

k21, k22 values for the Tryptophan aqueous solution and tryptophan solutions in the presence of different concentrations of SDS.

**Keywords:** Microscopic Dissociation Constants, Tautomeric Ratios, Tryptophan, Sodium Dodecyl Sulfate.

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#### Abstract No.189

##### Probing Crucial Amino Acids Role in Chondroitinase ABCI Enzyme Activity by Site-directed Mutagenesis

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Chondroitinase ABC I is an 112.5 kDa enzyme with 997 amino acid. This enzyme is a GalAG (galactosaminoglycan) depolymerizing lyase with a broad-specificity and depolymerizes a variety of GAG substrates, including C4S (chondroitin 4-sulphate), DS (dermatan sulphate), C6S (chondroitin 6-sulphate) and hyaluronic acid. Previous studies suggested that cABC1 enzyme promote neural system regeneration by degrading GAG chains followed by decreasing CSPG s inhibitory effect so there is a wide range of application of the enzyme in medicine. Cloning and expression of the enzyme have been done successfully in BL-21. Three Dimensional structure analysis with x-ray crystallography suggested that the enzyme has three major domains. The optimum pH and temperature in Tris buffer is pH=8 and 37 °C, respectively. In order to increase thermal stability we applied quick change mutagenesis. We use a thermophile strain *Thetaiotaomicron* WAL2926 as a template to guide us the best selection of amino acids for enzyme engineering. In this study, proline 485 has been substituted with alanine. Finally the activity and stability of the wild type and its variant has been discussed and proven.

**Keywords:** Chondroitinase ABC I, Galactosaminoglycan, Site Directed Mutagenesis Stability.

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#### Abstract No.190

##### Monoclonal Antibodies in the Treatment of many Diseases in Mice Experimentally Using Veterinary Clinic of Sina Gilan, Iran

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Monoclonal antibodies (MAB) have found increasing use experimental therapies. Great limitation of their use is that they are recognized by the patient as being of foreign origin and an antiglobulin response is provoked. Recombinant DNA technology offers the ability to convert these rodent antibodies into a more human form. There are currently several different strategies which can be adopted to generate humanized antibodies resulting in different degrees of humanization can be achieved ranging from chimeric antibodies with a combination of human constant regions with rodent variable regions to fully reshaped antibodies where the variable regions are also humanized. At present the available data on clinical use of chimeric and reshaped antibodies is very limited. The rat IgG2b antibody CAMPATH-1G has been shown to be both useful as an immunosuppressive antibody as well as in the treatment of lymphoid malignancies. The reshaped version, CAMPATH-1H, was successfully used to clear detectable malignant cells from the blood and bone marrow in two patients with B-cell lymphoma. A more sustained course of the human antibody (126 mg over 30 days and 86 mg over 43 days) was tolerated than had previously been used for the rat antibody. In the future in veterinary clinic of Sina Gilan, it is clear that a majority of monoclonal antibodies produced for therapy will be humanised for the reasons discussed above. As far as improvements in the abilities of these antibodies to interact with human effector mechanisms goes it seems that there is unlikely to be any major differences between chimeric and fully reshaped antibodies. Important about humanised antibodies is whether there are any sequences contained within the variable region frameworks, complementary determining regions or constant region allotypes which can be processed and presented as T-cell epitopes.

**Keywords:** Monoclonal Antibody, Chimeric Antibody, Recombinant DNA Technology, Humanized Antibody, Antiglobulin.

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