Carbon – Science and Technology

ISSN 0974 – 0546

http://www.applied-science-innovations.com

RESEARCH ARTICLE Received:10/03/2016, Accepted:15/04/2016

Exploitation of endophytic fungus as a potential source of biofuel

Nawed Anjum, Vijay Kant Pandey and Ramesh Chandra

Department of Bioengineering, Birla Institute of Technology, Mesra, Ranchi, India.

Abstract: Biofuel demand is unquestionable in order to reduce greenhouse gaseous emission which can lead to climatic changes and global warming effect. Finding sufficient supply of clean energy for the upcoming is one of the society's most daunting challenges and is directly linked with global stability, economic prosperity and quality of life. Endophytic microbes reside in the healthy part of the plant without causing any symptoms of disease. It is well known that the endophytic microbes produces wide variety of bioactive compound having, antibacterial, antifungal, antiviral, antitumor, antioxidant, anti-inflammatory, immunosuppressive drugs, and volatile organic compounds having similarity with conventional diesel fuel. Now the endophytic fungi, have also been known to possess a suitable lipid matrix at high concentrations and volatile organic compounds having similarity with conventional diesel fuel that make them promising sources for next generation biofuels. This would be more efficient and having lesser number of biosynthetic steps in production, can be brought to immediate use in the existing internal combustion engines without taking about any major modification in automobile design. The present article therefore aims to review the current status of research in the field of alternative source of energy emphasizing endophytic fungi as a source of biofuel precursor, in order to encourage and generate interest among research groups across India and the world for initiating and undertaking more enthusiastic and intensive research activity on endophytic fungi from the Indian subcontinent having the potential to make fuel-related hydrocarbons.

Keywords: Endophytic, Bioactive, Volatile, Biofuels, Hydrocarbons.

---

1 Introduction: Energy is a critical prerequisite for socio-economic development. Conventional fuels will continue to play a dominant role in the energy setting in the country in the following few decades. However, conventional fuel resources are limited, nonrenewable, polluting and, therefore, need to be used cautiously **[1]**. Fossil fuels like coal and oil have played a chief role in humanity's recent history that provided an immense energy source which has sustained much of society's development and industrialization. These fuels are still serving as the major source of energy for the world, and yet it is agreed that these traditional sources of energy cannot continue to power human growth into the future. The demand for oil production will endlessly increase and only rise as developing nations continue to grow. Furthermore, many experts predicted that the rate of world oil production has already peaked and that it will only reduce from now onwards as fewer and fewer oil reserves are discovered. This declining supply and mounting demand will raise the price of oil and other fossil fuels, and will ultimately make them economically unsustainable **[2]**. The use of fossil fuels poses additional problems most remarkably burning fossil fuels produces massive extents of the greenhouse gas carbon dioxide, which has a negative impact on the Earth's environment by contributing to global warming. Because of such concern, there is an urgent need to pursue the development of renewable energy sources, particularly microbial biofuels. Photosynthetic microorganisms, such as micro-algae and cyanobacteria can harness solar energy and store it as chemical energy in the biomass [3]. This energy can then be released through biochemical conversion. The structural and storage carbohydrates in biomass have low energy content and it is essential to concentrate the energy content further for fuel application. Anaerobic microbial fermentation is an effective and commonly used method for such biochemical conversion process. Useful, renewable fuels produced by microorganisms comprise hydrocarbon, ethanol, hydrogen and methane. Fuel produced by microbes has the potential for assisting to meet world increasing energy demands. Living organisms assimilate and concentrate energy in their biomass as well as products. Hence, biomass in its various forms is an attractive alternative source of energy [4].

The composition of structural and storage carbohydrates in biomass have low energy content, cannot be used as fuel directly. It is required to focus in the total energy content of the biomass for further fuel applications. Fuel produce commercially valuable originated from microorganism primarily depends on getting the microbes which can produce the desired fuel efficiently, additionally the amount of substrate they require for fermentation should be low and economical [5]. As per the current scenario, it is vital that the process for the production of synthetic fuels does not consume natural fuels more than what they produce. Endophytic bacteria living in plant tissues do not substantively harm or gain benefit other than residency [6]. The term endophyte (Gr. *endon*, within; *phyton*, plant) was first coined in 1866 by De Bary [7]. Plant endophytes are very abundant and broadly distributed. Nearly 300,000 plant species that exist on the earth, each individual plant is host to one or more endophytes. Only a few these plants have ever been absolutely investigated relative to their endophytic biology. Consequently, the opportunity to find new and useful endophytic microorganisms among host plants in different settings and ecosystems is great. Presently endophytes are gaining interest as they have been reported for the production compounds having biofuel potential [8, 9] which can serve as clean alternative source of energy.

2. Microbial diversity: The source for novel microorganisms

History of microbial biotechnology discloses that many desired materials like foods, beverages, pesticides and antibiotics were produced using microorganisms. Microorganisms are considered as miniatures of chemical factories. Until Pasteur showed that fermentation is initiated by living organisms and showed that lactic acid fermentation is caused by living organisms that people certainly began to explore microbes as a source for bioactive natural products. The major phase of traditional microbial biotechnology was the development of the acetone-butanol and glycerol fermentations, continued by processes yielding, for example, citric acid, vitamins and pharmaceutical important antibiotics. The birth of recombinant DNA technology glided microbial biotechnology to the modern era and headed to the production of various biopharmaceutical products, such as tissue plasminogen activator, blood clotting factors, erythropoietin, human growth hormone, interleukins and interferons through genetic engineering. In addition to recombinant DNA technology, another discovery which revolutionized the world are high throughput screening techniques for novel compounds, modern microbial biotechnology encompasses fermentation, microbial physiology, strain improvement, downstream processing, cell immobilization (enzyme engineering), metabolic engineering, cell fusion, bioreactor design, in vitro mutagenesis (protein engineering) and directed evolution of enzymes [10]. Microbial biotechnology has made substantial advances in recent years with an immense impact on the humans. The progresses are very fast and new scopes are being added day to day. Despite this escalation of research, it is estimated as only the 10% of microbial diversity have been explored till date, 90% of the microbial diversity is still remain unexplored. It is the new microbial biodiversity that will be the powerful force for new and enhanced biotechnological developments and applications in the sectors of food, chemicals, materials, energy, agriculture, pharmaceuticals, mining, environmental protection, etc.

3. Microorganisms as an alternative energy source:

Amongst the numerous matrices established as an alternative source of energy, approximately 90% are exclusively from plants [11]. Using plants increased the production both in developed and developing countries, leading into serious negative impacts on ecosystems in several cases [12]. One way to positively meet the demands of industry is to look for potential biodiesel precursors in organisms excluding plants. Microorganisms have several advantages over plants and animals. Microalgae, yeasts, fungi, and bacteria can accumulate high levels of lipids, and can be easily cultured and maintain, and do not compete with food production. Furthermore, they can be produced in large-scale fermentation processes, which make them a possibly alternative source of energy. Biohydrogen, Biomethanol and Biodiesel seem to be the most promising of the microbial biofuels moreover needing further active research and development.

3.1 Methane production: Methane (CH_4) is an energy-rich fuel which is utilized for the generation of mechanical, electrical and heat energy. Methane can be manufactured by anaerobic decomposition of waste materials and also it can be efficiently generation by using algal biomass. During the fermentation process, naturally occurring mixed anaerobic bacteria population is exploited for the production of methane, and cells are retained within the digester. A huge amount of organic matter is degraded, with a low yield of microbial cells, while about 90% of the energy available in the substrate is retained and can be effectively purified as gaseous products. The end product is a mixture of methane gas and CO_2.

3.2 Hydrogen production: Methane and its combustion product both are greenhouse gases. This leads to aiming more efforts on the development of clean energy sources. However, microbial production of Hydrogen by acidogenesis process during anaerobic degradation of organics is a promising energy alternative [13, 14]. In the acidogenic phase of anaerobically digesting organic wastes such as sewage sludge is used and hydrogen gas is produced. Anaerobic hydrogen-producing microorganism hydrolyze the degradable primary substrate such as proteins, lipids and polysaccharides and decompose to smaller molecules with the production to acetate and other saturated fatty acids, with the major end products as CO_2 and H_2. The generation of hydrogen using microorganisms, or cell-free systems based on microbial components, is still very much in its early stages. However, there are three possible routes of Hydrogen production are biophotolysis of water, photoreduction and fermentation [15]. Rich microbial species, belonging to the genera *Clostridium*, *Enterobacter* and *Bacillus*, are reported to produce hydrogen [16].

3.3 Ethanol production: The microbial production of ethanol has become an important source of a valuable fuel, particularly in regions of the world that have abundant supplies of plant residues. Fermentation production of fuel alcohol can be through microbial conversion of low cost agricultural substrates, high in starch and sugar content. Agricultural products, starch and sugars from plants are the most common used feedstocks utilized as the carbon source by the microbes [17], which can easily be processed to create the simple sugars needed for fermentation. Numerous microorganisms are capable of producing ethanol, but not all are suitable for industrial processes. The yeasts frequently used in industrial alcohol production comprise *Saccharomyces cerevisiae*, *S. uvarum*, *S. diataticus* etc. Yeast cultures, particularly *saccharomyces*, have been most thoroughly studied because they are very efficient in converting sugars into ethanol, i.e. cost competitive and are not as strongly inhibited by high ethanol concentrations as the other microbes. The ethanol productivity ranges between 1 and 2 g ethanol/h/g cells. Selected bacterial cultures because of their higher temperature tolerance were also studied for use in ethanol production processes. However, their yield of ethanol was not as high as in yeast fermentation [4]. Recently, the bacterium *Zymomonas mobilis* has been selected to achieve a high productivity of 2.5-3.8 g ethanol/h/g cells [18].

A major problem that outbreak ethanol fermentation by *S. cerevisiae* is that the fermentation of sugars by glycolysis also produces generates ATP [19]. Although this problem can be somewhat be overcome by immobilizing the yeast cells to delay cell growth, the consequence of slowing growth is that the intracellular ATP concentration rapidly rises, and the accumulation of this product of glycolysis downregulates the entire glycolytic pathway, thereby decreasing ethanol production [17]. The usages of yeast possess other problem as well, mainly the negative effects of environmental and intracellular stress which accumulate in the fermentation environment. The factor such as high concentrations of ethanol, high sugar concentrations which increase the osmotic pressure in the cells, temperatures as high as 38°C and high sodium ion concentrations will often be a greater source of stress when acting together, making the conditions for *S. cerevisiae* even worse. This stress reduces the yeast's overall viability, thereby hindering its ability to yield ethanol **[19]**.

Table (1): Lipid composition of Microorganism.

	Microorganism	**Lipid content (%)**	**References**
Microalgae	*Nannochoris* sp.	40-70	**[22]**
	Chlorella vulgaris	40-60	**[23]**
	Chlorella emersonii	63	**[23]**
	Chlorella minutissima	57	**[24]**
	Schizochytrium sp.	55-60	**[24]**
Bacteria.	*Gordonia* sp.	93	**[25]**
	Rhodococus opacus	96	**[25]**
Yeast	*Lypomices starkegi*	64	**[24]**
	Rhodotula glutinis	72	**[24]**
Fungi	*Aspergillus oryzae*	57	**[26,24]**
	Cunninghamella echinulate	40-47	**[27,28]**
			[28]
	Mortierella isabellina	68-86	**[29]**
	Mucor circinelloides	20	**[30]**

Some of these major problems could probably be overcome by using other organisms to perform the fermentation reaction. One key candidate is *Zymomonas mobilis*, an unusual anaerobic gram-negative bacterium which break down glucose through the Entner-Duodoroff (ED) pathway **[19]**. Additionally, since it has a usually lower concentration of ATP, which serves as less product-inhibition of the ED pathway and as a result flux through the pathway is overall increases **[17]**. *Z. mobilis* is also attractive because it produces ethanol more efficiently than *S. cerevisiae* and is capable of resisting certain stresses better. This collective difference allow *Z. mobilis* to form ethanol from glucose at as much as 97% of the theoretical yield, and does at a much faster rate than *S. cerevisiae* **[17]**. Inauspiciously, the bacterium can only utilize a few simple sugars as a carbon source to produce ethanol. Additionally, it is sensitive to acetic acid, a commonly found compound in industrial fermentation environments **[20]**. Eukaryotic microorganisms such as yeast, fungi, microalgae, and bacteria have been known to accumulate lipid, mainly as a storage material **[21]**. However, the extent of lipid accumulation varies, usually between species, from as little as 3-5% w/w, dry weight to in excess of 80% w/w, dry weight **[8]**. Santos *et al*., in 2011 listed several microorganisms that exhibit high levels of lipids presented in table 1 which is a good prospective for the production of biodiesel.

3. Endophytic fungi as a source of biofuel precursor:

An endophyte typically defined as a microorganism such as fungi or bacteria that expends either the part or complete of its lifecycle inside the healthy tissues of a living plant, typically causing no symptoms of disease **[31, 32]**. The importance of endophytes had been revealed over a long period as a chief source of bioactive compounds, as many of endophytes were exposed to produce novel bioactive metabolites having pharmaceutical activity. Endophytes are well known for the production of various classes of natural products and have been reported to display a wide array of biological activity and are grouped into various categories, which include terpenoids, alkaloids, phenolic compounds, steroids, lactones, lignans, quinones, etc. Some of these compounds are those produced by their respective host plants as well [33]. During the long co-evolution of endophytes and their host plants, endophytes have adapted themselves to their distinct microenvironments by genetic variation and mainly by uptake of some plant DNA into their own genomes through Horizontal gene transfer [34].

The ideal biofuel for instant use is one that functions within the setting of the existing setup, in particular with existing engines and distribution systems. For this chemical similarity to gasoline is required, which is an array of hydrocarbons with an average chain length of eight [35]. Fungi are a major global carbon recycler and have been recognized as producers of eight carbon (C8) volatiles for nearly 90 years [36]. Conceding endophytic microorganisms, endophytic fungi, consist mostly of ascomycetes [37] and fungi imperfect represent one of the largest and relatively unexplored resources of bioactive natural products [32] and volatile low molecular mass hydrocarbons [21]. Recently, endophytic fungi have been isolated and it has shown to produce bioactive volatile compounds with fuel potential [9]. Three focal chemical properties of biodiesel attributed to the fatty acid profile are iodine value, saponification value and high heating value. Several microbes are known to produce alkanes of chain length extending from C16 to C35. Few microbes, such as photosynthetic bacteria, make hydrocarbons such as pristane and phytane, while cyanobacteria are well known for the synthesis of 7- and 8-methylheptadecanes [38]. Furthermore Fungi, commonly synthesizes long chain hydrocarbons along with a series of low-molecular mass alcohols, terpenoids, esters, ethers, and ketones [39].

Several fungi are well-known to produce octane, 1-octene and lower molecular mass hydrocarbons [40-42]. An endophytic fungus, *Gliocladium roseum* (NRRL 50072), synthesizes a series of volatile hydrocarbons and hydrocarbon derivatives on an oatmeal-based agar under microaerophilic conditions as studied by solid-phase micro-extraction (SPME)-GC/MS. Many of these hydrocarbons produced by endophytic fungi are the major constituents in diesel fuel, including octane; 1-octene; heptane, 2-methyl; hexadecane; undecane, 4-methyl; nonane, 3-methyl; and benzene, 1,3-dimethyl along with the abundance of low molecular-mass esters, alcohols, ethers and fatty acids, so the volatiles of this fungus have been termed as 'myco-diesel' [40-42]. Microbes producing such volatile high-energy substances are of extreme importance given the world's general need for alternative fuel sources. Ranjan Kumar (2015) presented the potential of endophytic fungi as a potential to produce hydrocarbon related to biofuel [43] listed in Table (2).

Table (2): Endophytic fungal having with biofuel producing potential.

Native host plant	Endophytic fungus	Biofuel related hydrocarbons/ precursors produce	Reference
Acalypha indica	*Myrothecium inundatum*	Octane 1, 4-cyclohexadiene 1-methyl- and cyclohexane (1-ethylpropyl)	[44]
Ocimum sanctum and *Brassica junce*	*Colletotrichum* sp. and *Alternaria* sp.	Oleic acid , Linoleic acid, Palmitic acid, Stearic acid Linolenic acid	[45]
Jatropha curcas	*Colletotrichum truncatum* *Colletotrichum truncatum* *Colletotrichum truncatum*	Palmitic acid, Stearic acid Oleic acid, Linoleic acid	[46]

Various techniques such as Gas chromatography with a flame ionization detector (GC-FID), gas chromatography-mass spectrometry (GC-MS) and Nuclear Magnetic Resonance (1H and 13C NMR) are some commonly used examples which can be employed to determine the ester content existing in biodiesels [47, 48]. Through GC-MS analysis, it is possible to identify some of the esters obtained from the endophytic fungi and compare them based on the levels found in each of the microorganisms. Additionally, methyl esters such as palmitic, stearic, oleic, linoleic, and linolenic acids, which are considered the most important methyl esters in biodiesel from plants like soybeans, were identified **[49]**. Santos et. al. in 2011 also lists the characteristics of the methyl esters of the different species of endophytic fungi presented in table 3. Endophytic fungus exhibited comparatively lower levels of methyl ester concentrations. Amongst the endophyte samples, there were some fungi that were identified as promising sources of methyl esters, with similar concentrations of methyl esters seen from soybeans. Santos et. al. (2011) reported about the concentrations of methyl biodiesel **[14]** as presented in table 4.

Table (3): Composition of methyl esters present in different species of endophytic fungi.

Endophytic fungi	**Palmitic acid**	**Stearic acid**	**Oleic acid**	**Linoleic acid**	**Linolenic acid**
Xylaria (NICL3)	21.60%	2.45%	22.40%	43.79%	5.28%
Penicillium (PAOE)	33.25%	5.20%	13.80%	43.51%	2.94 %
Penicillium brasilianum	26.40%	6.04%	13.90%	44.60%	0.94%
Penicillium griseoroseum	29.90%	7.56%	10.10%	29.45%	2.10%
Xylaria (NICL5)	15.50%	8.76% -	26.50%	49.58%	7.78%
Trichoderma (T19)	53.05%		10.66%	1.41%	0.92%
Trichoderma (T25)	31.20%	3.39%	28.55%	23.33%	4.73%
Trichoderma (T27)	30.94%	1.59%	21.74%	13.00%	0.34%
Soy biodiesel	11.29%	3.54%	22.45%	54.62%	8.11%

Table (4): Concentration of biodiesel present in endophytic fungi.

Endophytic fungi	**Concentration of Biodiesel**
Xylaria (NICL3)	66.7 %
Penicillium (PAOE)	83.1%
Penicillium brasilianum	50.8%
Penicillium griseoroseum	40.5%
Xylaria (NICL5)	91.0%
Trichoderma (T19)	67.8%
Trichoderma (T25)	11%
Trichoderma (T27)	40.1%
Trichoderma harvezionum	40.1%
Soy biodiesel	90.7%

3. Conclusion: Microbial lipids can possibly be used as a raw material for biodiesel production using the well-known method for the production of methyl esters by transesterification reaction with methanol in the presence of a basic catalyst or acid catalyst. Microbial metabolism is extremely diverse, and can equally utilize and produce a wide variety of valuable molecules. Many microbial systems are also well characterized and now can be easily manipulate genetically, and scientific advances will drive these systems further and easier to work with in the future. Although there is no biofuel alternative currently existing which can solve all of the economic and environmental issues associated with fossil fuels. The genetically modified species are capable of efficiently make use of otherwise useless materials and byproducts thus makes microbial biofuels an appealing target for research. It is the needed to further investigate endophytic fungi aiming their lipid profiles and capability to produce volatiles having similarity with conventional fuel, using molecular biological tools so that it can serve as one of the alternative platform to compensate the raising future energy crisis. Many of the endophytes showed concentrations of methyl esters at high levels similar to those found in soybean. However till date the quantitative data obtained so far from different endophytic fungi did not exhibit methyl ester concentrations up to the satisfactory levels to be considered as suitable biofuels. Therefore, a search for different endophytic fungi having the potential to produce methyl ester followed by the optimization of the transesterification reaction and culture conditions (pH, temperature, inductive mediums) in order to maximize lipid production, would be very promising avenues for future research to increase biofuel production.

References:

[1] A.S. Malipatil, M. Bandi, IJRAME 2(9) (2014) 23-33.
[2] K. Aleklett, M. Hook, K. Jakobsson, M. Lardelli, S. Snowden, B. Soderbergh, Energy Policy 38(3) (2010) 1398-1414.
[3] D. Prabuddha, J. Banerjee, M.K Maiti, Bioresource Technology 102 (2011) 5815–5823.
[4] Y.K Lee, (L.E. Kun, Ed.) World Scientific Publishing Co. Pte. Ltd (2003) 655-670.
[5] K. Tanaka, N. Kashiwagi, T. Ogawa, *J. Chem. Technol. Biotechnol.* 42 (1988) 235-240.
[6] C.I. Kado, A. Balows, H.G. Truper, M. Dworkin, W. Harder, K.H. Schleifer, The Prokaryotes New York, N.Y: Springer-Verlag., 1 (1992) 659-74.
[7] A.D Bary, Germany: Springer-Verlag (1866).
[8] F. Santos, C. Florisvaldo, P.F. Taicia, N. Joanita, R.M. Marcos, E. Rodrigues-Fo, J. Microbiol. Biotechnol. 21(7) (2011) 728-733.
[9] G. A. Strobel, Biofuels 5 (2014) 447-455.
[10] L. D. Arnold, Tibtech 18 (2000) 26-31.
[11] T. P. Durrett, C Benning, J. Ohlrogge, Plant J. 54 (2008) 593-607.
[12] Q. Li, W. Du, D. Liu, Appl. Microbiol. Biotechnol. 80 (2008) 749-756.
[13] F. Christopher, In: Feldman D.L. (ed) The energy crisis. The Johns Hopkins University Press (1996) 230-242.
[14] R. Sparling, D. Risbey, H.M. Poggi-Varallado, Int. J. Hydrogen Energy 22 (1997) 563-566.
[15] M. J. Waites, N. L. Morgan, J.S. Rockey, G. Higton, Blackwell Science Ltd, (2001) 288.
[16] A. Nandi, M. Sengupta, Crit. Rev. Microbiol. 24 (1998) 61–84.
[17] F. W. Bai, W. A. Anderson, M. Moo-Young, Biotechnology Advances 26 (1) (2008) 89-105.
[18] M. Agrawal, Z. Mao, R.R. Chen, Biotechnology and Bioengineering 108(4) (2011)777-785.
[19] Y. Lin, S. Tanaka, Applied Microbiology and Biotechnology 69(6) (2006) 627-642.
[20] A. Mohagheghi, N. Dowe, D. Schell, Y. Chou, C. Eddy, M. Zhang, Biotechnology Letters 26(4) (2004) 321-235.
[21] S.B. Naik, Y.R. Krishnamurty, Curr. Sci. 98 (2010) 883-884.
[22] E.W. Becker, Cambridge University Press, London (1994).
[23] A.M. Illman, A.H. Scragg, S.W. Shales, Enzyme Microbial Technol. 27 (2000) 631-635.
[24] X. Meng, J. Yang, X. Xu, L. Zhang, Q. Nie, M. Xian, Renew Energy 34 (2009) 1-5.
[25] M. Gouda, S.H. Omar, L.M. Aouad, World J. Microbiol. Biotechnol. 24 (2008) 1703-1711.
[26] Y. Chisti, Biodiesel from microalgae. Biotech. Adv. 25 (2007) 294-306.
[27] S. Fakas, S. Papanikolaou, M. Galiotou-Panauotou, M. Komaitis, G. Aggelis, J. Appl. Microbiol. 105 (2008) 1062-1070.
[28] S. Fakas, S. Papanikolaou, A. Batsos, M. Galiotou-Panayotou, A. Mallouchos, G. Aggelis, Biomass Bioenergy 33 (2009) 573-580.
[29] S. Papanikolaou, M. Komaitis, G. Aggelis, Bioresour. Technol. 95 (2004) 287-291.

[30] G.L.F. Vicente, F.J. Bautista, R. Gutierrez, V. Rodriguez, R. Martinez, F. Rodriguez, Energy Fuels 24 (2010) 3173-3178.
[31] R.X. Tan, W.X. Zou, Nat. Prod. Rep. 18(4) (2001) 448-59.
[32] A.A.L. Gunatilaka, J. Nat. Prod. 69(3) (2006) 509-526.
[33] N. Anjum, R. Chandra, Asian J. Pharm. Clin. Res. 8(4) (2015) 233-238.
[34] K. Germaine, E. Keogh, G. Garcia-Cabellos, B. Borremans, D. Lelie, T. Barac, FEMS Microbiol Ecol. 48 (2004) 109-121.
[35] A.S. Sarpal, G.S. Kapur, S. Mukherjee, A.K. Tiwari, Fuel 80 (2001) 521-528.
[36] E. Combet, J. Henderson, D.C. Eastwood, K.S. Burton, Mycoscience 47 (2006) 317-326.
[37] R.K. Bhagobaty, S.R. Joshi, Mycobiology 39 (2011) 71-78.
[38] N. Ladygina, E.G. Dedyukhina, M.B. Vainshtein, Process Biochem. 41 (2006) 1001-1014.
[39] A.L. Sunesson, W.H.J. Vaes, C.A. Nilsson, G. Blomquist, B. Andersson, R. Carlson, Appl. Environ. Microbiol. 61 (1995) 2911-2918.
[40] R.K. Bhagobaty, Proc. Natl. Acad. Sci. 85(1) (2015) 21–25.
[41] B.J. McAfee, A. Taylor, Nat. Toxins 7 (1999) 283–303.
[42] G.A. Strobel, B. Knighton, K. Kluck, Y. Ren, T. Livinghouse, M. Griffin, D. Spakowicz, J. Sears, Microbiology 154 (2008) 3319–3328.
[43] K.B. Ranjan, Proc. Natl. Acad. Sci. 85(1) (2015) 21–25.
[44] D. Banerjee, G.A. Strobel, E. Booth, B. Geary, J. Sears, D. Spakowicz, S. Busse, Mycosphere 1 (2010) 229-240.
[45] P. Dey, J. Banerjee, M.K. Maiti, Bioresour Technol. 102 (2011) 5815-5823.
[46] S. Kumar, N. Kaushik, PLoS One 8(2) (2013) e56202.
[47] G. Knothe, J. Am. Oil. Chem. Soc. 83 (2006) 823-833.
[48] M.R. Monteiro, A.R.P. Ambrozin, L.M Liao, A.G Ferreira, Talanta 77 (2008) 593-605.
[49] Z. Helwani, M.R. Othman, N. Aziz, W.J.N. Fernando, J. Kim, Fuel Process Technol. 90 (2009) 1502-1514.

*****