Biotransformations are chemical reactions catalyzed by microbial cells (growing or resting) or enzymes isolated from microorganisms. Drug biotransformation is generally considered to detoxify the drug to form more polar metabolites which can be easily excreted. However, it can also lead to the formation of metabolites possessing greater pharmacological activity than the parent compound or, alternatively, it may prove to be more toxic [1]. Active metabolites may possess on-target activity (significant or entire contribution in pharmacological action) or off-target activity (unrelated to the activity of the parent drug). In some cases metabolites formed might reverse the action of the parent drug [2].

Animal models (hepatocytes, subcellular fractions, liver slices) have been used extensively for studying drug metabolism but microorganisms could be used for the production of mammalian metabolites too. Cytochrome P450-dependent enzymes have been discovered in a variety of yeast, bacteria and fungi possessing the capability to mimic mammalian metabolic reactions partially or completely. Mammalian biotransformation is generally categorized in the phase I and phase II reactions [1].

**Phase I Reactions.** This type of the reactions is called as functionalization since they introduce a functional group in the molecule resulting in a slight increase in hydrophillicity and may increase its pharmacological activity. These are further classified as:  
**Hydrolysis.**  
— Carboxylesterases, cholinesterases, organophosphatases, e.g. hydrolysis of procaine (used as local anaesthetic);  
— Peptidases;  
— Epoxide hydrolases:  
— detoxifying enzyme for epoxides (aromatic, unstable and reactive molecules);  
— formation of diols (accessible to phase II).  
**Reduction (azo- and nitro-reductions).**  
— enzymes of intestinal flora (especially in large intestine);  
—cytochrome P450 (usually oxidizing enzyme), has the capacity to reduce xenobiotics under low oxygen or anaerobic conditions;  
— interactions with reducing agents (reduced forms of glutathione, NADP).  
**Oxidations.**  
These reactions include hydroxylation, epoxidation, oxidation of alcohols and aldehydes, oxidative degradation of alkyl chains, oxidative deamination.  
**Phase-II Reactions.** This type of the reactions generally further increases the hydrophillicity of the drug and facilitates the excretion of the drug and its metabolites. They are classified on the basis of conjugation of drug molecule or phase—I metabolite with endoge-
Biotransformation is crucial for estimation of specific clinical parameters of the drugs. High bioavailability and clearance usually results from high metabolism, thus establishing the fact that metabolite studies are an important factor in drug designing [1]. Identification of active metabolite is necessary when a drug exhibits unexpectedly enhanced pharmacological activity in vivo [2]. Initially, the purpose of microbial biotransformation was to obtain more active or less toxic metabolites. Metabolites obtained through microbial transformation could help to correlate with those obtained through in vivo or in vitro animal models. When drug metabolism is studied, microbial biotransformation offers several advantages as compared to mammalian metabolism:

1. Simple and cheap maintenance of microbial cultures as compared to cell or tissue cultures or laboratory animals.
2. Facile repetitive screening process in which different strains are used to metabolize the drug.
5. Less toxic novel metabolites as compared to parent molecule.
6. Mild and ecologically harmless reaction conditions (normal pressure, low temperature, neutral pH) for sustainability.
7. Dependence on the nature of the biocatalyst and substrate prediction of the metabolic reactions.
8. Convenient scaling up of the metabolite production for pharmacological and toxicological evaluation, isolation and structure elucidation when parallel animal metabolic studies reveal the required information about the metabolites.
9. Generation of structural diversity in a chemical library through introduction of functional groups at various positions of a drug molecule thus in turn affecting the structure-activity relationships.
10. Suitable alternative where it is tedious to introduce a functional group by chemical methods, e.g. 11-α and or 11-β-hydroxylation of corticosteroids. It also liberates from use of hazardous chemicals and catalysts thus provide a relatively more safe and efficient method.
11. High stereospecificity of reaction due to the complex, three dimensional and asymmetric nature of enzyme enabling to recognize its substrate and even distinguish different stereochemical configurations of the substrate molecule [3].
12. High regiospecificity as an enzyme specifically attacks its substrate at the position where the reaction takes place [3].
13. Mostly mild incubation conditions.

Biotransformation undoubtedly is a phenomenon that engulfs the solutions to major economic and financial problems faced by pharmaceutical industries regarding the discovery and synthesis of new molecules having the desired characteristics to be launched as an active drug in the market. Although biotransformation encompasses various fields and objectives, the focus of this article aims at 3 main objectives: (1) lead expansion: obtaining more active or less toxic metabolites from bioactive molecules; (2) biosynthesis of precursors/intermediates involved in the production of bioactive molecules; (3) stereochemical reactions and resolution of racemic mixture.

LEAD EXPANSION: OBTAINING PHARMACOLOGICALLY IMPROVED METABOLITES FROM BIOACTIVE MOLECULES

Biotransformations enhance the molecular diversity around active core structures after initial screening or after selecting compounds for preclinical development. In the initial lead expansion phase, the biotransformations can be utilized as a tool for drug designing, leading to substitutions at positions difficult to access by synthetic approaches. These derivatives help refine the structure-activity relationships, potentially generating new ideas of compounds to be synthesized. Also, the biotransformations are propitiously combined with synthesis, as in most cases reactions can be applied to related structures, thus multiplying the number of available compounds for screening.

Metabolites exhibiting significant pharmacological activity or less toxicity as compared to parent molecule could be expediently used as leads for drug designing. Structural modification during lead optimization phase of drug discovery might improve desired properties of lead candidates. Accordingly, if a metabolite with ample pharmacological activity and less toxicity is discovered, it might serve as a lead with an additional benefit of advanced properties. Such approach was used in the discovery of ezetimibe, a cholesterol absorption inhibitor [2]. In this study an active metabolite (lead candidate) which was 30 times more potent than the parent molecule was further optimized to produce final drug candidate which was 400 times more potent than the original lead.

Metabolites apart from possessing specific inherent distinction in its chemical characteristics from the parent drug also acquire structural similarity to the parent molecule. Hence, these metabolites show certain pharmacological characteristics similar to the parent molecule. This is mostly observed in simple functionalization reactions, e.g. O-demethylation, N-demethylation, hydroxylation and dehydrogenation. A minor structural modification of the metabolite may cause loss of potency or modification of pharmacological activity of the parent drug. For example, O-dem-