

have very similar capacities of polymorphism detection in the alfalfa genotypes. The dendrogram groups the alfalfa genotypes analyzed into three groups.

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Genetic diversity of blackberry biotypes from the west area of Romania



Danci Marcel, Trinc Diana*, Petolescu Cerasela, Velicevici Giancarla, Danci Oana, David Ioan, Dragunescu Anca, Becherescu Alexandra, Alda Simion

Banats University of Agricultural Sciences and Veterinary Medicine, Timisoara 300645, Romania

E-mail address: trincdiana@yahoo.com (T. Diana).

The main objective of our study was focused on the assessment of the intra and interspecific variability of seven *Rubus fruticosus* genotypes, collected from the west area of Romania. RAPD technique is used to assess diversity within and between genotypes. We tested 16 RAPD primers and used 13 of them. The researches are focused on two major directions: assessment of the variability induced by in vitro culture and assessment of intergenotypical variability at molecular level. Axillary buds were used as explants. The greatest number of shoots was obtained with MS medium containing 2 mg/l BA and 0.5 mg/l GA3. Between regenerants and explants-donor plant there were no differences at the molecular level. The results achieved by us prove the applicability of these techniques in the analysis of somaclonal variation in the regenerated plants in *Rubus*. RAPD marker data proved to be a good method of assessing genetic relatedness among *Rubus* genotypes. The results revealed a high genetic polymorphism among different studied blackberry genotypes. Genetic similarities were calculated by the Dice formula. Construction of dendrograms was derived from the application of the UPGMA.

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The effects of explant type and plant growth regulators in *Hypericum perforatum* L. on callus induction and plant regeneration



Burcu Çetin*, Halil Ibrahim Koçak, Betül Kurtuluş

Dumlupınar University, Faculty of Art and Sciences, Department of Biology, Kutahya, Turkey

E-mail address: burcu.cetin@dpu.edu.tr (B. Çetin).

An efficient protocol of callus induction, multiple shoot regeneration and rooting stages was developed for in vitro propagation of *Hypericum perforatum* L. Being sterilized, the seeds of the *Hypericum perforatum* were germinated on the MS (Murashige and Skoog, 1962) medium. Shoot tips, nodes and leaves of the six-week-old plants were used as explants for indirect organogenesis studies. For callus initiation, explants were inoculated into solid MS medium supplemented with 6-benzylaminopurine (BAP; 0.5, 1 or 2 mg/l) or kinetin (0.5, 1 or 2 mg/l) in combination with 2,4-dichlorophenoxyacetic acid (2,4-D; 0.5 mg/l or 1 mg/l). The best callus initiation was obtained from node explants cultivated in the presence of 0.5 mg/l BAP and 0.5 mg/l Kinetin. Following the callus formation, the formed calli were cultured on the MS medium with different concentrations of BAP (1–2–3 mg/l) for shoot regeneration. The best shoot regeneration was observed in the leaf explants in eight weeks following the transfer of calli to the MS medium supplemented with 2 mg/l BAP. For rooting, the shoots were transferred to the

MS medium containing IBA, NAA and IAA (0.5–1 mg/l). The highest rooting (80%) was obtained on the MS medium with 1 mg/l IBBA.

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Renewables, biorafinery, bioenergy, biofuels, bioproducts

Sulfonated polysulfone and polyvinylidene fluoride membranes for direct methanol fuel cell



Monchai Pianmanakij*, Anuvat Sirivat, Kitipat Siemanond

Department of Petroleum, Petroleum and Petrochemical college, Chulalongkorn University, Bangkok, Thailand

E-mail address: monchaip.p@gmail.com (M. Pianmanakij).

Fuel cell is renewable alternative energy source which converts fuel into electricity. The direct methanol fuel (DMFC) consists of methanol, water and by products namely water and CO₂. Proton Exchange membrane (PEM) is a solid polymeric electrolyte to use in DMFC. The membrane supports proton transport between the cathode and anode. Nafion is widely used as the membrane in DMFC because it high proton conductivity and mechanical strength. Nevertheless, it has high methanol permeability and cost. High methanol permeability leads to a decrease in DMFC performances. Therefore, alternate polymer membranes were developed in this work, namely polysulfone and polyvinylidene fluoride. The polymer membranes were prepared for use as PEMs by a sulfonation process to improve electrical properties and characterized for the degree of sulfonation, ion exchange capacity, proton conductivity, water uptake and mechanical strength. The degree of sulfonation increased with increasing ratio of acid per polymer and affected both proton conductivity and water uptake. The properties of membranes were compared with a commercial membrane Nafion.

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Acinetobacter calcoaceticus, *Enterobacter asburiae* and *Pseudomonas aeruginosa* as producers of rhamnolipids



Alena Cejkova^{1,*}, Olga Schreiberova¹, Richard Jezdik¹, Josef Chudoba², Vladimir Jirku¹, Tomas Rezanka¹, Jan Masak¹

¹ Department of Biotechnology, Institute of Chemical Technology Prague, Czech Republic

² Laboratory of Mass Spectroscopy, Institute of Chemical Technology Prague, Czech Republic

E-mail address: alena.cejkova@vscht.cz (A. Cejkova).

Rhamnolipids are representatives of highly surface active biosurfactants which exhibit low toxicity, good biodegradability and high stability in a wide range of temperatures, pH and salinity. In addition to the traditional rhamnolipid producer *Pseudomonas aeruginosa*, we have demonstrated the possibility of using other microorganisms as producers of the important product usable in many industrial branches and in bioremediation of contaminated environment. Bioreactor cultivations with *Acinetobacter calcoaceticus*, *Enterobacter asburiae* and *P. aeruginosa* provided rhamnolipids production up to 4.5 g/l. The produced rhamnolipid congeners were analyzed by tandem mass spectrometry with negative electrospray ionization. Mono-rhamnolipid and di-rhamnolipid homologues containing one or two saturated or monounsaturated 3-hydroxy

fatty acids were found in all strains. Physiochemical characterization of rhamnolipids was evaluated by the emulsification test, oil displacement test and phenanthrene solubilization. The composition and properties of the produced rhamnolipids using above mentioned strains were different, which allows preparing appropriate mixtures depending on the intended applications. We have also demonstrated that all the applied producers allow the bioreactor cultivation in mixtures (ternary or dual cultures) while maintaining stable rhamnolipid production. It was confirmed that this is another way to prepare rhamnolipids of desired properties.

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Cyanobacterial strains, isolated from extreme conditions sources of Kazakhstan – Producers of biodiesel

Bolatkhon Zayadan^{1,*}, Fariza Sarsekeyeva¹, Kirill Mironov², Aizhan Userbayeva¹, Dmitry Los²

¹ Department of Biotechnology, Al-Farabi Kazakh National University, Almaty, Kazakhstan

² Laboratory of Intracellular Regulation, Institute of Plant Physiology, Russian Academy of Sciences, Moscow, Russia

E-mail address: zbolatkhan@mail.ru (B. Zayadan).

In recent years a systematic and orderly mastering new cultures of cyanobacteria for further practical use in biotechnology. In this regard, the aim of this work was the isolation, identification and determination of fatty acid composition of new strains of cyanobacteria from extreme conditions sources of Kazakhstan. Samples were isolated from lake Issyk (13 °C), hot spring Turgen (45 °C) and salinity lake Balkhash (salinity 4 g/l). The standard techniques of cultivation of phototrophic microorganisms were used. Fatty acids were derived using methyl esters and separated by GC–MS Ali-gent 7890GC. The results showed that 3 different strains were obtained: sp2 from Issyk, sp1 from Turgen and sp1 from Balkhash. Morphological characteristics and 16S rDNA gene were identified those strains as *Synechococcus elongatus*, *Cyanobacterium aponinum* and *Cyanobacterium stanieri*. Strain *Synechococcus elongatus* sp.I2 synthesized saturated 26.9% (generally palmitic) and unsaturated 73.1% (oleic, lenoic and lenoleic) FA. In *Cyanobacterium aponinum* sp.T1 strain advantageously synthesized saturated 62.8% (myristic and palmitic) and monoenoic 37.1% (palmitoleic) FA. Cells of strain *Cyanobacterium stanieri* sp.B1 characterized by accumulation mainly of saturated and monounsaturated FA thus dominated myristic (30.1%) and palmitoleic (39.7%) acids. All isolates based on the composition of the FA can be used as producers for the production of biodiesel.

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Environmental benefits of seaweed biomass as a bioenergy feedstock

Seong Rin Lim^{1,*}, Hye Rim Park², Kyung A. Jung², Ji Hye Jo³, Jong Moon Park²

¹ Department of Environmental Engineering, College of Engineering, Kangwon National University, Chuncheon, Republic of Korea

² School of Environmental Science and Engineering, and Division of Advanced Nuclear Engineering, POSTECH, Republic of Korea

³ Division of Resource Circulation, Korea Environment Institute, Republic of Korea

E-mail address: srlim@kangwon.ac.kr (S.R. Lim).

Bioethanol is produced from a diverse range of biomasses. Recently, many interests have been taken in marine seaweed as a new bioethanol feedstock. Thus, the objective of this study is to quantify the bioenergy potential of marine seaweed. We estimated potential ethanol yields of marine biomass. Red seaweeds have higher ethanol yields than brown seaweeds, even though realistic productivity of brown seaweeds was higher than for red seaweeds, due to advanced farming technology. For carbon-neutral ability, marine seaweeds have negligible carbon emissions in biomass production, compared to terrestrial biomasses, due to the difference for land use. Seaweed production has higher carbon sequestration potential than for terrestrial crops. The seaweed-based bioethanol does not require irrigation water in farming and can reduce the use of water in the fermentation process by using seawater. Marine seaweeds do not need fertilizers and pesticides in farming. Also, the seaweed-based ethanol helps establish the nutrient (nitrogen and phosphorus) cycles in nature by returning the nutrients in the seaweed fermentation residue to terrestrial ecosystems, due to high nutrient composition. Therefore, this study can contribute to examining the potential of marine seaweed as a sustainable bioenergy feedstock.

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Synthesis of methyl ester sulfonate surfactant from palm oil methyl ester by using UV or ozone as an initiator

Hattaya Tulathammakit*, Boonyarach Kitiyanan

The Petroleum and Petrochemical College, Chulalongkorn University, Bangkok 10330, Thailand

E-mail address: hattaya.tula@gmail.com (H. Tulathammakit).

Methyl ester sulfonate (MES) is a potential anionic surfactant+derived from palm oil methyl ester. MES shows several advantages, such as excellent cleaning properties, good biodegradability, and tolerance to hard water. Although alpha methyl ester sulfonate (α -MES) is commercially available, it still has some drawbacks, such as disalt formation and low water solubility. In order to overcome these drawbacks, MES was synthesized via sulfoxidation by using different initiators. Ozone is one of the most interesting initiators because of its highly oxidizing power, which can react with organic compound rapidly under mild conditions. In this work, the starting methyl ester was comprised of 36% C16 and 64% C18. The reaction was performed at 40 °C under atmospheric pressure in a photochemical reactor with 16 UV lamps (253.7 nm). The outlet product was transferred to separation and purification processes by using liquid extraction techniques. The structure of MES was confirmed by FT-IR and ESI-MS. When reaction time was fixed at