hours. No mortalities occurred in any group during the experimental period. Then samples were fixed in 10% formalin-salin, 5µm sections were prepared and stained with haematoxylin and eosin and studied by light microscope. Histopathological lesions in exposed fishes contained nuclear conformation, pyknosis and necrosis, vacuolated cytoplasm, margination, haemorrhage in hepatic blood vessels that were sever in upper levels of paraquat. These finding showed that paraquat have toxic effects in *Barbus sharpeyi* and result to histopathologic lesions in liver. These observations was same with findings in other researches.

Key words: Hepatic Lesions, Sublethal Toxicity, Paraquat, *Barbus Sharpeyi*

Evaluation of Freeze-Thawed Ghezel ram's spermatogonial cells colonization after co-culture with sertoli cells.

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Spermatogenesis is a complex and coordinated process that is dependent upon the type of stem cells known as spermatogonial stem cells. Few studies on culture of sheep spermatogonial stem cells are available. In this study, 3-5 tissue samples autopsied from testis of 2 month old male Ghezel rams after two stages of enzymatic digestion were divided into four groups: Group 1: 'Spermatogonial cells co-cultured with sertoli cells for 10 days, freeze-thawed in day 0'; group 2: 'Spermatogonial cells co-cultured with sertoli cells for 10 days, freeze-thawed in day 14'; group 3: 'Freeze-thawed spermatogonial cells in day 0 cultured for 10 days'; group 4: 'Freeze-thawed spermatogonial cells in day 14 cultured for 10 days' and control group: 'Spermatogonial cells co-cultured with day 0 fresh sertoli cells for 14 days'. The rate of colonization and colonies diameter were evaluated during these 10 days. Groups 3 and 4 did not show any signs of colony whereas in group 1 a higher rate of colonization was observed comparing to group 2 ($P < 0.05$). It can be concluded that freezing ram's spermatogonial cells is a proper way for their long-term preservation and after thawing these cells maintain their colonization ability *in vitro*. The best time for freezing sheep spermatogonial cells is after enzymatic digestion (day 0) and culturing the cells prior freezing leads to a decrease in their colonization ability.

Key words: *Spermatogonial Stem Cells, Sheep, Co-Culture with Sertoli Cells, Colonization*

The study of the prevalence of class I integrons and virulence plasmid in clinical strains of *Salmonella* Typhimurium

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Certain serovars of *Salmonella*, belonging to subspecies I of *Salmonella enterica*, carry virulence plasmid of *spv* operon carrier. This operon along with other genes has a major role in creating virulence and drug resistance. Considering the importance of the issue, this study examines the prevalence of class I integrons and virulence plasmid in clinical strains of *Salmonella* Typhimurium. 663 stool samples were collected from hospitals in Tehran. After performing biochemical tests on the samples, 37 genuses of *salmonella* were isolated. Then using specific primers ST11, ST15 and Fli15, Typ04 to identify genus and species respectively, 12 isolates from the total of 37 *salmonella*, were confirmed as *salmonella* Typhimurium. Finally in order to investigate the presence of class I integrons and virulence plasmid, Multiplex PCR test was performed, using Int and Vir primers. The results showed that from the total of 12 isolates, 4 isolates (33.3%) had the genes related to class I integron and 5 isolates (41.7%) had the virulence plasmid. The presence of virulence plasmid genes and class I integrons among Typhimurium strains isolated in this study shows that, considering the horizontal transfer of these genes, these strains can cause serious problems in public health, community health and treatment.

Key words: *Salmonella* Typhimurium, Virulence Plasmid, Integron.

Investigation of antioxidative properties of protein hydrolysate obtained from waste, in the Salmon (*Salmo salar*) filleting operation

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In this study antioxidant properties of Salmon (*Salmo salar*) filleting waste protein hydrolysates produced by trypsin was investigated. Protein hydrolysates under two different conditions of time and amount of enzyme and same temperature produced. Proximate composition, degree of hydrolysis, DPPH free radical scavenging activity, metal chelating activity and reducing power was measured. Maximum content of protein, minimum content of lipid, moisture and ash were $89/74 \pm 0/0$, $0/6 \pm 0/01$, $5/7 \pm 0/2$ and $3/5 \pm 0/2$ for the hydrolysates respectively, which with salmon waste has been significant different ($p < 0/05$). Maximum degree of hydrolysis, DPPH free radical scavenging, metal chelating activity and reducing power were $81/78 \pm 0/48$, $93/99 \pm 4/56$, $12/8 \pm 0/6$ and $0/42$, for the protein hydrolysates respectively. In conclusion say enzymatic hydrolysis of salmon wastes generated peptides with antioxidant properties and could be used as food additives after clinical approve.

Key words: Trypsin, Protein Hydrolysate, Salmon, Free Radical

Seroprevalence of bluetongue virus infection in sheep in Khoy in Iran by Competitive ELISA

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Bluetongue is one of the viral disease in ruminants. The type species of the genus Orbivirus within the family Reoviridae. It is transmitted by colicoides. This study was conducted on 200 sheep blood samples from 19 sheep flocks in 7 villages of Khoy in the West- North of Iran. 160 sheep are ewe and 40 are male. The objective was describing the prevalence and distribution of serum antibodies to Bluetongue virus (BTV) in a sample. Competitive ELISA was applied to detect antibodies. 134 samples (67%) were positive and 66 samples (33%) were negative. 23 of males and 111 of females were positive (57.50% and 69.37%, respectively). The difference prevalence of antibodies in serum between male and females was not significant($p>0.05$). The relationship between prevalence antibodies in serum and age-groups was significant ($\phi= 0.59$) and ($p<0.05$). From this study it is concluded that the bluetongue antibodies presence in the sheep sera from Khoy sheep flocks and can to create a disease.

Key words: *Seroprevalence, Bluetongue Virus, Sheep, Competitive ELISA, Khoy, Iran*