Enzyme-Mediated Synthesis of Heterocyclic Compounds

Deepshikha Rathore, Geetanjali, and Ram Singh

Abstract

Heterocyclic compounds are cyclic organic molecules possessing at least one atom other than carbon in the ring structure. They are a widely used class of organic compounds. The heterocyclic scaffolds represent the central framework of many biologically active molecules. The other applications, like agrochemicals, veterinary products, dyes, etc., also make them essential. Due to its high global demand, there is always a need for new and efficient methodology in synthesizing these molecules. The sustainable process with a minimal environmental impact is the need of the present day and biocatalysis supports this. Enzymes are biocatalysts and play a progressively significant role in the synthesis of heterocyclic molecules. This chapter discusses the utility of different enzymes for the synthesis of nitrogen-, oxygen- or both containing heterocyclic molecules.

Keywords

Enzyme • Biocatalysts • Heterocyclic compounds • Green synthesis • Sustainable synthesis

1 Introduction

Heterocyclic compounds are cyclic organic molecules possessing at least one atom other than carbon in the ring structure (Sabir et al. 2015). Their both properties, physical and chemical, are reliant on the presence of heteroatom(s). These compounds find their applications in almost all types of industries, including pharmaceuticals, agrochemicals, dyes, and pigments and others (Joule and Mills 2013; Rudi et al. 2005; Broughton and Watson 2004; Arunkumar 2015; Arora et al. 2012; Kozikowski 1984). Due to its increasing demand, researchers continuously keep an eye on their synthetic routes. They are always trying to develop various efficient and environmentally friendly methodologies (Busto et al. 2011; Feber 2004; Shoda et al. 2016). The conventional methods, like chemical catalysis, electrochemical, microwave-assisted, and using ionic liquids, solid and solution phase synthesis, are some of the well-known methods (Busto et al. 2011; Feber 2004; Shoda et al. 2016). Still, the development of a sustainable synthetic process is widely explored in organic synthesis. Biocatalysis has shown potential towards the synthesis of organic molecules, including heterocyclic molecules in a sustainable and environmentally friendly manner (Milner and Maguire 2012; Mane et al. 2018; Wu et al. 2019; Singh et al. 2006).

Biocatalysis is the use of bio-based catalysts or enzymes in organic synthesis (Dalal et al. 2016; Xie et al. 2013; Li et al. 2008; Xiang et al. 2013, 2014; Xue et al. 2012; Ding et al. 2015). Apart from the other advantages possessed by enzyme-catalyzed reactions, they also take care of the production of single enantiomers instead of racemic mixtures (Singh et al. 2006). The two fundamental properties of catalysts also apply to enzyme-catalyzed reactions: (i) increase in the rate of reaction; and (ii) remain non-consumed after the reaction. The focus of research revolves around the development of stereo-, regio- and chemo-selective reactions (Shoda et al. 2016; Kobayashi et al. 1997, 1996). Different classes of enzymes such as hydrolases, oxidoreductases, transferase, lyase, isomerase and ligase perform different types of reactions (Fig. 1) (Shoda et al. 2016; Singh et al. 2006; Groger and Asano 2012; Webb 1992).

In this chapter, the enzyme-catalyzed synthesis of pyrroles, indoles, phenazines, benzocarbazoles, benzimidazoles,

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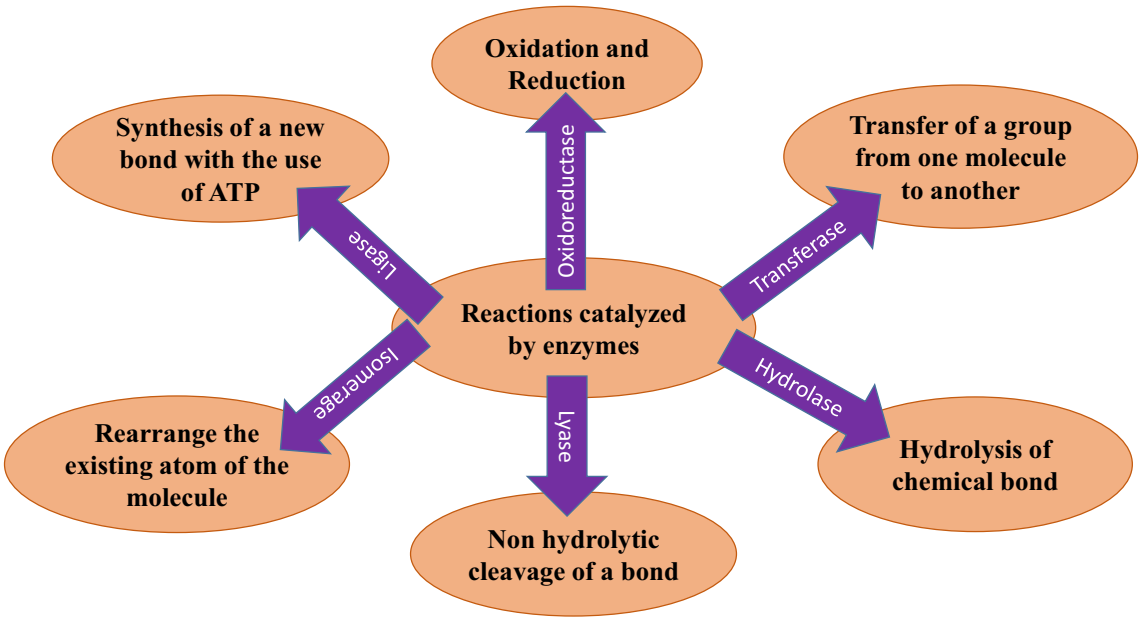


Fig. 1 Reactions catalyzed by enzymes in organic synthesis

pyrazoles, benzofurans, chromenes, dioxins, lactones and oxazolidinones have been discussed.

2 Enzyme-Mediated Synthesis of Heterocyclic Compounds

Enzyme-mediated synthesis is based on the ability of its active site to allow a particular substrate to enter into it and further get transformed into a suitable product (Yang et al. 2015; Kłossowski et al. 2013). Many name reactions like Morita–Baylis–Hillman reaction (Reetz et al. 2007), Michael addition (Zhang et al. 2017a), aldol addition (Li et al. 2008) and others (Hu et al. 2012; Wang et al. 2017) have been successfully carried out using enzyme-catalyzed method. Some of the important enzyme-mediated reactions have been discussed in this section towards the synthesis of heterocyclic compounds.

2.1 Nitrogen-Containing Heterocycles

Nitrogen heterocycles are mainly found in peptides and alkaloids, and these both are most widely spread in mostly all-natural products. The one, two and more number of nitrogen-containing heterocycles with 5-, 6- and 7-membered aromatic compounds is very important moiety in the field of medicine (Jampilek (2019); Vitaku et al. 2014; Singh and Geetanjali 2011). The synthesis of N-containing heterocyclic molecules has always been explored for sustainable processes (Veer and Singh 2019; Poonam 2019; Chauhan and Geetanjali 2000; Chauhan et al. 2003). There is always demand for the wide-structural diversity of N-heterocycles and hence exploration of synthetic protocols. They are prepared by synthetic reactions as well as via enzymatic methods. The biosynthetic pathways of naturally occurring heterocyclic compounds help in designing their enzymatic synthetic paths (Hemmerling and Hahn 2016; Junghanns et al. 1995).

2.1.1 Pyrroles

Pyrroles are five-membered ring, N-containing heterocyclic molecule which showed their utility in diverse biological activities, speciality polymeric materials, etc. (Trofimov et al. 2004; Bellina and Rossi 2006). The Paal–Knorr reaction has been used to synthesize the derivatives of N-substituted pyrrole in 60–99% yield catalyzed by α-amylase obtained from hog pancreas. This reaction was standardized using aniline (R=Ph) and 2,5-hexanedione as the starting materials (Fig. 2) (Zheng et al. 2013). The α-amylase from hog pancreas yielded N-phenyl-2,5-dimethylpyrrole in 94% yield; however, from *Aspergillus oryzae* gave the same product in 65% yield. This suggests that the sources of enzyme also play important role in product formation.

The authors have optimized the biocatalyst from eight different enzymes they tried for the reaction. The optimized enzyme, α-amylase from hog pancreas under mild reaction condition such as 50 °C temperature in methanol showed excellent activity using wide variety of primary amines (Zheng et al. 2013). The reactions showed high yields and better efficiency under mild reaction conditions.

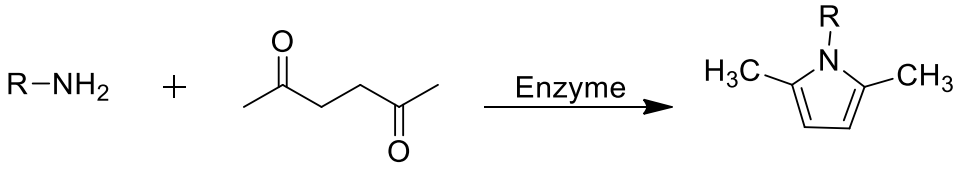


Fig. 2 Condensation of aniline and 2,5-hexanedione

2.1.2 Indoles

Spirooxindoles are indole derivatives present in many secondary metabolites and biologically important molecules (Ding et al. 2006; Galliford and Scheidt 2007). These molecules have been synthesized using enzymes as biocatalysts (Chai et al. 2011). The reaction of isatin, malononitrile and ethyl acetoacetate in the presence of lipase from porcine pancreas (PPL) in water–ethanol gave spirooxindole derivatives in 82–95% yield (Fig. 3) (Chai et al. 2011). The reaction condition with respect to reaction time, catalyst, temperature and solvent was optimized by the authors.

The use of others solvents like acetone, dichloromethane, hexane, chloroform, tetrahydrofuran, acetonitrile and dimethylformamide either did not promote the reaction or gave only moderate yield (Chai et al. 2011). Other commercially available hydrolytic enzymes were also evaluated for the reaction but PPL gave better results. The enzymes amano lipase M from *Mucor javanicus* and Amano lipase A from *Aspergillus niger* also gave the products in 82 and 84% yield, respectively. The lipase acrylic resin from *Candida antarctica* gave only trace of spirooxindoles. This was proposed that the specific spatial conformation along with the tertiary structure of PPL gave better yield of spirooxindole derivatives (Chai et al. 2011).

2.1.3 Phenazines

Phenazine is dibenzo annulated pyrazine with the formula $(C_6H_4)_2N_2$. Their derivatives are multifunctional in nature and successfully used in pharmaceuticals (Zhuo et al. 2013; Gamage et al. 2006), energy sectors (Okazaki et al. 2017; Lee et al. 2010), sensors (Pauliukaite et al. 2010), etc. Due to their broad applications, enzymatic synthesis has also been performed on this molecule synthesis (Sousa et al. 2014, 2018). Sousa et al. studied the use of Laccases, a multi-copper oxidase for the synthesis of phenazine and phenoxazinone frameworks from substituted aromatic amines (Fig. 4) (Sousa et al. 2014). This has been observed that the laccase-catalyzed reactions give only water as waste product when reactions are performed in aqueous solvent systems (Witayakran and Ragauskas 2009; Mikolasch and Schauer 2009). The reaction was performed under mild reaction condition using solvent methanol in phosphate buffer (pH 6–7) at temperature of 37 °C under aerobic conditions. The enzymatic oxidation of *meta*, *para*-disubstituted amine derivatives afforded phenazines. With the starting reagent, 1,2-diaminobenzene, 2,3-diaminophenazine was formed in 66% yield, whereas with *ortho*-aminophenol, phenoxazine formed in 83% yield (Sousa et al. 2014). The oxidation of 1-amino-2-naphthol with PPL at pH 7 gave 14H-dibenzo[a,j]phenoxazine-5,6-diol in 59% yield (Fig. 5) (Sousa et al. 2014).

This group further utilized the optimized protocol for a one-step asymmetric phenazines and phenoxazinones synthesis using spore coat protein A (CotA)-laccase enzyme as catalyst from *ortho*-substituted diamines and *ortho*-substituted hydroxylamines through aerobic oxidations (Sousa et al. 2018). Some of the other similar molecules synthesized using similar procedure with different reactants are given with their yields in Fig. 6 (Sousa et al. 2014, 2018).

2.1.4 Benzocarbazoles

Carbazole consists of two six-membered benzene rings fused on either side of a five-membered nitrogen-containing ring. This is based on indole structure where a second benzene ring is fused at the 2–3 position of indole. Carbazole derivatives possess pharmaceutical properties (Knölker and

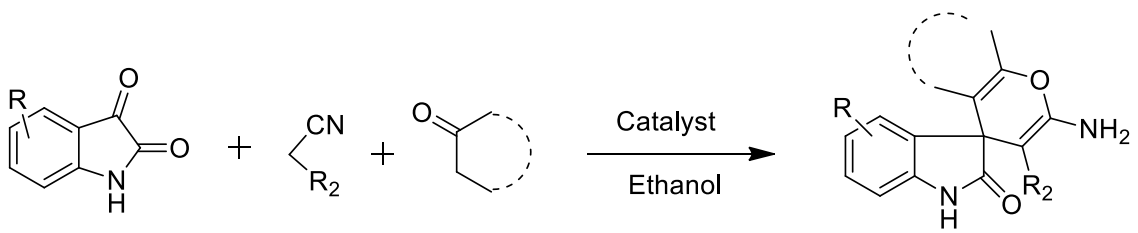


Fig. 3 Synthesis of different spirooxindole derivatives

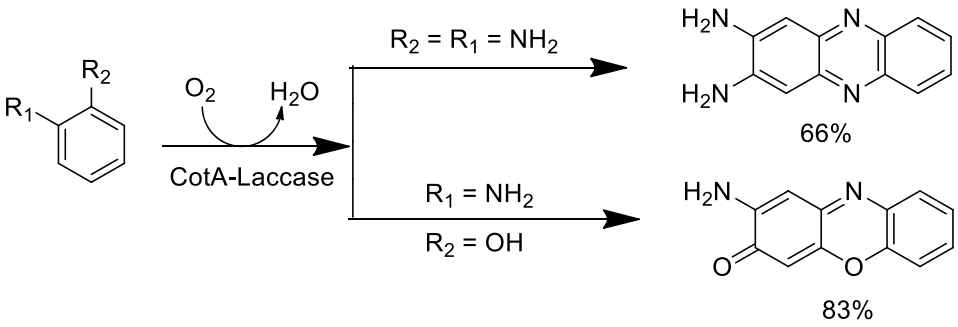


Fig. 4 Laccase-catalyzed reactions with substituted aromatic amines

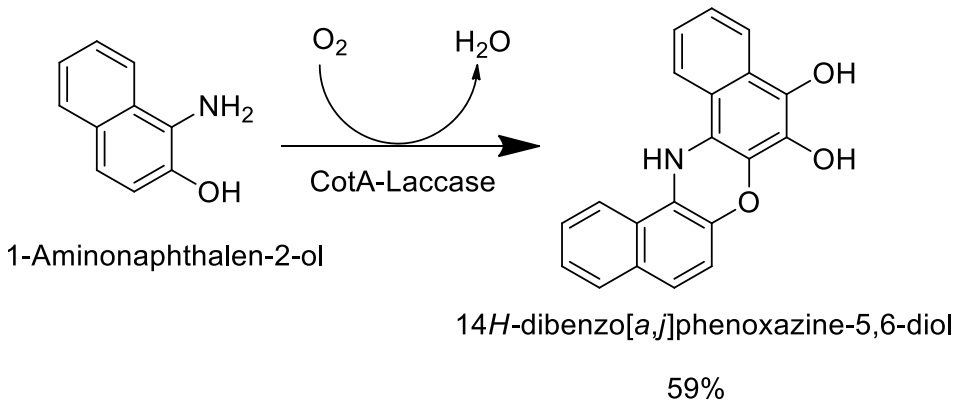


Fig. 5 Laccase-catalyzed synthesis of 14H-dibenzo[a,j] phenoxazine-5,6-diol

Reddy 2002; Pecca and Albonico 1971) and have applications in material sciences (Grazulevicius et al. 2003; Lia and Grimsdale 2010).

Sousa et al. performed the enzymatic synthesis of carbazoles using CotA laccase (Sousa et al. 2015). The oxidation of the *meta*, *para*-disubstituted arylamine 2,4-diaminophenyldiamine afforded benzocarbazole derivative in 74% yield and hence developed a clean method to construct in one-step C–C and C–N bonds (Fig. 7) (Sousa et al. 2015). The electrochemical behaviour of the target substrate plays essential role for product formation through an intramolecular oxidative coupling step (Sousa et al. 2015).

2.1.5 Benzimidazoles

Benzimidazole is a bicyclic heteroaromatic system which is composed of a benzene ring fused with an imidazole ring, i.e. five-membered ring with three number of carbon and two nitrogen is attached. It is occurring in nature as part of the vitamin B12 molecule with chemical formula $C_7H_6N_2$. This type of molecules possesses therapeutic properties including broad-spectrum anthelmintic, fungicidal or antimicrobial action (Salahuddin and Mazumder 2017).

Wang et al. in 2010 developed an efficient synthesis of bioactive compound in an ecologically and economically favourable way (Wang et al. 2010). In their study, they developed the enzyme-mediated synthesis of 2-alkylbenzimidazole. *o*-Phenylenediamine is used as primary reactant for the synthesis of the desired product (Fig. 8). In this synthesis, *o*-phenylenedimine reacted with the corresponding ester in the presence of immobilized lipase from *Mucor miehei* (MML) as a catalyst. This mixture was stirred at 50 °C for 60 h to complete the reaction and achieve the synthesis of benzimidazole derivatives (Wang et al. 2010). The reaction with other hydrolases like lipase acrylic resin from *Candida antarctica* B, Amano lipase M from *Mucor javanicus* and lipase from *Candida rugosa* gave low to poor yields. The spatial conformation of the lipases plays very important role in the product formation (Chai et al. 2011; Wang et al. 2010).

2.1.6 Pyrazoles

Pyrazoles are five-membered heterocyclic molecules possessing two nitrogen atoms in the ring. Pyrazole derivatives are biologically active compounds and also useful for other applications like dyes and luminophores (Mishra and Sasmal 2011; Stellrecht and Chen 2011). These molecules have been synthesized through enzyme-catalyzed reaction. Mane et al. used a whole cell biocatalyst, *Saccharomyces cerevisae* (Baker's yeast), with 1,3-dicarbonyl compound and hydrazines at room temperature to give the N-substituted pyrazole derivatives in 70–92% yield through oxidative cyclocondensation reaction (Fig. 9) (Mane et al. 2015). This synthetic method was found to be useful to a series of

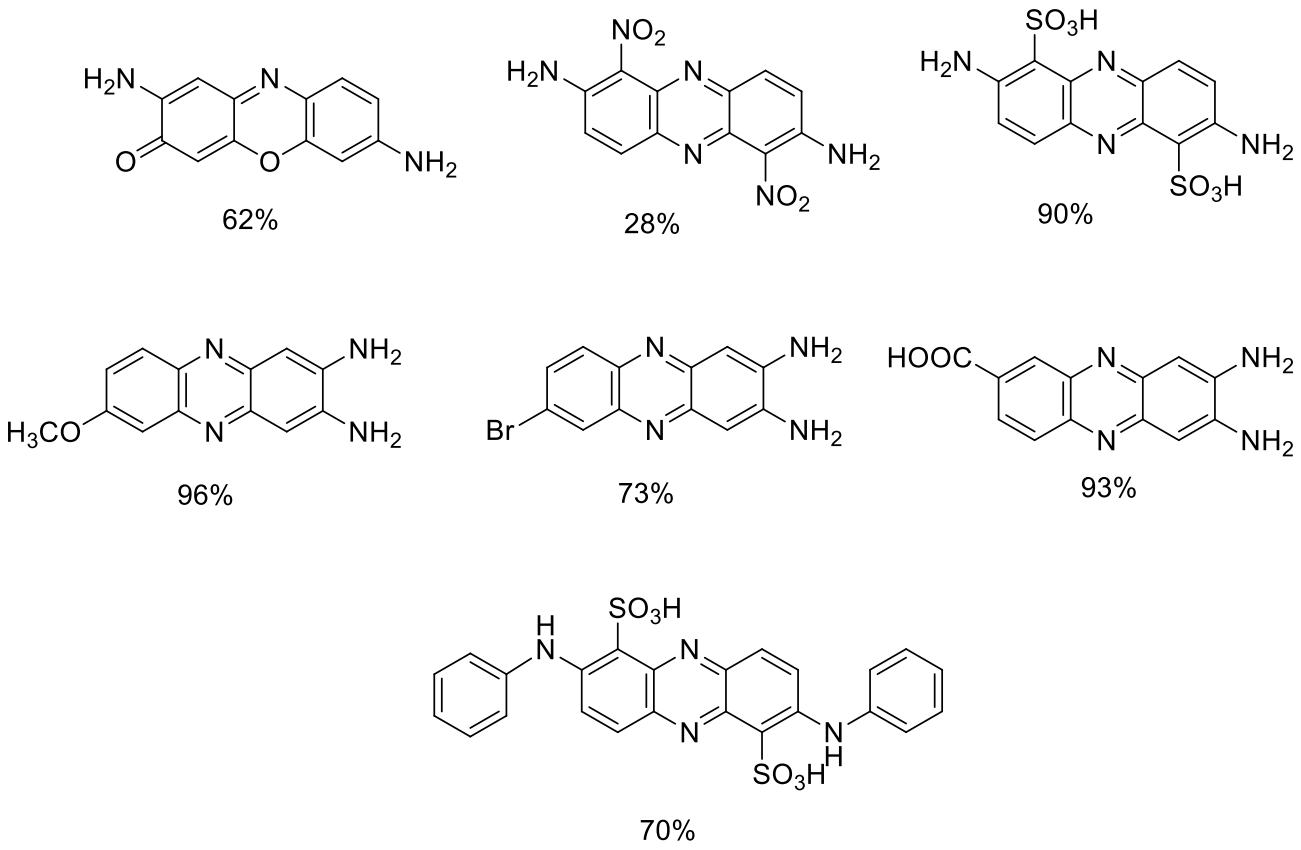


Fig. 6 Examples of laccase-catalyzed synthesis of some molecules

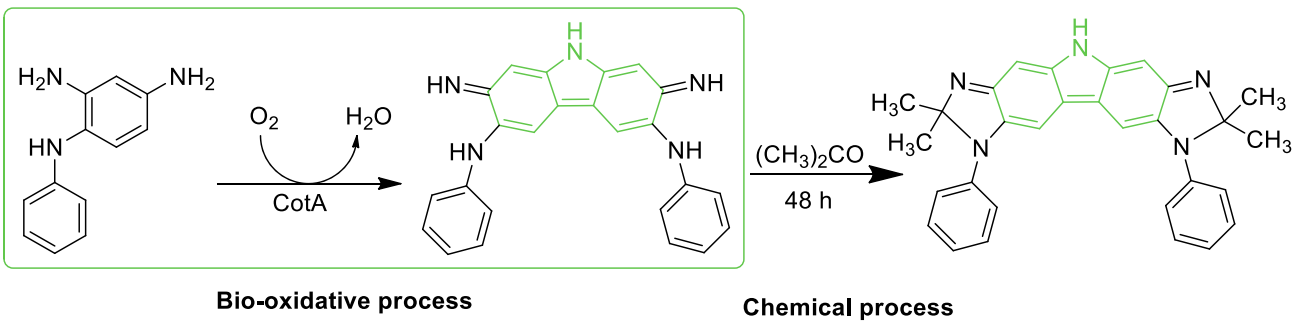


Fig. 7 Chemoenzymatic synthesis of benzocarbazole

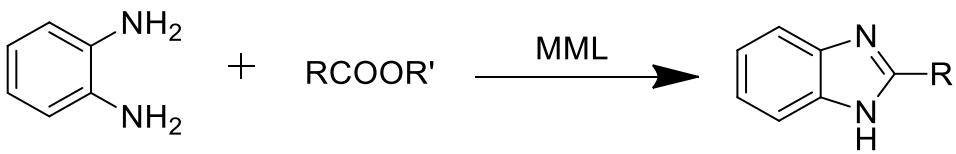


Fig. 8 Enzyme-mediated 2-alkylbenzimidazole synthesis

pyrazole derivatives where fermented Baker's yeast played role in efficient cyclocondensation 1,3-diketones with hydrazines/hydrazides. The study also suggested that the presence of the enzyme lipase in Baker's yeast accelerated the reaction leading to the desired pyrazole derivatives (Mane et al. 2015).

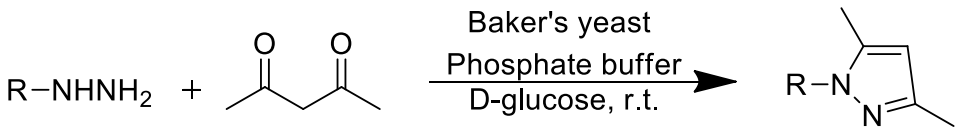


Fig. 9 Baker's yeast catalyzed synthesis of pyrazoles

Pyranopyrazoles are another important group of heterocyclic compounds. A four-component cyclocondensation reaction of hydrazine hydrate, malononitrile, ethyl acetoacetate and benzaldehyde afforded 6-amino-3-methyl-4-(3-nitrophenyl)-2,4-dihydropyrano[2,3-c]pyrazole-5-carbonitrile using lipase from fungi *Aspergillus niger* as catalyst (Fig. 10) (Bora et al. 2013). This method of synthesis was successfully utilized for different carbonyl compounds as one of the reactant yielding dihydropyrano[2,3-c]pyrazoles in 75–98% yield (Bora et al. 2013). The cyclic ketones also successfully gave spiro-substituted dihydropyrano[2,3-c] pyrazoles in 70–80% yield. The enzyme showed its utility towards wide range of substrates, reusability and mild reaction condition, i.e. room temperature and ethanol solvent. The study was also done on lipases from different sources such as *Pseudomonas cepacia*, Amano AK, *Penicillium camemberti*, *Porcine pancreas* and *Aspergillus niger* giving the yield of 65, 72, 75, 91 and 95%, respectively, for the product 6-amino-3-methyl-4-(3-nitrophenyl)-2,4-dihydropyrano[2,3-c]pyrazole-5-carbonitrile.

2.2 Oxygen-Containing Heterocycles

There is no doubt that oxygen-containing heterocycles play important role in industrial, medicinal and nutritional applications due to their diverse biological functions and natural abundance (Venkatachalam and Kumar 2019). Their synthetic methods are always being explored. There are many chemical synthetic methods to produce oxygen-containing heterocycles but due to toxicity and unfriendly approach towards environment and economy, there is demand for green synthesis. Enzyme-mediated synthesis is one of the methodologies to produce oxygen-containing heterocyclic compounds.

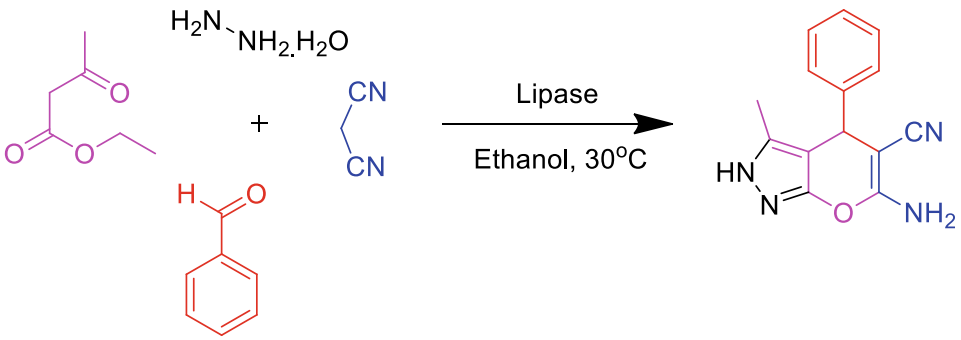
2.2.1 Chromenes

4H-chromene derivatives are shown their potential in various pharmaceutical activities (Zhang et al. 2018). They have been synthesized with the help of immobilized *mucor miehei* lipase (Fig. 11) through multi-component reaction using aldehydes, active methylene compounds and suitable nucleophile in 81–96% yield (Fig. 12) (Zhang et al. 2018). The immobilization of enzymes on magnetite nanoparticles (MNPs) has advantages for low mass transfer resistance, high specific surface area and easy separation from the reaction mixture in the presence of magnetic field (Vaghari et al. 2016; Hola et al. 2015). Zhang et al. used silica-coated MNPs as starting material whose surface was functionalized with 3-aminopropyltriethoxy silane to introduce amino groups. This was further treated with 2,4,6-trichloro-1,3,5-triazine (TCT) to develop support for covalent immobilization of enzyme (Zhang et al. 2018; Abbasi et al. 2016; Ranjbakhsh et al. 2012). Xu et al. also reported lipase-catalyzed synthesis of tetrahydrochromene derivatives using 1,3-dicarbonyl compound, aldehyde and malononitrile (Xu et al. 2011).

2.2.2 Dioxins

Dioxin is 6-membered heterocyclic, non-aromatic organic molecule which consists of four carbon atoms and two oxygen atoms. The molecular formula is $C_4H_4O_2$. It is also refereed to 1,4-dioxin or *p*-dioxin. There is an isomeric form which is 1,2-dioxin (o-dioxin) and is very unstable due to its peroxide nature. Agarwal et al. in 2014 proposed an enzymatic synthesis of polybrominated dioxins by using monooxygenase halogenase CPY450 Bmp7 as catalyst in

Fig. 10 Dihydropyrano[2,3-c] pyrazole synthesis



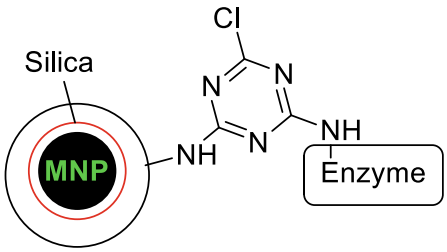


Fig. 11 Covalent immobilization of lipase enzyme

the reaction using bromocatechol as substrates (Fig. 13) (Agarwal and Moore 2014). Both bromocatechol and the electrophilic quinone, i.e. 3,5-dibromo-1,2-dibenzoquinone undergo coupling reaction in the presence of CYP450 Bmp7 enzyme as catalyst to produce dibenzo-p-dioxins. In this case, the benzoquinone provided both 1,4-dioxin oxygen atoms (Fig. 13) (Agarwal and Moore 2014). This reaction method is indicative of synthetic hetero-Diels–Alder coupling between orthoquinones and enamines leading to the formation of 1,4-benzodioxin frameworks. The whole reaction has very mild reaction conditions. The synthesis of desired dioxins can be achieved at room temperature. Excess bromine was quenched by the addition of sodium thiosulphate, and the reaction was extracted twice with the ethyl acetate. This methodology was simple, quite easy to handle and environment-friendly (Agarwal and Moore 2014).

2.2.3 Lactones

Lactones are cyclic esters and have potential applications in the field of synthetic intermediates, pharmaceutical molecules and polymers (Fischer and Pietruszka 2010). A monoclonal antibody (Fig. 14) was utilized as biocatalyst for the synthesis of -lactone (Kitazume et al. 1996). This antibody behaved as enzyme-like catalyst (abzyme) leading to the formation of carbon–carbon bond through the generation of carbanion and the internal nucleophilic attack on the carbonyl carbon to give -lactone (Fig. 15) (Kitazume et al. 1996). Drozdz et al. gave a chemoenzymatic method for the synthesis of lactone using catalyst acyltransferase from *Mycobacterium smegmatis* in high yield of 84–99% through Baeyer–Villiger (BV) oxidation method (Drożdż et al. 2016). The enzyme retained its activity even in harsh reaction condition like oxidation with 60% aq. H_2O_2 at 45 °C. The practical potential of this method was established by the use of different ketones as starting material to give their corresponding lactones (Drożdż et al. 2016).

2.2.4 Benzofuran

A five-membered ring possessing an oxygen atom fused with a benzene ring is known as benzofuran. Benzofuran derivatives have wide range of applications mainly in the field of pharmaceutical industries. They exhibited selective cytotoxicity against tumourigenic cell lines (Hayakawa et al. 2004), antiviral and antitumor and activities (Kim et al. 2006), and pharmaceutical agents (Murata et al. 2003; Murota et al. 1990). Kidwai et al. gave an enzymatic synthesis of this molecule (Kidwai et al. 2013). They studied the enzymatic oxidation of catechols/hydroquinones in aqueous solution using laccase as a catalyst and pyrazolin-5-ones as co-substrate (Fig. 16). Here, the enzyme laccase performs one-electron oxidation on catechol to quinone which undergoes 1,4-addition reaction with co-substrate to develop furan ring leading to benzofuro[2,3-c]pyrazolin-5-ones derivatives (Fig. 16) (Kidwai et al. 2013). The optimized synthetic process has been successfully extended towards the synthesis of a new series of benzofuropyrazole derivatives through the coupling of 3-methyl-1-phenyl-pyrazolin-5-one/3-methyl-pyrazolin-5-one and catechols/hydroquinones (Kidwai et al. 2013).

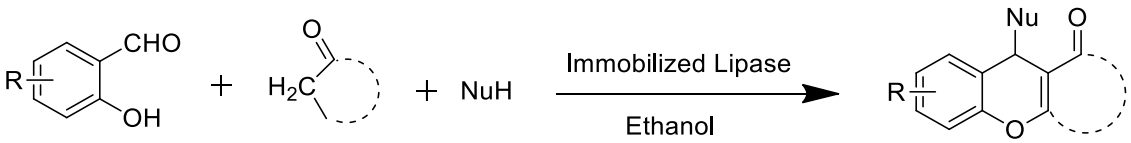


Fig. 12 Enzymatic synthesis of 4H-chromenes

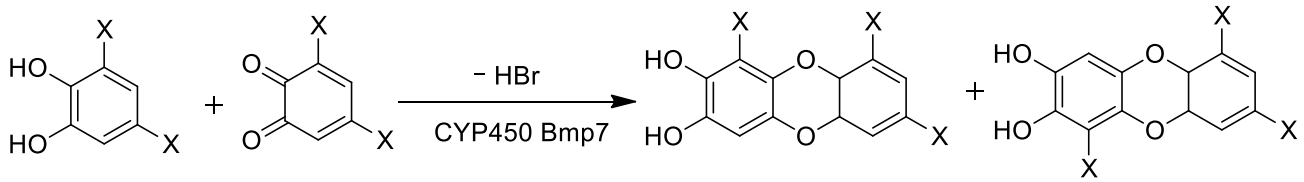


Fig. 13 Enzymatic synthesis of dioxin catalyzed by CYP450 Bmp7 (X=Br)

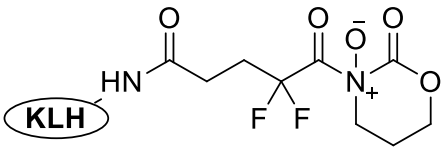


Fig. 14 Keyhole limpet haemocyanin (KLH) antibody

2.3 Nitrogen- and Oxygen-Containing Heterocycles

Heterocycles possessing nitrogen and oxygen in single ring or in a molecule are well-known moiety in medicinal chemistry. Their utility as immunomodulator, antifungal, psychotropical, antibacterial, neuro-related drugs, etc., has been established (Bhattacharya et al. 1991; Kakeya et al. 1998; Danielmeier and Steckhan 1995; Mishra et al. 2019). This section discusses the enzymatic synthesis of those heterocyclic molecules which have nitrogen and oxygen as heteroatoms.

2.3.1 Oxazolidinones

Oxazolidinones are nitrogen- and oxygen-containing five-membered heterocyclic molecules possessing varied applications (Kakeya et al. 1998; Danielmeier and Steckhan 1995). Yadav et al. studied the synthesis of 3-ethyl-1,3-oxazolidin-2-one using Novozyme 435 as catalyst from 2-aminoalcohol and dimethyl carbonate in 61–89% yield (Fig. 17) (Yadav and Pawar 2014). Among the eight immobilized lipases studied, the *Candida antarctica* lipase B (Novozyme 435) was considered as the choice of the catalyst for the reaction. The authors optimized the effect of various parameters like catalyst loading, temperature, agitation speed, solvent and mole ratio for the reaction (Yadav and Pawar 2014).

The lipase-catalyzed reaction was also utilized for the synthesis of enantioenriched oxazolidinone derivatives with excellent enantiopurities (Zhang et al. 2015). The reaction of 2-(methylamino)-1-phenylethanol and disubstituted carbonate as substrates yielded corresponding oxazolidinone in 46% yield with an absolute (S)-configuration as the major enantiomer (ee 92%) (Fig. 18) (Zhang et al. 2015). Different lipases such as from *Burkholderia* (*Pseudomonas*) *cepacia*, *Pseudomonas fluorescens* and *Candida antarctica* were studied. The immobilized *P. cepacia* gave better result and faster substrate transformation in chosen solvent *tert*-butyl methyl ether (Zhang et al. 2015). Various enzyme-mediated synthesized heterocyclic compounds have been represented in Table 1.

3 Summary and Outlook

Heterocyclic compounds are industrially essential molecules. Their history started in the eighteenth century, and since then, they developed themselves as both natural products and synthetic molecules. There is always a demand for the wide-structural diversity of heterocycles and hence

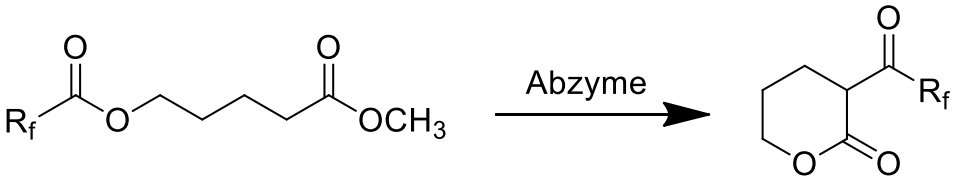
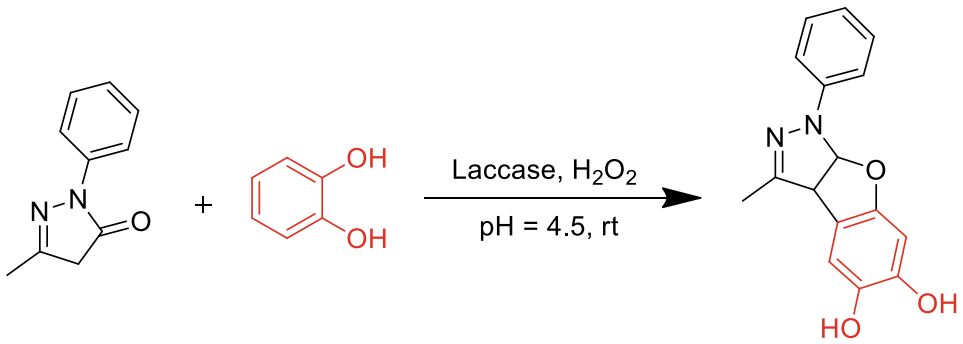


Fig. 15 δ-Lactone synthesis with abzyme catalyst

Fig. 16 Benzofuro [2,3-c] pyrazolin-5-ones synthesis



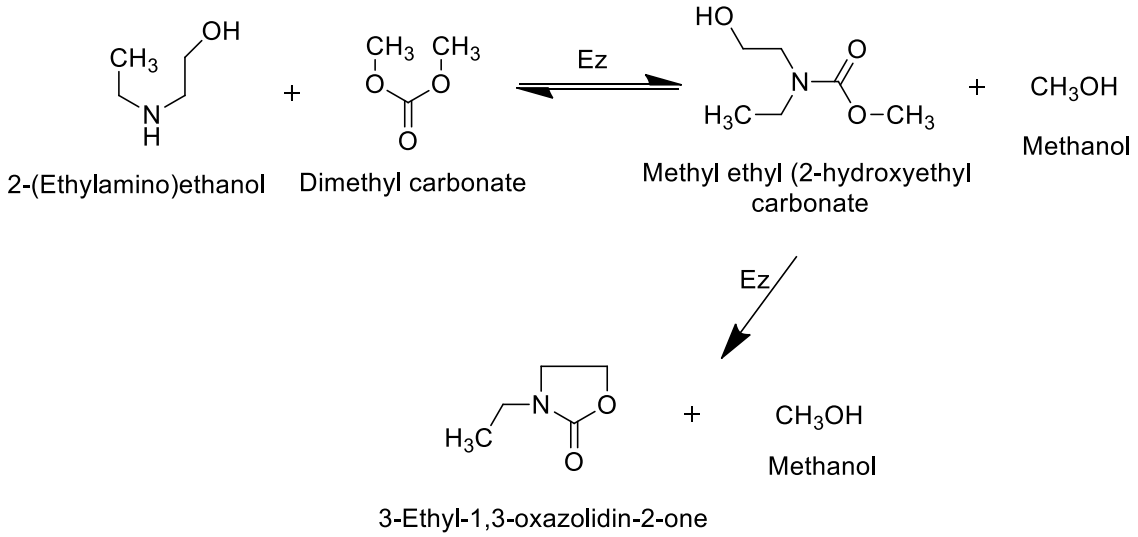


Fig. 17. 3-Ethyl-1,3-oxazolidin-2-one synthesis

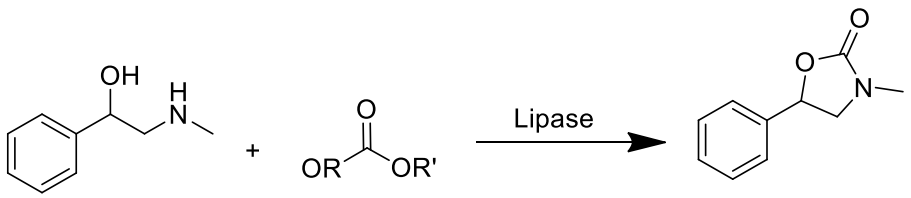


Fig. 18 Synthesis of oxazolidinone through lipase-catalyzed reaction

Table 1 Various enzyme-mediated synthesized heterocyclic compounds

S. No.	Heterocyclic compounds	Enzyme used for synthesis	References
1	*N*-Substituted pyrrole derivatives	Amylase from hog pancreas	Zheng et al. (2013)
2	Spirooxindole derivatives	Lipase from porcine pancreas	Chai et al. (2011)
3	Phenazine derivatives	Laccases	Sousa et al. 2014)
4	Phenoxazinone derivatives	Laccases	Sousa et al. (2014), Mihovilovic (2006), Bruyneel et al. (2009)
5	Carbazole derivatives	CotA laccase	Sousa et al. (2015)
6	2-Alkylbenzimidazole derivatives	Immobilized lipase from *Mucor miehei*	Wang et al. (2010)
7	Pyrazole derivative derivatives	*Saccharomyces cerevisae* (Baker's yeast)	Mishra and Sasmal (2011)
8	Pyranopyrazole derivatives	Lipase from *Aspergillus niger*	Bora et al. (2013)
9	4H-Chromene derivatives	Immobilized *mucor miehei* lipase	Zhang et al. (2018)
10	Tetrahydrochromene derivatives	Lipase	Xu et al. (2011)
11	Dioxin derivatives	Monooxygenase halogenase CPY450 Bmp7	Agarwal and Moore(2014)
12	δ-Lactone derivatives	Abzyme	Kitazume et al. (1996)
13	Lactone derivatives	Acyltransferase from *Mycobacterium smegmatis*	Drożdż et al. (2016)
14	Benzofuran derivatives	Laccase	Kidwai et al. (2013)
15	3-Ethyl-1,3-oxazolidin-2-one derivatives	Novozyme 435	Yadav and Pawar (2014)
16	Oxazolidinone derivatives	Lipase	Zhang et al. (2015)

(continued)

Table 1 (continued)

S. No.	Heterocyclic compounds	Enzyme used for synthesis	References
17	Indolyl 4H-Chromenes	Lipase	Zhang et al. (2017b)
18	Lactone	Engineered Baeyer–Villiger monooxygenase (BVMO)	Farhat (2020), Mihovilovic (2006)

exploration of synthetic protocols. Due to the environmental issues related to chemical synthesis, the researchers started looking for alternatives to chemical synthesis in accordance with green and sustainable chemistry. Out of the several modified methods, biocatalysis or using enzymes for synthesis showed potential. The enzymatic heterocyclic synthesis has contributed significantly to the structural diversity of heterocyclic compounds. This also allowed their applicability in various fields. No doubt, the chemical synthesis has also provided structural diversity, but for many asymmetric syntheses has better been performed using enzymatic methods.

This chapter discussed the enzymatic synthesis of nitrogen- and oxygen-containing or possessing both the atoms heterocyclic molecules. The synthesis of pyrroles, indoles, phenazines, benzocarbazoles, benzimidazoles, pyrazoles, benzofurans, chromenes, dioxins, lactones and oxazolidinones has been discussed. This has been observed that the enzyme-catalyzed reactions are more suitable than chemical reactions due to their high selectivity and mild reaction conditions. With the development of genetic engineering and enzyme engineering, the cost of enzymes continues to decrease. Enzyme activity and stability continue to increase. The superiority of enzymatic clean production is definitely promoting breakthrough development in this field. Enzyme-catalyzed biotransformation technology represents the future development of this synthetic industry.

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