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Poster Presentations

Nicolae Levandovschi et al., J Biotechnol Biomater 2017, 7:6 (Suppl)
DOI: 10.4172/2155-952X-C1-086

Rom-Agrobiofertil NP- biological fertilizer for a sustainable agriculture

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Statement of the Problem: Evolution of modern agriculture has led to extensive use of agricultural chemicals which have degraded the soil fertility, soil profile and rendered the soil unsuitable for plant growth. Biofertilizers are being recognized as an essential component for increasing the sustainable agricultural productivity and maintaining long term soil fertility in an eco-friendly manner. Rom-Agrobiofertil NP is a biological fertilizer based on three bacteria belonging to *Azospirillium lipoferum, Azotobacter chroococcum* and *Bacillus megaterium species*. Rom-Agrobiofertil NP contains living microorganism which, when applied to seed, plant surfaces, or soil, colonizes the rhizosphere and the interior of the plant and promotes growth by increasing the supply or availability of primary nutrients to the host plant.

Methodology & Theoretical Orientation: The product was applied to tomatoes (Prekos F1), cucumbers (Mirabelle F1) and cauliflower (Cornell F1), both in the seedling phase and after planting, as an aqueous solution sprayed on soil. Two treatments were applied on seedlings: 4 days before sowing and 15 days after emergence. During vegetation period were performed three applications, using 15 l/ha. Plant height, number of leaves and the length of the roots as well as the obtained yield were determined and recorded.

Findings: In tomato, cucumber and cauliflower seedlings the application of Rom-Agrobiofertil NP has improved plant height (5.6 -17.9%), number of leaves (> with 20.0-29.6%), number of roots (19.5–40.8%) and the length of the roots (21.2-33.1%). In tomato, cucumber and cauliflower crops, the effect has been reflected both in the growth and development of plants and in the obtained yield higher with 2.3% on tomatoes, 3.7% on cucumbers and 2.4% on cauliflower.

Conclusion & Significance: Additional obtained yield was significant in comparison with untreated controls, in all three vegetable crops

Biography

Nicolae Levandovschi has dedicated his last years in learning and discovering how bacteria works best. His major concern was to ensure the optimal composition of culture media, the right range of both temperature and pH and the exact moment of harvesting the bacterial culture. He applied this model to all the bacteria he was responsible for, as Chief of department of living or inactivated vaccines. The newer the project, the harder the challenge seems to be the pathway for improving range of products of Romanian company Romvac, in order to meet the requirements of Ph. Eur., GMP or EMA. As part of an experienced team, he has included in Romvac Co portfolio Rom-Agrobiofertil NP, a top product for good health of soil and rich crops in agriculture.

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Notes:

Leidy Yanira Rache et al., J Biotechnol Biomater 2017, 7:6 (Suppl)
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Cellulase and xylanase activities of *Xanthomonas axonopodis* pv. *manihotis (Xam)* strains collected from different regions of Colombia

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Statement of the problem: Microorganisms are an important source of plant cell wall-degrading enzymes. This is especially true for plant pathogenic bacteria because the production of plant cell wall-degrading enzymes is practically a prerequisite for pathogenesis. Recent studies characterized the activity of the enzymes produced by different bacteria, and their utilization in industrial processes, including the degradation of lignocellulosic biomass for biofuel production. Currently, the sources to obtain biofuel are not renewable, and the diversity of enzymes produced by native endogenous bacteria is becoming more importance. Thus, we propose to analyze the carboxymethyl cellulase (CMCe) and xylanase activities of the cassava pathogen, *Xanthomonas axonopodis* pv. *manihotis (Xam)* strains collected from different regions of cassava production in Colombia.

Methodology & Theoretical Orientation: We performed a screening of the enzymatic activity of 660 *Xam* strains maintained at the Natural History museum collection of the Universidad de Los Andes. The hydrolytic activity was determined using 0.2% Congo red and identifying clear halos. Strains showing statistically significant differences and the highest coefficient estimates were selected and individually analyzed for their hydrolytic activity.

Findings: A total of 34 out of 660 *Xam* strains showed significant differences, and the higher coefficient estimate of CMCe activity. The highest ranges of carboxymethyl cellulose degradation ranged from 6.269 to 4.992 cm2 in area, and the lowest between 1,71 and 0.445 cm2. A total of 46 out of 660 Xam strains showed significant differences and highest coefficient estimates of the xylanase activity. The highest ranges of Xylan degradation ranged from 0.3375 to 0.261 cm^2 in area, and the lowest between 0.2096 to 0.2 cm2. Differences in *Xam* hydrolytic activity were analyzed and related at the molecular level.

Conclusion & Significance: This study is an important approach to increase the knowledge on plant hydrolytic activities of *Xam* and to discuss the possible use of these enzymes in biotechnological processes

Biography

Leidy Yanira Rache has developed her studies in different areas of knowledge, such as in vitro plant tissue culture of several fruits species, gene flow in GM cotton, and pesticide degrading capabilities of bacteria. Currently, she is studying the population genetic diversity and cellulolytic activity of *Xanthomonas axonopodis* pv. *manihotis* from different regions in Colombia. The aims of the study are to propose control strategies to the blight caused by the bacteria, and to promote research for knowledge of not only native *Xam* species but also others native pathogenic bacteria in Colombia.

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Notes:

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Cytotype and molecular variability of Myriophyllum L

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Myriophyllum L. (watermilfoil) is the largest genus of the family *Haloragaceae* and belong the most species-rich genera of aquatic plants. Distinguishing of genus *Myriophyllum* is rather challenging. Despite the apparent taxonomic complexity, almost nothing is known about the cytogenetic structure of the genus in North America. In our study, we used flow cytometry and chromosome counting to recognize genome size and DNA ploidy level in species of genus *Myriophyllum*. We analyzed 294 European and 329 North American population samples. All species of genus *Myriophyllum* were found in both areas, except North American *M. alterniflorum*. Large cytotype variability (2x, 3x, 4x, 6x, 8x and 9x) was found in the USA and Europe. Cytotype variability was found in populations of *M. heterophyllum* where diploids and triploids were examined. Sympatric growth of diploid and triploid cytotypes was encountered in one population of *M. pinnatum*. Two populations of *M. aquaticum* in Europe (Hungary) had cytotype variability (6x and 8x) whereas all N. American populations were octoploids. Cytotype variability was also found in populations of *M. sibiricum* where hexaploids and nonaploids were observed. There was clear geografic isolation showing *M. sibiricum* as a hexaploids only in N. American and nonaploids in European populations. Only one nonaploid specimen of *M. sibiricum* was found in N. American populations and just one hexaploid of *M. sibiricum* in the European population. Nonaploids in USA and hexaploids in Europe of M. *sibiricum* were detected for the first time. Genome size (2C) ranged from 0.41 pg in diploid *M. humile* (2n=14) to 2.66 pg in nonaploid *M. sibiricum* (2n=63). These findings give evidence that a detailed study of cytotype composition. Last but not least, studies of ploidy variation have repeatedly proved necessary to elucidate the mechanisms of triggering the invasive behavior in plants.

Biography

T Kávová is a Molecular Geneticist with specialization on population-genetic studies. She is most interested in ecological topic with the implantation of new techniques in molecular analyses. Techniques that she perfectly controls range from basic lab work, through complete knowledge of molecular genetic techniques including sequencing to current flow cytometry specialization. The topic of her thesis is genetic and cytogenetic variability of *Myriophyllum L.* in the native and invasive area of the genus..

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Notes:

Dagmar Stehlikova et al., J Biotechnol Biomater 2017, 7:6 (Suppl)
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Design and testing of duplex-PCR primers for detection of bacterial spot of tomato

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This study presents development of duplex-PCR assay for specific detection of plant pathogenic bacteria of *Xanthomonas* genus causing bacterial spot of tomato and pepper. PCR primers for differentiation of *X. euvesicatoria* and *X. vesicatoria* were developed based on the DNA sequences of bacterial type strains. Primer pairs were designed and subsequently thoroughly tested and optimized for parallel detection of the bacteria. Specificity of the primers was tested on a large complex of bacterial strains pathogenic to tomato, pepper and related crops. Following the described protocol *X. euvesicatoria* and *X. vesicatoria* can be quickly and reliably identified in a single duplex-PCR assay.

Biography

Dagmar Stehlíková is a Molecular Geneticist with specialization on detection phytopathogenic bacteria. She is most interested in quarantine bacteria *Xanthomonas, Ralstonia, Burkholderia* etc. Techniques that she is expert in molecular methods are loop-mediated isothermal amplification, reatime PCR and multipex PCR.

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Notes:

Vlasta Pilizota et al., J Biotechnol Biomater 2017, 7:6 (Suppl)
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Production of extra jam from indigenous grape varieties

Vlasta Pilizota and **Ante Loncaric**
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Noah, Othello and Isabelle are grape varieties introduced in Europe in the 19th century due to their natural resistance to *Phylloxera*. Since these varieties were directly planted in winegrowers' fields they were called direct producers or direct producer wines. The term came to cover native American species (*Vitis aestivalis, V. labrusca, V. riparia, V. rupestris*) and hybrids obtained from interspecific crossings, either with each other, or with the European common species *Vitis vinifera*. Due to EU Regulation No. 1308/2013, and Croatian Law on Wine (NN 14/14), it is forbidden to produce wine from these grape variates. In this research, we investigated the possibility of exploitation of these grape variates for production of jams to encourage local producers to preserve these grape cultivars. The results of investigation showed that the highest soluble solids content had white Noah followed by red Noah, Isabelle and Othello, 22; 21; 19; 18 Brix, respectively. Othello and white Noah had the highest total acid content (1 g/100 g), while Isabelle had the greatest polyphenol content (2.04 mg/mL), and antioxidant activity (2073.93 μmol trolox/100 mL). The main polyphenols in white grape variate were gallic acid, epicatechin, quercetin and caffeic acid, while in red variates additional anthocyanins were identified (cyanidine-3-glucoside, cyanidine-3-rutinoside, malvidin-3-glucoside and delphinidin-3-glucoside). The soluble solids content of extra jams was set on 60-62%, which is according to the National (Croatian) Law (NN 46/07, 55/11), and directives for production of extra jams. Jam production caused decrease in AOA for 25; 31; 42 and 47% in black Noah, white Noah, Othello and Isabelle, respectively. The amounts of individual polyphenols were lower in jams compared to unprocessed grapes. The smallest change of color, caused by jam production, was measured in Othello (2.22) and Isabelle (2.75) grape varieties.

Biography

Vlasta Pilizota has her expertise in basic research of structure and composition of food (and/or food model systems) such as aroma composition of different food, thermo-physical and rheological properties of foodstuffs, inhibition of browning of fruits and vegetables, safety of minimally processed fruits and vegetables, improving the process for processing and preservation of foods, and the applied research for food processing – industry.

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M Shokry, J Biotechnol Biomater 2017, 7:6 (Suppl)
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Commercial polyester fabric repair of abdominal defects and hernias

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Hernias are common surgical affections of human and animals and their treatments may vary from simple herniorrhaphy to hernioplasty with prosthetic materials. This work was designed to study the feasibility of using commercial polyester fabric (CPF) as a prosthetic mesh for reconstruction of abdominal hernias and defects in experimental and clinically affected animals. The subjects of the experimental work were 12 dogs, 3 goats and 3 donkeys. Following anesthesia, an artificial abdominal defect, measured 5X12 cm in dogs and goats and 12X20 cm in the donkeys, was produced by resection of a piece of abdominal muscle in 3 different abdominal regions, such as the epigastrium (3 cases), the mesogastrium (12 cases) and the hypogastrium (3 cases). An appropriate piece of sterilized CPF was implanted either in 2 layers in small animals or in 4 layers (folded) in large animals in the abdominal defects. The techniques of implantation used, were reteroperitoneal (6 cases) in which the CPF was implanted between the internal rectus sheath and peritoneum, intra-peritoneal implantation either with (3 cases) or without (3 cases) mentalization; and double sandwish (3 cases) using 2 layers. Morphological and histo-pathological examinations were carried out on full thickness sections after euthanasia at 1, 2 ,3 ,4, 5 and 6 months postoperatively. The clinically affected cases comprised different species of animals at different locations. Gross examination of the abdominal wall at the experimental implantation site at monthly intervals for 6 months showed the unaltered integrity of the CPF and uniform infiltration of the prosthetic material with a layer of white connective tissue of variable thickness (4 to 10 cm). Histological features correlated well with the gross examination. Complications related to herniorrhaphy in the clinical cases have been rarely serious. In conclusion, CPF has excellent biocompatibility and no adverse histological changes. Low cost CPF therefore offers a worthwhile alternative prosthesis for mesh herniorrhaphy

Biography

M Shokry is currently a Professor Emeritus, Dept. of Vet Surgery, Faculty of Veterinary Medicine at Cairo University in Egypt. He has published more than 100 Articles in the various national and International journals.

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Phylogenetic relationships of the genus *Curcuma* L. based on cytogenetical and molecular inferences

Rama Rao and **Judith Mary Lamo**
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The genus *Curcuma* L. of the family *Zingiberaceae*, is pantropical in distribution and comprises of three subgenera viz. *C.* subg. *Curcuma, C.* subg. *Hitcheniopsis* and *C* subg. Ecomatae. Of the three subgenera, the subgenus *Curcuma* (Baker) K. Schum., contains highly complex polyploid taxa with overlapping morphological characters which contributed to taxonomic perplexity in the genus *Curcuma*. A combination of classical and molecular cytogenetics as well as molecular approaches, are essential for detailed scrutiny particularly polyploid complexes which may offer a reasonable taxonomic concept. To resolve some of these issues, fifteen species belonging to the *Curcuma* subg. *Curcuma* were taken up for cytogenetical (viz. chromosome count, male meiosis and heterochromatin banding pattern) and sequence targeted (viz. nrITS and cpDNA) studies. Mitotic study in root-tip cells, could resolved the species into three groups with 2n=42 (*C amada, C aromatica, C. comosa, C haritha, C mangga* and *C montana*), 2n=63 (*C aeruginosa, C caesia, C latifolia, C longa, C leucorrhiza, C sylvatica, C zanthorrhiza* and *C zedoaria*) and 2n=105 (*C raktakanta*) with male meiotic analysis showing a varying degree of chromosome association(s) suggesting the genus might have been affected by inter-specific crosses and thus confirming their allopolyploid nature. For phylogenetic investigation, a total of 27 *Curcuma* species which included 15 species collected exclusively from India and the remaining 12 world species retrieved from GenBank, were taken up for detailed analysis with *Alpinia galangal and Globba substrigosa* as outgroups. The studies could successfully resolve the species with respect to their infrageneric groups and could deduce some of the taxonomic discrepancy related to the genus *Curcuma. C. comosa, C. montana* and *C. latifolia* with similar morphological and floral traits demonstrated a close phylogenetic relationship. *C. sylvatica* was considered a variant derived from *C. amada* due to the mango-like aroma of the rhizome, etc. However, in light of our cytogenetical data on chromosome count and male meiosis we rule out the possibility of *C. sylvatica* (2n=63) being a variant of *C. amada* (2n=42). In depth, scrutiny of the species within the *Curcuma* clade based on morphology, cytology and molecular parameters could resolve the identity of some closely resembling *Curcuma* species when there is confusion with respect to their identity. Moreover, a close relationship between species within the *Curcuma* clade suggested that hybridization and subsequent chromosome doubling has played an important role in species diversification of *Curcuma*

Biography

Rama Rao has about 30 years of teaching and research experience in the field of Plant Genetics and Molecular Biology. He has mainly focus on characterization of genetic diversity of plant resources of Indian Thar Desert regions, later the North-east regions of India with focus on Meghalaya. Over these years, he has identified novel genotypes in *Vigna, Curcuma, Citrus* and bananas. He has published more than 105 research publications in journals of international repute.

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Antheraea mylitta cocoonase: A boon in silk industry

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Sericulture, both an art and a science of raising silkworms for silk production, has better prospects in developing countries as silk production is highly economical. Silkworm species vary in terms of the quality of silk they produce and the food plants they consume. Wild silkmoths include tasar silkworm, eri-silkworm, oak-tasar silkworm and muga silkworm. The Indian tasar silkworm. *Antheraea mylitta* is a natural fauna of tropical India, represented by 44 ecoraces. Wide distribution and polyphagy of this insect species has resulted in extensive variation in the population. Tasar cocoons are reported to be largest among all the silk-producing insects in the world. Cocoon, shelter for larva development to silk moth, contains fibrous protein, fibroin and is coated by the globular protein, sericin. The escape of the silk moth from cocoon requires the action of cocoonase enzyme secreted by the pupa. Cocoonase is a protease enzyme which hydrolyses sericin, soften cocoon and later they escape out. Seeking this vital function, the study focuses on the production of active recombinant *Antheraea mylitta* cocoonase and its post translation modification (PTM). PTM can significantly modulate the integral properties of protein affecting its stability, interaction and providing proper folding. Several PTMs such as phosphorylation, SUMOylation, myristoylation and glycosylation are being checked. Obtained detailed findings will be discussed

Biography

Dev Mani Pandey is interested in the research areas and scientific expertise includes: molecular biology, functional genomics, stress physiology and bioinformatics approaches on plants like rice, groundnut, medicinal plants etc., using recent biotechnological tools. He is also associated with Central Tasar Research and Training Institute, Ranchi, India for sericulture related research. He is also actively involved in research, teaching and other department and institute activities.

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Technological innovation against arbovirosis: Development of bioinsecticide against the mosquito *Aedes aegypti*

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Recently, several emerging and reemerging arboviruses have caused concern around the world. The *Aedes aegypti* mosquito is the vector of dengue, urban yellow fever, chikungunya and zika. Dengue is the most common arboviruses in the world, affecting more than 2.5 billion people a year. On the other hand, the zika virus brought an alarming fact since it was linked to babies born with microcephaly. Currently, Brazil is experiencing an outbreak of wild yellow fever in several regions of the country, which brings out the fear of the return of urban yellow fever. The most efficient way to combat all these diseases is through the control of the mosquito vector. This is mainly done by chemical insecticides, despite the emergence of resistance. In this sense, the search for new, safe and effective insecticides is necessary. Our research group has explored the biotechnological potential of natural products, especially the *Agave sisalana*. Preliminary studies showed that the *A. sisalana* crude extract has important larvicidal activity against *Ae. aegypti*. Through flow cytometry and histopathology, we observed that exposure to the crude extract of *A. sisalana* caused the necrosis of hemocytes of the exposed larvae, besides lysis of the intestinal cells and the peritrophic membrane. These results show the potential of *A. sisalana* as raw material for the production of an insecticide that helps control emerging and reemerging arboviruses transmitted by *Aedes aegypti*.

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Synthetic biology for detection of contaminants and for diagnose of disease: New biotechnology

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Despite the great advances on diagnostic technologies for disease and for detection of environmental contaminants (i.e. pesticides, heavy metals, hydrocarbons, and others), there is still a need for a non-endpoint diagnose in addition to better precision and real-time results. Therefore, herein we are presenting a novel approach based on synthetic biology to diagnose disease and for detection of environmental contaminants/pollutants. Our technology is based on a construct microbial DNA sensor. Thus, we developed three types of DNA sensors for early detection of diabetes (US Patent No.: 9,683,266 B2, June 20, 2017) Alzheimer's disease (patent pending), respectively. Also, we developed a microbial DNA sensor for detection of heavy metals in soil. Although microbial/molecular sensors have been used for detecting different biological molecules, chemicals, as well as contaminants, their sensitivity is limited. Therefore, we present here three types of sensors with higher sensitivity based on assemblage of different genetic parts which are cloned on benign bake yeast, Saccharomyces cerevisiae. The genetic parts were sequences related to proteins for detections of molecules such as glucose or beta-amyloid for diagnosing of diabetes or Alzheimer's disease, respectively. We also assembled a genetic building block for identification of specific heavy metals in soil. The diagnosis was based on the biofluorescence emitted by the mixture of the DNA sensor with patient blood plasma when the respective molecule or proteins have been detected. Hence, the degree of the diagnosed disease is based on the intensity of the fluorescence unit (FSU). Likewise, the microbial DNA metal sensor was able to identify different heavy metals in soil at very low concentrations, also based on the intensity of the fluorescence of the DNA sensor. The denoted technology brings great advantages, since it enables us to accurately classify diabetes patients in different groups (i.e. diabetic, pre-diabetic, normal), thus predicting development of the disease at early stages. In addition to early detection of the disease, the present technology also allows for earlier clinical intervention. Similarly, the technology enables us to identify metal contaminations which are undetectable under conventional methods. The above mentioned synthetic biology approach was effectively supported by a computational modeling. This new biotechnology applied to the medical and environmental fields facilitate the integration of different molecular techniques with physiological mechanisms at the cellular and molecular level on real time, based on the integration of biological sciences, engineering, and computational modeling for a more predictable biological process. This allows biology to become more effective at the industrial level not only for health solutions but also for economic benefit

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Effect of synthesis process variables on morphological and mechanical properties of vitreous carbon scaffolds for tissue engineering applications

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Vitreous carbon foams have been shown to promote bone cell adhesion, mineralization and proliferation. However, their low mechanical resistance as well as their high manufacturing cost restricts their utilization in the biomedical area. The purpose of this study was to develop bone tissue engineering scaffolds from vitreous carbon foams, which were fabricated through the template route using an economical and renewable precursor. Towards this, cellulose sponges were impregnated with a sucrose-based resin and then carbonized under inert atmosphere. The effect of the concentration of the components of the resin (HNO_3 and sucrose) on the mechanical and morphological properties of the resulting foams was determined. Moreover, the ability of the synthesized foams to promote cell adhesion was evaluated *in vitro* using human osteoblasts. Our results show that it was possible to produce vitreous carbon foams with highly interconnected polyhedral cells (cell size~1000 μm). Scaffold morphology was strongly affected by the concentration of the catalyst in the resin (HNO_3) due to its foaming effect, which lead to porous and irregular surfaces on the carbonaceous materials. Also, increasing the concentration of sucrose in the precursor resin favored the mechanical resistance of the resulting foams, reaching values close to the commercial foams. In conclusion, vitreous carbon foams with trabecular bone-like morphology were obtained from a non-toxic and renewable precursor. The fabricated foams were shown to be highly cytocompatible and to promote human osteoblast adhesion. Although the compressive strength of the foams is much lower than that of native bone, their high porosity will allow their reinforcement using an additional biocompatible phase (coating/filler). Therefore, the vitreous foams synthesized here could be used as the porous component of a composite biomaterial system for the treatment of bone defects

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Study heterotrophic growth of Chlorella sp under different carbon-to-nitrogen and carbon-to- phosphorous ratios

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Microalgae have caused interest in recent years because of their particular way of accumulating lipids. These microorganisms can be cultivated in autotrophic, mixotrophic, and/or heterotrophic way. Heterotrophic cultures decrease growth time, increase biomass concentration, and total lipid yields. Appropriate composition of the culture medium will favor the growth of the cells, for this reason commercial culture media have been modified to establish the effect of increasing or decreasing the concentration of some nutrients when producing biomass and lipids. However, to achieve good lipid yields, it is necessary to ensure a high concentration of biomass at an initial stage of cultivation. Lipids, being primary metabolites, can be induced by subjecting the microalgae to stress conditions depending on both the species and the abiotic factors. This work evaluated the heterotrophic growth of the native microalga *Chlorella* sp using glucose as carbon source and varying relations carbon/nitrogen and carbon/phosphorus to favor the production of biomass. In addition, the change of fatty acid composition changes with biomass production. Maximum biomass obtained was 9.25 g/L and 8.67 g/L for C/N of 25:1 and C/P of 200:1 during 7 days of cultivation, their productivities were 0.93 g/L*d and 0.99 g/L*d. Total fatty acid production was favored with C/N 50:1 and C/P 400:1 reaching 25.7% and 22% of total fatty acids in dry biomass, also higher fatty acid productivities in biomass of 41.16 mg/L*d and 24.32mg/L*d with C/N 10:1 and C/P 200:1. Low C/N and C/P ratios stimulated biomass production, biomass lipid productivity, and decreased total fatty acid production. High C/N and C/P ratios improved the production of total fatty acids. In this way, the maximum production of biomass must be reached for further achieving the stage of nutritional exhaustion due to the deficiency of N and P in the culture medium. This causes the elongation of polyunsaturated fatty acid chains.

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Effect of *Melissa officinalis* on rat hippocampal and cortex slices subjected to oxygen and glucose deprivation

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Cerebral stroke is 3rd leading cause of death after cancer and heart attack worldwide. Free radicals play key role in brain injury. The results of the present investigation demonstrate that Oxygen-glucose deprivation (OGD) followed by re-oxygenation led to cell damage/death via an increase in free radical's production in rat brain hippocampal and cortex slices compared with control (non-OGD) after 2h OGD followed by 1h reperfusion. *Melissa officinalis* at concentration 40 µg/ml displayed potential role in neuro-protection against OGD, followed by re-oxygenation in mitochondrial viability assays *in vitro*. In addition, *Melissa officinalis* decreased or slowdown the production free radical in the supernatant and slices homogenate of hippocampal and cortex at the end of 2h OGD followed by 1h reperfusion. Furthermore, higher concentrations of *Melissa officinalis* slightly showed neurotoxicity for hippocampal and cortex slices which could be due to a pro-oxidant effect.

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A dry method to preserve tear sample

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The tears overlay the epithelial cells of cornea and conjunctiva surface. It provides lubrication, protection and nutrition to the ocular surface. Tears is an important bio-fluid containing thousands of molecules, including proteins, lipids, metabolites, nucleic acids, and electrolytes. Tear fluid can be easily and noninvasively accessed, and it has become a useful resource for biomarker research of ocular and systemic diseases. According to a recent review, hundreds of potential specific molecular biomarkers in tears had been detected to associate with ocular diseases, such as, dry eye disease, keratoconus, Graves' orbitopathy. Other reports showed that tear can also reflect the states of breast cancer, prostate cancer and multiple sclerosis. Ideally tear samples should be stored by a simple, low-cost and efficient method along with the patient's medical record. The primary methods for collecting tears are using the Schirmer's strip and glass capillary tube, followed by flash-freezing at −80°C. But, cryopreservation of tears cannot absolutely prevent the degradation of proteins, as the samples contain various enzymes and hydrolases. Additionally, use of the required cold chain during sample transportation is challenging and costly. Here, we developed a novel Schirmer's strip based dry method which allows storage of tear samples in vacuum bags at room temperature. Using this method tear protein pattern can be faithfully preserved. LC-MS/MS analysis of proteins recovered from our dry method and from traditional wet method, indicating that there is no significant difference. This dry method facilitates sample transportation and makes it possible to store tear samples at large scale, which in turn increases the research pace of tear related diseases.

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Dihydroxyphenyl glyceric acid biopolyether of plant origin-prospective therapeutic agent

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The structure elucidation of main structural element of high-molecular water-soluble fractions from different species of comfrey Symphytum asperum, S.caucasicum, S.officinale, S.grandiflorum and bugloss Anchusa italica (Boraginaceae) was carried out. According to ^{13}C, ^{1}H NMR, APT, 1D NOE, 2D heteronuclear $^{1}H/^{13}C$ HSQC and 2D DOSY experiments the main structural element of these preparations was found to be poly[oxy-1-carboxy-2-(3,4-dihydroxyphenyl)ethylene] or poly[3-(3,4-dihydroxyphenyl) glyceric acid] (PDPGA). Thus, the polyoxyethylene chain is the backbone of the polymer molecule. 3,4-Dihydroxyphenyl and carboxyl groups are regular substituents at two carbon atoms in the chain. The repeating unit of this regular polymer is 3-(3,4-dihydroxyphenyl)glyceric acid residue. Most of the carboxylic groups of PDPGA from *Anchusa italica* and *Symphytum grandiflorum* unlike the polymer of *S.asperum*, *S.caucasicum* and *S.officinale* are methylated. The 2D DOSY experiment gave the similar diffusion coefficient for the methylated and non-methylated signals of PDPGA. Both sets of signals fell in the same horizontal. This would imply a similar molecular weight for methylated and non-methylated polymers. PDPGA is endowed with intriguing pharmacological properties as immunomodulatary (anticomplementary), antioxidant, anti-inflammatory, burn and wound healing properties. Then the basic monomeric moiety of this polymer, 3-(3,4-dihydroxyphenyl)glyceric acid (DPGA) was synthesized via Sharpless asymmetric dihydroxylation of *trans*-caffeic acid derivatives using a potassium osmate catalyst and a stoichiometric oxidant *N*-methylmorpholine-*N*-oxide. *S.caucasicum* PDPGA and synthetic DPGA exerted anti-cancer efficacy *in vitro* and *in vivo* against human prostate cancer (PCA) cells via targeting androgen receptor, cell cycle arrest and apoptosis without any toxicity, together with a strong decrease in prostate specific antigen level in plasma. However, our results showed that anticancer efficacy of PDPGA is more effective compared to its synthetic monomer. Overall, this study identifies *S.caucasicum* PDPGA as a potent agent agains.

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Screening and characterization of agarolytic bacteria from different sources

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Statement of the Problem: Agar, a complex polysaccharide produced by marine red algae, is used throughout the world in a variety of laboratory and industrial applications, owing to its jellifying properties. In India, more than 100 plant tissue culture laboratories (PTCL) are engaged in plant propagation programme and ~25 are in Maharashtra state. Once the plants are transferred to soil, the residual agar with low nutrient contents thus becomes waste agar. Around 1500 kg waste agar is generated per day in a medium sized PTCL which poses a problem of disposal, as it is dumped in pits and takes months for decomposition. Agarolytic microorganisms produce agarases, which catalyze the hydrolysis of agar. Most agarolytic bacteria described have been isolated from marine sources, though there are few reports from other sources as well.

Methodology & Theoretical Orientation: The aim of the present study is to isolate bacteria which is effective in degradation of agar. Collection of samples for screening of microbes through different sources including river water, spinach field soil and compost has been done. Pure bacterial isolates obtained from enrichment technique followed by repeated sub-culturing were screened for agar-degrading ability by streaking on agar media. Isolates were characterized by using biochemical and molecular methods. Bacterial genomic DNA isolation was carried out using CTAB method. 16S r-RNA of isolates were studied.

Findings: Total 7 strains were selected as the candidates that hydrolyze agar around colony, among which three each from river water and spinach soil and one from compost sample. Three strains of *Pseudomonas, Enterobacter* and *Aeromonas* were identified and characterized using 16S rRNA.

Conclusion & Significance: Investigation on the efficiency of these isolates from non-marine sources may yield useful information and their usage for accelerated degradation of waste agar from PTCL and thus would be valuable for the industry.

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Salinity responsive genes profiling in tomato utilizing RNA sequencing and quantitative real time PCR

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Soil salinity and scarcity of fresh water resources are two of the most environmental constraints that negatively affect plant growth and productivity worldwide. Tomato (*Solanum lycopersicum* Mill.) has been classified as moderately sensitive to salinity at all plant developmental stages. Identification of salinity response genes that control tomato salt tolerance will provide important guide lines for breeding programs and genetic engineering in tomato. In this study, Illumina RNA-sequencing and qPCR were achieved in two improved tomato genotypes (L46 and L56) for salt tolerance evaluation, in order to differentially expressed genes estimation the two genotypes were affected by salinity treatment 0.5 d sm-1 as a control and 9.6 d sm-1 as a salt stress. cDNA libraries were constructed and sequenced. About 13.3 million short reads (92 bp) were generated from cDNA libraries originated from leaves of both genotypes. Genotype L56 showed over expression of major salinity- responsive genes that aid in the salinity tolerance mechanism. Genotype L46 showed different group salinity responsive genes. In conclusion, the salt tolerant breeding genotype L56 is genetically robust, as it shows enhanced expression of salt-responsive genes in response to saline conditions. By contrast, the salt susceptible genotype L46 showed some potential genetic background. These genotypes have great potential for future breeding programs.

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