Microbial Transformation of Natural Products

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This article revises the current state of microbial transformation use in natural products. It represents the results of the most recent reports. Due to the importance of this transformation, there is still a strong need to intensify the direction of microbial transformation of the natural product compounds, besides exploiting more microorganisms that might be the used biocatalysts in producing the novel materials for pharmaceutical purposes.

Keywords:
Biotransformation, Biocatalysts, Natural products, Microbial transformation
INTRODUCTION

Microbial biotransformations are a combination of biochemical reactions to transform the structures of the phytochemicals and organic compounds, by exploiting microorganisms and their isolated enzymes, to develop a variety of useful constituents, through regio-stereoselectivity reactions (Baiping et al., 2010; Muffler et al., 2011). Biotransformation can be classified into two different approaches, the one that involves the transformation of substrates that are completely strange to the particular system which called "Xenobiotics," and the bio synthetically directed biotransformation in which the substrate tolerates a formal affiliation to a natural biosynthetic intermediate (de, 2011; Omar et al., 2012). In Biotransformation processes, an intact whole cell microorganism or isolated enzyme systems can be used, and each approach has its advantages and disadvantages. For instance, clean enzymes in biocatalysis could have selectivity for certain feedbacks, simple system and processes and better acceptance to co-solvents used to dissolve low-water soluble substrates. On the other hand, enzyme separation and cleansing is fairly costly and needs time and usually, it is more difficult to perform reactions need more than one enzyme (de, 2011; Roberts et al., 1994; Severiano et al., 2013).

Human has commonly used microbial biocatalysis since thousands of years ago for the bread making, dairy products and alcoholic drinks. Scientifically, Louis (1862) put the first scientific bases for the microbial transformation applications, when he used a pure culture of _Bacterium xylinium_ is used to transform the alcohol to acetic acid. Subsequently, several microbial transformations' experiments have been carried out, which showed that a one-step procedure might produce a remarkable product (de Carvalho and da Fonseca, 2006; de, 2011).

Microbial biotransformation by the whole cell microorganisms is often advantageous as compared to isolated enzymes; it is respected economically and ecologically a competitive tool for the biotechnological professionals in search of new techniques to manufacture clean valuable chemicals, pharmaceutical, and agrochemical compounds (Carballeira et al., 2009; Luo et al., 2013). Microbial transformation has been extensively used, to create new and useful metabolites of almost all classes of terpenes, Steroids and herbal extracts such as tea extracts as a substitute of chemical synthesis for preparation of pharmacologically active compounds (Chen and Chen, 2013; Omar et al., 2012). Biotransformation can be some times the only predictable technique to yield specific compounds, such as the hydroxylation of the non activated carbon atoms (de Carvalho and da Fonseca, 2006). In the literature there are numerous examples and cases of biotransformation directing to the creation and separation of chiral organic compounds.

The whole cell microorganism's bio-catalysis provides a bulky collection of enzymatic action and selectivity, such as the _oxygenases_, which are hard to separate, and need extensive and costly cofactor requirements. Additionally, using whole cells may provide other enzymes, e.g., oxido-reductases or hydrolytic enzymes that can have substrate specificities and product selectivity (Holland, 1998; Holland, 2001; Joyeau et al., 2013).

Nevertheless, finding of the appropriate microorganism to perform the favorite biotransformation reaction is still a big challenge. Therefore, traditional screening of a series of microbial strains is still the most usable practice.

Commonly, the importance of biotransformation technologies in general and microbial transformations in particular can primarily show the following three purposes: I. Medicinal herb fermentation processing either by microorganisms or plant cells, in order to harvest mass of secondary metabolites of the targeted plant. II. Constructing a microbial model for metabolic mechanisms of the herbal medication. In the direction of understanding the appropriate medicinal metabolites in human, this because microorganisms' enzymes could break down _Xenobiotics_ similarly as it happens by mammalian enzymes, e.g., hydroxylation, acetylation, N-dealkylation, and others (Asha and Vidyavathi, 2009) and III. Manufacturing and altering the effective constituents of the phytomedicine, enlarge the natural products collection, and to draw the routes of drugs' synthesis (Baiping et al., 2010).

So far massive work has been accomplished in biotransformation and numerous extensive reviews have been published (de Carvalho and da Fonseca, 2006; Muffler et al., 2011). As a compliment, this article will review the most recent findings which only covers the transformations of some natural products extracts and terpenoids that are more related to the current research.

**Biotransformation of Natural Products and Extracts**

The hydrolytic and reductive capacities of microorganisms (bacteria, yeast and fungi) have been identified decades ago, and at this time they are used in preliminary and manufacturing reactions. Different bioactive phytochemicals and herbal products have been exposed to the microbial bio-catalysis as an attempt to find further lively and fewer toxic products. Here is a review of the microbial conversion of some of the natural products and extracts; in addition to that we list some microbial transformation of bioactive compounds from medicinal plants' origin in Table 1.
Table 1: Examples of microbial transformation of some bioactive phytochemicals

<table>
<thead>
<tr>
<th>Phytochemicals</th>
<th>Structure</th>
<th>Origin and Bioactivity</th>
<th>Microbial catalyst</th>
<th>Transformed products</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Artemisinin</td>
<td><img src="image" alt="Artemisinin Structure" /></td>
<td>Artemisia annua Anti malaria</td>
<td><em>Mucor polymorphous, Aspergillus niger, Cunninghamella echinulata and others</em></td>
<td>3β-hydroxy Artemisinin and 3β-hydroxydeoxyartemisin 1α-hydroxydeoxyartemisin, deoxy artemisinin and 3α-hydroxydeoxyartemisin and others reviewed extensively by Omar et al., 2012</td>
<td>(Omar et al., 2012)</td>
</tr>
<tr>
<td>Codeine</td>
<td><img src="image" alt="Codeine Structure" /></td>
<td>Papaver somniferaum Narcotic analgesic</td>
<td><em>Pseudomonas testosteroni, And Streptomyces griseus</em></td>
<td>Codeine, 14β-hydroxy codeineone, N-Desmethyl-codeine and N-Demethylation</td>
<td>(Kunz et al., 1985)</td>
</tr>
<tr>
<td>Ursolic acid</td>
<td><img src="image" alt="Ursolic acid Structure" /></td>
<td>Anti Cancer</td>
<td><em>Nocardia sp. NRRL. 5646</em></td>
<td>Oleanolic acid methyl ester, oleanolic acid and ursolic acid methyl ester</td>
<td>(Zhang et al., 2005)</td>
</tr>
<tr>
<td>Acronycine</td>
<td><img src="image" alt="Acronycine Structure" /></td>
<td>Acronychia baueria Anti-tumor</td>
<td><em>Cunninghamella echinulata NRRL 3665, Streptomyces spectabilis NRRL 2494, Cunninghamella sps.</em></td>
<td>9-OH-acronycine, 3-OH methylacronycine, 1-OH acronycine, 9,11-diOH acronycine and 3-OH-methyl-11-OH acronycine</td>
<td>(Betts et al., 1974; Rosazza, 1978; Rosazza et al., 1978)</td>
</tr>
<tr>
<td>Colchicine</td>
<td><img src="image" alt="Colchicine Structure" /></td>
<td>Colchicum autumnale Gout relief Anti-tumor</td>
<td><em>Arthrobactus colchovorum and Streptomyces griseus ATCC 13968</em></td>
<td>N-Deacetyl colchicine, 2-O-Desmethyl colchicine and 3-O-Desmethyl colchicine</td>
<td>(Kyslíková et al., 2013; Zeitler and Niemer, 1969)</td>
</tr>
<tr>
<td>Compound</td>
<td>Chemical Structure</td>
<td>Sources/Properties</td>
<td>Metabolites/Characteristics</td>
<td>References</td>
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<tr>
<td>Progesterone</td>
<td><img src="image" alt="Progesterone" /></td>
<td>Pregnant mares / Hog ovaries Sex hormone</td>
<td>Thamnostylum piriforme ATCC 8992 and Mucor griseocyanus ATCC 1207 14a-OH progesterone, 9a-OH progesterone, 14a-OH progesterone, 7a, 14a, di-OH progesterone and 6b, 14a-OH progesterone</td>
<td>(Chantilis et al., 1996; Hu et al., 1995)</td>
<td></td>
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<tr>
<td>Betulinic acid</td>
<td><img src="image" alt="Betulinic acid" /></td>
<td>Doliocarpus schottianus Anti-HIV and Anti-melanoma agent</td>
<td>Bacillus megaterium ATCC 14581 and Cunninghamella Elegans ATCC 9244 3b, 7b-diOH-lup-20(29)-en-28-oic acid 3b, 6a,7b-triOH-lup-20(29)-en-28-oic acid and 1b, 3b, 7b-triOH-lup-20(29)-en-28-oic acid</td>
<td>(Kouzi et al., 2000)</td>
<td></td>
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<tr>
<td>Caffeine</td>
<td><img src="image" alt="Caffeine" /></td>
<td>Coffea arabica CNS stimulant</td>
<td>Pseudomonas putida and Pencillium roqueforti Theobromine 3-desmethyl caffeine and Biodegradation (Immidazole ring breakage)</td>
<td>(Fuhr et al., 1992; Kurtzman and Schwimme r, 1971)</td>
<td></td>
</tr>
<tr>
<td>Testosterone</td>
<td><img src="image" alt="Testosterone" /></td>
<td>Male human urine anabolic steroid Sex hormone</td>
<td>Mucor griseocyanus ATCC 1207, Thamnostylum piriforme ATCC 8992 and Botrytis cineraria 14a-OH testosterone, 14a-OH androst-4-ene-3,17-dione, 14a-OH progesterone, 9a-OH testosterone and 7b, 17b-dihydroxy androstan-3-one and</td>
<td>(Farooq and Tahara, 2000; Hu et al., 1995; Sivapathas undaram et al., 2001)</td>
<td></td>
</tr>
<tr>
<td>Taxol</td>
<td><img src="image" alt="Taxol" /></td>
<td>Taxus baccata Anti-tumor</td>
<td>Nocardioides albus 13-deacetylpaclitaxel</td>
<td>(Desai et al., 1998)</td>
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</tbody>
</table>
**Hongqu extract**

It is a transformed product that is fermented on rice by *Monascus purpureus* W. (Hongqumei), and it has been consumed as food and medicine in China since centuries ago. This product has several bioactive components with lower hyperglycemia, lower hypercholesterolemia, lower hyperpiesia, antimicrobial, and anti tumor activities, and numerous related bio-active compounds have been isolated from Hongqu, such as monascorubin, monacolins, notalin, and ergosterin (Baiping et al., 2010).

**Green tea and Yerba mate Extracts Biotransformation**

Green tea (*Camellia sinensis*) and yerba mate (*Ilex paraguariensis*) extracts have been biologically transformed. These two plants have some plenty constituents of the poly phenolic compounds that assumed to contribute in the health benefits of tea. Biotransformation of their extracts with *tannase* has improved the antioxidant power to 55% and 43% for the green tea and the yerba mate correspondingly (Macedo et al., 2011; Sang et al., 2011). Figure 1 illustrate the transformation of epigallocatechin gallate (EGCG) to epigallocatechin (EGC) and Galic acid.

**Biotransformation of Triterpenes**

Triterpenes biologically expected to be a multipurpose collection of terpenes. Oleanane, ursane, lupane, and dammarane–euphane carbon skeletons supposed to be the greatest vital triterpenoids structures. Terpenoids have been reported to have numerous biological effects like anti-inflammatory, hepato protective, analgesic, antimicrobial, anti mycotic, virostatic, immune modulatory and tonic effects. On the other hand, some triterpenoids display some disadvantages such as the hemolytic and cytostatic characteristics that limit their pharmacological practice, in addition to that some of them are poorly water soluble, which significantly limits its application. As consequence biotransformation of these triterpenes became a promising significant technique to overcome some of the triterpenes restrictions and to expand the range of usable triterpenes (Chen et al., 2013; Choudhary et al., 2011; Grishko et al., 2013; Iqbal Choudhary et al., 2013).

**Betulin**
Feng et al. (2013) in a most recent article have investigated the biotransformation of betulin to betulinic acid by Cunninghamella blakesleeana cells and the LC–MS analysis showed that betulin could be converted into at least five derivatives from cultured C. blakesleeana cells, betulinic acid showed to be the most significant (Feng et al., 2013). Figure 2 shows the transformation of betulin to betulinic acid.

Asiatic acid and Asiaticoside

Recently, a research group has published their biotransformation experiments of Asiatic acid, Asiatic acid considered one of the major important triterpenes of Centella asiatica that shows wound healing, anti-inflammation and anti tumours activity. Various types of microorganisms have been used in these experiments which afforded many derivatives (Guo et al., 2013; He et al., 2010; Huang et al., 2012).

Asiaticoside, also one of the major triterpenes saponins in Centella asiatica, and was commercially used as a wound-healing agent, owing primarily to its potent anti-inflammatory effects some asiaticoside derivatives possess strong neuroprotective effects against beta-amyloid-induced neurotoxicity by anti-apoptotic and anti oxidative injury mechanisms (Alfarra and Omar, 2013). Oxy-asiaticoside an intermediate derivative of asiaticoside and was used in for the tuberculosis treatment and wound lesions. Asiaticoside was transformed by the extracted enzymes produced by Fusarium oxysporum to derhamno-degluco-asiaticoside and derhamno-asiaticoside (Monti et al., 2005). However, to the best of our knowledge, up to this date there are no reports on the microbial transformation of Asiaticoside (Alfarra et al., 2013).

CONCLUDING REMARKS

Microorganisms catalysis or what is known in the technology age biotransformation have been extensively used since the early days of mankind for the making of dairy products, alcoholic beverages and bread. However, in this day and age there is a great interest and passion to trespass of whole-cell catalysts of microorganisms as natural reagents in organic synthesis, in addition, it is notable that the use of microbial transformation still gaining popularity in the research of the production of drug metabolites, carbohydrates, and amino acids. The exponential increase of the publications number still shows no indication of considerable achievements. Just few microorganisms was explored for the probable use as biocatalysts, even though many of the species not examined until now. Discovery of the specific microorganism that can convert the interested specific molecule still one of the difficulties that consumes time and effort in the research. There is a strong believe that significant attempt in the area of microbial transformation of drugs still in need. It is showed to be a very useful method to get considerable volumes of metabolites for pharmacological and toxicological studies, furthermore, concepts of microbial biotransformation assumed to contribute to resolve the problems that may occur from drug interactions and metabolism. Work on biotransformation area should include the exploring of some extremophilic microorganisms, the development of methods for the use and control of microbes, combinatorial methods, and the use of the recombinant engineered microorganisms.

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