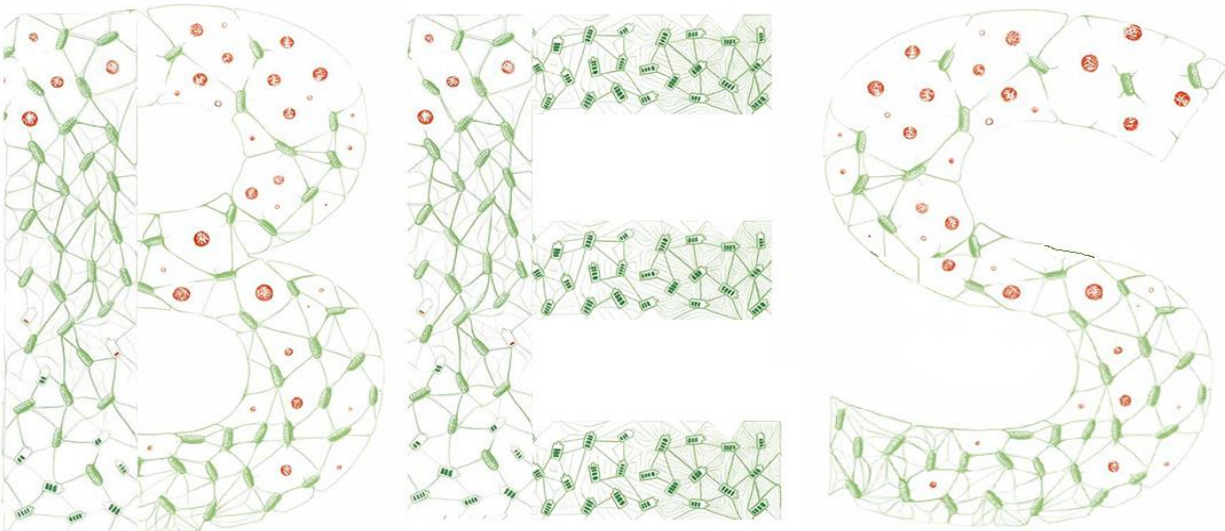


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Email: shyabiswas@biotechkiosk.com
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Editorial, Sales & Circulation Office

1330 NW 6th Street
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From the Publisher's Desk

Welcome to Biotechnology Kiosk!

The new issue of BK is now online for our readers with the regular features. This issue includes a perspective on the applications of bio-electrochemical systems for sustainable energy and editor picks along with a popular article.

We hope our readers will enjoy reading these news and views on the current cutting-edge topics that include latest research breakthroughs in different areas of medicine and biotechnology.

We look forward to receiving your feedback. We do hope that you will enjoy reading this issue of Biotechnology Kiosk. Please do write to us with your comments. Your suggestions are always appreciated.

Dr. Megha Agrawal & Dr. Shyamasri Biswas.

Editors-in-Chief, Biotechnology Kiosk





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Optimization of Bio-Electrochemical Systems

Abhijit Biswas*

USA Prime Biotech LLC, 1330 NW 6th St, Suite A-2, Gainesville, FL 32601, USA.



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Abstract

The ongoing research in the sustainable energy sector has shown tremendous potential of microbes or electro-active biofilms (EABfs). These biofilms act as the important component of bioprocessing technologies that are based on bio-electrochemical systems (BESs). EABfs exhibit unique characteristics including redox reactions and resilience against otherwise harmful products that make BESs promising for important applications in energy recovery in the form of electricity or hydrogen or even production of fuels or chemicals from CO₂. A deeper understanding of the mechanisms of EABfs characteristics is considered essential for the optimization of BESs for practical applications. To this end, a wide range of characterization techniques based on electrochemical, visual and chemical methods have been employed for the analyses of EABfs. These techniques can provide very valuable and wide-ranging information about EABfs that include performance, morphology and biofilm composition. Especially, significant attention has been paid to developing non-destructive visual techniques for EABfs characterization. The goal is to obtain in-situ information of EABfs functioning for industrial-scale development of BESs. Visual techniques are considered extremely useful for EABfs monitoring studies that can complement the information obtained with other characterization techniques. In this perspective, we have provided a short overview of various visual characterization techniques that have been proposed to study EABfs for the optimization of BESs.

Keywords: *Bio-electrochemical systems; electro-active biofilms; characterization; visual techniques*

*Corresponding Author

Dr. Abhijit Biswas

E-mail: abbtf@yahoo.com

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INTRODUCTION

The dual challenge of rapidly rising world population with increasing energy demands and simultaneous depletion of fossil fuel reserves has accelerated research and developments in sustainable energy and recovery technologies that include recovery of energy and nutrients from wastewater [1, 2]. To this end, bio-electrochemical systems (BESs) have been shown very promising to recover resources such as nutrients and energy from wastewater. BESs employ microorganisms and the microbes that act as catalysts can use external electron acceptors or electron donors as electrodes for chemical conversions [3-5]. Hence, BESs are capable of converting chemical energy into electrical energy (and vice-versa). BESs can be broadly divided into two major sustainable energy technology platforms - microbial fuel cells (MFCs) and microbial electrolysis cells (MECs) (Figure 1) [6].

Previous studies showed the potential of MFCs that were employed to convert organic wastes including low-strength wastewaters and lignocellulosic biomass into electricity. On the other hand, MECs were employed where electrical energy was used to produce hydrogen or other industrially useful products including caustic and peroxide. In addition, researchers also designed and implemented BESs to recover nutrients, metals or removal of recalcitrant compounds. Furthermore, solar energy was used to generate electricity by implementing photosynthetic micro-organisms along with higher plants. Thus, it is possible to realize a range of potential applications of BESs by utilizing the diversity on microbial and

enzymatic catalysts that are offered by nature (Figure 1) [6].

In addition to MFCs and MECs for energy recovery in the form of electricity or hydrogen, another type of energy technology platform has been considered, which is based on Microbial Electrosynthesis Cell (MES) (Figure 1) [6]. MES system is used for the production of fuels or chemicals from CO₂. All these technologies are governed by the working principle on electro-active microbial communities (such as electro-active biofilms). However, the difference in these technologies is that MFCs and MECs operate with exoelectrogens at the anode; whereas, MESs uses eletrotrophs at the cathode for operation [7-9].

BESs are made of electro-active biofilms (EABfs). These biofilms are essentially electro-active bacteria that develop on the surface on an electrode. During the conversion of chemical energy into electrical energy and vice versa, the electro-active bacteria acts as catalysis to promote the energy conversion. Since these biofilms play extremely important roles in BESs formation and operation, the current research has focused on studying the required operating conditions for bio-catalysis of EABfs. For the improvement of BESs performance and optimization, there have been studies on employing advanced materials and optimized electrode designs for desired interaction between EABfs and electrode surface. It is not only important to gain insights into the behavior of electro-active bacteria and EABfs with respect to the operational conditions that include electrode

designs and electrode current and potentia, but also characterization studies are

considered essential to better understand the mechanisms of EABfs [10-15].

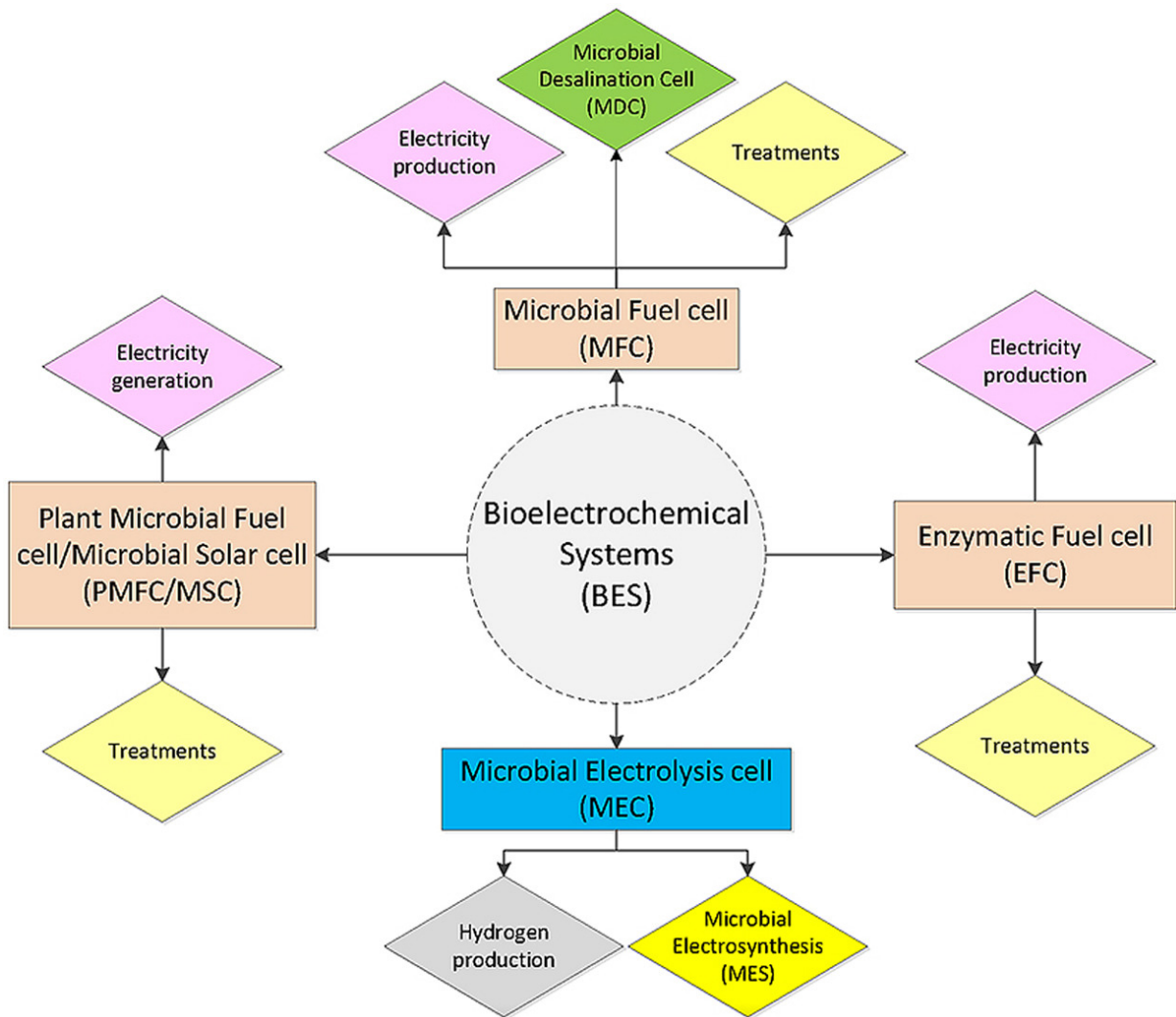


Figure 1: Bio-electrochemical systems and various applications [Source: Renewable Energy (2016)].

Important Connection between Electro-Activity and Biofilm Characteristics

Various studies have indicated that it is critically important to establish the relationship between electroactivity and biofilm characteristics for the optimization of

BESs. This is due to the reason that increased electroactivity has been found to result in efficient product formation in all BES operations. Therefore, a significant attention has been paid to generate new avenues that enable modifying the electrode surface to enhance cellular attachment by an increased

surface area. In addition, more robust biofilm formation by engineering strains is also suggested. All these efforts are aimed at enhancing biofilm formation on electrodes. It is believed that efficient design and optimization of BESs can be achieved by fine-tuning bio-electrochemical cell parameters. To this end, various research tools such as mathematical modeling that allows for scalability and investigation of microbe–electrode interactions and the effect of electrode type have been proposed. Additionally, electrode modifications, microbial composition along with understanding the cellular processes underlying electron exchange and product formation have been considered in all current and future research directions to optimize BESs for practical applications. Especially, research efforts are directed to improving Coulombic efficiency and optimizing biofilm formation along with bioprospecting for electroactive strains and electron exchange mechanisms. These efforts are expected to impact the development of optimization of BESs in the future [16-20].

Characterization of Electro-Active Biofilms: Conventional Techniques

Several types of conventional characterization techniques have been employed for the in-depth analysis of EABfs. These techniques can be employed for electrochemical, visual, and chemical analyses to study EABfs [21]. A wide range of information about EABfs including knowledge about microbial activity, biofilm structure, thickness and composition, mass transfer limitations and also conductivity can be obtained by employing these techniques [22-24]. For example, researchers have

employed electrochemical techniques to determine the general performance indicators of EABfs to reveal the relationship between electric current and potential. Some commonly used electrochemical techniques include Cyclic Voltammetry (CV), potentiostatic control, and Electrochemical Impedance Spectroscopy (EIS). These electrochemical techniques have been employed to study different stages of EABfs growth. This also includes obtaining information about microbial activity, information about redox active compounds along with charge storage. Researchers have also shown employability of chemical analyses in BESs primarily to estimate the concentration of substrate along with products in the bioreactor. Establishing the connection between these concentrations and the electrons exchanged at the electrode(s) is very important as it gives insights into the coulombic efficiency of electrodes [25-28]. Despite some usefulness of these conventional characterization methods, these techniques are usually destructive that often lead to the destruction of biofilm after performing a given analysis. It makes extremely difficult to monitor EABfs during the experiments [29]. We will discuss in the following section some relevant non-destructive visualization techniques for EABfs.

Non-Destructive Modern Visualization Techniques for Biofilm Characterization and Monitoring

It is known that EABfs are biological matrixes that comprise embedded electro-active bacteria. In this scenario, the biofilm composition and mechanical properties are usually unique that pose serious challenge to

characterize and study these films. This is due to the reason that as EABfs grow on an electrode, the matrix composition constantly changes with time. This makes characterizing EABfs and predicting their performance very difficult. To overcome these challenges, researchers have proposed visual techniques that can be used for multiple functions. These include detecting specific compounds that are present in the extracellular matrix and also to visualize the distribution of the biofilms as a function of time. Visual techniques can be leveraged to image the morphology and cellular density in the biofilm structure. The major advantage is the ability to gain insights into the 3D distribution of biofilms. This enables identifying species that comprise the biofilm and also mapping their disposition in the biofilm. Particularly, the combination of electrochemical and visual techniques is considered ideal that allows to obtain new information about EABfs. Furthermore, it is believed that in-situ visualization techniques for EABfs can be leveraged to perform monitoring of biofilm characteristics, and follow the growth of biofilm over time. To this end, some recognized visual techniques including Confocal Laser Scanning Microscopy (CLSM) and Optical Coherence Tomography (OCT) have been shown very useful to study EABfs along with monitoring film thickness, composition to localize microbial species [30, 31].

In addition to CLSM and OCT, there are some other visual techniques that have been shown in some studies. These include Raman Microscopy, Scanning Electron Microscopy (SEM), Scanning Transmission

X-ray microscopy (SAXM) and Magnetic Resonance Imaging (MRI) that have been employed to obtain valuable information on electrochemical data. Among various visualization methods, CLSM is considered very promising and useful for visualization, quantification, 3D imaging along with characterization of biofilm composition. MRI and OCT are also frequently employed that allow for 3D imaging. These techniques have the ability to determine the biofilm distribution and its volume without destroying the sample. In addition, Raman and STXM are also non-destructive methods for studying the biofilm composition [32-34].

Recently, light sheet fluorescence microscopy (LSFM) was shown for nondestructive, label-free and in-vivo imaging of large electro-active biofilm specimen. The technique was shown to function even at nontransparent surfaces. In this study, researchers demonstrated LSFM for label-free analyses of prokaryotes on electroactive biofilms (Figure 2) [31]. Biofilm growth was linked to the production of current serving as measure of metabolic activity in-vivo. This was done by monitoring with high spatial and temporal resolution. After 35 h of exponential growth, they showed a growth of homogeneous biofilm with a thickness of 9 μm . Subsequently, a stratification of the biofilm including the formation of 3D structures was conducted for several hours. During this process, light reflection was shown to be sufficient that was used to visualize the biofilm structure and development over time. Fluorescence staining was employed to confirm the final morphology [31].

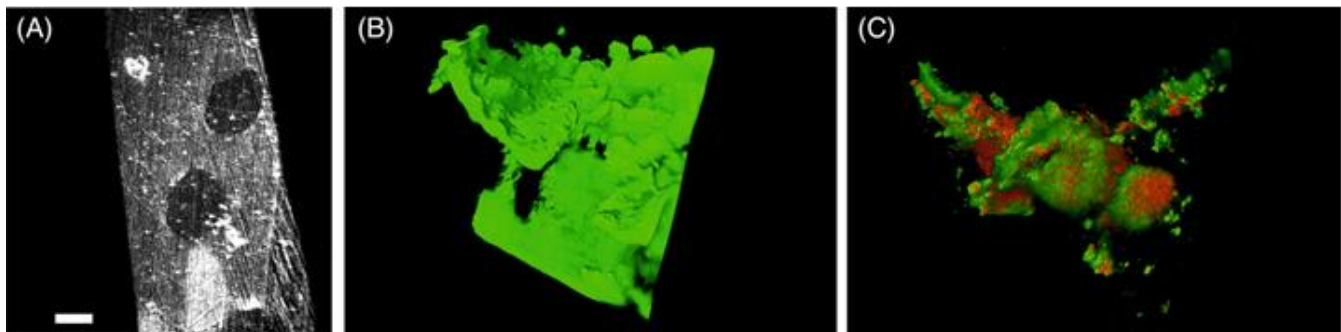


Figure 2: (A) Visualization of growth substratum is shown without any biofilm. (B) A comparison to a three-dimensional adult biofilm based on light reflection is shown. (C) Reanalysis of the same biofilm sample is shown after LIVE/DEAD nucleic acid fluorescent staining confirming the organization of the individual cells. The three-dimensional organization of the biofilm can be seen in reflection [Source: *Cytometry Part A* (2020)].

Conclusion

Bio-electrochemical systems (BESs) offer tremendous potential in today's sustainable energy sector. Optimization of BESs is hugely important research field that involves characterization to understand the formation of electro-active biofilms that are integral components of BESs. To this end, several very promising characterization techniques for electro-active biofilm visualization and 3D imaging on an electrode have been proposed. Such characterization of biofilms is considered critical to optimize BESs technology and take the field of sustainable energy to the next level. Additionally, combinatorial approaches including conventional characterization of biofilms and non-destructive visualization methods could pave the way to new breakthroughs in electro-active biofilms in the future, which could significant benefit the BESs technology in the future.

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Best Practices for Small Business Owners in Biotech

By Julie Morris

Juliemorris.org

Biotechnology is one of the most profitable, impactful, and life-affirming industries to operate in – albeit one of the most challenging. The competition is stiff, funding hard to come by, and medical enigmas are always tough to crack. You need to think on your feet and be decisive to survive and, indeed, thrive. [Biotechnology Kiosk offers advice](#) to small business owners on how they can be more successful in this demanding niche:

Focus on the talent from the get-go

Finding (and retaining) quality talent is one of the most challenging aspects of being in biotech. The talent pool is limited and promising candidates are likely to be snapped up by bigger organizations. As such, having a [fine-tuned and well-oiled hiring process](#) is critical to locating good people (who will make you successful). Some suggestions are using professional recruiters (or techniques), having a fast interviewing process, and making snappy (but informed) hiring decisions.

Prioritize [technical skills as well as team fit](#) – replacing talent because of a poor fit can be incredibly expensive down the road. Finally, recruit with an eye to the future – the skills you need now may not be relevant later.

Come up with strategies to retain talent

As a small company, you likely won't be able to match the salaries or monetary perks offered by larger organizations. A competitive employee reward program and development initiatives can help you to close this monetary gap. [Some suggestions from MTI Events](#) are allowing remote work, profit sharing, extra time off, and peer-to-peer recognition. A healthy working culture and growth opportunities can be just as attractive as money to talent.

Stay on top of your finances

Biotech is a risky business, as you likely know, where failures often outnumber successes. Having a steady flow of capital is essential to keep your research or clinical trials (if applicable) going and weather financial storms. As such, a significant portion of your time and energy should be dedicated to managing money: attracting investors, maintaining a healthy cash flow, and avoiding needless expenses. Understand investor sentiment better to align yourself with what

they prioritize (multiple drugs in phase 2 trials and a solid corporate philosophy, among other things). Make note of grants, tax breaks, and other benefits that you may be able to apply for. Finally, stay in touch with the local [investor culture and market conditions](#) to attract more funding.

Put yourself out there with marketing

Biotech often involves cutting-edge concepts and research that are not well-understood by the general public – including potential investors, employees, and customers. You can use marketing to clarify what you do, raise your profile, and better connect with your audience. This will make you more attractive to your various stakeholders. Some [high-impact marketing strategies for biotech businesses](#) include competitive analysis, market analysis, and paid marketing.

Understand that the FDA isn't your enemy

[According to a CAP report](#), the FDA approves an overwhelming majority of new drugs. Although the process may be laborious, it's faster than equivalent processes in other countries. Furthermore, the FDA is forthcoming and surprisingly helpful with its policies and processes. Being scientists themselves, FDA regulators are supportive, knowledgeable, and typically interested in the work you're doing – don't hesitate to approach them. Building a good relationship with your regulators can be helpful on several levels – it can help you avoid pitfalls and generally get your product through the door faster.



Be careful with your IP

R&D arrangements – where you tie up with other organizations to pool your talent and other resources to achieve goals – are the norm in biotechnology, especially for smaller companies. You should prioritize safeguarding your IP and avoid terms that may set you back or make it hard for you to enter into licensing deals and arrangements down the road. [TechCrunch offers some suggestions on protecting your IP](#).

Conclusion

To sum it up, focus on building a solid team, controlling your finances, and marketing yourself better. Other general business skills – time management, delegation, consistent goal setting, networking, and prioritization – will stand you in good stead.

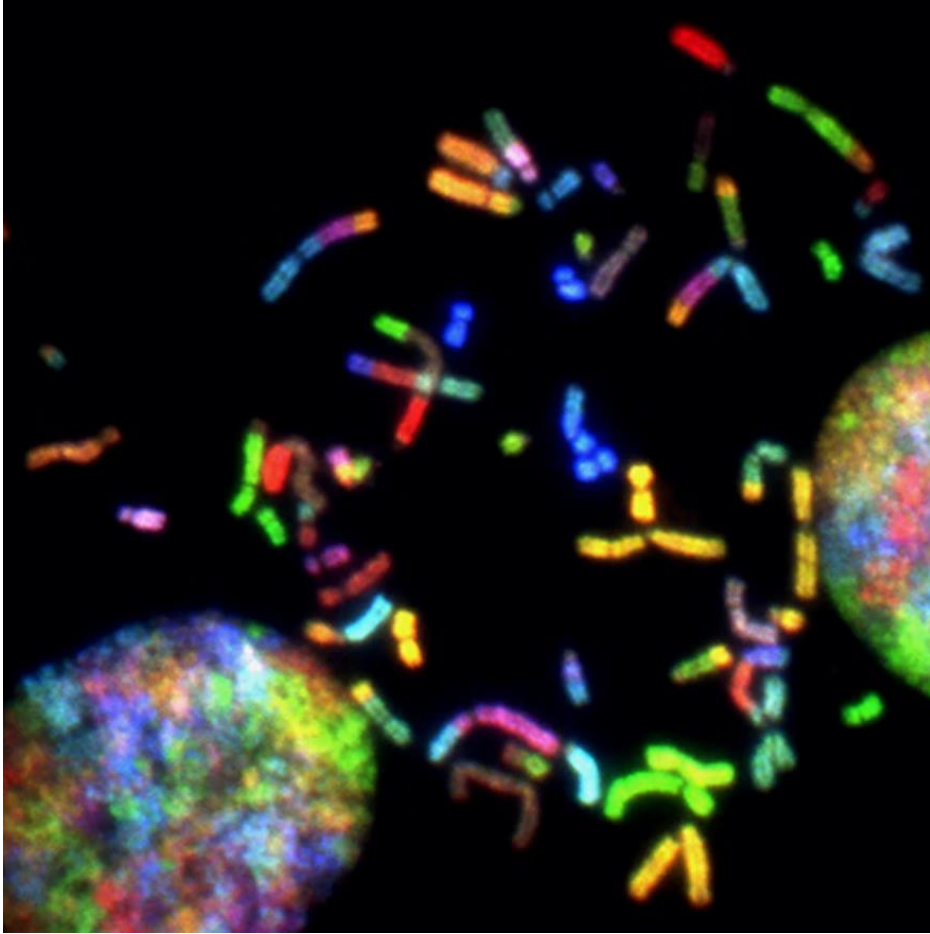


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Biotechnology Advances around the World

Editor's Picks

Every issue of Biotechnology Kiosk presents select latest research news picked by the editors-in-chief on significant research breakthroughs in different areas of biotechnology around the world. The aim is to promote further R&D in all of these cutting-edge areas of biotechnology. The editors have compiled and included the following innovations and breakthroughs to highlight the latest biotechnology advances.



Dr. Megha Agrawal
Co Editor-in-Chief



Dr. Shyamasri Biswas
Co Editor-in-Chief

Migraines

Migraines affecting the sleep cycle

Rapid Eye Movement (REM) sleep is known to be the stage of sleep that involves the most brain activity and vivid dreams. REM sleep is considered very important for learning and memory function.

In a recent study by researchers in the U.K., it is reported that adults and children with migraines may get less quality, REM sleep time compared to people who don't have migraines. Researchers conducted a meta-analysis and they published their work in *Neurology*[®], the medical journal of the American Academy of Neurology (Subjective Sleep Quality and Sleep Architecture in Patients With Migraine: A Meta-analysis. *Neurology*, 2021, DOI: <https://doi.org/10.1212/WNL.00000000000012701>). In this study, children with migraines were also found to get less total sleep time compared to their healthy peers but took less time to fall asleep.

In the meta-analysis, researchers included 32 studies, involving 10,243 people and the participants were asked to complete

a questionnaire to rate their own sleep quality. It asked about sleep habits that included how long it takes to fall asleep, total sleep time and also the use of sleep aids. Higher scores indicated worse sleep quality.

It was reported that adults with migraines overall had higher average scores on the questionnaire than people without migraines with a moderate amount of the difference due to the migraines. The difference was found even greater in people with chronic migraines.

Upon analyzing sleep studies, researchers found adults and children with migraines had less REM sleep as a percentage of their total sleep time than their healthy counterparts. Furthermore, when looking at children with migraines, it was found that they had less total sleep time, more wake time, and shorter time for sleep onset than children without migraines. This study paves the way to a better understanding of migraines and also how the migraines affect sleep patterns.

Alzheimer's disease

Prevention and treatment of Alzheimer's disease

The widely recognized hallmark feature of people living with Alzheimer's disease (AD) is thought to be the progressive accumulation of toxic protein deposits that occur within the brain called beta-amyloid. Despite the research advances made in AD, it is still not fully known about the origin of the amyloid and why it gets deposited in the brain.

Recently, a ground-breaking research by a team of researchers in Australia has discovered a likely cause of Alzheimer's disease. It is considered a significant finding that offers potential new prevention and treatment opportunities for progressive neurodegenerative diseases.

The study was published in the PLOS Biology journal (Synthesis of human amyloid restricted to liver results in an Alzheimer disease-like neurodegenerative phenotype. PLOS Biology, 2021; 19 (9): e3001358 DOI: <https://doi.org/10.1371/journal.pbio.3001358> . In this study, researchers tested on mouse models and they identified that a probable cause of AD was the leakage from blood into the brain of fat-carrying particles transporting toxic proteins.

In this study, it is reported that genetic modification of C57BL/6J mice engineered to synthesize human A β only in liver (hepatocyte-specific human amyloid (HSHA) strain) has marked neurodegeneration concomitant with capillary dysfunction, parenchymal extravasation of lipoprotein-A β , and neurovascular inflammation. Furthermore, researchers observed that the HSHA mice showed impaired performance in the passive avoidance test. This suggested impairment in hippocampal-dependent learning. Thus, this study paves the way to establishing a new theory and evidence of a lipoprotein-A β /capillary axis for onset and progression of a neurodegenerative process. Furthermore, this finding shows the possibility of addressing the abundance of these toxic protein deposits in the blood through a person's diet that could potentially lead to the development of some drugs that could specifically target lipoprotein amyloid. This could reduce the risk or slowing the progression of Alzheimer's disease.

Compiled and Edited by Dr. Megha Agrawal and Dr. Shyamasri Biswas.



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