Summary

Dihydropyrimidinone chemistry has been actively pursued recently. 3,4-Dihydropyrimidin-2(1H)-ones (DHPMs)\(^\text{\textsuperscript{a}}\) bear close relationship in their structure as well as cardiovascular effects with 1,4-dihydropyridines (DHPs), typified by nifedipine, amlodipine and nicardipine etc, which are potent calcium channel blockers. This has aroused considerable interest in these compounds during past one decade. While the DHPs are racemic, dihydropyrimidinones are inherently asymmetric. However, both of these categories of compounds depict identical receptor bound conformation, which has led to the binding site model and identification of conformational preferences depicting interaction with receptor sites, leading to the observed calcium channel binding effects. The increasing importance of scaffold decoration of heterocycles is attested by the fact that, compared to combinatorial approach and High Throughput Screening (HTS) approach, a good number of potent drug leads have been rationally designed, identified and evaluated for different types of biological effects.

DHPMs 4 are synthesized by simple one-pot cyclocondensation reaction of \(\beta\)-ketoester 1, aldehyde 2 and urea/thiourea 3 (Scheme 1). A number of high yielding variants of the traditional three-component Biginelli condensation employing a variety of catalysts, reagents and reaction conditions/techniques have been developed. Among appropriately functionalized DHPMs, several lead compounds with excellent calcium channel modulatory activity have also been identified.

![Scheme 1](image)

Although a large number of DHPM derivatives can be prepared in a one-pot Biginelli condensation (Scheme 1), systematically designed DHPMs have more often been obtained only through chemical functionalization of an appropriate site of the DHPM core. There are six possible sites (Figure 1) around the DHPM ring where modification/functionalization has been achieved. In this thesis, we have carried out some

\(^{a}\)Commonly referred to as Biginelli compounds
useful regioselective synthetic transformations on DHPMs, which were either not known or lacked practical utility. We have also develop an approach for transformation of Biginelli DHPMs to highly substituted pyrimidines and representative compounds have also been evaluated as modulators of cytostatic activity and inhibitors of \textit{Mycobacterium tuberculosis}. Also we have addressed N-1 and N-3 diversification for gaining access to enantiomerically pure DHPMs and have presented satisfactory characteristic data of both the enantiomers, which is extremely scanty in the literature. Further, using very simple chemical transformation, we have been able to append, organophosphorous as well as phosphorus heterocyclic groups at the N-3 position and evaluated them for calcium channel blocking activity. In yet another instance, while performing N-3 acylations, we isolated N-1,N-3 diacyl DHPM derivatives, which were found to transform to N-3 acyl derivatives through deacylation at N-1. Based upon this observation, we developed a useful protocol for “acyl group transfer” to various nucleophiles and have investigated scope and limitation of this reaction.

The thesis is presented in the form of eight chapters. Chapter 1 is introductory and a summary of the chapters 2-7 is presented below sequentially.

\textbf{Chapter 2. Review of Literature}

This chapter embodies a fairly comprehensive treatment to the subject including the chemistry, particularly the synthetic methods available for the synthesis of DHPMs, scaffold decoration methods used to create structural diversification at each of the six diversity oriented center around the DHPM core. The literature citations have been completed up to the year 2011.
Chapter 3. Regioselective oxidation of 3,4-dihydropyrimidin-2(1H)-ones to 1,2-dihydropyrimidin-2(1H)-ones and C-2 functionalization. In vitro evaluation as modulators of cytostatic activity and inhibitors of Mycobacterium tuberculosis

Oxidation of DHPM core is an important transformation as it conveniently results in the formation of 1,2-dihydropyrimidin-2(1H)-ones, which possess diverse pharmacological profile. For example, MKC-422 5, a HEPT (1-[(2-hydroxyethoxy)methyl]-6-(phenylthio) thymine) analogue is in clinical trials as anti-HIV drugs. Several nucleosides containing 5-substituted pyrimidine moiety have been shown to inhibit the growth of murine mammary carcinoma virus. Pyrimidine core with extended π-systems have interesting fluorescent properties and are useful in the development of advanced electronic and photonic materials. Unlike a facile in vitro and in vivo oxidation of DHPs, a key step in the initial metabolism of DHP-based drugs, DHPMs are resistant to ring oxidation. Further, the sensitivity of the methyl group at C-6 position to oxidizing agents, makes the selective oxidation of the dihydropyrimidinone ring troublesome.

A useful outcome of the oxidation of Biginelli DHPMs is the use of resultant 1,2-dihydropyrimidin-2(1H)-ones in obtaining C-2 substituted multifunctionalized tetrasubstituted pyrimidine derivatives, which form a basic skeleton of a wide range of biologically active molecules. Some of the representative drugs which contain pyrimidine core are Bay 41-4109 6 and Bay 39-5493 7 (non-nucleosidic inhibitor of hepatitis B virus), HAP-1 8. In literature, some methods have been reported to convert Biginelli DHPMs to C-2 substituted pyrimidines efficiently.

![Chemical structures](image)

In this chapter, we have presented the results of our investigation on:

(i) Highly regioselective oxidation of 3,4-dihydropyrimidin-2(1H)-ones at N3-C4 position
(ii) C-2 functionalization of 1,2-dihydropyrimidin-2(1H)-ones
(iii) Cytostatic and anti-tuberculosis activities of C-2 functionalized tetrasubstituted pyrimidines

267
We have found that 1,2-dihydropyrimidin-2(1H)-ones 10 could be obtained through the N3-C4 oxidation of DHPMs 9 with pyridinium chlorochromate (PCC) in dichloromethane using neutral reaction conditions and in a synthetically useful manner. The reaction requires 3.0 equiv. of PCC to drive the oxidation to completion; although, using 1.0 equiv. of the reagent, prolonged stirring was required. However, the modest excess of PCC is largely offset by the simplicity of the operation and reasonable good yields (55-71%) of the purified products. Further, no side product arising from C4/C6 dealkylation/oxidation etc. was observed.

Scheme 2. Synthesis of 1,2-dihydropyrimidin-2(1H)-ones 10 through PCC oxidation of 9.

The synthetic scope of oxidation of 9 by PCC under neutral reaction conditions has been established by using 9 with different substituents at N-1, C-4, C-5 and C-6 positions (Scheme 2). The plausible mechanism of dehydrogenation mediated by PCC has also been proposed.

In order to achieve the synthesis of C-2 functionalized tetrasubstituted pyrimidines 13 from 1,2-dihydropyrimidin-2(1H)-ones 11, introduction of a good leaving group at the C-2 position was required for activation towards reaction with nucleophiles. Thus, treatment of appropriate 11 with phosphorous oxychloride at 105°C, under solvent-less reaction conditions furnished corresponding C-2 chloro derivatives 12 (Scheme 3) in 85-93% yield. Reaction of 12 with ammonia and various N and O nucleophiles in absolute ethanol at 80°C, furnished corresponding C-2 substituted pyrimidines 13, in very good yields (65-97%).

Some of the compounds were evaluated for their activity against M. tuberculosis and modulators of cytostatic activity in cell culture. Although none of the compounds tested for inhibition of M. tuberculosis displayed very high activity, the nature of the C-2 substituent was found to modulate the cytostatic activity in cell culture.
Chapter 4. An efficacious chemoselective reduction of 1,2-dihydropyrimidin-2(1H)-ones using magnesium-methanol: Synthesis of 3,4-dihydropyrimidin-2(1H)-ones

Methods are available for creating N3-C4 double bond through dehydrogenation of DHPMs, but only few methods have been reported wherein the N3-C4 double bond of 1,2-dihydropyrimidin-2(1H)-ones has been selectively reduced. We envisaged that the reduction potential of alkali metals may be sufficiently high enough to add two electrons to the C=N bond and lead to smooth reduction of the C=N link. To achieve this goal, we have employed magnesium-methanol as the reagent for the selective reduction of N3-C4 double bond of the 1,2-dihydropyrimidin-2(1H)-ones.

1,2-dihydropyrimidin-2(1H)-ones 10, have been used as a precursors for carrying out the reduction of N3-C4 double bond. The reaction (Scheme 4) is initiated by addition of magnesium turnings and a crystal of resublimed iodine to a solution of 10 in methanol and the corresponding DHPM 9 are obtained in 60-76% yield.

Further, the transformation of 10 into 9, aided by magnesium in methanol is smooth, facile and highly selective to C=N link as other reducible functionalities such as ester and enamine ester and urea carbonyl remained unaffected. This constituted the first example of formation of Biginelli DHPMs 9 through chemoselective reduction of 10 (Scheme 4). A mechanism for reduction of 10 proceeding through the formation of free radical leading to the formation of DHPM 9 has been proposed.

Thus, the reduction of 1,2-dihydropyrimidin-2(1H)-ones with magnesium in methanol constitutes a highly efficacious protocol to effect highly regio- as well as chemoselective reduction of –C=N- link. Apart from simplicity of the procedure, no side product was formed rendering the reduction to be high yielding. The reduction reaction is tolerant to substitution pattern on the 1,2-dihydropyrimidin-2(1H)-one ring.

Scheme 4. Reduction of 1,2-dihydropyrimidin-2(1H)-ones 10 with Mg/MeOH.

Chapter 5. Chemical resolution of enantiomers of 3,4-dihydropyrimidin-2(1H)-ones using chiral auxiliary approach

DHPMs show pharmacological profile similar to classical DHP calcium channel modulators. In the inherently asymmetric DHPMs, the influence of the absolute configuration at the stereogenic C-4 center on biological activity is well documented. The proposed binding-site model for a series of DHPM calcium channel modulators based on a detailed structure-activity profile is shown in Figure 2. It has been established that calcium channel modulation (antagonistic vs agonistic activity) is dependent on the absolute configuration at C-4, whereby the orientation of the C-4 aryl group (R- vs S-configuration) acts as a “molecular switch” between antagonistic (aryl group, up) and agonistic (aryl group, down) activity and individual enantiomers show opposing biological activities. For example, only (R)-enantiomer of SQ 32,926 14 carries the therapeutically desired antihypertensive effect. For the \( \alpha_{1a} \)-selective adrenoceptor antagonist L-771, 688 15, the (S)-enantiomer is significantly more active than the (R)-enantiomer, and in case of mitotic

Figure 2. Proposed binding site model for DHPM (The receptor sensitive groups are on the ‘left hand side’ of the molecule).
kinesin Eg5 inhibitor monastrol 16, (S)-enantiomer is a more potent inhibitor of Eg5 activity.

Chemical as well as enzymatic resolution strategies have thus far been the methods of choice to obtain optically active DHPMs. Thus, synthesis of enantiomerically pure DHPMs is of considerable current interest and has been a formidable task. Recently, strategies to access enantiopure DHPM derivatives employing catalytic asymmetric synthesis have been reported and reviewed in this chapter.

In our approach, quenching of an anion of DHPM 17 (R³ = Me) with optically pure amino acid chloride resulted in the smooth isolation of diastereomers 18a-b (R³ = Me, R⁴ = CA) (Scheme 5). When metalated N-1 unsubstituted DHPM 17 (R³ = H) was reacted with chiral auxiliary, substitution proceeded at N-1 position. The distinction between the N-1 and N-3 substituted products was based on the ¹H NMR spectral analysis.

Scheme 5. Chemical resolution of enantiomers 19 from racemic DHPMs.
Deacylation of diastereomers 18a-b with lithium aluminium hydride (LAH) in THF led to the corresponding optically pure (S)- and (R)-enantiomer 19a-b (Scheme 5). The absolute configuration at C-4 position of 19 was assigned in analogy with the optical rotation and circular dichroism spectroscopic correlation with the known (S)-and (R)-enantiopure DHPMs.

Thus, in this investigation, inherently racemic DHPMs have been resolved using chemical resolution methodology through chiral auxiliary approach. DHPMs bearing both aryl and alkyl group at C-4 position could be efficiently resolved. Absolute configuration of the enantiomers has been assigned using circular dichroism (CD) spectral correlations.

Chapter 6. Synthesis and calcium channel binding studies of 3,4-dihydropyrimidin-2(1H)-ones bearing organophosphorous unit at N-3

N-3 substituted Biginelli DHPMs such as 14 and 15 are potent calcium channel blocking agents. Suitable molecular modification at N-3 position of the DHPM core thus holds potential for significantly improving the understanding of the structure-activity relationships of DHPMs and for designing efficacious calcium channel blockers. Recently, organophosphorous compounds especially phosphorous heterocycles have received widespread attention due to their ubiquity in biological systems and their potential to serve as novel pharmaceuticals. Among DHPMs, no work has yet been reported on the synthesis of DHPMs bearing an organophosphorous unit. Therefore, we have synthesized a series of DHPM derivatives bearing a dianaminophosphinyl, phosphonate as well as phosphorous heterocyclic unit at the N-3 position and have explored calcium channel binding activity of these compounds.

For systematic presentation of these results, the chapter is divided into the following two sections:

(i) Synthesis of DHPMs bearing organophosphorous or a phosphorous heterocyclic unit at N-3

(ii) Calcium channel binding studies of DHPMs bearing organophosphorous or a phosphorous heterocyclic unit at N-3

We envisaged that owing to more basic character of the N-3 of DHPMs, it should react with POCl₃ to yield a reactive dichlorophosphinyl derivative 21 (Scheme 6), a common intermediate for reaction with various nucleophiles. Upon treating 21 with different nucleophiles (ammonia, amines, ethanol, diamines and aminoaicloahols) in THF at room temperature, 22 (Y = NH₂) or 23 (X = O, NH), bearing appropriate
organophosphorous group at N-3 position have been obtained in synthetically useful manner (60-95%).

**Scheme 6.** Synthesis of N-3 substituted DHPM derivatives.

Representative compounds were compared against nifedipine 24 for their ability to relax a membrane-depolarization-induced contraction, which is almost exclusively dependent on the influx of extracellular calcium. The novel compounds and 24 were tested across a concentration range of 1 µm to 30 mM. 24 completely relaxed the KCl-induced contraction with an IC\textsubscript{50} value approximately 0.01 µM against 110 mM KCl. In contrast these compounds maximally relaxed the KCl-induced contractions by only 25% with the relaxation only significant at 10 µM.

Thus, a highly efficient method for the synthesis of N-3 substituted DHPM derivatives bearing a diaminophosphinyl, phosphonate as well as phosphorous containing heterocycles at N-3 position has been developed. Reactions depicted high regioselectivity and furnished products, substituted exclusively at N-3 position. Representative compounds have also been screened for their calcium channel binding properties. It has been found that phosphorous containing N-3 substituted DHPMs are less active in relaxing membrane depolarization induced contraction of vascular smooth muscle, as compared to nifedipine.
Chapter 7. Synthesis of N1,N3-diacyl-3,4-dihydropyrimidin-2(1H)-ones. Acyl group transfer to amines: Synthesis of primary, secondary and tertiary amides

The regioselective N-3 acylation of DHPMs is of considerable importance, since most of the pharmacologically attractive DHPM derivatives are N-3 acylated analogues. Although direct N-3 acylation of C-4 aryl-substituted DHPM derivatives has been achieved, the reports on the direct synthesis of N1,N3-diacyl DHPM derivatives are only scanty. We developed synthetic strategy to obtain exclusively or mainly 26 avoiding the formation of competitive 27.

Readily available DHPM 25 was transformed to N1,N3-diacyl-3,4-dihydropyrimidin-2(1H)-ones 26, through reaction with an anhydride or acid chloride at low temperature (Scheme 7). Corresponding N-3 acylated derivative 27 was also formed in minor amount along with 26.

Scheme 7. Synthesis of N1,N3-diacylated DHPM derivatives 26.

We reasoned, due to the inbuilt difference in basicity of N-1 (enaminoester nitrogen) and comparatively electron rich N-3 site of N1,N3-diacyl derivative 26, the acyl group at N-1 is expected to be more electrophilic and prone to acyl group transfer to nucleophilic sites. For capturing the released acyl group from the N1,N3-diacyl derivatives 26, during in situ conversion to corresponding monoacyl N-3 derivative 27 (Scheme 7), we treated 26 with ammonia gas and obtain primary amides. Reaction of 26 with primary amines 28 in THF at room temperature for 0.5 h furnished the corresponding secondary amides 29 in 60-96 % yield. The primary amines used include arylamines (aniline, 2-methoxyaniline and 2-methylaniline), alkylamines 28 (R^2 = butyl, n-butyl, sec-butyl and n-heptyl; R^3 = H) and biogenic amines such as β-phenethylamine, homoveratryamine, tryptamine and methyl tryptophanate (racemic as well as enantiomers). When 26 was
reacted with secondary amines such as diethylamine, morpholine, piperidine, N-methyl homoveratrylamine and N-methyl aniline in THF at room temperature for 0.5 h tertiary amides 29 (Scheme 8) were obtained in 70-93%.

![Scheme 8](image)

**Scheme 8.** Synthesis of primary, secondary and tertiary amides 27 ($R_2^3NCOR_1^1$) from ammonia, primary and secondary amines.

Thus, in this investigation, we have demonstrated an efficient method for the preparation of primary, secondary and tertiary amides by reacting N1,N3-diacyl DHPMs with ammonia, primary and secondary amines, respectively. The fact that the transformation is accomplished under neutral reaction conditions, acylation of acid or base sensitive amines can also be performed. Further, the yields are high and isolation of the products is straightforward.