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Specific charge separation of Cd doped TiO_2 photocatalysts for energy applications

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ABSTRACT

Titanium dioxide (TiO_2) nanostructures are well known for their excellency in photocatalytic activities. In this work, additive-free and Cd metal ion - incorporated titanium dioxide (TiO_2) nanoparticles have been prepared to employ a facile route of synthesis using a hydrothermal method. Metal-metal nanocomposites have been synthesized by incorporating cadmium (Cd) with the appropriate amount of TiO_2 nanoparticles. The properties of the derived materials had been investigated by employing various characteristic tools such as various techniques, including X-ray diffraction (XRD), Scanning electron microscopy (SEM), Fourier transform infrared spectroscopy (FTIR), UV-visible absorbance spectroscopy (UV-vis), and photoluminescence spectroscopy (PL) are all examples of advanced imaging techniques that may be used to study materials. Using a method known as a vibrating sample magnetometer, we measured the magnetic properties of bare and Cd-doped TiO_2 nanoparticles. The investigations on crystalline nature of samples are agreed well with the standard crystalline features of TiO_2 nanoparticles. Emerged grain sizes have been estimated for all samples of pure and additive incorporated TiO_2 samples. Morphological characterization revealed that different particle features varied with the compositional changes. Spectral and optical absorption spectra of the prepared nanoparticles ensured the yield of derived TiO_2 nanoparticles with the additive component. An evaluation of the photocatalytic activity of Cd doped TiO_2 nanoparticles under UV irradiation was made using the methylene blue (MB) degradation method. The photodegradation efficiency were studied under visible light which confirms that the material is gifted one for water-treatment technologies to meet the rising clean water shortage.

1. Introduction

Titanium dioxide (TiO_2) is a non-harmful semiconductor with a bandgap of 3.0 eV. Titanium oxide is a well-known inorganic metal oxide semiconductor with an n-type configuration [1]. Titanium dioxide could be employed in water parting and oxidation steps because of its unique optical properties and synthetic soundness [2]. Controlled doping with the appropriate elements would improve the appropriate electronic properties (specifically all around located band holes) in many of these TiO_2 gadget applications [3]. There is widespread recognition that TiO_2 combined with iron (III) impacts the charge transporter significantly [4]. The additive incorporation is to diminish the enactment energy of the anatase-to-rutile stage change, particularly in TiO_2. In comparison to traditional powder union strategies in the production of oxide powders, aqueous cycles in sol–gel synthesis have the capacity for immediate planning of glass-like fired powders and provide a low-temperature alternative. Cadmium (Cd) is a remarkable semiconducting medium widely used in various applications such as

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In Press, Corrected Proof What's this?

Cinnamon mediated- zinc oxide nanoparticles and its cryogenic effect on SARS-CoV-2 recovered HTPI patients – An in vitro cum pilot study

Parameswari R [a 1], R. Babujanarthanam [a], T.B. Sridharan [b], T. Yogesh [a], S. Nithyanandan [c], R. Gomathi [d], R. Ganeshmoorthy [e]

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Highlights

- 1. This is the first study on homeostatic status and its influence on fertility in male by SARS CoV-2.
- 2. Male infertility by hyper blood pressure as a side effects of SARS CoV-2.
- 3. SARS CoV-2 entry and possible mechanism on spermatogenesis by ACE-2 to homeostatic imbalance caused Testosterone reduction in Covid-19 recovered subjects.
- 4. Use of Zinc rich component and its importance in during and after Covid-19 associated infertility.

Abstract

Infertility affects more than 40–50% of couples, with men accounting for 20–30% of the problem. Spermatogenesis fails due to poor sperm quality. Although external factors of smoking, alcohol, heat, and metal exposure cause testicular damage, causes infertility than genetic factors. However, after infection with COVID-19, the SARS-CoV-2 virus enters the blood-testis barrier via ACE2 receptors influenced hypertension and disrupts spermatogenesis. Meanwhile, preserving sperm cells during treatment or medication and the

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Original article

An evaluation of the biological activity of zinc oxide nanoparticles fabricated from aqueous bark extracts of *Acacia nilotica*

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Abstract

The phyto nanotechnology strategy was executed for the genesis of zinc oxide nanoparticles by applying bark aqueous extract of *Acacia nilotica* (AN-ZnO NPs). The AN-ZnO NPs emergence was substantiated through a distinct peak at 350nm in the ultraviolet–visible spectroscopy. The fourier transform infrared spectroscopy exposed the phyto-organic moieties engaged as reduction and stabilization factors in fabricating nanoparticles. The crystalline trait, elemental proportions, spherical and hexagonal geometry of AN-ZnO NPs was witnessed by X-ray diffraction, energy-dispersive X-ray analysis and scanning electron

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Smoke toxicity effect of bio-fabricated mosquito coil for the sustainable management of mosquito vectors

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Highlights

- Plant powder based mosquito coil prepared.
- Biocoil as potential Larvicidal.
- More predation happens when added the boicoil in mosquito breeding sites.

Abstract

Mosquitoes are major disease-causing vectors. Control of adult mosquito vectors by synthetic chemical applications leads to chemical resistance and causes environmental pollution. Commercially available mosquito chemical-coils also provide health hazards like respiratory diseases probably to a larger extend than plant-based bio-coils. In our study laid smoke-exposed gravid females of *C. quinquefasciatus* a lower number of eggs, egg hatchability were also comparatively lower and the progeny production during the F1 generation was significantly lower. In the present study there was an 84.39% reduction of the population of bio-coil exposed mosquitoes compared to 90.13%, after the use of a chemical-coil. LC_{50} of bio-coil extract ranged from 201.595ppm to 374.395ppm. LC_{50} of achemical-coil extract ranged from 209.747ppm to 296.307ppm. Predation efficiency was lower in with chemical-coil treated aquatic environments than at bio-coil extract exposed breeding sites of *C. quinquefasciatus*. The predation efficiency against *C. quinquefasciatus* was 22.1% (I) with chemical-coil and 11.4% (II) with bio-coil in a natural setting, whereas it vely. Very low concentrations of bio-coil increased the predation on young mosquito instars where predators and prey co-exist. The feeding efficiency of *Poecilia*

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Original article

Chemical composition and mosquitocidal efficacy of panchagavya against Anopheles stephensi, Aedes aegypti and Culex quinquefasciatus

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ABSTRACT

Objectives: The current study looks towards reporting the chemical compounds present in the panchagavya (PG), free radicals scavenging and mosquitocidal activity of PG in the laboratory condition.
Methods: The existence of chemical compounds in the PG were studied by GC–MS analysis. Free radicals scavenging activity of PG was studied by using various invitro assays. Mosquitocidal efficacy of PG was studied by the experiment on larvicidal, pupicidal, adulticidal, fecundity, longevity, and ovicidal activity against *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus*.
Results: GC–MS analysis revealed fifteen chemical compounds present in the PG. Free radical scavenging was done by 2,2-diphenyl-1-picrylhydrazyl, 2,2′-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid), hydroxyl, and superoxide assays, and the IC_{50} was calculated as 37, 37.5, 35, and 38 µg/mL respectively. PG exhibited better larvae and pupae mortality against I-IV instar of *Cx. quinquefasciatus* (LC_{50}: 148.765, 162.534, 187.619, 210.835 and 234.624 ppm, LC_{90}: 286.636, 306.390, 350.276, 390.735 and 419.195 ppm). The highest adult mortality was found against *An. stephensi* (91.10 ± 1.74%) with the IC_{50} and IC_{90} values of 128.114 and 260.609 ppm. *An. stephensi* showed highly decreased fecundity and longevity even at a low concentration of PG. Inhibition of 100% egg hatchability of *An. stephensi* was obtained at 250 ppm followed by *Ae. aegypti*, and *Cx. quinquefasciatus* at 300 ppm respectively. On comparing with other mosquito vectors *An. stephensi* was effectively inhibited by PG at each stage of their life cycle.
Conclusion: The results provide the first proof that PG could be a successful natural agent for controlling different mosquito vectors. Furthermore, our findings pave the way for more research into the efficacy of natural materials' mosquitocidal activities.

1. Introduction

Mosquitoes are one of the most significant risks to public safety in the world, as they are carriers of many pathogenic microorganisms that cause various human diseases and those diseases are endangering human life and contributing to multiple morbidities and high mortality (Huang et al., 2019). Mosquito-borne diseases are widespread in more than 100 countries, triggering deaths of about 2 million people worldwide, and at least 1 million children suffer from mosquito-borne diseases per year, endanger-

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Standardization and Evaluation of an In-house Multiplex Real-time Polymerase Chain Reaction for Simultaneous Detection of Pathogenic Species of *Brucella*, *Rickettsia*, and *Leptospira* in Patients with Acute Febrile Illness

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Abstract

Background and Objectives: Acute febrile illness is caused by a wide range of etiologies. Brucella, Rickettsia and Leptospira as a cause of acute febrile illness have not been well studied. Identification by culture is laborious and diagnosis is often based on serological tests. We aimed to develop an in-house multiplex real-time PCR assay for the simultaneous detection of Brucella, Rickettsia and Leptospira and evaluate on samples collected from patients presenting with acute febrile illness. **Methods:** Samples (n=1101) were collected from patients presenting with acute febrile illness from different regions. An inhouse multiplex real-time PCR was developed for the specific detection of Brucella species, Rickettsia species and pathogenic species of Leptospira. The assay was evaluated on clinical samples. IgM ELISA was carried out on randomly selected samples (n=178). **Results:** The detection limit of the multiplex real-time PCR assay was 6.3, 43.7 and 1.2 genome copies per 10 µl of PCR input for Brucella, Rickettsia and Leptospira respectively. Among 1101 samples, Leptospira was positive in 1.36% samples and Rickettsia was positive in 0.36% samples. Among random samples, 37.1% of samples were positive for Brucella IgM, 19.6% were positive for Rickettsia IgM and 11.2% were positive for Leptospira IgM. **Interpretation and Conclusions:** The study showed a high seroprevalence of Brucellosis and Rickettsiosis among the samples. The in-house multiplex real-time PCR assay will be a useful tool in the syndromic diagnosis for a specific and comprehensive laboratory diagnostic testing.

Keywords: Acute febrile illness, *Brucella*, *Leptospira*, Multiplex real-time polymerase chain reaction, *Rickettsia*

Asian Pac. J. Health Sci., (2022); DOI: 10.21276/apjhs.2022.9.2.04

Introduction

Acute febrile illness is a common presentation and has a wide range of etiologies.[1] Differential diagnosis of acute febrile illness is difficult when it is based only on clinical features because most of the illnesses show non-specific symptoms such as the sudden onset of fever, headache, and malaise.[2] Pathogen-specific tests are expensive, and therefore, patients are often managed with empirical treatment resulting in unnecessary and/or overuse of antimicrobials thereby adding to a global burden of antimicrobial resistance.[3] The major bacterial cause of acute febrile illness and its prevalence remains poorly characterized in many parts of India. Emerging and reemerging bacterial infections, especially enteric fever, brucellosis, rickettsial infections, and leptospirosis as a cause of acute febrile illness, are often underestimated due to the non-availability of diagnostic resources in many health-care settings and expensive molecular assays. A syndromic diagnostic approach is needed for specific and comprehensive laboratory diagnostic assays.

Brucella, *Rickettsia*, and *Leptospira* are important groups of pathogens transmitted by animals causing mild-to-severe infections in humans.[4] These pathogens can be associated with significant morbidity and mortality especially in people with occupational risks such as animal exposure and dwelling in rural communities. Clinical presentation is very protean and often presents as an acute febrile illness with symptoms such as fever, malaise, and headache.

These pathogens are obligate intracellular bacteria, so a very low number of pathogens circulating in the blood and bacterial isolation from blood are difficult. Serological diagnosis is often

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Original article

Green synthesis and antimicrobial efficacy of titanium dioxide nanoparticles using *Luffa acutangula* leaf extract

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ABSTRACT

The present study deals with the synthesis of titanium dioxide (TiO_2) nanoparticles using *Luffa acutangula* leaf extract and explore the antimicrobial potential of synthesized nanoparticles. The biosynthesized TiO_2 nanoparticles were characterized using different spectroscopic and microscopic techniques. The absorption spectrum of synthesized TiO_2 nanoparticles were primarily characterized by Ultraviolet visible (UV–vis) spectrophotometer. The functional groups associated with the TiO_2 nanoparticles and the *Luffa acutangula* leaf extract were examined by Fourier Transform Infrared (FTIR) spectroscopy. The crystalline structure of nanoparticles were analyzed by X-ray diffraction (XRD) examination. Morphological characters were examined by Scanning Electron Microscopy (SEM) and Transmission Electron Microscope - Selective Area Electron Diffraction (TEM - SAED). Presence of elemental composition of synthesized TiO_2 nanoparticles were characterized by Energy Dispersive X-ray (EDX). The antimicrobial properties of the TiO_2 nanoparticles were observed to be highly toxic against bacterial strains are *Bacillus subtilis* (*B. subtilis*), *Escherichia coli* (*E. coli*), *Enterococcus faecalis* (*E. faecalis*), *Klebsiella pneumonia* (*K. pneumoniae*), *Staphylococcus aureus* (*S. aureus*) and *Pseudomonas aeruginosa* (*P. aeruginosa*) and the fungal strains are *Aspergillus flavus* (*A. flavus*), *Aspergillus niger* (*A. niger*), *Rhizopus oryzae* (*R. oryzae*) and *Sclerotium rolfsii* (*S. Rolfsii*). The zone of inhibition was estimated by disc diffusion assay and moreover, minimum inhibitory concentration was evaluated by the micro broth dilution assay. It can be concluded that titanium dioxide nanoparticles manifest a strong antimicrobial action and thus can be developed as a novel type of antimicrobial materials for the cure of microbial infections.

1. Introduction

Nanotechnology has attained vast attention over time, and it involves synthesizing and developing different nanomaterial (Kasinathan et al., 2016; Panimalar et al., 2022; Rathnakumar et al., 2019; Magdalane et al., 2018; Aina et al., 2018) and it is an accelerating field of recent research with desirable applications in medicine and electronic and has been developing very fast in recent generation, impacting on distinct areas such as environment and economy (Mani et al., 2021; Magdalane et al., 2021; Loo et al., 2018; Srinivasan et al., 2020). It is fundamentally concerned about the synthesis of nanoparticles (NPs) and their application in different fields of science, medicine, designing and imaging (Ocsoy et al., 2013; Jayakumar et al., 2022). Nanoparticles are characterized as building blocks of nanotechnology and the size ranging from 1 to 100 nm in diameter (Hoffmann et al., 1995; Fabrega et al., 2010; Fang et al., 2012). The foremost feature of nanoparticles is their surface area to volume aspect ratio, enabling them to combine with other particles easier. Moreover, metal nanoparticles have been intensively used in biology because they are biocompatible

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Original article

Green synthesis of gold nanoparticles using *Jatropha integerrima* Jacq. flower extract and their antibacterial activity

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ABSTRACT

Background: With the rise of antibiotic resistance, there is an increasing interest in discovering novel antimicrobial agents. Antibiotics could be replaced with metal-based nanoparticles that have long-lasting antibacterial properties. This study aimed to synthesize gold nanoparticles (AuNPs) and test their antibacterial effectiveness against a variety of human pathogens.
Methods: The AuNPs were characterized using UV, FT-IR, XRD, and TEM with EDX. By using the well diffusion and microdilution techniques, the impact of synthesized AuNPs was tested against *B. subtilis*, *S. aureus*, *E. coli*, and *K. pneumoniae*.
Results: The AuNPs were synthesized from *Jatropha integerrima* Jacq. flower extract. UV–vis spectrum showed a high peak at 547 nm; FT-IR revealed phenolic compounds in the plant extract were responsible for AuNP formation. XRD and SAED confirmed the crystalline nature while TEM revealed the shape to be spherical and DLS revealed the size to be 38.8 nm with the stability of −0.3 mV. The AuNPs exhibit maximal and minimal antibacterial activity towards *E. coli* and *B. subtilis*. The MIC of AuNPs against *B. subtilis*, *S. aureus*, *E. coli*, and *K. pneumoniae* were found to be 5.0, 10, 2.5, and 2.5 μg/mL, respectively.
Conclusion: Thus, synthesized nanoparticles might be a good alternative to develop an antibacterial agent against the selected human pathogens.

1. Introduction

Disease-causing microbes have become drug-resistant through the adequate usage of antibiotics (Maiti et al., 2014). Different forms of bacteria are developing resistance to currently available antibiotics, posing a severe threat to public health. Antibiotics are ineffective in treating illnesses caused by drug-resistant bacteria. Such antibiotic resistance needs a stronger alternative solution (Adil et al., 2019). In this situation, there is a higher need for novel bactericides to be developed to avoid a threat to public health.

As a result, it is critical to create effective and low-cost techniques for the synthesis of therapeutic agents to combat the aforementioned health issues. Nanotechnology is expanding possibilities, enabling new solutions to be developed using existing resources. Metals, semiconductors, and metal oxides are employed in nanotechnology for a broad range of applications in information, energy, environmental, and medicinal sectors (Heera and

Abbreviations: ATCC, American Type Culture Collection; AuNPs, Gold nanoparticles; DLS, Dynamic Light Scattering; EDX, Energy dispersive X-ray analysis; FTIR, Fourier Transform Infra-Red Spectroscopy; JIF-AuNPs, Synthesized gold nanoparticles; MIC, Minimum inhibitory concentration; MTCC, Microbial Type Culture Collection; PDI, Polydispersity index; SAED, Selected area diffraction pattern; SPR, Surface plasmon resonance; TEM, Transmission Electron Microscopy; UV–Vis, UV–Visible spectroscopy; XRD, X-ray Diffraction spectroscopy; ZOI, Zone of inhibition.

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Original Article

Isolation of bioactive compounds from lichen *Parmelia sulcata* and evaluation of antimicrobial property

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ABSTRACT

Background: Lichens were used as an ailment in the traditional medicine for treating various disorders for centuries. Since there is less evidence in the literature about the medicinal property of *Parmelia sulcata* (*P. sulcata*), we made a pioneer attempt to explore the antioxidant and antimicrobial properties of lichens.
Methods: In the present study, the three Samples were collected by using the column chromatography by elucidating the ethyl acetate extract of *P. sulcata*, and the samples were subjected to DPPH and ABTS assays to find the free radical scavenging activity, total phenols and flavonoids were estimated. The minimum inhibitory concentration was evaluated against the bacterial species (*Bacillus subtilis*, *Pseudomonas aeruginosa*, *Escherichia coli* and *Klebsiella pneumoniae)* and fungal species (*Candida albicans* and *Aspergillus fumigatus*) by the microdilution method. The best activity sample was analyzed using the Gas Chromatography–Mass Spectrometry (GC–MS), Fourier Transmission Infrared Spectroscopy (FT-IR) and Nuclear Magnetic Resonance (NMR).
Results: The results shown that all the samples contain phenols and flavonoids which are responsible for antioxidants, antibacterial and antifungal activities. Among that sample-3 shown best antimicrobial activity and it was analyzed and identified as 7-hydroxy-3-(2-methylbut-3-en2-yl)-chromen-2-one.
Conclusion: The outcome of the study suggests that sample-3 shown good antimicrobial activity and identified as 7-hydroxy-3-(2-methylbut-3-en2-yl)-chromen-2-one. It can be a resource for further studies.

Introduction

Lichens are complex organisms with symbiotic association between fungi (mycobiont) and alga or cyanobacteria (photobiont) [1,2]. Lichens are scientifically proved as the earliest colonizers of terrestrial habitats on the earth with a worldwide distribution from arctic to tropical regions [3]. As such, lichens dominate approximately 8% of the Earth's land surface [3,4], they are commonly seen in the plains to the highest mountains, even at extreme conditions [5]. Currently, about 20 000 lichen species are distributed throughout the world and their medicinal benefits are yet to be explored [6]. Lichens have a long lifespan, usually grows slowly up to a few centimeters per year; during the growth process, the lichens produce numerous bioactive compounds to be protected against different physical and environmental barriers [7].

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Original article

Phytosynthesis of silver nanoparticles from *Jatropha integerrima* Jacq. flower extract and their possible applications as antibacterial and antioxidant agent

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E. coli
Superoxide

ABSTRACT

Jatropha integerrima Jacq. flower extract was used for the synthesis of silver nanoparticles in the current study. Various spectroscopic analyses were used to characterize the synthesized nanoparticles (JIF-AgNPs). The antibacterial efficacy of JIF-AgNPs was studied by well diffusion and microdilution techniques. In addition, the impact of JIF-AgNPs on free radicals was evaluated. On the ultraviolet–visible spectrum, the nanoparticles exhibit the highest absorbance at 422 nm. Based on the Fourier transform infrared spectrum, phenols and amino acids were involved in capping the JIF-AgNPs. Crystalline sphere-shaped nanoparticles with an average size of 50.07 nm and zeta potential of −19.0 mV were confirmed by X-ray diffraction, transmission electron microscopy, and dynamic light scattering analysis respectively. The JIF-AgNPs exhibit the highest and lowest growth inhibitory activity towards *E. coli* and *B. subtilis*. The minimal inhibitory concentration of JIF-AgNPs against *E. coli*, *K. pneumoniae*, *S. aureus*, and *B. subtilis* were 2.5, 5.0, 5.0, and 7.5 μg/mL, respectively. The JIF-AgNPs exhibited significant radical scavenging activities against DPPH (IC_{50}-32.5 ± 0.06 μg/mL), hydroxyl (IC_{50}-25 ± 0.09 μg/mL), Superoxide (IC_{50}-42.5 ± 0.13 μg/mL), and ABTs (IC_{50}-33.5 ± 0.15 μg/mL). Thus, synthesized nanoparticles were a good alternative to develop an antibacterial and antioxidant agent.

1. Introduction

The value of nanotechnology in the field of therapeutic medicine has expanded in recent days due to its wide variety of applications, cost-effective and eco-friendly approach for the preparation of novel drug materials (Kalaimurugan et al., 2019). Nanoparticles (NPs) with a size of 1–100 nm are considered essential building blocks in nanotechnology. Metals like gold, silver, copper, and zinc have sparked interest in the production of NPs due to their superior magnetic, electrical, medicinal, and optical capabilities. Silver nanoparticles (AgNPs) are familiar among different nanoparticles for their antibacterial, antioxidant, and cytotoxic properties (Das et al., 2019). Silver nanoparticles are the most often employed nanosized particles in various nanotechnology disciplines, particularly in biomedical applications (Elisabeta Barbinta-Patrascu et al., 2020).

Antimicrobial resistance is one of the most pressing issues of our time resulting from the inappropriate usage of antibiotics (Mboya et al., 2018). Solving this challenge will require a multidisciplinary strategy, including developing novel antimicrobial drugs (Talapko et al., 2020). Although silver's antibacterial action has been known since ancient times, many scientists are currently reinvestigating it, and the medicinal use of silver is on the rise (Sim et al., 2018). Silver nanoparticles exhibit exceptional antibacterial action even at low concentrations, so their utilization has steadily increased in recent decades (Yin et al., 2020). Metal nanoparticles, particularly silver nanoparticles are characterized

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In vitro Evaluation of Antibacterial, Antioxidant, and Anticancer properties of Panchagavya

Sivaji Sathiyaraj[1], Gunasekaran Suriyakala[1], Arumugam Dhanesh Gandhi[2], Thirumalpur Neelakandan Baskaran[1], Ranganathan Babujanarthanam[3]*

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ABSTRACT:

Natural products have gained more attention in healthcare and treating various diseases because of their promising antibacterial, antioxidant and anticancer activity. The current research focused on reporting the antibacterial, antioxidant, and anticancer activity of panchagavya (PG) which is a natural formulation of five cow products. The antibacterial activity of PG was studied against *E. coli, P. aeruginosa, S. aureus, B. subtilis*, and the maximum zone of inhibition was found to be 19.67±0.44, 23.50±0.29, 14.17±0.60, and 17.17±0.17mm respectively. The in vitro antioxidant study revealed the free radical scavenging activity of PG by 2,2-diphenyl-1-picrylhydrazyl and hydrogen peroxide assays. The percentage of inhibition for the 2,2-diphenyl-1-picrylhydrazyl radicals was found to be 34.62, 49.58, 57.64, 69.12 and 75.79% while the hydrogen peroxide radicals was 23.27, 34.47, 46.53, 59.71 and 72.74% at 50, 100, 150, 200, 250µg/mL respectively. PG showed significant anticancer activity against the HepG2 cell line in a dose-dependent manner with IC_{50} value of 122 µg/mL. The findings of the present study may be useful for the discovery of novel antibacterial, antioxidant and anticancer agents from the natural cow products. However, further in vivo studies are needed to elucidate the mechanism of PG on cancerous cells.

KEYWORDS: Panchagavya, cow products, antibacterial, free radicals, HepG2.

INTRODUCTION:

Alexander Fleming's discovery of penicillin in 1928 was a pillar in the development of modern medicine[1]. Afterward, excessive antibiotic use is consequently detrimental to human health, the ecosystem, and the environment. It could also increase the occurrence of drug-resistant pathogens. Resistance to antibiotics is a major global concern that is rapidly increasing in morbidity, mortality, and health care in both hospitals and the community[2,3].

Therefore, because of the evidence of the rapid global spread of resistant clinical isolates, the need to discover new antimicrobial agents is of utmost importance. However, the record of rapid, widespread emergence of resistance to newly introduced antimicrobial agents indicates that even new families of antimicrobial agents will have a short life expectancy[4].

Oxidative stress has been involving in the selection of resistant bacterial strains since reactive oxygen species (ROS) revealed to be an essential driving force[5,6]. The role of oxidative stress in the antibiotic-mediated cell death process is undefined and subject to discussion[7]. Bactericidal antibiotics with unique targets in bacterial cells have been shown to promote the development of harmful reactive oxygen species (ROS) under aerobic conditions, leading to the killing of these drugs[8]. Besides, many research groups have presented compelling reasons against the ROS-mediated antibiotic killing of bacteria[9,10]. Concerning the development and function of antibiotic-induced ROS in bacterial resistance, several studies have attempted to solve the role of ROS and the response to oxidative stress in cell killing following drug therapy[11]. The capacity to form biofilms and thus sessile biofilm communities is one of the bacteria survival strategies. Oxidative stress has recently been reported to contribute to the phenomenon of selection of pro-biofilm variants and H_2O_2 resistance since ROS is an important driving force in the selection of variants of *P. aeruginosa* strains[6].

Free radicals can be neutralized by antioxidants and thus antioxidants protect humans from infection and degenerative diseases caused by the free radicals[12,13]. It is possible to classify antioxidants into two main groups, natural and synthetic. Synthetic antioxidants, however, induce or facilitate harmful health effects in humans, such as mutagenesis and carcinogenesis. Therefore, it is essential to substitute synthetics with naturally occurring antioxidants that are capable of preventing diseases associated with free radicals[14,15]. For both developed and developing nations, cancer remains a

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Original Article

Biosynthesis, characterization, and antibacterial activity of gold nanoparticles

Sivaji Sathiyaraj[a], Gunasekaran Suriyakala[a], Arumugam Dhanesh Gandhi[b], Ranganathan Babujanarthanam[a,*], Khalid S. Almaary[c], Tse-Wei Chen[d], K. Kaviyarasu[e,f]

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ABSTRACT

Background: In recent decades focus of research has been toward an alternative antibacterial agent because of growing bacterial resistance and side effects of antibacterial agents. In the current study, the antibacterial activity of gold nanoparticles has been evaluated on selected human pathogens.
Methods: In this study, we used *panchagavya* (PG) to synthesize gold nanoparticles, and the resulting nanoparticles (PG-AuNPs) were characterized by several spectroscopic techniques. In addition, antibacterial activity of PG-AuNPs against *Escherichia coli*, *Bacillus subtilis*, and *Klebsiella pneumoniae* were studied by well diffusion method.
Results: The synthesis of AuNPs was affirmed by a colour change, which was further validated by UV–vis spectra with a maximum absorption peak at 527 nm. Bandgap energy was calculated as 2.13 eV by *Tauc method* from the UV result. The presence of amino acids and proteins in PG was responsible for the conversion of gold ions to AuNPs, according to FTIR analysis. (111), (200), (220), and (311) crystallographic planes were observed by XRD; further crystalline nature was validated by SAED analysis. The size and zeta value were found to be 53.29 nm and -9.8 mV respectively. Spherical shaped nanoparticles and elemental structure of PG-AuNPs were confirmed by HRTEM and EDS analysis. The antibacterial activity of PG-AuNPs showed the maximum and minimum zone of inhibition against *K. pneumoniae* (17.12 ± 0.14 mm) and *B. subtilis* (11.42 ± 0.58 mm).
Conclusion: Antibacterial activity of PG-AuNPs was found to be strong against gram negative bacteria and moderate against gram positive bacteria. Based on the result, it was concluded that PG-AuNPs could be used to combat antibiotic drug resistance. Besides, in vitro and in vivo toxicity studies of PG-AuNPs should be conducted.

Introduction

Nanomaterials are becoming more widely used in medicine due to their drug delivery mechanism in cancer therapy, as well as their availability, material properties, and capacity to improve drug selectivity against cancer cells [1]. Gold nanoparticles (AuNPs) have generated increased interest among diverse metallic nanoparticles because of their unique qualities, which include nano size, low toxicity, comparatively simple fabrication, and precise targeting [2]. The antibacterial property of AuNPs has recently been a major research topic, making them a good candidate for antibiotic complementation. The antibacterial activity of AuNPs is mediated by the development of holes in the bacterial cell wall, resulting in cell death due to the loss of cell contents. Furthermore, AuNPs can inhibit multidrug-resistant pathogens by attaching to bacte-

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Biosurfactants for a Sustainable Future: Production and Applications in the Environment and Biomedicine

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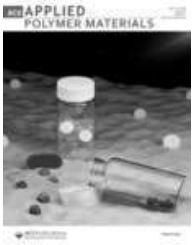

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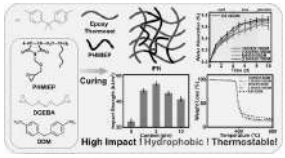

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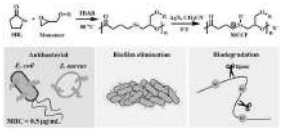

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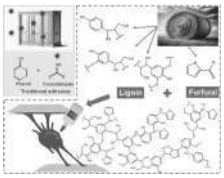

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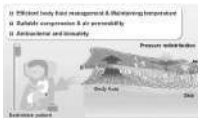

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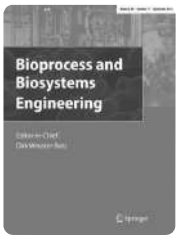

Bioprocess and Biosystems Engineering

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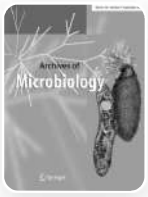
Bioprocess and Biosystems Engineering is an international peer-reviewed forum for discussion at the interface between engineering and biological sciences in the development of efficient bioprocesses.

- Reviews multidisciplinary approaches for integrative bioprocess design.
- Takes into account the environment and constraints of technical production processes.
- Publishes original articles, short communications and invited review articles.

Bacillus megaterium–induced biocorrosion on mild steel and the effect of *Artemisia pallens* methanolic extract as a natural corrosion inhibitor

Original Paper Published: 20 June 2020

Volume 202, pages 2311–2321, (2020) Cite this article



Archives of Microbiology

Aims and scope

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Seenivasan Kokilaramani, Mohamad S. AlSalhi, Sandhanasamy Devanesan, Jayaraman Narenkumar, Aruliah Rajasekar & Muthusamy Govarthanan

Abstract

Methanolic extract of *Artemisia pallens* (MEAP) (Asteraceae) was explored as greenbiocorrosion inhibitor for mild steel 1010 in 1.5% sodium chloride environment. *Bacillus megaterium* SKR7 induces the development of biofilm on the metal surface and forms the pitting corrosion. MEAP was showed (25 ppm) optimum inhibition effect of biocorrosion and further corrosion rate was highly reduced (0.3335 mm/year) than the control system

Anti-bacterial and anti-biofilm properties of green synthesized copper nanoparticles from *Cardiospermum halicacabum* leaf extract

Research Paper Published: 04 May 2020

Volume 43, pages 1649–1657, (2020) Cite this article

Bioprocess and Biosystems Engineering

Aims and scope

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Parthipan Punniyakotti, Pratheep Panneerselvam, Dhandapani Perumal, Rajasekar Aruliah & Subramania Angaiah

Abstract

In the present study, a copper nanoparticle (Cu NPs) was synthesized by a green synthesis method with *Cardiospermum halicacabum* leaf extract. The surface area of Cu NPs was measured with dynamic light scattering (DLS). UV–Vis spectrum clearly illustrates the typical absorption peak of Cu NPs. The crystalline property of Cu NPs was confirmed from the XRD pattern. TEM analysis clearly indicates the average particle size of synthesized Cu

Electrokinetic (EK) and Bio-electrokinetic (BEK) Remediation of Hexavalent Chromium in Contaminated Soil Using Alkalophilic Bio-anolyte

Original Paper Published: 16 May 2019

Volume 50, pages 330–338, (2020) Cite this article

Indian Geotechnical Journal

Aims and scope

Submit manuscript

Raja Kumaresan Sarankumar, Adikesavan Selvi, Kadarkarai Murugan & Aruliah Rajasekar

Abstract

The present investigation deals with remediation of chromium (Cr(VI)) by electrokinetic (EK) and bio-electrokinetic (BEK) processes. The experiments were conducted using spiked concentration of 1 g/kg of chromium in the soil. The results showed a maximum chromium removal of 40.12% and 90.4% by EK and BEK, respectively, at the end of 7 days. Chromium-resistant alkalophilic bacterial strain, *Bacillus licheniformis* SR3, was used as bio-anolyte counterpart in BEK. During the process, the variations in electrical gradient and pH were

studied and compared between EK and BEK. Chromium removal was further confirmed by UV spectroscopy, FTIR and XRD studies. The results of FTIR showed notable difference in the intensities of the peak, thus confirming the effective remediation by BEK. The obtained results have found to support BEK integrated system as an effective remediation option for cleaning up chromium-contaminated soil environments.

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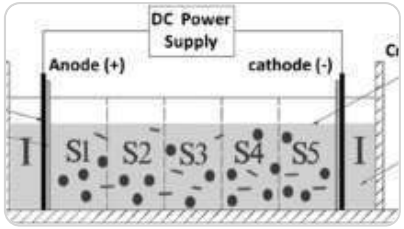

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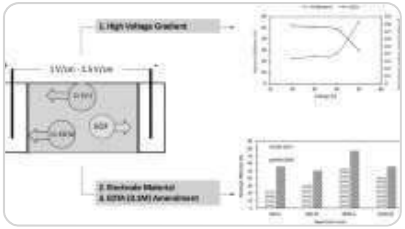

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Ethics declarations

Conflict of interest

All authors declare that they have no conflict of interest.

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Original Article

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Abstract

Background:

An in-house multiplex real-time polymerase chain reaction (PCR) was developed in two cocktails for the identification of six *Toxoplasma gondii*, *Rubella virus*, cytomegalovirus, herpes simplex virus (1 and 2), and *Treponema pallidum* (syphilis) (TORCH-S) agents, which causes congenital infection among pregnant women.

Objective:

Standardization and validation of an in-house multiplex real-time PCR assay for the detection of TORCH-S infection.

Methods:

This study was conducted from February 2017 to February 2019. Primers specific for *T. gondii*, *Rubella virus*, cytomegalovirus, herpes simplex virus (1 and 2), and *T. pallidum* were designed using Primer3 software (https://bioinfo.ut.ee/primer3-0.4.0/). The primer sequences obtained were subjected to BLAST analysis using BLAST database. Synthetic DNA was obtained to use as positive control templates for all the six TORCH-S agents. The lower limit of the detection was performed using plasmid construct for each virus serially diluted from 10^{-1} to 10^{-9}.

Results:

An in-house multiplex real-time PCR was standardized and validated in two cocktails for TORCH-S agents, cocktail-1 (HSV1, rubella, and *T. gondii*), and cocktail-2 (HSV2, CMV, and *T. pallidum*). The lower limit of the detection for HSV1, rubella, and *Toxoplasma* were 60.7 copies/10 μl input, 76.4 copies/10 μl input, and 34.4 copies/10 μl input and for HSV2, CMV, and *T. pallidum* were 80.8 copies/10 μl input, 166 copies/10 μl input, and 43.7 copies/10 μl input, respectively.

Conclusion:

TORCH-S infection is one of the significant reasons for irregular pregnant outcomes. It is absolutely important to screen TORCH-S infection for women who had the histories of abnormal pregnancies to prevent birth defects and perinatal complications. This multiplex real-time PCR assay provides a rapid, sensitive, and specific technique to detect these six TORCH-S agents.

INTRODUCTION

TORCH-S is a medical acronym for a set of perinatal infections with known adverse impacts on fetal development and pregnancy outcomes. The full form of TORCH-S is *Toxoplasma gondii*, *Rubella virus*, cytomegalovirus, herpes simplex virus (1 and 2), and *Treponema pallidum* (syphilis).

Toxoplasmosis is caused by the parasite *T. gondii*. Infected pregnant women are often presented with symptoms such as mild or asymptomatic, making the diagnosis difficult.[1] The organism is transmitted hematogenously to the placenta. When this occurs, an infection may be transmitted to the fetus transplacentally or during vaginal delivery.[23] The clinical implications of infection due to toxoplasmosis in pregnant women could lead to spontaneous abortions, stillbirths, intrauterine growth retardation, preterm deliveries, or fetal damage.[4] Worldwide, 190,100 infants born with congenital toxoplasmosis.[5]

The clinical manifestations of rubella include a mild exanthema that is frequently accompanied by adenopathy and occasionally arthralgia. It can cause fetal death or congenital rubella syndrome (CRS) during the first trimester, which is characterized by multiple defects to cataracts, congenital heart defects, neurological problems, hepatomegaly, and splenomegaly.[6] It has been estimated that during 1996–2010; globally 105,000 infants with CRS were born every year, among them 38% of which were from India.[7]

Cytomegalovirus (CMV) is the most common cause of congenital infection and complicates approximately 1% of every single live birth.[8] Primary maternal CMV carries a 30%–40% risk of vertical transmission. Moreover, congenital CMV is a frequently identified viral cause of mental retardation and is the leading nongenetic cause of neurosensory hearing loss.[91011]

Herpes simplex virus (HSV1 and HSV2) infections are transmitted from pregnant women to their neonates. It causes even death in infants and is associated with lifelong infection. HSV-1 and HSV-2 both can be responsible for neonatal, which can lead to spontaneous abortions, intrauterine growth retardation, preterm labor, and congenital and neonatal herpes infections.[12] The greatest risk of transmission to the fetus and the newborn occurs in case of an initial maternal infection contracted during the second half of pregnancy. Worldwide, herpes simplex virus infections are common, are transmitted from pregnant women to their neonates, and can cause infection or death.[1314]

Congenital syphilis, the infection caused by the spirochete *T. pallidum*, can be transmitted sexually or from mother to child *in utero* during the second or third trimesters of pregnancy.[1516] The most maternal syphilis infections are asymptomatic, but still result in poor pregnancy outcomes in more than 50% of cases.[17] Congenital syphilis could lead to spontaneous abortions, stillbirths, intrauterine growth retardation, preterm deliveries, or fetal damage. If the infection is untreated, it may even lead to complications such as early fetal loss, preterm birth, and low birth weight.[18]

The TORCH-S IgM assays have some limitations in their specificity producing up to 10% false positives. It has been reported that the routine practice of screening for IgM class antibodies during pregnancy may lead to numerous false-positive results, which can cause needless worry as well as unnecessary follow-up testing and treatment. Because of the crucial medical interventions required both for the mother and the child, it is important to have assays with extremely high sensitivity and specificity. In contrast to serological testing, the polymerase chain reaction (PCR) assay allows for earlier testing as viral genomic material seen in blood and body secretions of the infected mother, the PCR testing improves the specificity of the results.[19]

Serological assays which are the mainstay of diagnosis have several limitations. First, individuals may vary in their antibody response to the antigens used in the assay both temporarily and in magnitude. Second, antigenic cross-reactivity could be a problem causing false positives. Third, the IgM assays could be falsely negative in the presence of an excess of specific IgG. Furthermore, the rheumatoid factor could interfere while not using IgM capture format assays of rheumatoid factor absorbent for the pretreatment of sera. In contrast, genomic material will be present very early in infection and the

entire duration of active pathogen replication. The choice of highly conserved specific sequences will overcome pathogen cross-detection by PCR. Hence, we standardized an in-house multiplex real-time PCR assay and validated using plasmid control. This assay can simultaneously detect the following 6 common TORCH-S agents: *T. gondii*, *Rubella virus*, cytomegalovirus, herpes simplex virus (1 and 2), and *T. pallidum* (syphilis) in cocktail-1 (HSV-1 rubella, and *T. gondii*) cocktail-2 (HSV2, *T. pallidum*, and CMV). In the recent years, the real-time PCR has improved the diagnostic of viral and bacterial infections, being a powerful tool for the detection and quantification of RNA or DNA. Real-time PCR is increasingly used in diagnostics due to its high sensitivity and good reproducibility. This study was approved for ethical clearance by Sri Narayani Hospital and Research Centre Ethical Committee (No: IEC/IRB No: 21/04/06/11, dated: 04/06/2011).

MATERIALS AND METHODS

This study was cross-sectional in nature and conducted from February 2017 to February 2019 in Sri Narayani Hospital and Research Centre (SNH and RC), Sripuram, Vellore, Tamil Nadu, India. The laboratory study was carried out in Sri Sakthi Amma Institute of Biomedical Research, a unit of SNHandRC.

Selection of target genes

The first step in the development process of an in-house multiplex real-time PCR assay is the choice of a nucleic acid target. A literature review often reveals which target is the most suitable for each particular assay. For TORCH-S, specific and conserved nucleic acid target sequence was selected.

Primer and probes design

Selection of the target sequence has been done, next to find potential primers and probes targeting regions of the corresponding sequence using Primer3 software (https://bioinfo.ut.ee/primer3-0.4.0/). Primer3 is one of the most widely used primer design software; it is a frequently updated and open-source project and used many web-based applications to develop useful functions for primer and probe designing. The Primer3 software is widely used for designing PCR primers, hybridization and sequencing primers.

Validation of primers and probes

The amplicon's specificity was confirmed using the BLAST program. This software searches different databases for sequence similarity and returns a collection of gapped alignments with links to complete database entries. Both the maximum identity and query coverage should be 100%. The “expectation value” (E-value) is a measure of statistical significance that is given to each alignment produced by BLAST. It is an indication of the likelihood of discovering the match by chance. The E-value is a commonly used metric for determining the likelihood of a biological connection. Smaller E-values indicate a higher probability of an underlying biological connection. Sequences having E-values of ≤0.01 are most often found to be homologous. Validated-Primer and probes were custom manufactured commercially by Eurofins Genomics (Germany) often shipped and received in a lyophilized state. Primers and probes were resuspended in TE (10 mMTris•Cl, 1 mM EDTA, pH 8.0) to provide a stock solution of 100 μM. The resuspended primers and probes were aliquoted in 0.6 ml tubes. This reduces the number of freeze/thaw cycles that the master primer aliquots go through and also reduces the chances of contaminating the primary source for the primers and probes.

Development of gene constructs for positive control and lower limit of detection

Synthetic DNA was obtained to use as positive control templates for all the six TORCH-S agents. A positive control is necessary, when amplifying a target sequence to confirm whether the primer set or primer–probe set works. The synthetic genes were designed such that the final synthetic gene length for all the constructs was 250 bp. The specific genomic region for each of the six TORCH-S agents was obtained commercially from Eurofins Genomics (Germany). The synthetic gene was obtained subcloned into a vector (vector: PCR 2.1, Cloning: TOPO-TA). These constructs were used to establish a lower limit of detection as well as to serve as positive control templates for all our real-time PCR assays. The nucleotide sequences of real-time primers used for two mixes are shown in Table 1.


Table 1:
Primers and probes information for real-time polymerase chain reaction target genes

Evaluation and performance of real-time multiplex

The real-time PCR amplification was performed in multiplex for each agent as 25 μl reaction using QuantiTect a multiplex NR kit (Qiagen, Hamburg, Germany). Positive control (10 μl), forward and reverse primers (300 nM), and probe (200 nM) were used with initial polymerase activation (PCR) for 15 min at 95°C and followed by 45 cycles of denaturation at 94°C for 45 s. Annealing/extension was at 60°C for 75 s. Amplification and detection were performed on a real-time PCR system (Rotor-Gene Q) using the Taqman principle. The cutoff for real-time PCR endpoints was determined as amplification within the 40^{th} cycle with a fluorescence intensity or more with a typical sigmoid amplification.[2021]

Determination of efficiency and limit of detection

Construction of a standard curve from a serial dilution of the plasmid control is the most effective way to evaluate assay performance. The standard curve gradient may be used to determine the assay's efficiency. The technical assay dynamic range may be determined from the same experiment by running a broad variety of sample concentrations and verifying that they reach a limiting dilution. Figure 1 shows how to use a standard curve to determine an assay's technical dynamic range and efficiency. The plasmid construct was serially diluted 10-fold in TE buffer (pH 8.0) in the concentration ranging from 10^{-1} to 10^{-9}. Each dilution was tested in triplicates by real-time PCR. Appropriate negative controls were used replacing the template with nuclease-free water and included as every third sample. The PCR runs were validated only if the controls were satisfactory. Amplification shown in the highest dilution in at least two replicates of the triplicates tested at each dilution was taken as the lower limit of detection as plasmid copies per microliter. The approximate number of plasmid copies per microliter of DNA suspension was thus established. The calculation of the plasmid copy number of the agents tested was done according to the standard methods.[22] The primers' specificity was determined by testing them with heterologous plasmids.


Figure 1:
An example of high reproducibility and wide range of detection using a serial dilution of linearized plasmid.

Multiplex standardization

Multiplex real-time PCR uses a different set of primer pairs in the same reaction for simultaneous amplification of multiple selected target regions. Standardization of multiplex real-time PCR assays was performed using plasmid controls obtained commercially from Eurofins Genomics (Germany); uniplex real-time PCR was performed to ensure the specificity of the primers and probes. In all channels, each pathogen plasmid control was detected only for the specific set of primers and probes.

Efficiency

To establish meaningful comparisons between various samples, the amplification efficiency of PCR reactions was evaluated. To assess the efficiency of amplification in a particular primer set, each virus included in the research was serially diluted from the original control, and a standard curve was created. The efficiency was calculated according to the following formula: E= (10 [−1/pendiente]) − 1.

Diagnostic of sensitivity and specificity

An assay of analytical sensitivity was performed in uniplex and multiplex reactions to evaluate the behavior of the assay in response to changes in the quantity of nucleic acids. For each virus included in the research, these tests were conducted in triplicate serial dilutions in base 10 ($1\ 10^{-1}$ to $1\ 10^{-9}$) from the original control. To establish the starting concentration, all of the controls were measured.

RESULTS

Standardization

Standardization of an in-house multiplex real-time PCR assays was performed using synthesized plasmid control. Initially, the assay was performed in uniplex to ensure the specificity of the primers and probes. The uniplex PCR assay result for each virus was detected only for the specific set of primers and probes used. The lowest Ct value for the *T. gondii* was 16.49, 17.07 for Rubella, 17.77 for HSV1, 18.28 for HSV2, and 16.97 for CMV, and the highest for *T. pallidum* was 18.65. (Multiplex PCRs were tested to verify that cross-reactions were avoided and that results were similar to those obtained for uniplex reactions). Amplification plots for cocktail-1 and cocktail-2 are shown in Figure 2.

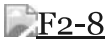
Figure 2:
Amplification plot for cocktail 1 and cocktail 2: Amplification plot for HSV1 (ROX, orange) Rubella (Cy5, red) and *Toxoplasma gondii* (FAM, green) positive controls in cocktail 1 and Amplification plot for CMV (FAM, green), HSV2 (Cy5, red) and *Treponema pallidum* (ROX, orange) positive controls in cocktail 2.

Efficiency

Efficiency was calculated for each virus in multiplex PCRs reactions, obtaining efficiencies of 90% for rubella, 99% for *T. gondii*, CMV, and *T. pallidum* obtained same efficiency 93% HSV1 and HSV2 virus showed the lowest efficiency (89%).

Determination of efficiency and limit of detection

The inhouse realtime PCR assay had a detection limit of 166 genome copies per 10 µl of PCR input for CMV, *T. gondii* show the lowest copy number 34.4 genome copies per 10 µl of PCR input. The copy numbers indicating high assay sensitivity to detect and also the low copy numbers. The lower limit of

detection for TORCH-S agents was determined. The lower limit of detection established for the TORCH-S agents is shown in Table 2.


Table 2:
The lower limit of detection for *Toxoplasma gondii*, *Rubella virus*, cytomegalovirus, Herpes simplex agents was determined. The lower limit of detection established for the *Toxoplasma gondii*, *Rubella virus*, cytomegalovirus, Herpes simplex agents

DISCUSSION

The multiplex real-time PCR assay has many advantages compared to other diagnostics methods, including speed, quantitative measurement, lower contamination rate, higher sensitivity, and higher specificity.[23] There are several methods that exist for the fast identification of viral infections. However, the molecular tests have shown excellent performance and may be an option for the diagnostic of routine within the laboratory.[24]

This research discusses the creation of six real-time multiplex PCRs that may be used in conjunction to identify six congenital infections. Using commercially produced plasmid controls, tests for lower limit of detection, efficiency, specificity, and sensitivity were evaluated, establishing the robustness of the standardized multiplex real-time PCR.

The technologies, uniplex and multiplex real-time PCR, were shown to have high sensitivity. For each pathogen, the lowest dilution suggests signal for the multiplex system spans from Ct 16.49 to Ct 18.24. The results were fairly comparable for both kinds of tests, but the Ct values in the multiplex PCR assays were greater. When multiple primers and probes are employed in the same mixture, this situation may be caused by a kinetic reaction.

The specificity tests revealed that the primers and probes are specific for the pathogens studied (Cocktail1 and 2), and no crossreactions were observed when genetic material from technically in each cycle the number of copies of genetic material doubles in realtime PCR. The Ct value for each dilution (1:10) obtained around 3.32.[25] By graphing the Ct values against dilution, the slope (m) reflects this value, and the line equation is calculated. It is a major problem since the PCR reaction efficiency should be between 90 and 100 percent (slope between 3.32 and 3.6).[21] If the efficiency is 100%, Ct values of 3.32 shows in each dilution for each real-time PCR cycle. When the slope reaches <−3.6, the effectiveness of the real-time PCR starts to decline. Furthermore, the value of R2 for a standard curve indicates how well the experimental data fits the regression line, i.e., how linear the data are. As a result, R2 should ideally be more than 0.9925. The slopes of various pathogens for multiplex real-time PCRs in our research vary from 3.34 and 3.63.

The efficiency of PCR amplification is often expressed as a percentage, i.e., the proportion of amplified genetic material in each cycle, with most pathogens having efficiencies around 90%. The lowest efficiency was found for HSV1 and HSV2 virus with a percentage of amplification of genetic material in each cycle of 89%. In contrast, higher efficiency was found for *T. gondii* with a value of 99%

The PCR efficiency is sufficient, since for 100% efficiency, with efficiencies of 90% achieved in our research. A molecular interaction in PCR efficiency makes a significant variation in the amount of end product. This scenario requires a large number of cycles to detect a certain quantity of genetic material.[26] Aside from this variation, the findings show that multiplex real-time PCR has excellent agreement between results for each run, ensuring reliable results.

The efficiency of real-time PCR varies according to assay performance; intramolecular interactions decrease real-time PCR efficiency; competing reactions and reagent quantity are additional factors in efficiency variance.

CONCLUSION

The in-house multiplex real-time PCR assays used in this research performed well in detecting six TORCH-S agents closely linked with congenital infections. The TORCH-S multiplex real-time PCR assay has benefits in that it does not need post-PCR processing and may be utilized in rapid diagnostic procedures to identify TORCH-S infective agents.

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Nil.

Conflicts of interest

There are no conflicts of interest.

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Keywords:

High-risk pregnancy; real-time polymerase chain reaction; *Toxoplasma gondii*; *Rubella virus*; cytomegalovirus; herpes simplex virus (1 and 2) and *Treponema pallidum* (syphilis) infections

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Original Article

Seroprevalence of TORCH-S Infections among Pregnant Woman: A Study from Vellore District (South India)

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Abstract

Introduction: TORCH-S agents include *Toxoplasma gondii*, Rubella virus, *Cytomegalovirus*, Herpes simplex virus (HSV) (1 and 2), and *Treponema pallidum* (syphilis) which are transmissible in utero at various stages of gestation. **Description of the Hypothesis Tested:** TORCH-S agents are known to cause adverse fetal outcomes and pregnancy loss. **The Approach Used:** Pregnant women attending a multispecialty hospital for regular antenatal care and high-risk pregnant women with a bad obstetric outcome from a rural area of Vellore District were recruited. A total of 180 pregnant women recruited from two centers were used. Pregnant women were evaluated for their serological status (IgM and IgG) against TORCH-S agents using commercial enzyme-linked immunosorbent assay kits available for respective pathogens. **Results:** Among the samples (n = 180) collected, IgM antibodies were positive in 3 (1.66%) for *Toxoplasma gondii* and 1 (0.55%) for HSV1. IgG antibodies were positive in 14 (7.77%) women for *T. gondii*, 152 (84.44%) for Rubella virus, 110 (61.11%) for CMV, 125 (69.44%) for the HSV-1 (16.66%), 30 were positive for HSV-2, and 5 (2.77%) women were positive for *Treponema pallidum*. In the 17–25-year age group, the number of IgG positives for *T. gondii* and HSV-2 were lower compared to other pathogens. **Conclusions:** The study reports a high prevalence of IgG to TORCH-S agents in pregnant women indicating a high risk among these populations. Routine screening for TORCH-S agents among antenatal women is warranted as timely diagnosis, and proper intervention could help initiate appropriate management. Information of these infections could help the clinicians for appropriate counseling on the potential for adverse fetal outcomes and preventive measures to the mothers.

Keywords: Bad obstetric history, enzyme-linked immunosorbent assay, high-risk pregnancy, IgG, IgM, TORCH-S infections

Introduction

TORCH-S is a medical acronym for a set of perinatal infections with known adverse impact on fetal development and pregnancy outcome. This includes infections with *Toxoplasma gondii*, Rubella virus, *Cytomegalovirus*, Herpes simplex virus (HSV) (1 and 2), and *T. pallidum*.

Toxoplasmosis is a major cause of congenitally acquired infection and it has been documented as the main reason of bad obstetric history (BOH), primarily leads to fetal death and morbidity of the newborns. Toxoplasmosis among pregnant women could lead to spontaneous abortions, stillbirths, intrauterine growth retardation, preterm deliveries, or fetal damage.[1] Among pregnant women during the 1st week of pregnancy, rubella virus infection causes devastating problems, leading to hearing loss, cataracts, congenital heart defects, neurological problems, hepatomegaly, and splenomegaly and are collectively known as congenital rubella syndrome (CRS).[2] About 40%–50% of *Cytomegalovirus* infected pregnant women could transmit the virus to the fetus. Among pregnant women, the *Cytomegalovirus* transmission from mother to fetus could occur even if the mother was infected earlier before pregnancy.

Among pregnant women, genital herpes has been documented with spontaneous abortions, intrauterine growth retardation,

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preterm labor, congenital, and neonatal herpes infections.[3] *T. pallidum* can be transmitted to the infant by an infected pregnant mother or at birth through contact with maternal lesions.[4] If the infection is untreated, it may lead to complications such as early fetal loss, preterm birth, and low birth weight.[5]

The diagnosis of TORCH-S infections mainly depends on serological testing as these maternal infections are initially asymptomatic and the clinical diagnoses are unpredictable. The presence of IgM antibodies indicates a recent infection and the presence of IgG antibodies indicates past infection. The detection of the IgM and IgG antibody is the best approach for the identification of TORCH-S infections. The present study was to evaluate the serological status of pregnant women against TORCH-S agents by detection of IgM and IgG using enzyme-linked immunosorbent assay (ELISA).

Materials and Methods

Pregnant women were recruited prospectively for the cross-sectional study using a random sampling method. A total of 140 serum samples from pregnant women attending Department of Obstetrics and Gynaecology at Sri Narayani Hospital and Research Centre (SNHRC), Sripuram, Vellore, a multispecialty hospital mainly serving rural and periurban populations of Vellore district. Forty serum samples were collected from pregnant women attending upgraded Primary Health Center (PHC), Ussoor, in the rural area of Vellore District. The pregnant women (n = 140) recruited from SNHRC were otherwise asymptomatic who came for routine checkups. The women (n = 40) identified as a high-risk group were recruited from the PHC during the special camp organized for women with "high-risk" pregnancy. Permission for collecting the samples from the PHC was obtained through the Department of Public Health, Vellore (District office) and Directorate of Public and Preventive Medicine, Chennai (Head office). A clinical questionnaire and written consent were obtained from each pregnant woman. This study was approved for ethical clearance by SNHRC Ethical Committee (No: IEC/IRB No: 21/04/06/11, dated: 04/06/2011).

Inclusion criteria

Pregnant women attending antenatal clinic at SNHRC and high-risk pregnant women camp at Ussoor PHC who gave consent to participate in the study were included in the study.

Exclusion criteria

Women who did not give informed consent to participate and had conditions irrelevant to the proposed study clinical groups were excluded from the study.

This study was conducted from March 2017 to February 2019. Demographic details of the pregnant women attending SNHRC (n = 180) including maternal age, domiciliary status, occupation, and educational level were collected. Clinical information including hemoglobin level and history of abnormal pregnancies such as two or more consecutive spontaneous abortions, intrauterine fetal death, congenital malformation, and stillbirth were collected using a structured standard questionnaire. Information on pregnant women attending upgraded PHC (n = 40) was very limited. Two milliliter of venous blood was collected into BD Vacutainer® serum tubes. Serum was separated and stored immediately at −80°C deep freezer until tested.

TORCH-S IgG and IgM enzyme-linked immunosorbent assay

TORCH-S IgG and IgM antibodies were detected from the serum by commercially available ELISA test-kit (Calbiotech Inc., Canada). The test was performed according to the manufacturer's instructions. Briefly, the serum was diluted at 1:21 with dilution buffer for all the TORCH-S ELISA except for *T. pallidum* IgM assay, the dilution was 1:51. The negative control, positive control, and calibrator control were added into the appropriate wells. After loading the sample into a 96-well plate, the plate was covered with an adhesive and incubated for 20 min at room temperature, for *T. pallidum* IgM assay alone, it was incubated for 45 min at 37°C. After washing thrice with 300 µl of 1X wash buffer, 100 µL of the conjugate solution was dispensed in each well. Then, the plate was covered again and incubated for 20 min at room temperature and 45 min at 37°C for *T. pallidum* IgM assay. After washing thrice with 300 µl of 1X wash buffer, 100 µL of TMB substrate was dispensed into each well, avoiding the formation of bubbles and incubated for 10 min at room temperature in dark and for *T. pallidum,* the plate was incubated in dark for 15 min at room temperature. The reaction was stopped using 100 µl of stop solution.

TORCH-S antibody index was calculated by dividing the value of each sample by calibrator values. ELISA results were recorded using a microplate reader (Mindray MR-96A, Germany), as a measure of optical densities (OD) of the reaction intensity of TORCH-S IgG and IgM antibodies at a filter wavelength of 450 nm. Cut-off points and antibody index calculations were done according to manufacturers' recommendation to categorize seropositive (antibody index >1.1), borderline positive (antibody index 0.9–1.1), and seronegative (antibody index <0.9) samples. In this study, all serum samples with intensity of antibody index 0.9–1.1 (borderline positives) were not considered.

Results

In our study, out of 180 random samples tested by IgM assay for the TORCH-S agents, three samples (1.66%) were positive for *T. gondii* and one sample (0.55%) was positive for HSV-1. Rubella virus, CMV, HSV-2, and *T. pallidum* were negative among the sample tested.

The study population included 180 pregnant women in the age group of 17–41 years. The median age of the study population was 26 years. Women from the rural area were 127 (70.55%) and from the periurban area were 53 (29.44%).

IgM-positive status for *T. gondii* was found in three women recruited from SNHRC and among the three, one was also

positive for HSV-1, whereas none of them was positive in the high-risk pregnant women recruited from PHC. Among the samples from collected SNHRC, the IgG-positive status was found 13 for *T. gondii*, 117 for Rubella virus, 82 for CMV, 93 for HSV1, 30 for HSV2, and 5 for *T. pallidum,* whereas the high-risk pregnant women group were positive 1,352,832 for *T. gondii*, Rubella virus, CMV, and HSV-1 IgG. HSV-2 and *T. pallidum* IgG were negative in all the 40 high-risk pregnant women from PHC. No statistical difference was observed in the prevalence of IgG-positive status for *T. gondii*, Rubella virus, CMV, and HSV-1 between women recruited from SNHRC and PHC [Table 1].

Among the 180 samples tested for IgG, 14 (7.77%) samples were found to be positive for *T. gondii*, 152 (84.44%) samples were positive for Rubella virus, 110 (61.11%) samples were positive for CMV, 125 (69.44%) samples were positive for HSV-1, 30 (16.66%) were positive for HSV-2, and 5 (2.77%) were positive for *T. pallidum*. The three individuals who were IgM positive for *T. gondii* and HSV-1 were in the third trimester. The IgM-positive samples for *T. gondii* and HSV-1 were also positive for IgG for respective pathogens.

Seroprevalence of TORCH-S among women was satisfied by different age group 17–25, 26–35, and >36. All IgM positives were in the age group of >36 years. Among the seven women who were in the age group of >36 years, three were positive for *T. gondii* and one was positive for HSV-1. Among IgG positives, *T. gondii* positives followed by HSV-2 were low in all three age groups compared to other pathogens tested. In the 17–25-year age group, the number of IgG positives for *T. gondii* and HSV-2 were lower compared to other pathogens. This difference was statistically significant ($P < 0.0001$). The difference in the proportion of IgG positives between 17–25 and 26–35-year age group was statistically not significant for any of the pathogens tested. The age group of >36 was too small in sample size for statistical analysis. The serological evidence of specific IgM and IgG antibodies against TORCH-S infections among pregnant women stratified by age groups is shown in Table 2, respectively.

In our study, women from the rural population were high compared to the periurban population. The seroprevalence of IgG for all the pathogens tested among the rural and periurban population is shown in Figure 1. No significant difference was observed between rural and periurban population for any of the pathogens tested for IgG.

IgG-positive status for *T. gondii*, Rubella virus, CMV, and HSV-1 was observed in women from all the first, second, and third trimester groups. HSV-2 IgG and *T. pallidum* were seen in women in the second and third trimester only. The IgG seroprevalence for *Toxoplasma gondii*, Rubella virus, CMV, and HSV-2 was higher in the third trimester compared to other stages. The IgG-positive status for HSV-1 was higher in women at the second trimester [Table 3]. The difference, however, was not statistically significant.

One sample was exposed to all the TORCH-S agents; two samples showed mixed infections to *T. gondii*, Rubella virus, CMV, HSV-1, and HSV-2. Five samples showed mixed infections to *T. gondii*, Rubella virus, CMV, HSV-1. Seven samples showed mixed infections to Rubella virus, CMV, HSV-1, and HSV2 and 40 samples showed mixed infections to Rubella virus, CMV, and HSV-1. The IgM positives were also positive by IgG ELISA.

During the study period, among the pregnant women recruited from SNHRC, two of them developed complications later during pregnancy that leads to abortion and stillbirth. They were each in the first and second trimester, respectively. The former was positive for Rubella virus IgG and the latter was positive for Rubella virus, CMV, and HSV. HSV-2 and *T. pallidum* IgG were not observed in women at the first and second trimesters, whereas pregnant women in the second and third trimester were IgG positive for all the pathogens tested.

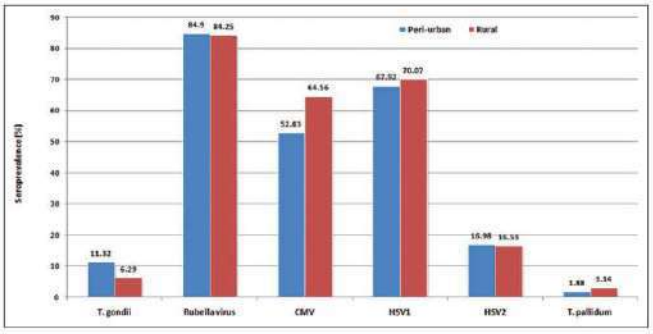

Figure 1: Seroprevalence of IgG for TORCH-S agents stratified by the domiciliary status expressed in percentages

Table 1: Seroprevalence of TORCH-S among women recruited from two centers

Study centers	*Toxoplasma gondii*, *n* (%)	Rubella virus, *n* (%)	CMV, *n* (%)	HSV-1, *n* (%)	HSV-2, *n* (%)	*Treponema pallidum*, *n* (%)
SNHRC (*n*=140)						
IgM	3 (2.14)	-	-	1 (0.71)	-	-
IgG	13 (9.28)	117 (83.57)	82 (58.57)	93 (66.42)	30 (21.42)	5 (3.57)
PHC (*n*=40)						
IgM	-	-	-	-	-	-
IgG	1 (2.5)	35 (87.5)	28 (70)	32 (80)	-	-

SNHRC: Sri Narayani Hospital and Research Centre, PHC: Primary health center, IgM: Immunoglobulin M, IgG: Immunoglobulin G, HSV-1: Herpes simplex virus-1, HSV-2: Herpes simplex virus-2, CMV: Cytomegalovirus

Table 2: Seroprevalence of TORCH-S among women stratified by different age groups

Immunoassay	Age group in years (*n*)	*Toxoplasma gondii*, *n* (%)	Rubella, *n* (%)	CMV, *n* (%)	HSV-1, *n* (%)	HSV-2, *n* (%)	*Treponema pallidum*, *n* (%)
IgG positives	17-25 (80)	5 (6.25)	65 (81.25)	80 (100)	77 (96.25)	8 (10)	2 (2.5)
	26-35 (93)	7 (7.52)	82 (88.17)	53 (56.98)	92 (98.92)	19 (20.43)	3 (3.22)
	>36 (7)	2 (28.57)	5 (71.42)	5 (71.42)	6 (85.71)	3 (42.85)	-
IgM positives	17-25 (80)	1 (1.25)	-	-	-	-	-
	26-35 (93)		-	-	1 (1.07)	-	-
	>36 (7)	2 (28.57)	-	-	-	-	-

IgM: Immunoglobulin M, IgG: Immunoglobulin G, CMV: Cytomegalovirus, HSV-1: Herpes simplex virus-1, HSV-2: Herpes simplex virus-2

Table 3: Serological evidence of specific immunoglobulin G antibodies against TORCH-S infection among pregnant women shown by gestational age

Pregnancy stage (*n*)	*Toxoplasma gondii*, *n* (%)	Rubella virus, *n* (%)	CMV, *n* (%)	HSV-1, *n* (%)	HSV-2, *n* (%)	*Treponema pallidum*, *n* (%)
First trimester (16)	1 (6.25)	5 (3.1.25)	11 (68.75)	12 (75)	-	-
Second trimester (38)	2 (1.81)	27 (71.05)	19 (50)	23 (60.52)	4 (10.52)	3 (7.89)
Third trimester (110)	11 (10)	96 (87.27)	72 (65.45)	77 (70)	26 (23.63)	2 (1.81)

Among the 180 samples, the gestational age of 16 women was not available. Of these, 14 (7.77%) were positive for Rubella, 8 (4.44%) were positive for CMV, and 13 (7.22%) were positive for HSV-1 IgG. IgG: Immunoglobulin G, CMV: Cytomegalovirus, HSV-1: Herpes simplex virus-1, HSV-2: Herpes simplex virus-2

Discussion

TORCH-S infections contribute to prenatal, perinatal, and postnatal morbidity and mortality where treatment or prevention is possible for most of the pathogens.[6] The prevalence of these infections in India has been documented in a piecemeal manner but only a very few studies exist on the prevalence of these infections as a syndromic diagnosis among pregnant women.

We looked at the seroprevalence of *T. gondii*, Rubella virus, CMV, HSV (1 and 2), and *T. pallidum* (TORCH-S) infections among pregnant women recruited from the rural and periurban population of Vellore district. The demonstration of IgM and IgG revealed the status of infections among pregnant women. Our study population included individuals recruited from our study center as well as from PHC during a special camp organized for high-risk pregnant women with BOH.

Among 180 individuals tested, *T. gondii* IgM was positive in 1.7% and HSV IgM was positive in 0.5%. On testing IgG for *T. gondii,* Rubella virus, CMV, HSV-1, HSV-2, and *T. pallidum*, a prevalence of 8%, 84%, 61%, 69%, 17%, and 3%, respectively, was observed.

In a recent study in the neighboring state of Telangana, South India, IgG seropositivity for *T. gondii*, Rubella virus, CMV, and HSV was reported to be 28%, 84%, 92%, and 61%, respectively.[7] Our study showed a comparatively low prevalence of *Toxoplasma gondii* and CMV and the prevalence rates of Rubella virus and HSV were near equal. This indicated previous exposure to TORCH pathogens in the population. In the states of Uttar Pradesh and Maharashtra, 19%, 29%, 7%, and 8% samples were found to be IgM seropositive for *T. gondii*, CMV, HSV, and Rubella virus among the antenatal cases (n = 162).[8] In our study, the IgM positivity was <2% for *T. gondii* and <1% for HSV-1 indicating the low prevalence of current infection among these populations. In a similar study carried out at pregnant women in the first trimester, specific IgM antibodies were found to be positive in 19%, 30%, 3%, and 33% cases for *T. gondii*, Rubella virus, CMV, and HSV-2 infections, respectively. In our study, we recruited pregnant women of all three stages of pregnancy. IgG-positive status among women in the first trimester was positive for *T. gondii*, Rubella virus, CMV, and HSV-1. The *T. gondii* and HSV-1 IgM-positive women were in the third trimester. IgG-positive status for all the pathogens was positive among women at the second and third trimesters. Toxoplasmosis poses a little risk of fetal transmission (<6%) in early pregnancy, whereas the rate of transmission ranges from 60% to 81% in the third trimester.[9] In our study, samples were collected randomly in a cross-sectional manner at one point during their regular checkups. Therefore, the prevalence data does not reflect the stages of pregnancy at which the infection was acquired. In general, pregnant women seek different health-care centers for antenatal and postnatal care. Follow-up on pregnancy outcome was also impractical in our study for this reason.

Acute and chronic *T. gondii* infection depends on serological data by the presence of IgM and IgG, respectively. However, distinguishing an acute from a chronic infection is difficult as IgM is reported to persist for several months to years following an acute infection.[10]

T. gondii and HSV-1 IgM positives in our study were also positive for IgG. Transmission to the fetus occurs predominantly in women who acquire their primary infection during gestation, and identification of the onset of the

infection is important. Therefore, other diagnostic tools such as IgA and IgG avidity detection are suggested.[11,12] These tests were not carried out in our study and are considered a limitation.

CRS among neonates occurs when a pregnant mother gets infected within the first 20 weeks of pregnancy. This could lead to cardiac, cerebral, ophthalmic, and auditory defects in the neonates.[13] A systematic review carried out in India shows that 38% of pregnant women are susceptible to Rubella virus infection in India.[14]

Determination of susceptibility to Rubella virus infection among the pregnant women in the population is therefore warranted to reveal the true risk of CRS. A recent study carried out in samples collected from different states of India reported 83% of Rubella virus IgG seropositivity among pregnant women.[15] The overall Rubella virus IgG seropositivity in our study was 71%. Another recent study[16] from Lucknow has reported 88% of Rubella virus IgG positivity with no difference in the age group which corroborates with our findings. In our study, CMV IgG positivity was seen in all women tested under the age group of 17–25 years. Early studies on HSV-1 and HSV-2 IgG positivity were low and significantly associated with increasing age. Little or no information in the literature exists on the prevalence of HSV-1 and HSV-2 in the population. Our study found a very high prevalence of IgG positivity for HSV-1 which is often neglected during screening for pregnant women. No studies exist on the prevalence of IgG to *T. pallidum* in pregnant women in India. We report here for the first time, a low seroprevalence of IgG of 3% in the population. The limitations of the study include lack of follow-up on pregnancy outcome among these pregnant women and detailed clinical workup. Testing newborns of these mothers were also not carried out and are considered a limitation. This indicates the necessity to screen pregnant women for TORCH-S pathogens early during the pregnancy and initiates necessary preventive and treatment options.

Conclusion

TORCH-S infections that include *T. gondii*, Rubella virus, *cytomegalovirus,* and HSV-1 and HSV-2 have long been known to be associated with bad obstetric outcomes. The infection presents with mild morbidity in mothers but has serious consequences to the fetus. It is therefore important to recognize the maternal infections and subsequently fetal monitoring if the infection is established. This will help the clinicians to counsel such mothers on preventive measures on the adverse fetal outcomes. Screening for pregnant women for TORCH-S agents could, therefore, reduce the incidence of adverse pregnancy and prevent birth defects.

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Conflicts of interest

There are no conflicts of interest.

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Evaluation of *Syzygium aromaticum* aqueous extract as an eco-friendly inhibitor for microbiologically influenced corrosion of carbon steel in oil reservoir environment

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carried out with and without bacterial strains and clove extract. The results obtained from weight loss and AC impedance studies indicate that these corrosion causing bacterial strains accelerated the biocorrosion reaction and biofilm playing a key role in this process. However, the addition of clove extract into the corrosive medium decreased the corrosion current and increased the solution and charge transfer resistance. The significant inhibition efficiency of about 87% was archived in the mixed consortia system with clove extract. The bioactive compounds were playing an important role in the antibacterial activity of the clove extract. It was revealed that clove extract has both biocidal and corrosion inhibition properties.

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Ethics declarations

Conflict of interest

The authors declare no competing financial interest.

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Research Article

Characterization of Biosurfactant From *Pseudomonas Stutzeri* SJ3 for Remediation of Crude Oil-Contaminated Soil

Sekar Harikrishnan, Singaram Jayalakshmi, Mohamad S. Alsalhi, and 3 more

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Abstract

In the present work, production of biosurfactant was studied from the bacterial strains isolated from the soil samples collected from oil contaminated sites in Karaikal ONGC, Puducherry, India. Six morphologically different hydrocarbonoclastic bacterial strains (SJ1-SJ6) isolated on oil agar plates were further screened for biosurfactant production. Based on the screening methods results of 26 mm oil displacement zone, positive results of drop collapse test, 68.14% emulsification index (E24) and 79.2% of bacterial adherence percentage, the isolate SJ3 was selected as the most potent strain and it was identified as *P. stutzeri* using standard biochemical and 16S rRNA gene sequencing-based methods. Optimization of the *P. stutzeri* strain showed 36 h incubation, 150 rpm agitation, pH 7.5, 37^{o}C, 1% salinity, 2% glucose as carbon source and 1% yeast extract as nitrogen source were the ideal conditions for growth and the biosurfactant production was found to be

1. Introduction
2. Materials And Methods
3. Result And Discussion
4. Conclusion
Declarations
References

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Biosurfactants for a Sustainable Future: Production and Applications in the Environment and Biomedicine

Chapter 7

Microbial Surfactants are Next-Generation Biomolecules for Sustainable Remediation of Polyaromatic Hydrocarbons

Punniyakotti Parthipan, Liang Cheng, Aruliah Rajasekar, Subramania Angaiah

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Summary

Polyaromatic hydrocarbons (PAHs) are considered as dangerous contaminants in water and soil, which are highly toxic and also carcinogenic to living organisms including humans. The concerns on the PAHs removal are increased due to the difficulties in their removal from contaminated water and soil. Bioremediation technology is the most promising, cost-effective, and eco-friendly approach to remove the hydrocarbons by using potential microorganisms. Nevertheless, the existing bioremediation technology has important limitations such as poor efficiency of microbial communities in the field and lesser bioavailability of pollutants. Biosurfactants are surface-active molecules, cost-effective and eco-friendly molecules synthesized by different types of microorganisms, which enhance the hydrocarbon degradation through increasing their mobility, micelle formation, increasing bioavailability to bacteria degrading microorganisms. The addition of biosurfactant producing microorganisms into the bioremediation process showed remarkable advantages in the remediation of PAHs. Impacts of the bioavailability on the PAHs degradation are discussed in this chapter.

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Citing Literature

Facile synthesis of reduced graphene oxide using *Acalypha indica* and *Raphanus sativus* extracts and their in vitro cytotoxicity activity against human breast (MCF-7) and lung (A549) cancer cell lines

Original Article Published: 05 March 2021

Volume 11, article number 157, (2021) Cite this article

Parthipan Punniyakotti, Rajasekar Aruliah & Subramania Angaiah

Abstract

In the present study, an eco-friendly approach is adapted for the synthesis of reduced graphene oxide (rGO's) by a simple hydrothermal reaction using two plant extracts namely *Acalypha indica* and *Raphanus sativus*. After the hydrothermal reaction, GO turns into a black color from brown color, which indicates the successful reduction of graphene oxide. Further, various characterization techniques such as UV–Vis spectroscopy, Raman spectroscopy,

Fourier transform infrared spectroscopy (FT-IR), and X-ray diffraction is used to confirm the physicochemical properties of synthesized rGO's. Raman analysis confirms the reduction of GO by noticing an increase in the I_D/I_G ratio significantly. Field emission scanning electron microscopy and transmission electron microscopy clearly show the morphology and crystalline nature of rGO's. FT-IR spectrum confirms that the bioactive molecules of the plant extract (i.e. polyphenols, flavonoids, terpenoids, etc.) playing a key role in the elimination of oxygen groups from the GO surface. Further, the synthesized rGO's are tested for their potential against human lung and breast cancer cell lines. A significant cancer cell inhibition activity is obtained even in the less concentration of rGO's with IC_{50} values for lung cancer cell lines are 38.46 µg/mL and 26.69 µg/mL for AIrGO and RSrGO, respectively. Similarly, IC_{50} values for breast cancer cell lines are 35.97 µg/mL and 33.22 µg/mL for AIrGO and RSrGO, respectively.

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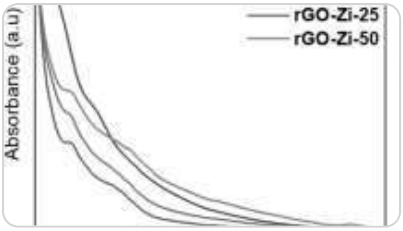

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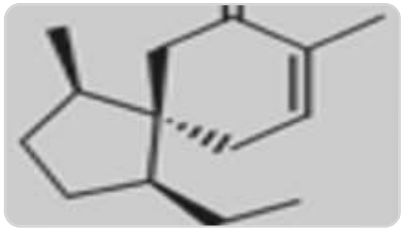

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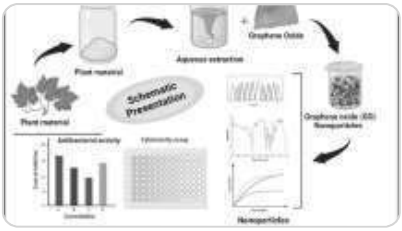

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Ethics declarations

Conflict of interest

The authors declare no competing financial interest.

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Reduced graphene oxide | Green synthesis | A549 human lung cancer cell | Cytotoxicity

ACS APPLIED POLYMER MATERIALS

Wettability Alteration of the Oil-Wet Carbonate by Viscosity-Augmented Guar Galactomannan for Enhanced Oil Recovery

Sandeep Rellegadla, Shikha Jain, Jitendra S. Sangwai, Meeta Lavania, Banwari Lal, Lisa Gieg, Aruliah Rajasekar, Achinta Bera, and Akhil Agrawal*

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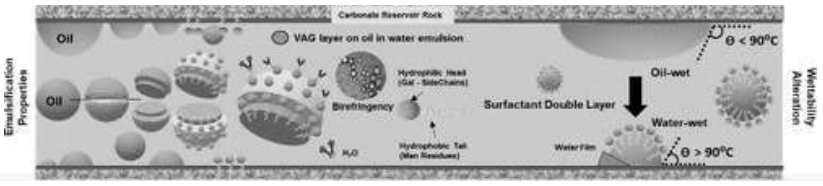
SI Supporting Info (1) »

SUBJECTS: Inorganic carbon compounds, Lipids, Petrochemicals, Polymers, Wetting

Abstract



The exhaustion of oil reserves encourages enhanced oil recovery (EOR) from mature oil fields. Hence, several polymers, surfactants, and nanofluids are developed, improving the recovery of additional oil from such mature wells. However, increased cost, harsh reservoir environment, and lower stability presents a significant challenge in developing a multicharacteristic injecting fluid, addressing all the past issues during oil recovery. Besides, almost all the earlier studies have demonstrated only higher viscosifying properties of polymers and rarely inspected the polymers' capabilities in altering surface wettability. In the present study, a guar galactomannan solution called viscosity-augmented guar (VAG) was prepared by the heat treatment of crude guar and removal of insoluble impurities. The resulting VAG solution demonstrated stable viscosity at higher temperatures (65 °C) under changing reservoir conditions. Additionally, the wettability alteration potential of VAG was evaluated by investigating the changes in the contact angle of oil-wet carbonate surfaces when treated with the VAG solution. Experimental results showed a reduction in the contact angle of the oil-wet surface to 81° after treatment with VAG from original surface values of 102° with the brine solution, indicating wetting transition from oil-wet to intermediate water-wet conditions. Moreover, the Fourier transform infrared spectroscopy and thermogravimetric analysis revealed the VAG's ability to displace oil components from the oil-wet surface. Additionally, emulsification studies showed a birefringence phenomenon of oil-water emulsions with VAG, reinforcing a proposed mechanism for wettability alteration by forming structured films oriented around the oil droplets in emulsions. The EOR potential of VAG showed an additional oil recovery of 7.31% of original oil in place compared to 3.59 and 2.98% by commercial polymers like xanthan gum (XGU) and partially hydrolyzed polyacrylamide, respectively. Overall, the favorable results of the VAG polymer are promising for promoting EOR, and for the first time, this study shows a polymer behavior in altering surface wettability of reservoir rocks and adds up different dimensions for future studies.

KEYWORDS: viscosity augmented guar, polymer Rheology, wettability alteration

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acsapm.1c00059.

- Experimental procedures, XRD analysis data, intrinsic viscosity values, kinetics of thermal degradation, qualitative wettability test results and polymer flooding ORSB setup, and recovery data and procedures including crude oil standard preparation (PDF)

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Bacterial community analysis of biofilm on API 5LX carbon steel in an oil reservoir environment

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Punniyakotti Elumalai, Mohamad S. AlSalhi, Sanjeet Mehariya, Obulisamy Parthiba Karthikeyan, Sandhanasamy Devanesan, Punniyakotti Parthipan & Aruliah Rajasekar

Abstract

This study aimed to characterize the biofilm microbial community that causes corrosion of API 5LX carbon steel. API 5LX carbon steel coupons were incubated with raw produced water collected from two oil reservoir stations or filter-sterilized produced water. Biofilm 16S rRNA amplicon sequencing revealed that the bacterial community present in the biofilm was dominated by Proteobacteria, including *Marinobacter hydrocarbonoclaustics* and *Marinobacter alkaliphilus*. Electrochemical analysis such as impedance and polarization results indicated

that Proteobacteria biofilm accelerated corrosion by ~ twofold (2.1 ± 0.61 mm/years) or ~ fourfold (~ 3.7 ± 0.42 mm/years) when compared to the control treatment (0.95 ± 0.1 mm/years). Scanning electron and atomic force microscopy revealed the presence of a thick biofilm and pitting corrosion. X-ray diffraction revealed higher amounts of the corrosion products Fe_2O_3, γ-FeOOH, and α-FeOOH, and confirmed that the microbial biofilm strongly oxidized the iron and contributed to the acceleration of corrosion of carbon metal API 5LX.

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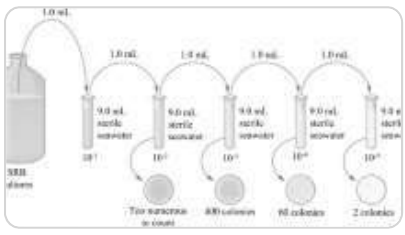

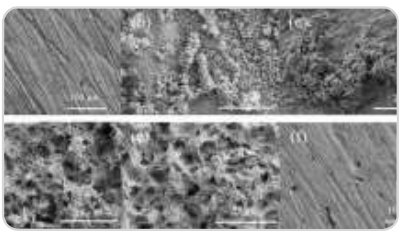

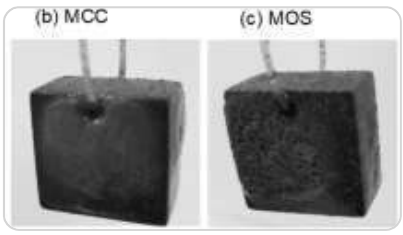

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Ethics declarations

Conflict of interest

The authors declare that they have no conflict of interest.

Additional information

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Electronic supplementary material

Below is the link to the electronic supplementary material.

Supplementary file1 (DOCX 133 kb)

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Biogenic Nanoparticles and Strategies of Nano-bioremediation to Remediate PAHs for a Sustainable Future

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Biotechnology for Sustainable Environment

Punniyakotti Parthipan, Chandar Prakash, Dhandapani Perumal, Punniyakotti Elumalai, Aruliah Rajasekar & Liang Cheng

Abstract

Polyaromatic hydrocarbons (PAHs) are considered as dangerous contaminants in water and soil, which are highly toxic, and also carcinogenic to living organisms including humans. The concerns on the PAHs removal are increased due to the difficulties in their removal from contaminated water and soil. Bioremediation technology is the most promising, cost-effective, and eco-friendly approach to remove the hydrocarbons by using potential

microorganisms. Nevertheless, the existing bioremediation technology has important limitations, such as, poor efficiency of microbial communities in the field, and lesser bioavailability of pollutants. To overcome these issues, advanced nano-biotechnology could be used. In recent studies, functionalized biogenic nanomaterials have shown possible PAH removal efficiency by adsorbing/desorbing them. Also, nano-sized photocatalysts can be used for photocatalytic oxidation of adsorbed or separated PAHs. Combining these integrated approaches will make a significant impact on the bioremediation of PAH contaminants. Nano-bioremediation could play an important role in mobility, micelle formation, and increasing bioavailability, which will assist in the removal/utilization of PAHs by biological (i.e., using microorganisms) or physicochemical (i.e., photocatalysis) methods.

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Bacillus megaterium–induced biocorrosion on mild steel and the effect of *Artemisia pallens* methanolic extract as a natural corrosion inhibitor

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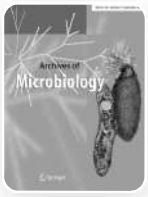

Archives of Microbiology

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Seenivasan Kokilaramani, Mohamad S. AlSalhi, Sandhanasamy Devanesan, Jayaraman Narenkumar, Aruliah Rajasekar & Muthusamy Govarthanan

Abstract

Methanolic extract of *Artemisia pallens* (MEAP) (Asteraceae) was explored as greenbiocorrosion inhibitor for mild steel 1010 in 1.5% sodium chloride environment. *Bacillus megaterium* SKR7 induces the development of biofilm on the metal surface and forms the pitting corrosion. MEAP was showed (25 ppm) optimum inhibition effect of biocorrosion and further corrosion rate was highly reduced (0.3335 mm/year) than the control system

(0.009 mm/year). The electrochemical study has supported the results with a higher value of total resistance (34 Ω cm^2) when compared to control systems. It reveals the formation of a protective layer on the metal surface and reduces the adsorption of biofilm. This was due to the antimicrobial effect of MEAP. Overall, the results recognized that MEAP used as a green corrosion inhibitor for MS 1010 with 83% inhibition efficiency.

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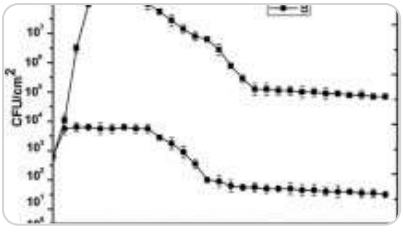

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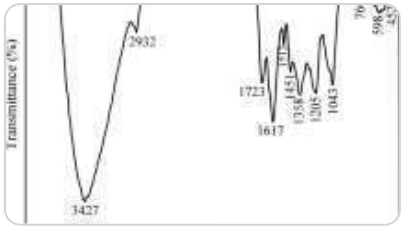

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Ethics declarations

Conflict of interest

All the authors listed in the manuscript declare no conflict of interest in this publication. The tannery cooling tower water and biofilm samples were collected from the authorities of Ranipet tannery effluent treatment Co. Ltd., Tamil Nadu, India with proper permission.

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Bioprocess and Biosystems Engineering

Aims and scope

Submit manuscript

Parthipan Punniyakotti, Pratheep Panneerselvam, Dhandapani Perumal, Rajasekar Aruliah & Subramania Angaiah

Abstract

In the present study, a copper nanoparticle (Cu NPs) was synthesized by a green synthesis method with *Cardiospermum halicacabum* leaf extract. The surface area of Cu NPs was measured with dynamic light scattering (DLS). UV–Vis spectrum clearly illustrates the typical absorption peak of Cu NPs. The crystalline property of Cu NPs was confirmed from the XRD pattern. TEM analysis clearly indicates the average particle size of synthesized Cu

NPs was in the range of 30–40 nm with hexagonal shape. Energy-dispersive spectroscopy confirms the major strong peaks of Cu NPs. FTIR analysis confirms the existence of various functional biomolecules over the metal nanoparticles and they are playing an important role in the formation of Cu NPs. The antibacterial and anti-biofilm analyses were carried out to confirm their aptitude for biomedical applications. Interestingly, Cu NPs control the development of biofilm by attaching over the cell wall and disturb their growth and development.

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Electronic supplementary material

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Original Paper Published: 16 May 2019

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Raja Kumaresan Sarankumar, Adikesavan Selvi, Kadarkarai Murugan & Aruliah Rajasekar

Abstract

The present investigation deals with remediation of chromium (Cr(VI)) by electrokinetic (EK) and bio-electrokinetic (BEK) processes. The experiments were conducted using spiked concentration of 1 g/kg of chromium in the soil. The results showed a maximum chromium removal of 40.12% and 90.4% by EK and BEK, respectively, at the end of 7 days. Chromium-resistant alkalophilic bacterial strain, *Bacillus licheniformis* SR3, was used as bio-anolyte counterpart in BEK. During the process, the variations in electrical gradient and pH were

studied and compared between EK and BEK. Chromium removal was further confirmed by UV spectroscopy, FTIR and XRD studies. The results of FTIR showed notable difference in the intensities of the peak, thus confirming the effective remediation by BEK. The obtained results have found to support BEK integrated system as an effective remediation option for cleaning up chromium-contaminated soil environments.

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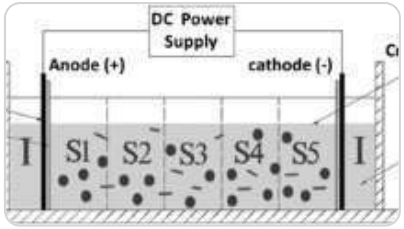

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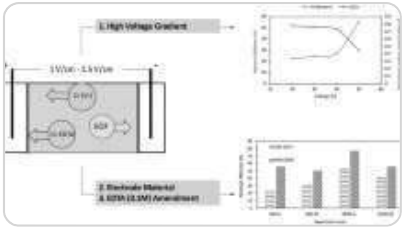

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A Decennials Study of the Trend of antibiotics studies in China

TONG YU, Adharsh Rajasekar, Songhe Zhang

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Abstract

Antibiotics are one of the greatest inventions in human history and are used worldwide on an enormous scale. Besides its extensive usage in medical and veterinary arenas to treat and prevent the infection, its application

from its inception (1991) to December 6, 2021, from the Web of Science Core Collection database. However, with few authors and institutions focusing on long term studies, we found the quality of contributions were uneven. Studies mainly focused on areas such as food science, clinical research, and environmental studies, including Molecular biology, genetics and environmental, ecotoxicology, and nutrition, which indicate possible primary future trends. Our study reports on including potentially new keywords, studies' milestones, and their contribution to antibiotic research. We offer potential topics that may be important in upcoming years that could help guide future research.

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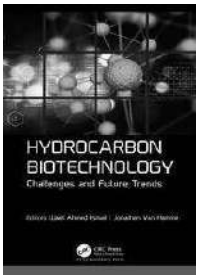

Biosurfactants-Mediated Remediation of Hydrocarbon Pollution

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This article is part of the Research Topic
Biosurfactants - A Next Generation Biomolecules for Enhanced Biodegradation of Organic Pollutants
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Editorial: Biosurfactants—A next generation biomolecules for enhanced biodegradation of organic pollutants

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Editorial
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Liverpool, United Kingdom

Editorial on the Research Topic
Biosurfactants—A next generation biomolecules for enhanced biodegradation of organic pollutants

Microorganisms can produce a range of amphipathic molecules (containing both hydrophobic and hydrophilic moieties) that accumulate at interfaces between hydrophobic and hydrophilic phases. When biosurfactant accumulations reach threshold concentration "micelles" arise which act to reduce the surface and interfacial tensions to form emulsions. Biosurfactants differ greatly in their structures, which affects their specific functions/applications. Different types of biosurfactants are used in many industries/applications including detergents, pharmaceuticals, oil recovery, and bioremediation. Biosurfactants have numerous advantages over chemical surfactants in industrial bioprocess applications including (i) reduced toxicity, (ii) easily biodegradable, (iii) stable at higher pH and temperature, and (iv) result in greater foaming (Parthipan et al., 2017; Prakash et al., 2021). Furthermore, they can be synthesized using cheap raw materials such as agro-wastes and industrial waste materials that reduce the production costs and reduce waste disposal and associated environmental problems (Parthipan et al., 2021).

The focus of this Research Topic was to explore bioremediation applications of biosurfactants from hydrocarbon-degrading microbes. Deployment of biosurfactants in the bioremediation of hydrocarbon contaminants is gaining attention due to their significant outcomes, such as increasing bioavailability, effective removal efficiency, biocompatibility, reducing the uses of toxic chemicals (chemical surfactant), etc. (Rahman et al., 2003). Hydrophobic hydrocarbons (petroleum hydrocarbons and other environmental pollutants) can cause adverse

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Christina Nikolova and Tony Gutierrez

from the environment (Lee et al., 2018). Conventional remediation methods such as chemical (chemical oxidation (with harmful oxidants), solvent extraction/washing), physical (incineration and *in-situ* thermal desorption), or biological (with ineffective microbes) methods have many limitations such as higher cost, slow process, and ineffectiveness (Hentati et al., 2021).

Biosurfactants can enhance the bioavailability of hydrocarbon molecules, thus improving hydrocarbon biodegradation. This Research Topic aimed to explore recent advancements in this research area with a focus on the identification of novel strains with effective biosurfactant producing capability and the impact of biosurfactants on the degradation of the different hydrophobic organic pollutants and heavy metal mitigation.

In the study "Identification of four secreted aspartic protease-like proteins associated with sophorolipids synthesis in *Starmerella bombicola* CGMCC 1576" by Liu et al. used site-directed deletion mutagenesis for the improvement of sophorolipids production by the yeast strain *Starmerella bombicola*. The sophorolipids production level was increased to 90% with 60.71 g/L in the strain Δ*sapl* after removing the coding genes clusters (*sapl1, sapl2, sapl3*, and *sapl4)* under the ammonium sulfate as a nitrogen source. However, no increase in sophorolipids production was observed in Δ*sapl* at yeast extract condition. Compared to that of the wild-type strain, the expression levels of the key genes for sophorolipids synthesis are upregulated in Δ*sapl* under ammonium sulfate conditions. Overall study, summarize that *sapl* gene cluster suppressed the key genes involved in the sophorolipids synthesis under ammonium sulfate condition by restraining the expression of the key genes involved in sophorolipids synthesis.

Optimization of the growth parameters could help microbes to produce their metabolites with the highest capability. A small change in their

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Genomic Insights Into the Lifestyles of Thaumarchaeota Inside Sponges

Markus Haber, Ilia Burgsdorf, Kim M. Handley, Maxim Rubin-Blum and Laura Steindler

producing *Bacillus* sp. SHA302 strain and optimize biosurfactant production using the Plackett-Burman and response surface methodology. Different parameters such as pH, carbon source, and nitrogen source were optimized and obtained 5.74 ± 0.52 g/L of biosurfactant with 29.2 ± 0.71 mN/m surface tension activity. Increasing biosurfactant concentration leads to a decrease in the surface tension and finally achieving critical micelle concentration where micelles are formed. The highest lead and zinc release rate was observed (53.8% ± 1.4 and 39.3% ± 1.7, respectively) with the highest CMC with acid treatment. While increasing the concentration, heavy metals in adsorption sites form complexes (anionic biosurfactants form non-ionic complexes with metallic cations) with the biosurfactant and are further released into the soil solution.

A review paper published on this Research Topic "Biosurfactant: A Next-Generation Tool for Sustainable Remediation of Organic Pollutants" by **Sharma et al.** summarized the types of biosurfactants, the importance, and advantages of the biosurfactant on environmental applications. The detailed properties of the biosurfactant and their role in the bioremediation of organic pollutants are elaborated.

Oil spillage in marine environments are very common due to the widespread exploration and transport of crude oil from offshore locations. Safe burning is one of the effective methods to remove oil spillage. In a study "Bioherder generated by *Rhodococcus erythropolis* as a marine oil spill treating agent" by **Yu et al.** described that the addition of herding agents at the oil slick edges may swiftly diminish the oil-water interfacial tension (<1 mN/m) around the oil slick edges, so negative spreading coefficient was generated, which enables the thickening of the oil slicks to reach a new equilibrium. For effective burning, oil floating on the sea surface needs to be 2–3 mm thick to counter heat loss at sea and to provide sufficient oil vaporization to maintain oil burning.

costs can be decreased by utilizing low-cost substrates, such as waste and by-products (sugarcane molasses, steep corn liquor, and soy waste). Also, production costs could be further reduced through the development of engineered strains and innovative bioreactors.

As mentioned earlier, screening and identification of potential biosurfactant-producing strains will assist and make a significant impact on environmental clean-up technology. In a study, "polyphasic analysis reveals potential petroleum hydrocarbon degradation and biosurfactant production by rare biosphere thermophilic bacteria from deception island, an active Antarctic volcano" by Schultz et al. isolated 50 different thermophilic bacterial strains with biosurfactant production capability, and among them, 13 strains were effective emulsifiers.

Overall, these five different studies demonstrated the effectiveness and importance of biosurfactants in the bioremediation of organic and inorganic pollutants. The outcomes of this Research Topic clearly indicated how biosurfactants are important for environmental and industrial applications. The major drawbacks in biosurfactant usage are problems connected with their synthesis, including a lack of efficient microorganisms, higher production costs, and low yield. Hence, improvements in surfactant chemistry, biotechnology, and remedial technology are essential to the future application of biosurfactants.

Author contributions

All authors listed have made a substantial, direct, and intellectual contribution to the work and approved it for publication.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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