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physical and chemical properties which makes it suitable polymeric supports for enzyme immobilization. Firstly, Polyaniline polymer was activated with glutaraldehyde as a bifunctional agent and then LPO was immobilized on it by covalent bonding. After that the polyaniline polymer surface topography with enzyme immobilized on it and bare polyaniline polymer surface topography were investigated with AFM.

**Results and Conclusions:** The AFM images demonstrate significant difference between bare polyaniline polymer surface topography and enzyme-polyaniline surface topography which establish that the process of enzyme immobilization was performed successfully.

**Keyword:** Immobilization, Lactoperoxidase, Polyaniline polymer, AFM

#### P-2-63915-Histological study of interactions between scaffolds prepared from mice kidneys and mesenchymal stem cells

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**Background:** International reports suggest that graft shortage has become the world's social crisis and a large number of dialysis patients who do not have the possibility for transplantation lose their lives. These limitations have increased efforts to rebuild tissues by tissue engineering techniques. Extracellular matrix (ECM) is a dynamic and vital compartment of all tissues and organs. Decellularization is a promising technique to produce natural scaffolds for tissue engineering.

**Objectives:** In this study, for preparation of natural scaffolds, physical and chemical procedures were employed.

**Materials & Methods:** In physical method, slow freezing and freeze-thaw were used and in chemical method kidneys were treated with 1% sodium dodecyl sulfate (SDS) for 48 hours. After sterilizing the scaffolds, they were cultivated with adipose derived mesenchymal stem cells (ASCs) for 25 days and studied histologically by different staining methods.

**Results:** After staining and observing the paraffinized sections with light microscope, it was shown that cell components were completely removed from the scaffolds. The observations from the scaffolds seeded with MSCs showed attachment and migration of the cells into the decellularized kidneys.

**Conclusions:** The results indicate that the scaffolds prepared from mice kidneys could induce proliferation, migration and probably, differentiation of ASCs. Further studies are required to confirm these results.

**Keywords:** Scaffold, mice kidney, Mesenchymal stem cells, Extracellular matrix, Decellularization

#### P-2-81712-Biodiesel production by immobilized lipase of *Aspergillus oryzae* from Canola and olive oil

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**Background:** Biodiesel is an alternative diesel fuel because of its favorable properties, environmental benefits and the fact that is derived from the renewable biological resources. Biodiesel is the only alternative fuel. Biodiesel is produced through a reaction known as transesterification such as enzymatically and chemically. Some advantages of lipase over the chemical-catalyzed reactions include no by-products, easy product removal, mild

reaction conditions and catalyst recycling. Immobilization of lipases has several advantages for industrial applications.

**Method and Materials:** Lipase immobilized by adsorption Anion-exchange method on the resin. And transesterification process carried out in three temperatures 25, 37 and 45°C in four molar ratios of oil: alcohol, 1:2, 3, 4 and 5 with agitation speed of 200 rpm for 60 hours. Alcohols molar ratios add step by step. Canola and olive oil methanol and ethanol used as substrate. Produced biodiesel analyzed by Gas chromatography method. Methyl/ ethyl myristate use as internal standard.

**Result:** Maximum yields of biodiesel for canola and olive oil obtained at 45°C at 1:4 molar ratio of oil: alcohol for methanol were 90% and 93% and for ethanol were 85% and 87% respectively. Temperature and molar ratio of oil: alcohol, have positive effect on biodiesel production. When used immobilized lipase less amount of enzyme needed and recovery of enzyme from reaction mixer is possible.

**Conclusions:** Result showed that transesterification of canola and olive oil with methanol had better yield than ethanol.

**Keyword:** Biodiesel, Immobilized lipase, Canola oil, Olive Oil, Gas Chromatography

#### P-2-98539-Purification and kinetic studies of recombinant benzoate dioxygenase from *Rhodococcus sp*

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**Background:** Benzoate dioxygenase from *Rhodococcus ruber* UKMP-5M was able to catalyze the oxidation of benzene ring to catechol and derivatives. Benzoate dioxygenase catalyzes the NADH-dependent oxidation of benzoate to less toxic compounds such as 1-carboxy-1,2-cis-dihydrocyclohexa-3,5-diene.

**Objectives:** The objective of this study was purification and characterization of recombinant benzoate dioxygenase from *Rhodococcus sp*.

**Materials & Methods:** The purified benzoate dioxygenase was characterized kinetically. The end product of benzoate dioxygenase reaction was identified by gas chromatography mass spectrometry (GC-MS).

**Results:** The gene *benA* from *R. ruber* UKMP-5M was amplified at 64°C. The PCR product was expressed into *Escherichia coli* as a protein expression host. The bacterium was induced with 0.5 mM isopropyl  $\beta$ -D-thiogalactoside (IPTG) at 22°C to produce benzoate dioxygenase. The crude protein was analyzed by sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) and western blotting. The benzoate dioxygenase was purified by ion exchange chromatography. The size of purified benzoate dioxygenase from *R. ruber* UKMP-5M was 25 kDa. The benzoate dioxygenase activity was 5.21 U/mL. The optimal pH and temperature were 8.5 and 25°C, respectively. The maximum velocity ( $V_{max}$ ) and Michaelis constant ( $K_m$ ) were 7.36 U/mL and 5.58  $\mu$ M, respectively.

**Conclusions:** As was identified by GC-MS analysis, cyclohexane dione was the product obtained from the benzoate dioxygenase reaction.

**Keywords:** Benzoate dioxygenase - *Rhodococcus ruber* - Purification - Kinetic

#### P-2-2064951-Partial purification and characterization of anticoagulant factor in *Echiscarinatus* venom

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