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Quest

Editorial

Mars has always been a symbol of our wanting for other worlds. The myth of its magical experience was shattered into pieces until India created a history on 24th September 2014, becoming the first country to successfully get a spacecraft into the Martian orbit on its maiden attempt. The careful planning and execution of the Mars Mission and the development of the cryogenic engine and GSLV Mark II brought about a turnaround.

The power of hard work resulted in victory, power means 'Shakti' which means Lord Amba, who is been worshipped on the auspicious period of Navratri. For India it is double celebrations this time with the Durga Ashtami's beginning and its MOM's success.

With this let's hope for more successes and the energy to fulfil them throughout. Congratulating you all. Jai Hind!

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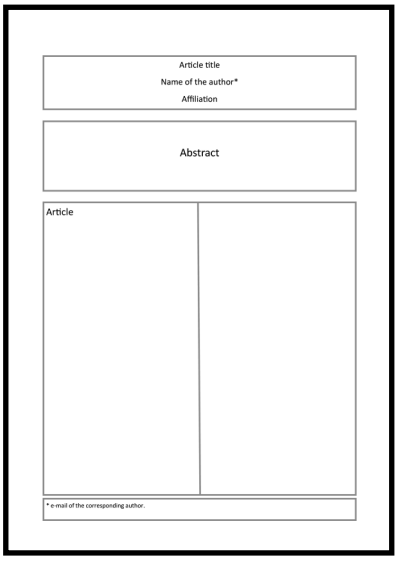
Index

Notice to Authors

Manuscripts submitted to Quest should adhere to below mentioned criteria.
Research News: About 400 words (1 page)
Research Article: About 2000 words (4 pages)

Common for all: -
Font: Calibri
Font Size: 14
Columns: 2
Line Spacing: 1
Margin: Narrow
References: 1) In text citing, S No, Superscript.
2) Author's name (s), *Journal name*, **Volume No**, Page No**,** (year).
3) Maximum number of references should not exceed than 25.



Book Review Competition held on 9th August on the occasion of World Book Lover's Day celebration

1st Prize: Naisargi Bhatt, IGBT, Sem-V
2ndPrize: Venkata Anand Parnandi, M.Sc. Microbiology, Sem-III
3rd Prize: Rajvi Gohil, IGBT, Sem-I

***Title of the Book*: The Last Lecture**
Author Name: Randy Pausch With Jeffery Zaslow
Published Year: 2008
Name Of Publication: Hyperion

Summary
"We cannot change the cards we are dealt, just how we play the hand" –RANDY PAUSCH
The LAST LECTURE is an inspiring and motivational book, that may land you a reality check about life, or maybe even just a good cry; through stories and aphorisms about a professor of computer science at Carnegie Mellon University, Pennsylvania.

Randy Pausch had been diagnosed with ten tumors in his liver. The university where he worked, offered a lecture series for students wherein the educators shared advise based on lessons learned from their lives. Randy had his chance to share before he died. "What wisdom would we impart to the world if we knew it was our last chance?"

He was a father of three young children, and married to the woman of his dreams. "I knew what I was doing that day," he writes in the book's introduction, "Under the ruse of giving an academic lecture, I was trying to put myself in a bottle that would one day wash up on the beach for my children."

His wife, Jai was not enthusiastic as Randy for his lecture. She knew, he would pour a lot of time and energy into the lecture. But then she relented when she saw how much it meant to him to have a recording of his life lessons for their three children. It was his chance to say goodbye to many people in his working world.
"I've never understood pity and self-pity as an emotion. We have a finite amount of time whether short or long it doesn't matter. Life is to be LIVED" he says.

Throughout the book, we revisit Randy Pausch's fulfillment of his childhood dreams, the stories that illustrates themes such as dreaming big, hard work, perseverance, sacrifice, courage, a positive outlook and dealing with adversities. Rather than focusing on his last moments, it was a speech about living, each day as though it was your last.
"Experience is what you get when you didn't get what you wanted."

It's all about how you frame that experience, he explains. It's not about what happens to you bout about how you react to what happens to you that makes a difference.
Randy Pausch is the main character of this book. After aggressive treatment, he still couldn't get better and he knew his time on earth was limited. But, he believed in moving forward which takes, the flow of story at peak and rules over the heart.

"It's not how hard you hit, it's how hard you get hit.. and keep moving forward." The ones who keep getting up and keep moving forward are the ones who win in the end. Life is hard,

but compared to what? He discusses about the disease and its effects on the remainder of his life in the chapter entitled 'Its About How to Live Your Life.' He points out at the end of the lecture that we all have a finite time. He advices to count blessings and be thankful, that attracts more to be thankful for. Complaining cannot help us achieve our goals.

Moral Of The Story

'Time is all you have. And you may find one day, that you have less time than you think' – RANDY PAUSCH

Never take time for granted. Each day you wake up is another day you are supposed to be here. There is a reason for your life. Hope to fill the pages of your 'Once upon a time' story right and vow to make each moment count. To die with dignity and grace is in everyone's "to do" list but to treasure your "sheer existence" has a whole host of other emotions that rule your life. The book needs to be shared for generations to come.

-Contributed By: Naisargi Bhatt, M.Sc. IGBT, Sem-V.

***Title of the Book*: Inferno**

Author Name: Dan Brown
Published Year: 2013
Name Of Publication: Doubleday

Summary

"Inferno" is Dan Brown's fourth book in the "Robert Langdon" series. Who is this "Robert Langdon"? Robert Langdon is the protagonist of the book who has appeared in other works by Dan Brown. He is a Professor of religious iconology and symbologyat Harvard, author of six books of iconology/symbology, almost always wears a Harris tweed jacket and loafers, and loves his vintage Mickey Mouse wristwatch.

The best thing about Dan Brown's books is that the readers are greeted with a rich treasure trove of historical information which he covers in incredible detail. He also includes a lot of real-world artistic pieces which he uses in very interesting and imaginative ways. All in all, one can always learn something unique in his books. They are such a fan favorites because his impenetrable plot is rather gripping and makes the readers eagerly turn the pages in order to find out what happens next as they are kept on the edge of their seats.

Now, about *"Inferno"*, this book was inspired by "The Divine Comedy", an epic poem written by Dante Alighieri and considered to be one of the world's greatest works of literature. The plot for "Inferno" is a roller-coaster ride. The story begins with the protagonist, Robert Langdon, waking up in a hospital in Italy with a head wound and retrograde amnesia, leaving him unable to recall when and how he got into his current predicament. When he is attacked by a female assassin Vayentha, Langdon narrowly escapes with the help of Dr. Sienna Brooks, a beautiful woman with a mysterious past. They find a small high-tech projector concealed inside a cylinder with a biohazard sign on it. The projector shows a modified version of Botticelli's "Map of Hell". Langdon finds a clue hidden in the image and so he and Sienna head into the city of Florence.

All the while, they have three different factions chasing after them: the punk assassin Vayentha, a researcher from the World Health Organization (WHO) and a mysterious global organization known only as *The Consortium*,

which specializes in fulfilling odd and eccentric requests of rich and powerful people. Langdon and Sienna have to evade all the three as they try to capture or kill them while trying to figure out the clue hidden in the image.

The story takes us in a whirlwind journey through the cobbled streets and magnificent cathedrals of Florence, and then through the beautiful canals and waterways of Venice. Finally the journey ends in the grand HagiaSopiaand the Yerebatan Cistern, located in the splendorous and majestic Istanbul.
The villain is a somewhat cliché mad-scientist named Bertrand Zobrist, a billionaire geneticist and Dante enthusiast. While considered to be one of the most brilliant minds of the generation, Zobrist advocated the stopping of humanity's growth because of population explosion, and even engineered a way to do so, as he wanted humanity to survive and not destroy itself. "Evil always thinks it's doing right" is a quote that is best applicable here.

Personally, the thing I like the best about "Inferno" is that it deals with a problem which is an actual concern, namely "Population Explosion". The current world population is more than 7 billion as of now, and with the developments in medicine and health care, the average life-span is also increasing. We are not the only sentient life on this planet as we share this world with billions of other species.
In thriller stories, the hero always saves the day. The world returns to its normal status, maybe a little jaded, but still quite functional. But the hero always succeeds in whatever audacious and harebrained plan he has to foil the conspiracy. But not in this one. Langdon attempts to stop the mass dispersal of a dangerous virus that will sterilize 1/3 of the world's population permanently, and he fails spectacularly. And with this, the world has changed, irrevocably and without a shadow of doubt, in ways that we possibly cannot imagine.

If anyone asks me about my favourite chapter of the book, I cannot give an honest answer, because I've never actually paid attention to such things. When I picked up the book, I could not put it down until I finished reading it. And when I was reading it, I cannot for the life of me remember which chapter was the best or the most exciting. Quite honestly, the entire book was compelling!

Moral of the story

Changing everyone's DNA without their permission is treating a symptom without addressing any causes.

Human population is increasing because of the lifestyles of all humans. What would a "morally obligated", "compassionate" human do, when faced with the personal choice to remain childless or have a child?

We are faced, as always, with a world full of survival pressures. As an individual, I believe that at some point in the future, I will be faced with an environment in which a serious pressure to survive is exerted on me – it may be triggered by a single human or a group of humans. Climate change is happening, triggered by humans. My DNA may be modified by a human or a group. What would my response be, if I were able to prevent an event like this?

Likelihoods of events like these will vary, but

they will vary between “already happened” and “highly likely”. Humans will be able to exert these very real and serious pressures on one another. Can we equip humanity with the framework and tools to choose, both individually and as a group? Will all of us be able to respond to these situations so that humanity survives?

-Contributed by: Venkata Anand Parnandi, M. Sc. Microbiology, Sem-III.

***Title of the Book*: The Secret**
Author Name: Rhonda Byrne
Published Year: 2006
Name Of Publication: Atria books, Beyond words publishing

Summary
"The Secret" says it all. The book basically emphasizes on 'The law of attraction'. The author has describe the law of attraction as a natural law which determines the complete order of the Universe and of our personal lives through the process 'like attracts like'.

The pages of the book become interesting as the present traces back to the past!
The mechanism of The Law claims that as we think and feel, a corresponding frequency is sent to the Universe that attracts back to us, events and circumstances on that same frequency.

The author tries and recreates the importance of positivity. The theory the book puts forward is complex yet so simplified. In the book, the 'feelings' of an individual are taken into consideration of the good feelings and the bad feelings.

Proponents of the law that the book claims say that desirable outcomes such as health,wealth and happiness can be attracted by changing one's thoughts and feelings!

Moral of the story
The Secret is an essential need in today's world as most people grieve due to their own creation of circumstances. The book takes a huge step to make the world realize its potential.

The backbone of the book is none other than an individual's thoughts and feelings. This book is one in a million and changes the reader's life with an unimaginable, unexpressed explanation!!!

-Contributed by: Rajvi Gohil, M.Sc. IGBT, Sem-II.

Plant Microbial Fuel Cell: A Renewable Approach

Today world's energy markets are dominated by a large increase in energy demand because of population and strong economic growth. It is well recognized that alternative sources of energy are very urgently required. Wind power, solar power and hydropower are usually called as renewable because they make use of energy sources that are renewed and for that reason they won't be depleted. Importance of electricity in our daily life is undeniable so conservation and proper use is very essential. Hence world's research looking towards bioelectricity production which can be produced using different biological fuel cells[1]. Fuel cell is another renewable approach to the world in which different types of fuel cell work in diverse manner as an example; Plant microbial fuel cell makes use of solar power and balances the energy level of the world. For minimizing the negative environmental impact, harvesting the energy from renewable and sustainable resources is the most critical challenges for society. Microbial fuel cells applications have been recently developed in industrial field which is also called as biological fuel cell also which are used for bioelectricity production from different plants, biomass and waste water using mediator free microbial fuel cell or mediator microbial fuel cell.

Significance of microbial fuel cell

Microbial fuel cell (MFC) is a bio electrochemical system that gives energy using microorganism and by their catalytic reaction they convert chemical energy into electrical energy. The electrical activity of microorganisms is an interesting and informative area of science. Type of MFC is mainly depending on and is a clean and reproducible way for energy production. MFC are being constructed using variety of materials and its system operated under a range of different environment like differences in temperature, pH, electron acceptor, electrode surface areas, and reactor size and operation time. Using microbial fuel cell technology variety of waste water can be oxidized like starch, glucose, acetate, pyridine, cellulose, other complex substrates, all industrial waste water, petroleum contaminants etc. According to new research conducted, new microbial fuel cell designs are more capable than hydrogen production technologies. Power productions from plants are a new concept and have very bright future in upcoming years. Today numbers of companies have emerged to commercialize microbial fuel cells and give many modified technologies to the world. Companies generate many aspects of MFC technology. MFC technology is promising technology as a renewable source of energy and highly efficient at commercial scale and meet the need for alternate energy source. MFC works in a very renewable, sustainable and efficient manner so its importance increases in the world to fulfill the needs of people.

Types of fuel cell

Fuel cell's applications categorize into three different broad areas: Portable power generation, Transportation services and Stationary power generation. All fuel cell works in different conditions, on different principle and all have different characteristics. There are main three types of fuel cell. In chemical fuel cell different combinations of chemicals are used to catalyze the reactions which include Phosphoric acid fuel cell (PAFC), Hydrogen fuel cell (HFC) and Solid oxide fuel cell (SOFC). Where as in *Biochemical fuel cell* it is electrochemical power generator in which fuel source is or-

ganic matter, air is oxidant at cathode and at anode oxidation of bioorganic matter by microorganism which include enzymatic bio fuel cell, Hydrogen fuel cell. In Biological fuel cell plants and microorganisms are used to produce electricity in which Microbial fuel cell (MFC), Photosynthetic algal microbial fuel cell (PAMFC), Plant microbial fuel cell (PMFC), Mediator less plant microbial fuel cell (ML-PMFC), Mediator plant microbial fuel cell (MP-MFC) are included. Microbial fuel cells are of two types: 1) Single chamber MFC (It has an anode and cathode compartment in that cathode expose directly to the air and external circuit is connecting the anode and the cathode) 2) Dual chamber MFC (It is prepared up of two separate compartments and they are connected by proton exchange membrane). Every fuel cell has two electrodes, one positive and one negative called, anode and cathode respectively. It has an electrolyte, which carries electrically charged particles from one electrode to other and there is also catalyst which speeds the reaction at electrodes[2].

Plant microbial fuel cell

Plant microbial fuel cell (PMFC) uses living plants and bacteria to generate bio-electricity in very clean, renewable, sustainable and efficient in bio-electricity production. PMFC constructed in single and dual chamber type of fuel cell. PMFC uses organic waste matter as fuels and readily available microbes as catalysts and does not require highly regulated distribution system. It has high conversion efficiency as compared to enzymatic fuel cell therefore it gives stable electricity production and microbial system increases electrical potential. This fuel cell system operates with no pollution and has remarkable long term stability. The P-MFC can probably be combined with other applications of biomass on the similar surface area, so no competition with food or feed production needs to occur. This adds to the social acceptance of the P-MFC. The economic possibility of the P-MFC will be determined both by the power output and the costs of the materials used in the system. PMFC technology is multi-disciplinary that provides scope for strengthening research across disciplines therefore with further improvements in design, cost and performance efficiency its scale up its areas in many applications. PMFC harvest solar energy as electricity by combination of electricity generation by bacteria through oxidation of compounds.

Classification of plant microbial fuel cell

Plant Microbial fuel cells are classified into basically two types.

A) Mediator Microbial fuel cell: Sometimes electrons are unable to transfer to the anode and cathode due to electrochemically inactive nature of the fuel cell so mediators assist the reactions. The electrons transfer from microbial cells to the electrode is done by mediator such as methyl blue, humic acid, methyl viologen, Neutral red, thionine, ferricyanide. Ferricyanide is also commonly used as mediator means act as an electron acceptor in the cathode chamber increases the power density. Mediators are expensive and toxic phenolic compounds so use of mediators are generally avoided. B) Mediator free microbial fuel cell: Plant microbial fuel cells which consists of active indigenous microorganisms which make cell electrochemically active so it has capacity to pass electrons to the electrode without any mediator. Some microorganisms have pili or some have enzymes that facilitate the transfer electrons to the electrode. This is a new area of research for bioelectricity production. Plant microbial fuel cell is commonly called mediator free microbial fuel cell which

gives stable electricity production[3] (Kaku, *et al.,* 2008).

Construction of plant microbial fuel cell

Plant microbial fuel cell is designed for power production with relatively low investment costs. For the construction, pre cultivation of plants was done using seeds of selected plants. Seeds were sterilized by immersion in 10% H_2O_2 for 15 minute followed by germination in humid perlite. Seeds were planted in soil that was taken in pots. Plants were cultivated at ambient temperature without adding any additional nutrients (fertilizers). If cultivation of plants take long time then additional nutrients are required like manure, urea etc. It has been observed that even without treatment of H_2O_2 seeds were growing at faster rate. Pots, glass chamber, plastic or wooden box can be used as an apparatus but it should be durable.

Electrodes were placed to the fuel cell at different positions. Stainless steel, copper plates, copper wire, lead oxide, carbon cloth can be used as an electrodes in the construction. In the PMFC, at the bottom an anode was placed which was overlaid with a layer of soil and plants were cultivated in that region. Cathode was submerged half in soil and half in water layer. At the both end of apparatus cathode was placed. As anode and cathode different material verity of metals can be used with different plants to check better power output. For electricity measurement closed circuit is required, which was created by joining anode, cathode and specific resistance with copper wire. Positions of plants should be in such a way that it has direct contact with anode or should be in the nearby area of anode so its exudates could directly react with anode. Root exudates release proton and electrons that react with oxygen that was placed at cathode.

Mechanics of plant microbial fuel cell

During growing season living plants transport organic matter to the soil in which organic carbon enters the soil as rhizodeposits. Several groups of organics such as water soluble, low molecular weight, high molecular weight, gases, mucilage covering roots set as rhizodeposits. Root exudates which also called as or-

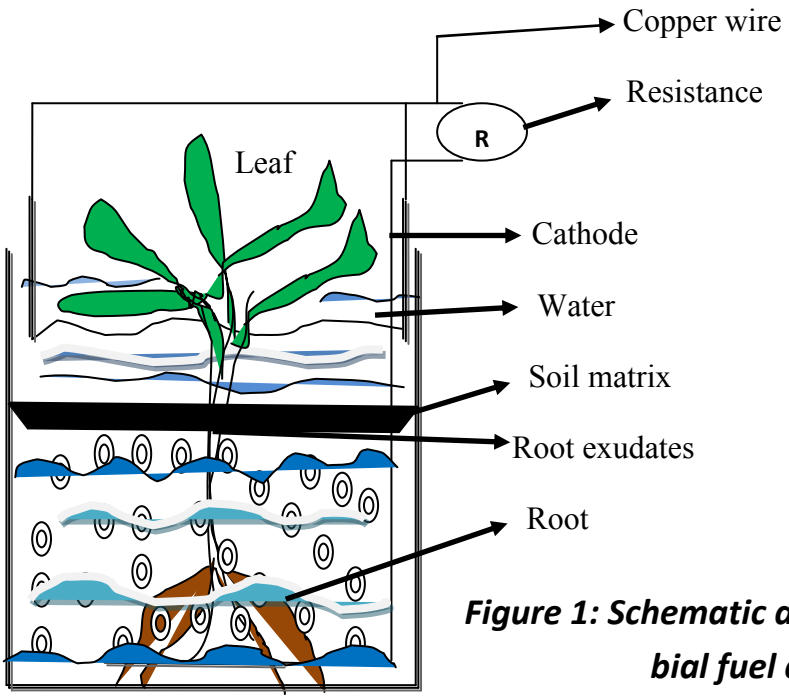


Figure 1: Schematic diagram of plant microbial fuel cell (PMFC)

ganic substances are released by roots into environment i.e., carbohydrates, vitamins, amides, amino acid, aliphatic acids, sterols, enzymes which have important role in nutrient accumulation[4]. Plant microbial fuel cell makes use of naturally occurring processes around the roots of plants that directly generate electricity in which plants that can grow in water logged are used to avoid in coming oxygen from the air to the cathode. At anode microbial catalyzed oxidation of reduced compounds are responsible for release of electrons that pass through an electrical circuit. The electrons arrived at cathode electrode and react with oxygen which present on anodic electrode and water is generated therefore bio-electricity produced through flow of electrons in the circuit. Plant microbial fuel cell with combination of algae also generates good electrical potential through electrochemically active indigenous bacteria and cost effective electrodes[5].

Application of plant microbial fuel cell

PMFC include renewability of materials, energy balancing, social acceptability, environmental performance, economic feasibility. Green electricity roof is typically suitable for developed countries and urbanized areas. Decentralized electricity production in developing countries is interesting because of low voltage application that could be powered with a PMFC. Applying PMFC in developing areas offers the opportunity of economic growth in the poorest areas of the world therefore PMFC technology is nearing application in society and it is time to put expectations of the environmental performance of the system to the test. Production of bio-electricity with a product, which is familiar with, plants, will offer them an opportunity to increase profit. Moreover, applying the PMFC as a decentralized system for electricity production will offer 1.2 billion people around the world that don't have access to electricity to develop economically and socially. PMFC is a technology that has high novelty and could develop in novel markets, technology and markets will co-evolve.

Plant microbial fuel cell gives a new idea to green biotechnology and fulfils the scarcity of electricity at global level and it can be consider as renewable, sustainable and efficient bio-electricity producing technology. Plant has different capacity and characteristics like root length, root thickness, organic matter concentration, water holding capacity, life span value, tolerance capacity, resistance capacity towards environmental factors etc affect the bio-electricity production therefore surface area of electrodes, selection of place of electrodes and concentration of root exudates play a vital role in bio-electricity production.

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-Contributed By: Swati Narolkar and Dhwani Gandhi

Effect of Solar Radiation on Bacteria

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Abstract: Microorganisms often regulate their gene expression at the level of transcription and/or translation in response to solar radiation. In this review, it is cited that how bacteria regulate their gene expression at both transcription and translation levels to enable biomarkers identification and comparison of gene regulation from one bacterial species to another. Also the use of solar radiation to disinfect the bacteria has been explained showing its benefits in water treatment.

Introduction

A wide diversity of tolerances to damaging radiation is exerted by Bacteria and they are simplest model organisms for examining their response and strategies of defense in terms of gene regulation. Sunlight is a source of all the radiation ranging from UV radiation, visible light and infrared, we experience different kinds of radiation on Earth, including the ionizing radiations such as gamma rays. Exposure of microorganisms to solar radiation leads to direct and indirect damage to the cell. Nucleic acid is likely to damage at exposure of cell in radiation. Pyrimidine bases is dimerized by the exposure of UVB and causing the formation of two major photoproducts, cyclobutane pyrimidine dimers (CPDs) and pyrimidine (6-4) pyrimidone photoproducts (6-4 PPs) [1]. Ionizing radiation leads to severe DNA/RNA damage such as double/ single strand breaks, base modifications[2.] quences of DNA lesions are either an inhibition of the progression of the polymerase during DNA replication and transcription or a lesion bypass with disincorporation that could eventually lead to mutations[1,2]. Similarly, strand breaks or oxidative damage to protein-coding RNAs or non-coding RNAs might cause errors in protein synthesis or deregulations of gene expression. The net biological effect of damaging radiation depends upon the balance between the rate of radiation-induced damage and both the efficiency of how the cell protects itself against damage accumulation as well as the rate at which that damage is repaired. However some damage are not repaired in the cell such as oxidative proteins and lipids damage and the level of accumulation of protein damage in the cell plays a pivotal role in bacterial radioresistance[5] and until now, have been much less studied than DNA damage.

Photoreactivation can be adversely affected by the UV-A and visible light as reactive oxygen species (ROS) generated due to harmful oxidative stress [1,2]. The resulting DNA lesions generated by oxidative stress include base and sugar lesions, strand breaks, DNA-protein cross-links and base-free sites[3]. The conse-

A wide range of methods exists to characterize the changes in the transcriptome and proteome of bacteria. Among the several transcriptome profiling methods, microarray and RNA-seq are the two most popular methods employed for studying gene expression and regulation of mRNA synthesis under damaging radiation and/or repair conditions. While

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microarray has been extensively used as transcriptomic approach, today the RNA-seq method is becoming a preferred method[6-8], mainly because it does not depend on the previous characterization of the reference transcriptome. A stable isotope labeling is the most comprehensive gel-free approach for measuring overall protein abundance and can be performed by in vitro (ICAT, iTRAQ, ICPL post-digest) and in vivo (e.g., metabolic labeling) approaches[9-12] .

Bacterial Disinfection for Water Treatment: Exposure to solar radiation is a common method used for bacterial disinfection for water treatment. The growth of E. coli is inhibited by continuous UVA radiation with a subsequent adaptation to stress[13]. Transcriptomic approach was used to assess short-time stress and UVA light adapted growth. More genes (i.e., 312) were expressed in the cells irradiated for a short time (1 h) than in UVA-adapted cells (50 h). The involvement of oxidative stress was confirmed with the induction of alkylhydroperoxidase reductase, the enzyme that converts lipid hydroperoxides to their corresponding alcohols. The decontamination with UV can also be used to remove terrestrial bacteria associated with spacecraft to avoid taking a risk for further bacterial contamination.

Marine Bacteria to Solar Radiation

The oceans are estimated to contain more than 1029 bacteria[14], where those microorganisms are fundamental components of the aquatic biogeochemical cycles. Solar ultraviolet radiation (UVR, 280–400 nm) has been shown to reach significant depths in many marine ecosystems, influencing a large part of the surface of the water column, where phytoplankton productivity takes place[15]. Marine bacteria present at the surface of oceans are exposed to the full spectrum of solar radiation. Both UVB and UVA can have important detrimental effects on bacterial activity, phytoplankton photosynthesis and photochemical transformation of dissolved organic matter. Lately, environmental changes related to the depletion of the stratospheric ozone layer[16] raise concerns about the response of aquatic microorganisms that could be significantly altered by the increasing level of damaging UVR.

A transcript profiling methodology was used to elucidate the expression patterns of the cyanobacterium Synechocystis sp. strain PCC 6803, in order to investigate changes in gene expression induced by irradiation with UVB and high-intensity white light. Several families of transcripts were found to be altered by both high intensity white light and UVB, with a subsequent down-regulation of the genes involved in the light-harvesting system, photosynthesis, photoprotection, and the heat shock response[17].These two profiles comparisons also corroborated the regulation of many pathways, including the synchronized induction of D1 protein recycling and a coupling between decreased phycobilisome biosynthesis and increased phycobilisome degradation. However, the gene expression profiles produced by high-intensity white light and UVB differed mostly in the regulation of several transcriptional processes, and in the regulation of the ribosomal protein transcripts, which are only repressed by UVB radiation[18].

Even though we would have expected to observe a greater level of resistance in oligotrophic bacteria, which are hypothetically better adapted to cope with UVB than copiotrophic prokaryotes, we observed a response that was

more subtle.

UV-Induced Protein Damage

It is noteworthy that the abundance of proteins can change not only as a result of gene expression, but also by increasing/decreasing protein stability and turnover, that can be in turn modulated by the level of proteins lesions. Proteins are important targets of damaging radiation and it seems that the ability to protect proteins against oxidation distinguishes radiation resistant bacterial species from radiation sensitive ones. Solar radiation can generate a wide range of protein damage due to oxidative stress, such as amino acid modifications, carbonyl group formation, fragmentation, formation of protein-protein cross -links, and formation of S–S bridges. A recent review presented modifications induced by radiation regarding sulfur containing amino acids[19]. Carbonylation is one of the radiation-induced damage and is an irreversible oxidative process unlike methionine sulfoxide and cysteine disulfide bond formation[20].This carbonylation is closely associated with the production of aberrant protein isoforms[21].The rapid carbonylation of mistranslated or otherwise aberrant proteins points to an important physiological role of carbonylation in protein quality control. Since carbonylated proteins are more susceptible to proteolytic degradation than their non-oxidized counterparts[22], the rapid carbonylation of an erroneous protein may ensure that it is directed to the proteolyse process. Biochemical analysis revealed that carbonyl groups in the active center of a protein trigger its degradation. Thus, carbonylation may act as a signal ensuring that damaged proteins enter the degradation pathway rather than the chaperone/repair pathway since carbonylation is an irreversible/unrepairable modification. However, highly carbonylated proteins can sometimes form high-molecular-weight aggregates that are proteolysis-resistant.

Carbonyl derivatives are mainly formed on the amino-acid side chains of proline, arginine, lysine, and threonine can also be formed by secondary reactions with reactive carbonyl compounds on carbohydrates (glycoxidation products), lipids, and advanced glycation/lipoxidation end products. In bacteria, it was recently demonstrated that oxidative damage is the cause, rather than a consequence of radiation-induced cell death. This was demonstrated for both Escherichia coli and the radiation resistant bacterium, Deinococcus radiodurans, where ionizing radiation resistance was dependent on the level of protection against protein carbonylation. In this way, sensitive bacteria would sustain lethal level of protein damage at radiation doses that elicit relatively little DNA damage, and that extreme resistance in bacteria would be dependent on protein protection. It was reported in E. coli that the cells with low concentrations of carbonyl products remain reproductively competent, whereas cells with a high carbonyl load become genetically dead (unculturable). Diverse vital cellular functions like transcription, translation apparatus, transport systems, amino acids synthesis and degradation, transport systems, TCA cycle, glycolysis, chaperone functions and catalase were found to be targeted by UVA radiation in E. coli. Proteins involved in metabolism, transcription, transport/folding and protein synthesis may therefore be the cellular functions that are most often affected by stress induced carbonylation, at least in certain bacteria. However, at present it is not clear what fraction of the vul-

nerable enzymes becomes modified during oxidative stress in different bacterial species, and whether such modifications typically interfere with protein function. This information is necessary in order to estimate the impact that this damage might have on cell viability.

Conclusion

As Climate change with increasing levels of UVB radiation reaching the Earth's surface, the knowledge of the impact of UV radiation on marine bacterial distribution, activity and gene regulation, is essential for understanding/predicting the possible alteration of biogeochemical cycling of elements in marine surface layers.

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Hairy Root Culture: A natural bio-factory to produce secondary metabolites production

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Abstract: The hairy root culture system is potential approach for the production of secondary metabolites, because it has genetic and biosynthetic stability and their fast growth offers easy culture and genetic manipulation, and, most importantly, an increase ability to synthesize useful metabolites. Hairy roots can be produced by transformation with the soil bacterium *Agrobacterium rhizogenes*, resulting in the so-called hairy roots disease. Hairy roots are induced when a plant is infected by an *A. rhizogenes*, by a part of a root inducing (Ri) plasmid in bacteria, called transfer DNA (T-DNA), which is transferred into the plant cell and expressed therein. The interest in hairy roots is mainly due to their ability to grow fast without needing an external supply of auxins. The focus of the present review is the hairy root culture technology used for natural bioactive compound for secondary metabolites production from medicinal plants.

Introduction

In the recent years, the interest in medicinal plants has increased in a great deal. Medicinal plants are the most important source of life saving drugs for the majority of the world's population. Medicinal plants potential source of secondary metabolites production and fine chemicals. Medicinal plants have been recognized as valuable source of therapeutic components for centuries, and about 60% of world's population are known to use traditional medicines derived from medicinal plants[1]. In recent years market of plant products expand rapidly and this trend will continue in the 21st century because more and more people prefer natural products.

Many of these products are difficult to synthesize chemically or difficult to produce in large amounts. In this perspective plant tissue culture technology holds promise specially plant cell cultures has been looked at as a potential alternative for efficient production of natural bioactive compounds. Manipulation of the plant genome by introducing foreign genes has become a core tool in plant biology. Targets include enhancement in productivity by increasing resistance to abiotic and biotic stresses as well as fundamental studies such as identification and characterisation of key regulatory genes. Plant transformation methods in use employ *Agrobacterium*, microprojectile bombardment, microinjection and electroporation of protoplasts[2]. Among these, *Agrobacterium*-mediated plant transformation is the most extensively used method for enhance the secondary metabolites production from medicinal plants.

Hairy root is a plant disease caused by *Agrobacterium rhizogenes* Conn., a Gram-negative soil bacterium. When the bacterium infects the plant, the T-DNA between the TR and TL regions of the Ri-plasmid in the bacterium is transferred and integrated into the nuclear genome of the host plant. The transformation process produces a valuable by-product, hairy root, which will form at or near the site of infection[3]. In addition, opines are produced and

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serve as specific food for the bacteria. Hairy roots grow rapidly, show plagiotropic growth, and are highly branched on phytohormone-free medium. The transformed root is highly differentiated and can cause stable and extensive production of secondary metabolites, whereas other plant cell cultures have a strong tendency to be genetically and biochemically unstable and often synthesize very low levels of useful secondary metabolites[4]. The hairy root system is stable and highly productive under hormone-free culture conditions. The fast growth, low doubling time, ease of maintenance, and ability to synthesize a range of chemical compounds of hairy root cultures offer additional advantages as continuous sources for the production of valuable secondary metabolites[5]. Hairy roots are also a valuable source of phytochemicals that are useful as pharmaceuticals, cosmetics, and food additives. These roots can also synthesize more than a single metabolite and, therefore, prove economical for commercial production purposes[6].

Plant Hairy root culture – A natural source of Secondary metabolites

Plant hairy root cultures are natural source of secondary metabolites. It is characterized by a high yield production compare to normal plant root production of secondary metabolites production. Normally, root cultures need an exogenous phytohormone supply and grow very slowly, resulting in poor or negligible secondary metabolite synthesis. In plant tissue culture, hairy root culture has revolutionized the role of secondary metabolites synthesis. This hairy root cultures ability to synthesize a range of chemical compounds offers an additional advantage as a continuous source for the production of valuable secondary metabolites. These culture very fast growth and also ease to maintenance culture in *in vtro* condition[7]. Hairy roots also offer a valuable source of root derived phytochemicals that are useful source of medicinal properties, pharmaceuticals and cosmetics[8]. Hairy root culture techniques can also synthesize one single metabolite and therefore prove economical for commercial production purposes (Table 1). Hairy root culture of many plant species have been widely studied for the *in vitro* production of secondary metabolites. Uma Maheswari *et al.* (2011) reported *the A*. *rhizogene* ATCC15834 transformed hairy root culture in Coleus forskohlii biomass of hairy roots increases quickly in culture[9]. Jinefer et al. (2012) study the adventitious root cultures and hairy root cultures are highly effective in producing highly valuable roots in *Boerhaavia diffusa*[10]. Satdive et al. 2007 got the increase production of azadirachtin by hairy root culture of *Azadirachta indica*[11]. Weathers et al., (1994) reported in the hairy roots in *Artemisia annua* using *Agrobacterium rhizogenes* reports indicate that content of artemisinin in hairy roots can be upto 0.4 % g/g DW[12]. Chunxian Yang et al., (2011) study the improvement of tropane alkaloids production in hairy root cultures of *Atropa belladonna*[13]. He is study suggested that produced tropane alkaloids at higher levels in the best line T3 produced 2.2 mg/g dry weight (DW) hyoscyamine, which was about 11 times more than that in non-transgenic hairy root cultures and 24 times more than that in the wild type. Xiaozhong Lan and Hong Quan (2013) reported the hairy root culture of *Przewalskia tangutica* for enhanced the production of pharmaceutical tropane alkaloids[14].

Establishment of hairy root cultures

For the production of hairy root cultures, the

Table: 1– Commercial production of secondary metabolites production from hairy root

Plant	Secondary metabolites
Amsonia elliptica	Indole alkaloids [15]
Cassia obtusifolia	Anthraquinone [16]
Catharanthus tricophyllus	Indole alkaloids [17]
Datura candida	Scopolamine, Hyoscyamine [18]
Rauvolfia micrantha	Ajmalicine, ajmaline [19]
Panax ginseng	Ginsenoside [20]
Papaver somniferum	Morphine,sanguinarine [21]

explants material is inoculated with a suspension of *A. rhizogenes*. The bacterial suspension is generated by growing bacteria in Yeast Mannitol Broth (YMB) medium for 2 days at 25°C under shaking conditions. Thereafter, pelleting by centrifugation (5 x 10 rpm; 20 min) and resuspending the bacteria in YMB medium to form a thick suspension (approx. 10^{10} viable bacteria/ml). Transformation may be induced in aseptic seedlings or surface sterilized detached leaves, leaf-discs, petioles, stem segments, from greenhouse grown plants by scratching the leaf midrib or the stem of a plantlet with the needle of a hypodermic syringe containing a small (about 5-10 ul) droplet of thick bacterial suspension of *rhizogenes.* Wounded plant cell releases phenolic substances and sugar (1); which are sensed by *Vir* A, *Vir* A activates *Vir* G, *Vir* G induced for expression of *Vir* gene of Ri-plasmid (2); *Vir* gene produces all the *Vir* -protein (3); *Vir* D_1 and *Vir* D_2 are involved in ssT-DNA production from Ri-plasmid and its export (4) and (5); the ssT-DNA (associated with *Vir* D_1 and *Vir* D_2) with *Vir* E_2 are exported through transfer apparatus *Vir* B (6); in plant cell, T-DNA coated with *Vir* E_2 (7); various plant proteins influence the transfer of T-DNA + *Vir* D_1 + *Vir* D_2 + *Vir* E_2 complex and integration of T-DNA to plant nuclear DNA(8). (LB= left border; RB= Right border; pRi = Ri plasmid, NPC = nuclear pore complex)

Genes responsible for hairy root formation

The agropine-type Ri-plasmid consists of two separate T-DNA regions known as the TL-DNA and TR-DNA. Each of the T-DNA fragments is separated from each other by at least 15 kb of non-integrated plasmid DNA. These two fragments can be transferred separately during the infection procedure. The TR-DNA of the agropine type Ri-plasmid carries genes encoding auxin synthesis (tms 1 and *tms* 2) and agropine synthesis (*ags*). The mannopine type Ri -plasmids contains only one T-DNA. TL-DNA region consists of four root locus (*rol*) genetic loci, *rol* A, *rol* B, *rol* C, and *rol* D, which affect hairy root induction. In particular, *rol* B seems to be the most important in the differentiation process of transformed cells and also function as induction of hairy roots by hydrolyzing bound auxins leading to an increase in

the intracellular levels of indole-3-acetic acid. Gene *rol* A involved in development of hairy root morphology, *rol* B is responsible for protruding stigmas and reduced length of stamens; *rol* C cause's inter-node shortening and reduced apical dominance.

Genetic manipulation of secondary metabolites

The stable introduction of foreign genetic information into the plants represents one of the significant developments in recent advances of plant biotechnology including high volume production of several biologically active natural compounds. Genetically manipulation of plant secondary metabolites used the *Agrobacterium rhizogenes.* It is causative agent of hairy root disease in several plants, has emerged as an important alternative to intact plants as well as cell cultures for the production of secondary metabolites. Hairy roots have been reported to yield higher amounts of secondary metabolites than cell suspension cultures and in some cases, intact plant roots. *Agrobacterium rhizogenes*-mediated hairy root cultures is the other feasible method, as these grow fast, are genetically stable and capable of synthesizing much more secondary metabolites than normal roots and other organs. Secondary metabolite productions in Ri-transformed plants are at levels, comparable to or even greater than that in non transformed plant in many cases, whereas in some plants reduction of specific secondary metabolite is also reported. Hairy root cultures produce secondary metabolites over successive generations without losing genetic or biosynthetic stability. Tylophora india shoot, tylophorine content was 20–60 % higher than that in the control using *Agrobacterium rhizogenes*. Similarly, in A4 transformed plants of *B. monnieri*, the content of four bacopa saponins (bacopasaponin D, bacopasaponin F, bacopaside II, and bacopaside V) were up to five times higher than non-transformed plants of same age. Ri-transformed plants of *Plumbego indica* are also reported to have an increased plumbagin content compared to non-transformed plants. Sevon et al. (1997) Ri -transformed plants of *Hyoscyamus muticus* showed reduced alkaloid production and same was in case of transgenic plants of *D. myoporoides, D. leichhardtii* for scopolamine and hyoscyamine .

***Agrobacterium rhizogenes* strain for secondary metabolites production**

A. rhizogenes wild strains are characterized by

Table 2: Different strains of Agrobacterium rhizogenes use for hairy root culture bellow.

Plant Name	Secondary metabolites	Strain
Salvia sclarea	**Ortonaphtoquinone diterpens**	**ATCC 15834** [22]
Coleus forskohlii	**Forskolin**	**ATCC18534 ,** [23] **MTCC533**
Datura innoxia	**Scopolamine**	**AR-1855** [24]
Artemisia **annua**	**Artemisin**	**A4,LBA 9402, K_{599},** [25]

the following four steps. 1. Chimiotactism induced movement of agrobacteria towards the plant cells. 2. Binding of the bacteria to the surface components of the cell wall.3.Activation of the virulence (*vir*) genes. 4. Transfer and integration of the transfer-DNA (T-DNA) into the plant genome. Different strain of *A. rhizogenes* used for hairy root culture for secondary metabolites production shown in the Table 2.

Application of hairy root culture

Hairy root cultivation has been reported to be the ideal production technologies and is being researched extensively. Hairy roots once established can be grown in a medium with low inoculums with a high growth rate. Biomass of hairy roots increases quickly in culture so it would be reasonable to expect hairy root culture of secondary metabolites for large scale production. Their fast growth and genetic and biosynthetic stability offer an additional advantage for their use as an alternative to plant cell suspension cultures, for production of secondary metabolites of interest. Hairy roots in particular are more stable artificial roots and are obtained by genetic transformation of different plant parts of medicinal plants using *Agrobacterium rhizogenes.* Hairy root culture if properly maintained and subculture at regular intervals will remain stable with regard to the secondary metabolites. Hairy root cultures (HRC) have allowed a deep study of plant metabolic pathways and the production of valuable secondary metabolites and enzymes, with therapeutic or industrial application. Furthermore, the potential of HR cultures is increasing continuously since different biotechnological strategies such as genetic engineering, elicitation and metabolic traps are currently being explored for discovery of new metabolites and pathways, as well as for increasing metabolites biosynthesis. Hairy roots have been applied in a wide range of fundamental studies of plant biochemistry, molecular biology, and physiology, as well as for agricultural, horticultural, and large-scale tissue culture purposes. In addition, hairy roots offer promise for phytoremediation because of their abundant neoplastic root proliferation. Recent progress in the scaling-up of hairy root cultures is making this system an attractive tool for industrial processes.

Prospects and limitation

The major advantages of hairy root cultures includes (i) synthesis of bioactive secondary metabolites independently from climatic and soil conditions; (ii) negative biological influences that affect secondary metabolites production in the nature are eliminated (microorganisms and insects) (iii) to select cultivars with higher production of secondary metabolites; (iv) with automatization of cell growth control and metabolic processes regulation, cost price can decrease and production increase Hairy roots are complicated biocatalysts when it comes to scaling up and pose unique challenges.

Conclusion

Medicinal plants have a very rich source of different phytochemical constitution, , so that consumption of herbal medicines and medicinal plants is widespread and increasing. The main source of medicinal plants raw material is represented by natural and wild fields. The use of controlled environments overcomes cultivation difficulties and thus facilitating the manipulation of plants to produce bioactive compounds. Hairy root culture is being considered as an alternative to agricultural processes for producing valuable phytochemicals.

Many secondary metabolites are obtained by direct extraction from the plant grown natural habitat, several many factor responsible for alter their yield. The use of hairy root culture has overcome several inconveniences for the production of these secondary metabolites. Organized cultures, and especially hairy root cultures, can make a significant contribution in the production of secondary metabolites production. Hairy roots are unique in their genetic and biosynthetic stability and their fast growth offers an additional advantage to use as a continuous source for the production of valuable secondary metabolites.

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