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*

### 3.1 Introduction

In the previous chapter (Chapter 2), a number of synthetic methods for DHPMs, a heterocyclic system of remarkable pharmacological profile has been compiled and pre-eminence of appropriately functionalized DHPMs in the area of calcium channel modulators has been established. In general, DHPMs and their derivatives have been found to possess equivalent or even superior calcium channel binding properties than the traditional DHP based drugs. This has fuelled considerable interest in creating diversity in these molecules to search for better calcium channel modulators. In addition, these molecules also act as mitotic kinesin inhibitors, α1a-adrenergic receptor antagonists, hepatitis B virus replication inhibitors and depict a variety of other biological effects.

The DHPM core constitutes a key part of polycyclic guanidine containing marine alkaloids such as batzelladine A. DHPMs also feature in some very active natural products such as dehydrocrambine A and Sch 575948 which are potent inhibitors of HIV glycoprotein gp120-CD4 receptor interaction. Consequent to their diverse pharmacological profile, synthetic investigations on DHPMs have received extensive attention by both synthetic organic chemists as well as medicinal chemists.

Oxidation of DHPM core is an important transformation as it conveniently results in the formation of 1,2-dihydropyrimidin-2(1H)-ones, which also possess diverse pharmacological profile. For example, pyrimidines such as MKC-422, a HEPT (1-[(2-hydroxyethoxy) methyl]-6-(phenylthio)thymine) analogue among others is in clinical trials as anti-HIV drug. 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 DHPs which undergo a facile *in vitro* and *in vivo* oxidation, a key step in the initial metabolism of DHP-based drugs, structurally similar DHPMs are resistant to

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Φ 3,4-dihydropyrimidin-2(1H)-ones
similar oxidation. Additionally, the methyl group at C-6 position is also sensitive to oxidizing agents, which makes the selective oxidation of the dihydropyrimidinone ring troublesome. Oxidation of 2 ($R^1 = \text{Ph}; R^2 = \text{Et}; R^3 = \text{Me}; R^4 = \text{H, Me}$) with $\text{SeO}_2$ (Scheme 1) in refluxing dioxane does not exclusively furnish 3 ($R^1 = \text{Ph}; R^2 = \text{Et}; R^3 = \text{Me}; R^4 = \text{H, Me}$), but also affords carboxylic acid 3 ($R^1 = \text{Ph}; R^2 = \text{Et}; R^3 = \text{COOH}; R^4 = \text{H, Me}$), along with aldehyde 3 ($R^1 = \text{Ph}; R^2 = \text{Et}; R^3 = \text{CHO}; R^4 = \text{H, Me}$). Overall transformation of 2 into 3 has been envisioned to be proceeding through initial dehydrogenation of the dihydropyrimidinone ring, which in turn activates the methyl group at C-6, thus facilitating oxidation of the methyl group to aldehyde or carboxyl group. Treatment of dihydropyrimidinones lacking a methyl group at C-6 position, such as 2 ($R^1 = \text{Ph}; R^2 = \text{Et}; R^3 = \text{H, Ph}; R^4 = \text{Me}$) with $\text{PCl}_5$ in $\text{POCl}_3$ (Scheme 1) resulted in the formation of the dehydrogenated products 3 ($R^1 = \text{Ph}; R^2 = \text{Et}; R^3 = \text{H, Ph}; R^4 = \text{Me}$) along with appropriate 2-chloropyrimidines 4 ($R^1 = \text{Ph}; R^2 = \text{Et}; R^3 = \text{H, Ph}$) which were also formed from 3 ($R^1 = \text{Ph}; R^2 = \text{Et}; R^3 = \text{H, Ph}; R^4 = \text{Me}$) by a N-1 dealkylation followed by chlorination sequence.  

$$\begin{align*}
\text{R}^2 \text{O} & \quad\quad \text{R}^1 \\
\text{O} & \quad\quad \text{R}^3 \\
\text{Cl} & \quad\quad \text{N} \\
\end{align*}$$

$$\begin{align*}
\text{R}^2 \text{O} & \quad\quad \text{R}^1 \\
\text{O} & \quad\quad \text{R}^3 \\
\text{N} & \quad\quad \text{R}^4 \\
\end{align*}$$

$$\begin{align*}
\text{R}^2 \text{O} & \quad\quad \text{R}^1 \\
\text{O} & \quad\quad \text{R}^3 \\
\text{N} & \quad\quad \text{R}^4 \\
\end{align*}$$

![Scheme 1](image)

Dehydrogenation of Biginelli compound 5 ($R^1 = \text{Ph}, R^2 = \text{CN}$) (Scheme 2) has been achieved using palladium on charcoal. The reaction was conveniently carried out in diphenyl ether at 230°C. However, DHPM esters of the type 2 ($R^1 = \text{Me}, R^2 = \text{R}^3 = \text{alkyl}, R^4 = \text{H}$) could not be dehydrogenated by this method. C-4 unsubstituted DHPM 5 ($R^1 = \text{H}, R^2 = \text{COOEt}$) has been dehydrogenated using bromine in acetic acid to obtain the corresponding 1,2-dihydro derivative 6 ($R^1 = \text{H}, R^2 = \text{COOEt}$).

$$\begin{align*}
\text{R}^2 \text{R}^1 \text{H} & \quad\quad \text{Pd/C} \\
\text{Me} & \quad\quad \text{Me} \\
\text{NH} & \quad\quad \text{NH} \\
\text{CO} & \quad\quad \text{CO} \\
\text{R}^2 \text{R}^1 \text{H} & \quad\quad \text{Pd/C} \\
\text{Et} & \quad\quad \text{Et} \\
\text{O} & \quad\quad \text{O} \\
\text{NH} & \quad\quad \text{NH} \\
\text{CO} & \quad\quad \text{CO} \\
\end{align*}$$

![Scheme 2](image)

A number of different oxidizing agents such as $\text{NaNO}_2$ in acetic acid, $\text{KMnO}_4$, $\text{MnO}_2$, DDQ, Chloranil, $\text{FeCl}_3$, $\text{RuCl}_3/O_2$ in $\text{AcOH}$, $\text{Br}_2$, and sulfur were...
inefficient for the conversion of DHPMs to corresponding pyrimidinone derivatives. Use of Eynde’s procedure to obtain pyrimidinones was not only multistep, but also furnished the desired products in moderate yield. Reagents such as CuCl₂/TBHP/K₂CO₃ and Jones reagent have also been employed for the oxidation of DHPMs. In these protocols, the desired dehydrogenation was attended by dealkylation of secondary alkyl or benzyl group at C-4 of DHPM. Using a combination of hydrated Co(NO₃)₂/K₂S₂O₈ in aqueous acetonitrile, the formation of only C-6 dealkylated product (R¹ = Ar; R² = Et, Me; R³ = H; R⁴ = H, Me) was observed. Use of ceric ammonium nitrate along with NaHCO₃ in acetone under argon atmosphere at low temperature led to regioselective oxidation of (R¹ = Ar; R² = Et, Me; R³ = Me; R⁴ = H, Me) to (R¹ = Ar; R² = Et, Me; R³ = Me; R⁴ = H, Me) (Scheme 1). Unusual product (R¹ = Ph) was obtained when the reaction was carried out at higher temperature in acetic acid.

The aryl group at the C-4 position of DHPMs was essential for the oxidative-dealkylation as C-4 alkyl DHPMs under similar set of reaction conditions afforded unusual product (R¹ = H) (Scheme 2).

Kappe et al. have employed conc. HNO₃ for the regioselective oxidative dehydrogenation of DHPMs to corresponding pyrimidinones. A combination of tert-butylhydroperoxide and (diacetoxyiodo)benzene afforded a clean and efficient oxidative dehydrogenation of DHPMs. Similarly, combination of ultrasound and heat has been used for the oxidation of DHPMs to their corresponding pyrimidin-2(1H)-ones by using potassium peroxydisulfate in aqueous acetonitrile. In another reaction protocol, photocatalytic oxidation has been used for the regioselective oxidation of DHPMs using a TiO₂/O₂ system under UV irradiation. Likewise, oxidation could also be achieved by using nitrosonium tetrafluoroborate.

A useful outcome of the oxidation of Biginelli DHPMs is the use of the resultant 1,2-dihydropyrimidin-2(1H)-ones in obtaining C-2 substituted multifunctionalized pyrimidines 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 (non-nucleosidic inhibitor of hepatitis B virus), HAP-1, N-cyanooiminopyrimidine (potent antimalarial agent), hypocholesterolemic agent Rosuvastatin (Ca²⁺ salt) (an HMG-CoA reductase inhibitor) and potent anticancer drug Gleevec (a tyrosine kinase inhibitor).
Methods have been reported to convert Biginelli DHPMs to C-2 substituted pyrimidines efficiently. For example, pyrimidines bearing sulfone moiety at C-2, were obtained through treatment of 2-thioxopyrimidines with Oxone® and were reacted with nucleophiles\(^{38}\) to obtain 2-substituted pyrimidines. Alternatively, palladium(0)-catalyzed copper(I)-mediated coupling of boronic acids with cyclic thioamides has also been used.\(^{39}\) Kang \textit{et al.} have reported two step procedure for the efficient conversion of Biginelli DHPMs to pyrimidines via PyBroP-mediated coupling.\(^{40}\) Similarly, pyrimidines have also been synthesized via palladium catalysed Suzuki [Boronic acid, Pd(OAc)\(_2\), PPh\(_3\), satd. aqueous Na\(_2\)CO\(_3\)-dioxane (4:6 v/v), 110°C]/Sonogashira [Pd(PPh\(_3\))\(_4\), CH\(_3\)CN, Et\(_3\)N, CuI, reflux] cross-coupling and Eschenmoser sulphide contraction (K\(_2\)CO\(_3\), PPh\(_3\), acetone, 80°C) coupling reactions.\(^{41,42}\) However, the development of a general protocol for obtaining C-2 elaborated pyrimidines through regioselective C-2 functionalization of DHPMs is of considerable importance, since following such a route, using a pre-functionalized DHPM and oxidising it at N3-C4 position selectively, the resulting pyrimidine would retain the substitutent pattern around the pyrimidine core. Such derivatives could also be evaluated for biological effects such as cytostatic and anti-tuberculosis activities owing to structural similarities with active heterocyclic molecules. To achieve this aim, we have explored the reaction sequence involving initial selective oxidation of DHPMs 2 to corresponding 1,2-dihydropyrimidin-2(1H)-ones 3, followed by
C-2 halogenation and finally coupling with various nucleophiles to afford tetrasubstituted pyrimidine derivatives 14 (Scheme 3).

Scheme 3

The results of this investigation are presented in the following sections:

3.2 Synthesis of tetrasubstituted 1,2-dihydropyrimidin-2(1H)-ones

3.2.1 Synthesis of C-4 substituted and C-4 unsubstituted DHPM precursors

3.2.2 Oxidation of 3,4-dihydropyrimidin-2(1H)-ones using pyridinium chlorochromate

3.3 C-2 functionalization of 1,2-dihydropyrimidin-2(1H)-ones

3.4 Cytostatic and anti-tuberculosis activities of C-2 functionalized tetrasubstituted pyrimidines

3.2 Synthesis of tetrasubstituted 1,2-dihydropyrimidin-2(1H)-ones

Pyridinium chlorochromate (PCC) has been widely employed for selective transformation of primary alcohols to aldehydes.\(^4^3\) The use of PCC is not limited to oxidation reaction alone, but several rearrangements and useful conversions have also been mediated by PCC, which makes it a versatile reagent in organic synthesis.\(^4^3\) PCC has been found to be remarkably efficient in oxidising Hantzsch DHPs,\(^2^6\) which led us to employ PCC for the oxidation of structurally similar DHPMs as detailed in the following subsections.

3.2.1 Synthesis of C-4 substituted and C-4 unsubstituted DHPM precursors

DHPM derivatives 2 were obtained through the traditional three-component Biginelli condensation reaction of the corresponding aldehydes with ethyl acetoacetate and urea (Scheme 4).\(^7^b\) Considerable variation in the nature of substituents (R\(^1\), R\(^2\), R\(^3\) and R\(^4\)) was introduced so as to understand the scope and limitations of the PCC mediated oxidation\(^\ast\) of DHPMs. Thus, DHPMs 2a-l (Scheme 4) were synthesized in good to

\(^\ast\) oxidative-dehydrogenation
excellent yield from the corresponding aromatic/aliphatic aldehydes, ethyl acetoacetate and urea/N-methyl urea by running reactions in an anhydrous solvent and catalysed by HCl and their identity could be easily established by comparison (TLC, m.p. and $^1$H NMR spectrum) with authentic samples. The key $^1$H NMR chemical shifts and the yields of the products 2a-l are presented in Table 1.

For the synthesis of C-4 unsubstituted DHPMs 2m-n, 1,3-oxazinane 15- a formaldehyde equivalent, was employed in place of formaldehyde (Scheme 4). Thus, an equimolar solution of 15, ethyl acetoacetate 16 and urea 17 (R$^4$ = H) in anhydrous acetonitrile containing trifluoroacetic acid was refluxed till completion (TLC). Extractive workup and purification of the residue furnished a compound at Rf: 0.6 (ethyl acetate:hexane/80:20) (TLC), which melted at 256-257°C. The $^1$H NMR (DMSO-d$_6$) spectrum (Figure 1) of this product showed signals at δ 1.15 (t, 3H, J 7.2 Hz, CH$_3$), 2.12 (s, 3H, C$_6$-CH$_3$), 3.86 (s, 2H, CH$_2$), 4.02 (q, 2H, J 7.2 Hz, CH$_2$), 6.97 (br, 1H, D$_2$O exchangeable, NH), 8.80 (br, 1H, D$_2$O exchangeable, NH). The characteristic features of this spectrum included a 2H singlet at δ 3.86, assigned to CH$_2$ (C-4) and a deuterium exchangeable broad singlet at δ 6.97, due to N3-H. The peaks in its $^{13}$C NMR (DMSO-d$_6$)
Table 1: Yields and $^1$H NMR (CDCl$_3$) chemical shifts of the DHPM 2a-l (Scheme 4).

<table>
<thead>
<tr>
<th>DHPM</th>
<th>C6-R$^1$</th>
<th>N1-R$^2$</th>
<th>N3-H</th>
<th>C4-H</th>
<th>C5-R$^2$</th>
<th>Yield (%)</th>
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<tr>
<td>2a</td>
<td>2.35 (s, 3H)</td>
<td>7.80 (br, 1H)</td>
<td>5.59 (br, 1H)</td>
<td>5.39 (d, 1H)</td>
<td>1.16 (t, 3H) 4.08 (m, 2H)</td>
<td>88</td>
</tr>
<tr>
<td>2b</td>
<td>2.51 (s, 3H)</td>
<td>-</td>
<td>5.70 (br, 1H)</td>
<td>5.38 (d, 1H)</td>
<td>1.18 (t, 3H) 4.10 (q, 2H)</td>
<td>92</td>
</tr>
<tr>
<td>2c</td>
<td>2.32 (s, 3H)</td>
<td>8.50 (br, 1H)</td>
<td>5.98 (br, 1H)</td>
<td>5.38 (d, 1H)</td>
<td>3.61 (s, 3H)</td>
<td>94</td>
</tr>
<tr>
<td>2d</td>
<td>0.98 (t, 3H) 1.63 (m, 2H) 2.65 (m, 2H)</td>
<td>8.62 (br, 1H)</td>
<td>6.36 (br, 1H)</td>
<td>5.37 (d, 1H)</td>
<td>1.15 (t, 3H) 4.05 (m, 2H)</td>
<td>85</td>
</tr>
<tr>
<td>2e</td>
<td>2.32 (s, 3H)</td>
<td>8.32 (br, 1H)</td>
<td>5.85 (br, 1H)</td>
<td>5.34 (d, 1H)</td>
<td>1.16 (t, 3H) 4.07 (m, 2H)</td>
<td>84</td>
</tr>
<tr>
<td>2f</td>
<td>2.35 (s, 3H)</td>
<td>7.95 (br, 1H)</td>
<td>5.70 (br, 1H)</td>
<td>5.36 (d, 1H)</td>
<td>1.20 (t, 3H) 4.01 (m, 2H)</td>
<td>82</td>
</tr>
<tr>
<td>2g</td>
<td>2.53 (s, 3H)</td>
<td>-</td>
<td>6.12 (br, 1H)</td>
<td>5.50 (d, 1H)</td>
<td>1.20 (t, 3H) 4.12 (m, 2H)</td>
<td>83</td>
</tr>
<tr>
<td>2h</td>
<td>2.37 (s, 3H)</td>
<td>8.10 (br, 1H)</td>
<td>6.10 (br, 1H)</td>
<td>5.51 (d, 1H)</td>
<td>1.07 (d, 3H) 1.23 (d, 3H) 4.96 (m, 1H)</td>
<td>83</td>
</tr>
<tr>
<td>2i</td>
<td>2.32 (s, 3H)</td>
<td>8.43 (br, 1H)</td>
<td>6.12 (br, 1H)</td>
<td>4.39 (m, 1H)</td>
<td>1.28 (t, 3H) 4.17 (m, 2H)</td>
<td>87</td>
</tr>
<tr>
<td>2j</td>
<td>2.27 (s, 3H)</td>
<td>8.48 (br, 1H)</td>
<td>6.16 (br, 1H)</td>
<td>4.37 (m, 1H)</td>
<td>3.70 (s, 3H)</td>
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<tr>
<td>2k</td>
<td>-</td>
<td>6.91 (br, 1H)</td>
<td>5.78 (br, 1H)</td>
<td>4.51 (m, 1H)</td>
<td>0.90 (t, 3H) 3.92 (q, 2H)</td>
<td>82</td>
</tr>
<tr>
<td>2l</td>
<td>2.28 (s, 3H)</td>
<td>7.15 (br, 1H)</td>
<td>5.38 (br, 1H)</td>
<td>4.30 (m, 1H)</td>
<td>0.88 (t, 3H) 4.18 (m, 2H)</td>
<td>80</td>
</tr>
</tbody>
</table>

Spectrum (Figure 1) appeared at $\delta$ 14.3, 17.4, 40.6, 59.1, 94.4, 148.7, 152.8 and 165.4 and corroborated well with its $^1$H NMR spectral assignment. The peak at m/z 185 (M$^+$+1) in its EIMS spectrum corresponding to the molecular formula C$_8$H$_{12}$N$_2$O$_3$ and correct microanalytical analysis (vide experimental) confirmed the structure, 5-ethoxycarbonyl-6-methyl-3,4-dihydropyrimidin-2(1H)-one 2m assigned to this compound, isolated in 85% yield.

Likewise, 5-ethoxycarbonyl-1,6-dimethyl-3,4-dihydropyrimidin-2(1H)-one 2n was isolated in 80% yield, when N-methyl urea 17 (R$^4$ = Me) was used along with 15 and 16 in the acid (TFA) catalyzed condensation reaction. The structure of 2n was established from the spectral data and correct microanalytical analysis (vide experimental).
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.

Figure 1. $^1$H NMR (300 MHz, DMSO-$d_6$) spectrum and $^{13}$C NMR (75 MHz, DMSO-$d_6$) assignments of 2m.

3.2.2 Oxidation of 3,4-dihydropyrimidin-2(1H)-ones using pyridinium chlorochromate

After synthesizing DHPMs 2a-n (Scheme 4), experiments were run in order to determine the suitable conditions for the oxidation reaction (Scheme 5). Bearing in mind the ester functionality in 2, initially we employed neutral reaction conditions using PCC adsorbed on a solid support analogously to the reported oxidation of DHPs.\(^{26}\) Such supported systems were readily prepared by adding a solid support [neutral alumina, silica gel (60-120, 100-200 and 230-400 mesh), or montmorillonite K-10 clay] to a solution of PCC in acetone and by subsequent removal of the solvent under reduced pressure. The orange powders so obtained were dried at 100°C and could be stored for several weeks without any decrease in their efficiency as adjudged in the oxidation experiments. However, when solid supported PCC was used in oxidative dehydrogenation, the reactions neither proceeded to completion (TLC), even after prolonged (48 h) stirring at room temperature nor did the process offer any other operational advantage, thus PCC was used as such without adsorbing on any solid support.

Scheme 5. Oxidative dehydrogenation of 2 to 3 using PCC.
Initially, the oxidation of 5-ethoxycarbonyl-6-methyl-4-phenyl-3,4-dihydropyrimidin-2(1H)-one 2a (R¹ = Ph, R² = Et, R³ = Me, R⁴ = H) with equimolar quantity of PCC in DCM was performed, however, the reaction did not proceed to completion. As the literature reports suggest,²⁸,⁴⁵ in similar oxidations, up to 5.0 molar excess of the oxidant has been employed to drive the reaction to completion. Therefore, for optimizing the reaction conditions for obtaining the products in a synthetically useful manner, when excess of PCC (3.0 equiv.) was employed in the oxidation of 2a using anhydrous DCM as solvent, under neutral reaction conditions, the reaction proceeded to completion (TLC) during 26 h, upon stirring at room temperature. The reaction mixture was filtered over celite bed for removing any suspended material and solvent removed under reduced pressure. The resulting residue was chromatographed to obtain a product at Rf: 0.4 (ethyl acetate) (TLC).

¹H NMR (CDCl₃) spectrum (Figure 2) of this product depicted signals at δ 0.93 (t, 3H, J 7.1 Hz, CH₃), 2.62 (s, 3H, C₆-CH₃), 4.05 (q, 2H, J 7.1 Hz, CH₂), 7.46 (m, 3H, ArH), 7.59 (m, 2H, ArH), 13.67 (br, 1H, D₂O exchangeable, NH). The salient features of this spectrum included the disappearance of the 1H doublet at δ 5.39 corresponding to C₄-H as well as N₃-H (δ 5.59, br) of 2a. The ¹³C NMR (CDCl₃) spectrum (Figure 2) showed signals at δ 13.4, 61.6, 111.5, 128.0, 128.4, 130.8, 158.3 and 166.1.* In its EIMS♥, parent ion peak appeared at m/z 281 (M⁺+23), corresponding to the molecular formula C₁₄H₁₄N₂O₃+Na of the expected product. Based on the spectral data as well as correct microanalytical analysis (*vide experimental*), the structure, 5-ethoxycarbonyl-6-methyl-4-phenyl-1,2-dihydropyrimidin-2(1H)-one 3a (Scheme 5) has been assigned to this compound. It was obtained in 63% isolated yield (Table 2). Any further increase in the equiv. of PCC (up to 5.0 equiv.) did not improve the yield and/or reduce the reaction time.

A comparison of the ¹H NMR spectra of 3a with that of the precursor DHPM 2a depicted downfield shifts in the resonances of the N1-H and C6-Me protons. This may be attributed to the activation of 3a as shown in Scheme 6.

* The variability in the number of peaks in the ¹³C NMR spectrum may be attributed to the presence of magnetically equivalent carbons in the molecule.
♥ Mass equivalent to sodium atom was invariably found to be added to the mass corresponding to parent ion during recording of EIMS.
Regioselective oxidation of 3,4-dihydropyrimidin-2(1H)-ones to 1,2-dihydropyrimidin-2(1H)-ones and C-2 functionalization.

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Figure 2. $^1$H NMR (400 MHz, CDCl$_3$) spectrum and $^{13}$C NMR (100 MHz, CDCl$_3$) assignments of 3a.

Scheme 6. Comparison of resonances of N1-H and C6-Me protons in 2a and 3a.

After optimising the reaction conditions (PCC, 3.0 equiv./DCM/r.t.) for the oxidative dehydrogenation of DHPMs 2, we have further investigated the scope of oxidation of DHPMs 2 by PCC under neutral reaction conditions, by using a number of DHPMs substituted with different substituents around the DHPM core (N-1, C-4, C-5 and C-6) (Table 2).

Oxidation of 5-ethoxycarbonyl-1,6-dimethyl-4-phenyl-3,4-dihydropyrimidin-2(1H)-one 2b was accomplished by using little excess (4.0 equiv.) of PCC in anhydrous DCM (Scheme 5). A product at Rf: 0.5 (ethyl acetate) (TLC), which showed a parent ion peak at $m/z$ 295 (M$^+$+23) in its EIMS spectrum, corresponding to the molecular formula C$_{15}$H$_{16}$N$_2$O$_3$+Na was isolated. The $^1$H NMR (CDCl$_3$) spectrum (Figure 3) of this product showed signals at $\delta$ 0.87 (t, 3H, J 6.9 Hz, CH$_3$), 2.55 (s, 3H, C6-CH$_3$), 3.65 (s, 3H, N1-CH$_3$), 4.00 (q, 2H, J 7.2 Hz, CH$_2$), 7.42 (m, 3H, ArH), 7.58 (m, 2H, ArH). The absence of
Table 2. Synthesis of 1,2-dihydropyrimidin-2(1H)-ones 3 through PCC oxidation of 2 (Scheme 5).

<table>
<thead>
<tr>
<th>Entry</th>
<th>2</th>
<th>3</th>
<th>Substituent (ring position)</th>
<th>Reaction time (h)</th>
<th>Isolated yieldsa,b (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>2a</td>
<td>3a</td>
<td>Ph Et Me H</td>
<td>26</td>
<td>63</td>
</tr>
<tr>
<td>2.</td>
<td>2b</td>
<td>3b</td>
<td>Ph Et Me Me</td>
<td>16</td>
<td>60b</td>
</tr>
<tr>
<td>3.</td>
<td>2c</td>
<td>3c</td>
<td>Ph Me Me H</td>
<td>18</td>
<td>71</td>
</tr>
<tr>
<td>4.</td>
<td>2d</td>
<td>3d</td>
<td>Ph Et n-Pr H</td>
<td>23</td>
<td>70</td>
</tr>
<tr>
<td>5.</td>
<td>2e</td>
<td>3e</td>
<td>4-MeOC₆H₄ Et Me H</td>
<td>25</td>
<td>60</td>
</tr>
<tr>
<td>6.</td>
<td>2f</td>
<td>3f</td>
<td>3,4,5- (MeO)₃C₆H₂ Et Me H</td>
<td>23</td>
<td>60b</td>
</tr>
<tr>
<td>7.</td>
<td>2g</td>
<td>3g</td>
<td>4-NO₂C₆H₄ Et Me Me</td>
<td>25</td>
<td>60b</td>
</tr>
<tr>
<td>8.</td>
<td>2h</td>
<td>3h</td>
<td>3-NO₂C₆H₄ i-Pr Me H</td>
<td>21</td>
<td>55b</td>
</tr>
<tr>
<td>9.</td>
<td>2i</td>
<td>3i</td>
<td>Me Et Me H</td>
<td>20</td>
<td>69</td>
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<tr>
<td>10.</td>
<td>2j</td>
<td>3j</td>
<td>Me Me Me H</td>
<td>20</td>
<td>65</td>
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<tr>
<td>11.</td>
<td>2k</td>
<td>3k</td>
<td>Me Et Ph H</td>
<td>19</td>
<td>71</td>
</tr>
<tr>
<td>12.</td>
<td>2l</td>
<td>3l</td>
<td>C₅H₁₁ Et Me H</td>
<td>20</td>
<td>62</td>
</tr>
<tr>
<td>13.</td>
<td>2m</td>
<td>3m</td>
<td>H Et Me Me</td>
<td>20</td>
<td>79</td>
</tr>
<tr>
<td>14.</td>
<td>2n</td>
<td>3n</td>
<td>H Et Me Me</td>
<td>18</td>
<td>63</td>
</tr>
</tbody>
</table>

* Based on isolated purified products.

b 4.0 equiv. of PCC, required for complete conversion.

C4-H (δ 5.38) and N3-H (δ 5.70) resonances and slight downfield shift of C6-Me signal (from δ 2.51 to 2.55) compared to the precursor 2b indicated the incorporation of N3-C4 double bond. The peaks in its 13C NMR (CDCl₃) spectrum (Figure 3) appeared at δ 13.2, 17.9, 33.0, 61.7, 111.5, 127.7, 128.1, 130.3, 138.2, 148.2, 155.7, 158.5 and 166.9 and corroborated well with its 1H NMR spectral data. From the spectral and correct microanalytical analysis (vide experimental) structure, 5-ethoxycarbonyl-1,6-dimethyl-4-phenylpyrimidin-2(1H)-one 3b (60%, Table 2) has been assigned to this compound.

Similarly, oxidation of DHPMs 2c and 2d with PCC under similar set of reaction conditions, furnished 5-methoxycarbonyl-6-methyl-4-phenylpyrimidin-2(1H)-one 3c and 5-ethoxycarbonyl-6-propyl-4-phenylpyrimidin-2(1H)-one 3d, respectively in 71% and 70% yield (Table 2) and the structures were assigned on the basis of spectral data and correct microanalytical analysis (vide experimental).
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.

Likewise, oxidation of 5-ethoxycarbonyl-6-methyl-4-(4-methoxyphenyl)-3,4-dihydropyrimidin-2(1H)-one 2e, having electron donating groups on the aryl ring, under similar set of reaction conditions furnished a compound [Rf: 0.3 (ethyl acetate) (TLC)] which showed a peak at m/z 311 (M+23) in its EIMS spectrum, corresponding to the molecular formula C_{15}H_{16}N_{2}O_{4}Na. The $^1$H NMR (CDCl$_3$) spectrum (Figure 4) of this product depicted signals at $\delta$ 1.05 (t, 3H, J 7.2 Hz, CH$_3$), 2.58 (s, 3H, C6-CH$_3$), 3.86 (s, 3H, OCH$_3$), 4.12 (q, 2H, J 7.2 Hz, CH$_2$), 6.94 (d, 2H, J 8.6 Hz, ArH), 7.61 (d, 2H, J 8.4 Hz, ArH), 13.76 (br, 1H, D$_2$O exchangeable, NH). Obviously, the absence of C4-H ($\delta$ 5.34) and N3-H ($\delta$ 5.85) resonances and the downfield shift of C6-Me signal (from $\delta$ 2.32 to 2.58) in the spectrum indicated the incorporation of N3-C4 double bond. The peaks in its $^{13}$C NMR (CDCl$_3$) spectrum (Figure 4) appeared at $\delta$ 13.7, 55.4, 61.6, 100.0, 113.9, 130.1, 141.7, 153.1 and 166.6 and corroborated with the $^1$H NMR spectral data of this compound. From the spectral and correct microanalytical data (vide experimental) structure, 5-ethoxycarbonyl-6-methyl-4-(4-methoxyphenyl)pyrimidin-2(1H)-one 3e (60%, Table 2) has been assigned to this compound. Similarly PCC oxidation of 5-ethoxycarbonyl-6-methyl-4-(3,4,5-trimethoxyphenyl)-3,4-dihydropyrimidin-2(1H)-one 2f, furnished 5-ethoxycarbonyl-6-methyl-4-(3,4,5-trimethoxyphenyl)pyrimidin-2(1H)-one 3f in 60% yield (vide experimental).
Figure 4. $^1$H NMR (400 MHz, CDCl$_3$) spectrum and $^{13}$C NMR (100 MHz, CDCl$_3$) assignments of $3e$.

Similar oxidation of 5-ethoxycarbonyl-1,6-dimethyl-4-(4-nitrophenyl)-3,4-dihydropyrimidin-2(1H)-one $2g$, bearing an electron-withdrawing NO$_2$ group on the C-4 aryl ring using PCC (4.0 equiv.) in anhydrous DCM furnished a white crystalline solid at Rf: 0.5 (ethyl acetate) (TLC), which melted at 128-130$^\circ$C. Its $^1$H NMR spectrum (Figure 5) recorded in CDCl$_3$ depicted signals at $\delta$ 0.93 (t, 3H, $J$ 7.2 Hz, CH$_3$), 2.61 (s, 3H, C6-CH$_3$), 3.69 (s, 3H, N1-CH$_3$), 4.03 (q, 2H, $J$ 7.2 Hz, CH$_2$), 7.75 (d, 2H, $J$ 8.7 Hz, ArH), 8.27 (d, 2H, $J$ 8.7 Hz, ArH). Formation of the double bond between N3-C4 positions was inferred from the absence of C4-H ($\delta$ 5.50) and N3-H ($\delta$ 6.12) signals of $2g$ in the $^1$H NMR spectrum of the product. $^{13}$C NMR spectrum (Figure 5) recorded in CDCl$_3$ displayed

Figure 5. $^1$H NMR (300 MHz, CDCl$_3$) spectrum and $^{13}$C NMR (75 MHz, CDCl$_3$) assignments of $3g$. 
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.

signals at δ 13.4, 18.2, 33.3, 61.9, 111.1, 123.4, 128.8, 144.3, 148.7, 155.2, 160.1, 166.0 and 169.0. Additionally, correct microanalytical analysis (vide experimental) and mass spectrum (m/z 340, M^+23), corresponded to the molecular formula C_{15}H_{15}N_{3}O_{5}+Na and led to the identification of 5-ethoxycarbonyl-1,6-dimethyl-4-(4-nitrophenyl)pyrimidin-2(1H)-one 3g (60%, Table 2).

Likewise, oxidation of 5-isopropoxycarbonyl-6-methyl-4-(3-nitrophenyl)-3,4-dihydropyrimidin-2(1H)-one 2h, under similar reaction conditions, furnished the corresponding 1,2-dihydropyrimidin-2(1H)-one 3h in 55% yield (Table 2) which was adequately characterized using spectral and microanalytical data (vide experimental).

When DHPM having alkyl group appended at the C-4 position, 5-ethoxycarbonyl-4,6-dimethyl-3,4-dihydropyrimidin-2(1H)-one 2i, was reacted with PCC under similar set of reaction conditions (Scheme 5), a single product (m.p. 134-136°C), Rf: 0.2 (ethyl acetate) (TLC) was obtained. The ^1H NMR (CDCl₃) spectrum (Figure 6) of this product showed signals at δ 1.39 (t, 3H, J 7.2 Hz, CH₃), 2.57 (s, 6H, 2×CH₃), 4.36 (q, 2H, J 7.0 Hz, CH₂), 13.58 (br, 1H, D₂O exchangeable, NH). In addition to other signals, an unsplit singlet at δ 2.57 corresponding to C4-Me and disappearance of signal of N3-H (δ 6.12), indicated the incorporation of double bond between N3-C4 positions. The peaks in its ^13C NMR (CDCl₃) spectrum (Figure 6) appeared at δ 14.2, 61.6, 111.3, 158.1 and 165.3 and corroborated well with ^1H NMR spectral data. In its EIMS spectrum, a peak at m/z 219 (M^+23) corresponding to the molecular formula C₉H₁₂N₂O₃+Na and correct microanalytical data (vide experimental) confirmed the structure, 5-ethoxycarbonyl-4,6-

Figure 6. ^1H NMR (400 MHz, CDCl₃) spectrum and ^13C NMR (100 MHz, CDCl₃) assignments of 3i.
dimethylpyrimidin-2(1H)-one 3i (69%, Table 2). Oxidation of 2j to corresponding 5-methoxycarbonyl-4,6-dimethylpyrimidin-2(1H)-one 3j was analogously achieved in 65% yield.

Likewise, the reaction of 5-ethoxycarbonyl-6-phenyl-4-methyl-3,4-dihydropyrimidin-2(1H)-one 2k with PCC depicted similar reaction pattern and furnished a single product, 5-ethoxycarbonyl-6-phenyl-4-methylpyrimidin-2(1H)-one 3k (71%, Table 2). The confirmation of the structure 3k was based on the spectral as well as microanalytical data reported Section 3.5.4.

The effect of increasing the alkyl chain length at C-4 position of DHPM on the oxidation reaction with PCC was also investigated. Reaction of 5-ethoxycarbonyl-6-methyl-4-pentyl-3,4-dihydropyrimidin-2(1H)-one 2l with PCC in anhydrous DCM, at room temperature furnished a compound at Rf: 0.4 (ethyl acetate) (TLC). Its $^1$H NMR (CDCl$_3$) spectrum (Figure 7) depicted signals at $\delta$ 0.94 (t, 3H, $J$ 7.3 Hz, CH$_2$), 1.31 (t, 3H, $J$ 7.1 Hz, CH$_3$), 1.41 (m, 2H, CH$_2$), 1.70 (m, 4H, 2×CH$_2$), 2.66 (s, 3H, C6-CH$_3$), 2.97 (t, 2H, $J$ 7.5 Hz, CH$_2$), 4.29 (q, 2H, $J$ 7.1 Hz, CH$_2$), 13.51 (br, 1H, D$_2$O exchangeable, NH). The absence of a multiplet ($\delta$ 4.30) corresponding to C4-H of 2l and a signal corresponding to N3-H ($\delta$ 5.38) indicated the oxidation of N3-C4 link. $^{13}$C NMR spectrum (Figure 7) recorded in CDCl$_3$ displayed signals at $\delta$ 13.8, 19.2, 22.2, 24.8, 39.0, 62.0, 107.8, 158.3 and 163.6. Additionally, correct microanalytical data (vide experimental) and mass

![Figure 7](image_url)
spectrum (m/z 253, M+1), corresponding to the molecular formula C_{13}H_{20}N_{2}O_{3} and led to the assignment of structure, 5-ethoxycarbonyl-6-methyl-1-pentylpyrimidine-2(1H)-one 3I (62%, Table 2) to this compound.

Likewise, when C-4 unsubstituted 5-ethoxy-6-methyl-3,4-dihydropyrimidin-2(1H)-one 2m was reacted with PCC (3.0 equiv.) in anhydrous DCM, at room temperature under neutral reaction conditions, a product (m.p. 210-211°C) was obtained at Rf: 0.5 (ethyl acetate:hexane/80:20) (TLC). In its 1H NMR (DMSO-d6) spectrum (Figure 8), signals appeared at δ 1.28 (t, 3H, J 7.12 Hz, CH3), 2.54 (s, 3H, C6-CH3), 4.23 (q, 2H, J 7.0 Hz, CH2), 8.74 (s, 1H, CH), 12.46 (br, 1H, D2O exchangeable, NH). In addition to other signals, an unsplit 1H singlet at δ 8.74 was assigned to the C4-H. Additionally, the absence of N3-H resonance (δ 6.97) of 2m indicated the formation of C4-N3 double bond. The peaks in its 13C NMR (DMSO-d6) spectrum (Figure 8) appeared at δ 14.0, 60.4, 106.0, 155.5 and 163.4 and corroborated well with 1H NMR spectral data. The peak at m/z 183 (M+1) in its EIMS spectrum corresponded to the expected molecular formula C_{8}H_{10}N_{2}O_{3}, which was also supported by correct microanalytical analysis (vide experimental). The structure, 5-ethoxycarbonyl-6-methyl-1,2-dihydropyrimidin-2(1H)-one 3m was assigned to this compound, isolated in 79% yield (Table 2). It was found that oxidation of C-4 unsubstituted DHPM 2m, was completed in comparatively shorter time (20 h, Table 2) to furnish 1,2-dihydropyrimidin-2(1H)-one 3m, compared to the C-4 substituted DHPM 2a (26 h).

**Figure 8.** 1H NMR (400 MHz, DMSO-d6) spectrum and 13C NMR (100 MHz, DMSO-d6) assignments of 3m.
Likewise, oxidation reaction of 5-ethoxycarbonyl-1,6-dimethyl-3,4-dihydropyrimidin-2(1H)-one 2n with PCC using optimized reaction conditions, furnished the corresponding product, 5-ethoxycarbonyl-1,6-dimethylpyrimidin-2(1H)-one 3n in 63% yield (Table 2). The structure of 3n was established from the spectral and microanalytical data (vide experimental).

The $^1$H NMR spectra of all the products (3a-n) showed a characteristic broad signal due to the enolizable –OH or N1-H proton around $\delta$ 12-13 ppm. Further, the identity of C-4, C-6 and substituted aromatic carbons was difficult to establish in the $^{13}$C NMR spectrum due to possible tautomerisation of N1-H to N3-H in solution as shown in structures 18 and 19 (Scheme 7). N1-alkyl substituted 1,2-dihydropyrimidin-2(1H)-ones do not exhibit such tautomerisation in solution and their $^{13}$C NMR spectrum displayed peaks corresponding to the number of carbon atoms.

![Scheme 7. Possible tautomerism in 3a-n.](image)

3.2.3 Proposed mechanism of reaction

The mechanism of dehydrogenation mediated by PCC is not very certain as the precise nature of the reduced chromium product has not been investigated, presumably it is chromium dioxide (Cr$^{4+}$) or some other intermediate formed from chromium dioxide and pyridinium chloride. The transformation of DHPMs 2 to 1,2-dihydropyrimidin-2(1H)-ones 3 might involve two electron-transfer processes as proposed in Scheme 8. The dehydrogenation mechanism may initially involve single electron-transfer process and abstraction of hydrogen from C-4 of DHPM 2 to form the radical 20, which further gets converted into carbocation 21 through single electron-transfer process. Finally, abstraction of N3-H from carbocation 19 might lead to the formation of 1,2-dihydropyrimidinone 3.

![Scheme 8. Proposed mechanism for the formation of 3.](image)
3.2.4 Conclusions

Thus, these few reactions demonstrate a single step facile transformation of 3,4-dihydropyrimidin-2(1H)-ones 2a-n to corresponding 1,2-dihydropyrimidin-2(1H)-ones 3a-n, in a highly regioselective manner. Compared to the literature methods, no side product was obtained and the PCC mediated oxidation protocol was tolerant to substitution variation around the DHPM core (C-4, C-5 and C-6). The method has operational advantage as the reaction was facile and the isolation/purification of the products was easy.

3.3 C-2 functionalization of 1,2-dihydropyrimidin-2(1H)-ones

The 2-aminopyrimidine structural subunit is present in a number of both natural products as well as synthetic compounds with interesting biological properties. This structural motif, containing a guanidine moiety, has been extensively examined as a drug-like scaffold. Of particular interest are derivatives possessing an aryl ring at the C-4 position and an electron-withdrawing substituent such as an ester or amide group at C-5 position. Several 2-aminopyrimidines of this type show interesting biological activities, for example as inhibitors of rho-associated protein kinase, glycogen synthase kinase 3 (GSK3), and of N-type calcium channels. Notably, the 2-amino-4-arylpurimidone subunit is also found in important drugs such as the hypocholesterolemic agents Rosuvastatin 12 (an HMG-CoA reductase inhibitor) and the potent anticancer drug Gleevec 13 (a tyrosine kinase inhibitor).

In general, 2-aminopyrimidine heterocycles are often constructed by condensation reactions of enones with suitable guanidine or related nitrogen containing building blocks. This approach, however, is of restricted use because of the limited availability of substituted guanidines. Alternatively, the 2-amino group on the pyrimidine can be introduced by displacement of a good leaving group at the C-2 position with a primary or secondary amine. This approach has been adopted by us for the conversion of Biginelli DHPMs to multifunctionalized pyrimidines via oxidation, halogenation followed by reactions with various N and O nucleophiles and the results are presented in the following sections. The overall synthetic protocol is outlined in Scheme 8.

3.3.1 Synthesis of 1,2-dihydropyrimidin-2(1H)-one precursor

The precursor 1,2-dihydropyrimidin-2(1H)-ones 3a-c were obtained as described (Section 3.2.2) through PCC mediated oxidation of 3,4-dihydropyrimidin-2(1H)-ones 2 (Scheme 5) (Table 2).
3.3.2 Synthesis of 5-alkoxycarbonyl-6-methyl-2-chloropyrimidines

In order to achieve the objective of C-2 functionalization to form tetrasubstituted pyrimidines 14 (Scheme 3) from 1,2-dihydropyrimidin-2(1H)-ones 3, introduction of a good leaving group at the C-2 position of pyrimidines was needed which could be efficiently displaced by nucleophiles. Keeping in mind the low dissociation energy of carbon-halogen bond, chlorination of 3 using phosphorous oxychloride (POCl$_3$) was attempted. Thus, 5-ethoxycarbonyl-6-methyl-4-phenylpyrimidin-2(1H)-one 3a (Scheme 9) was taken in phosphorous oxychloride (POCl$_3$), and reaction mixture was refluxed at 105°C until completion (TLC, 30 minutes). After the complete consumption of the starting compound, the excess POCl$_3$ was removed under reduced pressure. After purification with column chromatography, a viscous compound was obtained at Rf: 0.6 (ethyl acetate:hexane/10:90) (TLC).

![Scheme 9. Synthesis of tetrasubstituted pyrimidine-5-carboxylate derivatives 14.](image-url)

The $^1$H NMR spectrum (Figure 9) of this compound recorded in CDCl$_3$ depicted signals at $\delta$ 1.07 (t, 3H, J = 7.2 Hz, CH$_3$), 2.61 (s, 3H, C6-CH$_3$), 4.20 (q, 2H, J = 7.2 Hz, CH$_2$), 7.47 (m, 3H, ArH), 7.65 (m, 2H, ArH). The characteristic feature of the spectrum included the absence of the signal at $\delta$ 13.67 corresponding to N1-H. $^{13}$C NMR spectrum (Figure 9) recorded in CDCl$_3$ displayed signals at $\delta$ 13.5, 22.4, 62.1, 124.3, 128.3, 128.5, 130.6, 136.2, 160.4, 166.1, 166.9 and 168.5. In its EI mass spectrum, the compound depicted two peaks at $m/z$ 276 and 278 (M$^+$) in 1:3 isotopic ratio, as expected for the product.
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. (C_{14}H_{13}N_{2}O_{2}Cl). Based upon the spectral as well as correct microanalytical data (vide experimental) structure, 5-ethoxycarbonyl-6-methyl-4-phenyl-2-chloropyrimidine 4a was assigned to this compound. It was isolated in 91% yield.

Figure 9. $^1$H NMR (300 MHz, CDCl$_3$) spectrum and $^{13}$C NMR (75 MHz, CDCl$_3$) assignments of 4a.

Similar reaction of 5-methoxycarbonyl-6-methyl-4-phenylpyrimidin-2(1H)-one 3b with POCl$_3$ furnished 5-methoxycarbonyl-6-methyl-2-chloropyrimidine 4b in 93% yield. The structural assignment was based on spectral as well as microanalytical data, reported in Section 3.5.5.

Likewise, under similar set of reaction conditions C-4 unsubstituted derivative, 5-ethoxycarbonyl-6-methyl-1,2-dihydropyrimidin-2(1H)-one 3c upon reaction with POCl$_3$ furnished a viscous compound at Rf: 0.6 (ethyl acetate:hexane/25:75) (TLC) after workup and column chromatographic purification. Its $^1$H NMR (CDCl$_3$) spectrum depicted signals at δ 1.44 (t, 3H, $J$ 7.2 Hz, CH$_3$), 2.84 (s, 3H, C6-CH$_3$), 4.42 (q, 2H, $J$ 7.2 Hz, CH$_2$), 9.02 (s, 1H, CH). The absence of signal corresponding to N1-H indicated the chlorination at the C-2 position of 3c. In its $^{13}$C NMR (CDCl$_3$) spectrum, signals appeared at δ 14.0, 24.6, 63.6, 122.0, 162.0, 162.8, 164.7 and 172.5 and corroborated well with $^1$H NMR spectral data. Additionally, correct microanalytical data (vide experimental) and mass spectrum ($m/z$ 200.5, M$^+$), corresponded to the molecular formula C$_8$H$_9$N$_2$O$_2$Cl and led to the assignment of structure, 5-ethoxycarbonyl-4-methyl-2-chloropyrimidine 4c (85%) to this compound.
3.3.3 Synthesis of 5-alkoxycarbonyl-6-methyl-2-aminopyrimidines through nucleophilic displacement reactions of 5-alkoxycarbonyl-6-methyl-2-chloropyrimidines

3.3.3.1 Reactions with ammonia

Experiments were run in order to determine the suitable conditions for the incorporation of NH$_2$ group at the C-2 position of pyrimidines 4 (Scheme 9) through displacement of the C-2 chloro group. Initially, neat 5-ethoxycarbonyl-6-methyl-4-phenyl-2-chloropyrimidine 4a was treated (r.t. to reflux) with liquor ammonia, but no reaction was observed during 12 h and 4a was recovered as such after removing the reagent under reduced pressure (Table 3). Even reacting 4a with liquor ammonia in a sealed vessel yielded no success. However, when a solution of 4a in dry dichloromethane was heated in liquor ammonia, under pressure in a sealed vessel, a number of spots were observed on TLC. In yet another attempt, anhydrous ammonia gas* was purged through a solution of 4a in dry THF for 1 h at -78°C (ethyl acetate-liquid nitrogen slush) followed by stirring at room temperature, reaction was observed but it did not proceed to completion. Finally, when compound 4a was treated with dry THF saturated with ammonia gas at room temperature for 1 h, a clean reaction was observed which completed (TLC) in 2 h. The outcome of these attempts is compiled in Table 3.

Under optimized (NH$_3$/THF, r.t., 2 h) reaction conditions, 4a completely disappeared from the reaction mixture. Removal of solvent under reduced pressure and recrystallization of crude product from methanol, furnished a product at Rf: 0.7 (ethyl acetate:hexane/80:20) (TLC).

The product in its $^1$H NMR (CDCl$_3$) spectrum (Figure 10), showed signals at $\delta$ 0.94 (t, 3H, J 7.2 Hz, CH$_3$), 2.48 (s, 3H, C6-CH$_3$), 4.05 (q, 2H, J 7.2 Hz, CH$_2$), 5.70 (br, 2H, D$_2$O exchangeable, NH$_2$), 7.42 (m, 3H, ArH), 7.51 (m, 2H, ArH). Appearance of broad (D$_2$O exchangeable) singlet at $\delta$ 5.70, indicated the incorporation of NH$_2$ group at C-2 position of 4a. Further its $^{13}$C NMR (CDCl$_3$) spectrum (Figure 10) displayed signals at $\delta$ 13.4, 22.6, 61.1, 116.4, 127.7, 128.3, 129.5, 138.6, 161.8, 166.5, 167.5 and 168.4. In its mass spectrum, a peak appeared at 258 (M$^+$+1), corresponding to the molecular formula C$_{14}$H$_{15}$N$_3$O$_2$ of the expected product. From the above spectral and correct microanalytical

* evolved by using standard assembly by warming 30% aqueous ammonia solution and dried over KOH pellets

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data (vide experimental) structure, 5-ethoxycarbonyl-6-methyl-4-phenyl-2-aminopyrimidine 14a was assigned to this compound (80%).

**Table 3.** Optimization of reaction conditions for the synthesis of 14a.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Reaction conditions</th>
<th>Observations</th>
<th>Yield of 14a</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Liquor ammonia, r.t. to reflux, 12 h</td>
<td>no reaction</td>
<td>-</td>
</tr>
<tr>
<td>2.</td>
<td>Liquor ammonia, refluxed under pressure in sealed vessel, 12 h</td>
<td>no reaction</td>
<td>-</td>
</tr>
<tr>
<td>3.</td>
<td>DCM, Liquor ammonia, refluxed at 120°C under pressure in sealed vessel, 12 h</td>
<td>number of spots were observed on TLC</td>
<td>-</td>
</tr>
<tr>
<td>4.</td>
<td>NH3 gas, THF/-78°C, left overnight</td>
<td>reaction incomplete, starting recovered in the reaction</td>
<td>10%</td>
</tr>
<tr>
<td>5.</td>
<td>THF saturated with NH3 gas, r.t.</td>
<td>reaction completed within 2 h</td>
<td>80%</td>
</tr>
</tbody>
</table>

**Figure 10.** $^1$H NMR (300 MHz, CDCl$_3$) spectrum and $^{13}$C NMR (75 MHz, CDCl$_3$) assignments of 14a.

Likewise, reaction of 5-ethoxycarbonyl-6-methyl-2-chloropyrimidine 4c with THF saturated with ammonia gas, furnished the corresponding product, 5-ethoxycarbonyl-6-methyl-2-aminopyrimidine 14b in 75% yield. The structure of 14b was established from the spectral and microanalytical data (vide experimental).
3.3.3.2 Reactions with primary and secondary amines

In order to incorporate various alkyl/arylamino or ethoxy substituents at the C-2 position of 4 by displacing chloro group, reactions were performed with different nucleophiles as depicted in Scheme 9. To determine the most favourable reaction condition, reaction of 5-ethoxycarbonyl-6-methyl-4-phenyl-2-chloropyrimidine 4a was performed with benzyl amine (1.5 equiv.), using different reaction conditions (Table 4). When 4a was treated with benzyl amine in DCM or CH$_3$CN at room temperature, no reaction was observed. Using a mild base triethylamine (Et$_3$N) in THF or in ethanol at room temperature, reaction was observed but did not proceed to completion. Likewise, when 1,4-dioxane was used as solvent along with Et$_3$N as a base, reaction completed in 24 h. However, without a base, when 4a and benzylamine were heated at 80$^\circ$C in absolute ethanol (20 ml) reaction was completed within 2 h (Table 4). The resulting reaction mixture was distilled under reduced pressure to remove ethanol and crude product was purified by column chromatography to afford the desired product (m.p. 70-72$^\circ$C), Rf: 0.7 (ethyl acetate:hexane/20:80) (TLC).

Table 4. Optimization of reaction conditions for the reaction of 4a with benzylamine.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Reaction conditions</th>
<th>Observations</th>
<th>Yield of 14c</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>DCM/r.t.</td>
<td>no reaction</td>
<td>-</td>
</tr>
<tr>
<td>2.</td>
<td>CH$_3$CN/r.t.</td>
<td>no reaction</td>
<td>-</td>
</tr>
<tr>
<td>3.</td>
<td>Et$_3$N/THF/r.t.</td>
<td>reaction did not proceed to completion</td>
<td>5%</td>
</tr>
<tr>
<td>4.</td>
<td>EtOH/r.t.</td>
<td>reaction did not proceed to completion</td>
<td>7%</td>
</tr>
<tr>
<td>5.</td>
<td>Et$_3$N/dioxane/r.t.</td>
<td>Reaction completed in 24 h.</td>
<td>85%</td>
</tr>
<tr>
<td>6.</td>
<td>EtOH/80$^\circ$C</td>
<td>Reaction completed in 2 h.</td>
<td>92%</td>
</tr>
</tbody>
</table>

The $^1$H NMR (CDCl$_3$) spectrum (Figure 11) of the product depicted signals at $\delta$ 0.95 (t, 3H, J 7.2 Hz, CH$_3$), 2.49 (s, 3H, C6-CH$_3$), 4.05 (q, 2H, J 7.2 Hz, CH$_2$), 4.71 (d, 2H, J 6.0 Hz, CH$_2$), 5.63 (br, 1H, D$_2$O exchangeable, NH), 7.34 (m, 8H, ArH), 7.53 (m, 2H, ArH). The characteristic feature of this spectrum was the appearance of doublet at $\delta$ 4.71, which upon deuterium exchange was converted to a singlet. Further, appearance of signals corresponding to 5 additional aromatic protons indicated the incorporation of benzyl group on the pyrimidine core. Further its $^{13}$C NMR (CDCl$_3$) spectrum (Figure 11) displayed signals at $\delta$ 13.5, 22.9, 45.3, 61.0, 115.5, 127.2, 127.5, 127.9, 128.2, 128.5, 129.4, 138.9, 161.0 and 168.8. From the $^1$H NMR data, as well as supporting $^{13}$C NMR spectral data and
correct microanalytical and MS [m/z 348 (M$^+$+23)] the product was identified as 5-ethoxycarbonyl-6-methyl-4-phenyl-2-benzylaminopyrimidine 14c (92%, Table 5).

Likewise, reaction of 5-methoxycarbonyl-6-methyl-4-phenyl-2-chloropyrimidine 4b with benzylamine under similar reaction conditions, depicted similar reaction profile and furnished a single product, 5-methoxycarbonyl-6-methyl-4-phenyl-2-benzylaminopyrimidine 14d (85%, Table 5). The assignment of structure of 14d was based on spectral and microanalytical data reported in Section 3.5.7.

Similarly, C-4 unsubstituted derivative, 5-ethoxycarbonyl-6-methyl-2-chloropyrimidine 4c was reacted with benzyl amine (1.5 equiv.) in absolute ethanol (20 ml) at 80°C (Scheme 9). A product at Rf: 0.8 (ethyl acetate:hexane/20:80) (TLC) was isolated, which melted at 93-95°C.

Its $^1$H NMR spectrum (Figure 12) recorded in CDCl$_3$ displayed signals at $\delta$ 1.36 (t, 3H, J 7.2 Hz, CH$_3$), 2.66 (s, 3H, C6-CH$_3$), 4.31 (q, 2H, J 7.2 Hz, CH$_2$), 4.70 (d, 2H, J 6.0 Hz, CH$_2$), 5.94 (br, 1H, D$_2$O exchangeable, NH), 7.29 (m, 5H, ArH), 8.85 (s, 1H, CH). In this spectrum presence of a doublet at $\delta$ 4.70, which was converted to a singlet upon D$_2$O exchange and peaks corresponding to 5 additional aromatic protons, indicated the incorporation of benzyl group on the pyrimidine core. Additionally, its $^{13}$C NMR (CDCl$_3$) spectrum (Figure 12) displayed signals at $\delta$ 14.3, 24.7, 45.3, 60.4, 113.2, 127.4, 127.5, 128.6, 138.4, 161.1, 162.2 and 165.4, which corroborated well with $^1$H NMR spectral data. The peak at $m/z$ 272 (M$^+$+1) in its EIMS spectrum corresponded to the molecular formula.
C\textsubscript{15}H\textsubscript{17}N\textsubscript{3}O\textsubscript{2}. Additionally, the correct microanalytical data (vide experimental) confirmed the structure, 5-ethoxycarbonyl-6-methyl-2-benzylaminopyrimidine 14e (82\%, Table 5).

Figure 12. \textsuperscript{1}H NMR (300 MHz, CDCl\textsubscript{3}) spectrum and \textsuperscript{13}C NMR (75 MHz, CDCl\textsubscript{3}) assignments of 14e.

In order to further broaden the scope of this protocol for the synthesis of tetrasubstituted pyrimidine derivatives, primary alkyl amines were condensed with chloropyrimidine derivatives 4. Reaction of butyl amine with 4a in absolute ethanol (Scheme 9) furnished an exclusive product at Rf: 0.8 (ethyl acetate:hexane/40:60) (TLC), which showed a parent ion peak at \textit{m}/\textit{z} 336 (M\textsuperscript{+}+23) in its EIMS spectrum, corresponding to the molecular formula C\textsubscript{18}H\textsubscript{23}N\textsubscript{3}O\textsubscript{2}+Na. The \textsuperscript{1}H NMR (CDCl\textsubscript{3}) spectrum (Figure 13) showed signals at $\delta$ 0.94 (m, 6H, 2×CH\textsubscript{3}), 1.42 (m, 2H, CH\textsubscript{2}), 1.59 (m, 2H, CH\textsubscript{2}), 2.48 (s, 3H, C\textsubscript{6}-CH\textsubscript{3}), 3.49 (m, 2H, CH\textsubscript{2}), 4.04 (q, 2H, $J$ 7.2 Hz, CH\textsubscript{2}), 5.31 (br, 1H, D\textsubscript{2}O exchangeable, NH), 7.40 (m, 5H, ArH), 7.53 (m, 2H, ArH). The characteristic features of this spectrum included presence of three multiplets, corresponding to the protons of the butyl chain and broad singlet at $\delta$ 5.31 of NH, which upon D\textsubscript{2}O exchange converted the multiplet at $\delta$ 3.49 into triplet.

Further, its \textsuperscript{13}C NMR (CDCl\textsubscript{3}) spectrum (Figure 13) displayed signals at $\delta$ 13.5, 13.7, 19.9, 23.0, 31.7, 41.0, 60.9, 114.9, 127.8, 128.1, 129.3, 139.2, 161.2, 166.1 and 168.9 (Figure 12), which supported the \textsuperscript{1}H NMR spectrum. The NMR spectral data was further supported by the correct microanalytical data (vide experimental) and confirmed the structure, 5-ethoxycarbonyl-6-methyl-4-phenyl-2-butylaminopyrimidine 14f (97\%, Table
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.

5), assigned to this compound. Similarly reaction of 4a with 3-aminopropanol under similar set of reaction conditions furnished 14g (75%, Table 5).

Likewise, reaction of 5-ethoxycarbonyl-6-methyl-4-phenyl-2-chloropyrimidine 4a with iso-propylamine under optimized reaction conditions, furnished the corresponding C-2 elaborated pyrimidine derivative, 5-ethoxycarbonyl-6-methyl-4-phenyl-2-isopropylaminopyrimidine 14h in 87% yield (Table 5). The structure of 14h was established from the spectral and microanalytical data (vide experimental).

When binucleophilic 2-hydroxyaniline was reacted with 4a under similar set of reaction conditions (Scheme 9), a single compound [RF: 0.5 (ethyl acetate:hexane/30:70) (TLC)] having m.p. 145-147°C was obtained. Its 1H NMR (CDCl₃) spectrum (Figure 14) depicted signals at δ 0.98 (t, 3H, J 7.2 Hz, CH₃), 2.57 (s, 3H, C₆-CH₃), 4.09 (q, 2H, J 7.2 Hz, CH₂), 6.87 (m, 1H, ArH), 7.06 (m, 3H, ArH), 7.27 (br, 1H, D₂O exchangeable, NH), 7.45 (m, 3H, ArH), 7.56 (m, 2H, ArH), 10.23 (br, 1H, D₂O exchangeable, OH). The salient features of the 1H NMR were the appearance of D₂O exchangeable NH and OH resonances at δ 7.27 and 10.23, respectively and peaks for additional 4 aromatic protons, and indicated the incorporation of 2-hydroxyaniline at C-2 of the pyrimidine core. Further, a broad 1H singlet of OH at δ 10.23 confirmed that C-2 chloro of 4a was replaced by NH.
Table 5. Synthesis of C-2 elaborated pyrimidine derivatives 14 (Scheme 9).

<table>
<thead>
<tr>
<th>Entry</th>
<th>14</th>
<th>R¹</th>
<th>R²</th>
<th>X</th>
<th>R³</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>14c</td>
<td>Ar</td>
<td>Et</td>
<td>NH</td>
<td>Bn</td>
<td>92</td>
</tr>
<tr>
<td>2.</td>
<td>14d</td>
<td>Ar</td>
<td>Me</td>
<td>NH</td>
<td>Bn</td>
<td>85</td>
</tr>
<tr>
<td>3.</td>
<td>14e</td>
<td>H</td>
<td>Et</td>
<td>NH</td>
<td>Bn</td>
<td>82</td>
</tr>
<tr>
<td>4.</td>
<td>14f</td>
<td>Ar</td>
<td>Et</td>
<td>NH</td>
<td>n-Bu</td>
<td>97</td>
</tr>
<tr>
<td>5.</td>
<td>14g</td>
<td>Ar</td>
<td>Et</td>
<td>NH</td>
<td>(CH₂)₃OH</td>
<td>75</td>
</tr>
<tr>
<td>6.</td>
<td>14h</td>
<td>Ar</td>
<td>Et</td>
<td>NH</td>
<td>i-Pr</td>
<td>87</td>
</tr>
<tr>
<td>7.</td>
<td>14i</td>
<td>Ar</td>
<td>Et</td>
<td>NH</td>
<td>2-OHC₆H₄</td>
<td>85</td>
</tr>
<tr>
<td>8.</td>
<td>14j</td>
<td>Ar</td>
<td>Et</td>
<td>NH</td>
<td>4-OHC₆H₄</td>
<td>94</td>
</tr>
<tr>
<td>9.</td>
<td>14k</td>
<td>Ar</td>
<td>Et</td>
<td>NH</td>
<td>3-NH₂C₆H₄</td>
<td>75</td>
</tr>
<tr>
<td>10.</td>
<td>14l</td>
<td>Ar</td>
<td>Et</td>
<td>CH₂CH₂(1H-indol-3-yl)</td>
<td>65</td>
<td></td>
</tr>
<tr>
<td>11.</td>
<td>14m</td>
<td>Ar</td>
<td>Et</td>
<td>N</td>
<td>-(CH₂)₂-CH₂-(CH₂)₂-</td>
<td>68</td>
</tr>
<tr>
<td>12.</td>
<td>14n</td>
<td>Ar</td>
<td>Et</td>
<td>N</td>
<td>-(CH₂)₂-O-(CH₂)₂-</td>
<td>70</td>
</tr>
<tr>
<td>13.</td>
<td>14o</td>
<td>Ar</td>
<td>Et</td>
<td>O</td>
<td>Et</td>
<td>87</td>
</tr>
</tbody>
</table>

EtOH, 80°C

¹H NMR spectrum was further supported by ¹³C NMR (CDCl₃) spectrum (Figure 14) which displayed signals at δ 13.5, 22.8, 61.3, 115.5, 116.6, 121.7, 121.8, 127.9, 128.3, 129.6, 131.7, 138.6, 151.8, 158.8, 166.0, 167.3 and 168.6. Also in its mass spectrum, a molecular ion peak appeared at m/z 372 (M⁺+23), which corresponded to a molecular formula C₂₀H₁₉N₃O₃+Na. On the basis of these assignment as well as supporting microanalytical data (vide experimental) structure, 5-ethoxycarbonyl-6-methyl-4-phenyl-2-(2-hydroxyphenylamino)pyrimidine 14i (85%, Table 5) has been assigned to this compound. Likewise, the reaction of 4-hydroxyaniline with 4a furnished 14j in 94% yield (Table 5).

Similarly, reaction of 3-aminoaniline with 5-ethoxycarbonyl-6-methyl-4-phenyl-2-chloropyrimidine 4a in absolute etanol at 80°C (Scheme 9), yielded the corresponding tetrasubstituted pyrimidine derivative 14k, 5-ethoxycarbonyl-6-methyl-4-phenyl-2-(3-aminophenylamino)pyrimidine (94%, Table 5). The structure 14k has been analogously assigned on the basis of spectral data and correct microanalytical analysis (vide experimental).
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.

Using tryptamine as nucleophile, displacement of the 2-chloro substituent of 4a using standard reaction conditions allowed incorporation of the biogenic amine at the C-2 position leading to the formation of 14l in 65% yield (Table 5).

The scope of this methodology was further extended by using cycloalkylamines as nucleophiles. Therefore, when piperidine was reacted with 5-ethoxycarbonyl-6-methyl-4-phenyl-2-chloropyrimidine 4a (Scheme 9), it resulted in the formation of a product at Rf: 0.5 (ethyl acetate:hexane/10:90) (TLC) and which melted at 80-82°C. A molecular ion peak appeared at m/z 326 (M+1) in its mass spectrum, which corresponded to a molecular formula C_{19}H_{23}N_{3}O_{2}+Na expected for the product. Further, its ¹H NMR (CDCl₃) spectrum (Figure 15) depicted signals at δ 0.93 (t, 3H, J 7.2 Hz, CH₃), 1.64 (m, 6H, CH₂), 2.48 (s, 3H, C6-CH₃), 3.88 (t, 4H, J 4.8 Hz, 2×CH₂), 4.02 (q, 2H, J 7.2 Hz, CH₂), 7.38 (m, 3H, ArH), 7.57 (m, 2H, ArH). The characteristic features of this spectrum included multiplet at δ 1.64 and triplet at δ 3.88, which corresponded to the CH₂ groups of piperidine.

Its ¹H NMR spectrum was well supported by its ¹³C NMR (CDCl₃) spectrum (Figure 15) which displayed signals at δ 13.4, 23.1, 24.7, 25.7, 44.5, 60.7, 113.1, 128.0, 128.2, 129.1, 139.7, 160.1, 165.6, 166.8 and 169.1. The supporting spectral as well as the correct microanalytical analysis (vide experimental), led to the assignment of structure, 5-
ethoxycarbonyl-6-methyl-4-phenyl-2-(piperidin-1-yl)pyrimidine 14m (68%, Table 5) to this compound.

Likewise, the reaction of 5-ethoxycarbonyl-6-methyl-4-phenyl-2-chloropyrimidine 4a with morpholine under similar reaction conditions, depicted similar reaction pattern and furnished a single product, 5-ethoxycarbonyl-6-methyl-4-phenyl-2-morpholinopyrimidine 14n (70%, Table 5). The assignment of structure 14n was based on spectral and microanalytical data recorded in Section 3.5.7.

This reaction was further explored by carrying out a neat reaction of 4a with absolute ethanol. The reaction was performed at 80°C and the viscous compound at Rf: 0.3 (ethyl acetate:hexane/10:90) (TLC) was isolated. Its 1H NMR (CDCl3) spectrum (Figure 16) depicted signals at δ 1.03 (t, 3H, J 6.9 Hz, CH₃), 1.45 (t, 3H, J 6.9 Hz, CH₃), 2.57 (s, 3H, C6-CH₃), 4.15 (q, 2H, J 7.2 Hz, CH₂), 4.50 (q, 2H, J 7.2 Hz, CH₂), 7.44 (m, 3H, ArH), 7.63 (m, 2H, ArH). The characteristic features of this spectrum included the appearance of additional triplet and quartet at δ 1.45 and 4.50, respectively, corresponding to the presence of ethoxy group in the product.

Further, its 13C NMR (CDCl3) spectrum (Figure 16) displayed signals at δ 13.4, 14.3, 22.6, 61.4, 63.5, 119.6, 128.1, 128.9, 129.9, 137.7, 163.9, 166.3, 168.2 and 169.1. Additionally, correct microanalytical analysis (vide experiment) and mass spectrum (m/z 309, M+23), corresponding to the molecular formula C₁₆H₁₈N₂O₃+Na, led to the assignment of structure, 5-ethoxycarbonyl-6-methyl-4-phenyl-2-ethoxypyrimidine 14o (87%, Table 5) to this compound.
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.

3.3.3.3 Conclusions

Formation of C-2 functionalized pyrimidine derivatives 14 (Scheme 9, Table 5), evidently proceeded through nucleophilic displacement reactions and demonstrate very high regioselectivity under the reaction conditions employed. Thus, the transformation of DHPMs to C-2 elaborated pyrimidines (Scheme 9) represents a synthetically useful protocol owing to the observed regioselectivity, substituents as well as reagent tolerance and ease in execution of the reaction as well as isolation of the products. Besides, the yields are also in general high.

3.4 Cytostatic and anti-tuberculosis activities of C-2 functionalized tetrasubstituted pyrimidines

3.4.1 Cytostatic activity in cell cultures

While screening pyrimidine derivatives for their activity against a broad variety of DNA and RNA viruses (including HIV) in the appropriate cell culture models, we had observed that while none of the C-2 alkyl/aryl/amine substituted dihydropyrimidinone derivatives showed appreciable activity against any of the investigated viruses at subtoxic concentrations, C-2 amine substituted pyrimidine derivatives 14c and 14d were markedly

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Figure 16. \( ^1\text{H} \) NMR (300 MHz, CDCl\(_3\)) spectrum and \( ^{13}\text{C} \) NMR (75 MHz, CDCl\(_3\)) assignments of 14o.

**3.3.3.3 Conclusions**

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**3.4 Cytostatic and anti-tuberculosis activities of C-2 functionalized tetrasubstituted pyrimidines**

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While screening pyrimidine derivatives for their activity against a broad variety of DNA and RNA viruses (including HIV) in the appropriate cell culture models, we had observed that while none of the C-2 alkyl/aryl/amine substituted dihydropyrimidinone derivatives showed appreciable activity against any of the investigated viruses at subtoxic concentrations, C-2 amine substituted pyrimidine derivatives 14c and 14d were markedly

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\(^V\) Performed in collaboration with Professor Dr. Jan Balzarini, Rega Institute for Medical Research, Katholieke Universiteit Leuven, 10 Minderbroedersstraat, B-3000 Leuven, Belgium
cytostatic against MDCK cell cultures (IC$_{50}$: 1.2 µg/ml and 0.9 µg/ml, respectively). Compound 14d was more cytotoxic (MCC: 4-10 µg/ml) to confluent cell cultures (i.e., human embryonic lung cells, feline Crandell kidney cells) than 14c (MCC: ≥100 mg/ml) in these cell cultures. Therefore, representative members of the newly synthesized compounds were evaluated for their inhibitory effect against the proliferation of murine leukemia (LI210), murine mammary carcinoma (FM3A), human T-lymphocytes (CEM) and human cervix carcinoma (HeLa) cells (Table 6). Whereas, pyrimidine derivatives bearing amino (14a-c, 14f-h), morpholino (14n) and ethoxy (14o) substituents at the C-2 position (Table 5) depicted a marginal cytostatic activity of the test compounds (IC$_{50}$: 86-

Table 6. Inhibitory effect against the proliferation of murine leukemia (L1210), murine mammary carcinoma (FM3A), human T-lymphocyte (CEM) and human cervix carcinoma (HeLa) cells.

<table>
<thead>
<tr>
<th>Compound</th>
<th>L1210 (µM)</th>
<th>FM3A (µM)</th>
<th>CEM (µM)</th>
<th>HeLa (µM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>14a</td>
<td>402 ± 139</td>
<td>336 ± 177</td>
<td>382 ± 166</td>
<td>216 ± 29</td>
</tr>
<tr>
<td>14b</td>
<td>245 ± 34</td>
<td>432 ± 96</td>
<td>207 ± 45</td>
<td>183 ± 23</td>
</tr>
<tr>
<td>14e</td>
<td>378 ± 83</td>
<td>306 ± 6</td>
<td>480 ± 29</td>
<td>147 ± 13</td>
</tr>
<tr>
<td>14f</td>
<td>164 ± 13</td>
<td>192 ± 12</td>
<td>162 ± 40</td>
<td>138 ± 2</td>
</tr>
<tr>
<td>14g</td>
<td>238 ± 9</td>
<td>213 ± 0</td>
<td>150 ± 33</td>
<td>213 ± 14</td>
</tr>
<tr>
<td>14h</td>
<td>132 ± 28</td>
<td>174 ± 2</td>
<td>118 ± 44</td>
<td>86 ± 45</td>
</tr>
<tr>
<td>14i</td>
<td>58 ± 4</td>
<td>52 ± 4</td>
<td>43 ± 3</td>
<td>50 ± 11</td>
</tr>
<tr>
<td>14j</td>
<td>24 ± 9</td>
<td>24 ± 2</td>
<td>13 ± 4</td>
<td>24 ± 5</td>
</tr>
<tr>
<td>14k</td>
<td>46 ± 3</td>
<td>43 ± 1</td>
<td>46 ± 8</td>
<td>50 ± 2</td>
</tr>
<tr>
<td>14l</td>
<td>28 ± 6</td>
<td>34 ± 4</td>
<td>31 ± 3</td>
<td>23 ± 9</td>
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<tr>
<td>14m</td>
<td>43 ± 2</td>
<td>55 ± 1</td>
<td>72 ± 41</td>
<td>47 ± 3</td>
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<tr>
<td>14n</td>
<td>&gt; 500</td>
<td>&gt; 500</td>
<td>≥500</td>
<td>≥500</td>
</tr>
<tr>
<td>14o</td>
<td>251 ± 45</td>
<td>239 ± 54</td>
<td>120 ± 66</td>
<td>135 ± 71</td>
</tr>
</tbody>
</table>

*IC$_{50}$ a 50% inhibitory concentration
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.

500 µM), more bulky aryl group containing derivatives such as 14i-l, bearing groups such as 2-hydroxyaniline (14i), 4-hydroxyaniline (14j), 2-aminoaniline (14k) and tryptamine (14l), rendered a significantly higher antiproliferative activity (IC₅₀: 13-58 µM) (Table 6). The C-2 piperidine substituted pyrimidine 14m also showed higher cytostatic activity. 4-Hydroxyaniline substituted pyrimidine 14j showed highest cytostatic activity (IC₅₀: 13 µM) against CEM cells. It is also worth noticing that the cytostatic activity of the individual test compounds was independent of the nature of the tumor cell line (either murine versus human, or carcinoma versus leukemia).

3.4.2 Anti-tuberculosis activity against Mycobacterium tuberculosis H₃₇Rv strain

As the literature precedence attests there is only one report on the evaluation of aniline pyrimidines as antituberculosis agents but there is no report on a similar testing of 2-aminosubstituted pyrimidine-5-carboxylate derivatives such as 14. While isoniazid (Table 7) acts through inhibition of cell-wall synthesis and inhibits InhA- a NADH specific enoylase-reductase involved in the biosynthesis of fatty acids in mycobacteria, moxifloxacin is a broad spectrum antibacterial agent that acts through inhibition of the topoisomerase II (DNA gyrase) and topoisomerase IV, required for bacterial DNA replication, transcription, repair, and recombination. PA-824 kills nonreplicating M. tuberculosis by intracellular NO release.

The mode of action of the related drugs led us to test selected 14 (Table 5) for their anti-tuberculosis activity against M. tuberculosis.

From the minimum inhibitory concentrations (MICs) in Table 7, it can be deduced that the presence of a N-benzyl group at C-2 position (14c) or better a 3-aminoaniline substituent (14k) seems to be useful for significant antitubercular activity. The presence of an ethyl ester, rather than methyl ester substituent at C-5 position of the pyrimidine core (14c vs 14d) led to improvement in MIC values. Replacing the 3-aminoaniline substituent in 14k with 2-hydroxyaniline, piperidine or morpholine substituent at C-2 position to form 14i, 14m and 14n, respectively, only raised the MIC, without a significant effect on % inhibition. Replacing 3-hydroxypropyl amine substituent from C-2 position of 14g with n-butylamino substituent (14f) saw both an increase in % inhibition as well as marginal decrease in MIC. However, none of these derivatives were found to be significantly active.

∞ Performed in collaboration with Professor Dr. Kelly Chibale, Department of Chemistry and Institute of Infectious Disease and Molecular Medicine, University of Cape Town, Rondebosch 7701, South Africa
Table 7. Anti-TB activity (MIC) against \textit{M. tuberculosis} for selected pyrimidine-5-carboxylate 14 derivatives.

<table>
<thead>
<tr>
<th>Compound</th>
<th>% inhibition at 128 µM</th>
<th>MIC (µM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>14c</td>
<td>93</td>
<td>63.8</td>
</tr>
<tr>
<td>14d</td>
<td>3</td>
<td>&gt; 128</td>
</tr>
<tr>
<td>14f</td>
<td>99</td>
<td>117.7</td>
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<tr>
<td>14g</td>
<td>57</td>
<td>&gt; 128</td>
</tr>
<tr>
<td>14i</td>
<td>92</td>
<td>121.8</td>
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<tr>
<td>14k</td>
<td>99</td>
<td>31.2</td>
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<tr>
<td>14m</td>
<td>86</td>
<td>&gt; 128</td>
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<tr>
<td>14n</td>
<td>96</td>
<td>125.3</td>
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<tr>
<td>Isoniazid</td>
<td>-</td>
<td>0.12</td>
</tr>
<tr>
<td>Moxifloxacin</td>
<td>-</td>
<td>0.47</td>
</tr>
<tr>
<td>PA-824</td>
<td>-</td>
<td>0.48</td>
</tr>
</tbody>
</table>

3.4.3 Conclusions

While the nature of the C-2 substituent was found to modulate the cytostatic activity in cell cultures, none of the compounds tested for inhibition of \textit{M. tuberculosis} displayed useful activity. These results have opened scope for further investigation in this area. It would now be imperative to further explore the C-2 position on the pyrimidine to potentiate the cytostatic activity of the test compounds. Further, work in this direction is in progress.

3.5 Experimental

3.5.1 General information

Melting points were determined in open capillaries and are uncorrected. \textsuperscript{1}H NMR (300 and 400 MHz) spectra were recorded on multinuclear spectrometer Jeol FT-AL-300 and Bruker-400 MHz instruments, using commercial deuterated solvents and the chemical shifts are reported in part per million (δ) relative to tetramethylsilane (TMS, δ 0.0) used as internal reference standard. To overcome the solubility problem, wherever required, either
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.

A mixture of CDCl$_3$ and DMSO-d$_6$ [approximately 9:1 (v/v)] or alone DMSO-d$_6$ were used. Data are reported as follows: chemical shifts (multiplicity [singlet (s), doublet (d), double doublet (dd), triplet (t), quartet (q), broad (br), and multiplet (m)], integration, coupling constant [Hz] and assignment). $^{13}$C NMR spectra were recorded at 75 MHz on Jeol FT-AL-300 and at 100 MHz on Bruker-400 instruments, and the chemical shifts are reported in parts per million (δ unit) downfield from tetramethylsilane as the internal standard (CDCl$_3$, δ 77.0). EI mass spectra (MS) were recorded on Bruker Daltonics esquire 3000 spectrometer. IR spectra were recorded on Shimadzu FTIR 8400 S spectrophotometer. Elemental analyses were performed on FLASH EA 112 (Thermo Electron Corporation) analyzer and the results are quoted in %.

For monitoring the progress of a reaction and for comparison purpose, thin layer chromatography (TLC) was performed on Merck (60F$_{254}$, 0.2 mm) using an appropriate solvent system. The chromatograms were visualized under UV light. The compounds were purified using flash chromatography using silica gel (60-120 and 100-200 mesh) and mixtures of ethyl acetate/hexane as eluent.

### 3.5.2 Materials and methods

The solvents: acetonitrile (P$_2$O$_5$), MeOH/EtOH (Na metal followed by Mg treatment), benzene and tetrahydrofuran (THF) (Na-benzophenone ketyl), dichloromethane (DCM) and 1,4-dioxane (CaCl$_2$) and amines (KOH pellets) were adequately dried and drawn under N$_2$ atmosphere using hypodermic glass syringes. Pyridinium chlorochromate (PCC) was purchased from Aldrich Chemical Company. The low temperature was achieved by using slush of liquid nitrogen with appropriate solvent or alternatively using dry ice.

Compounds 2a-1 were synthesized according to the literature reports and their identity could be easily established by comparison (TLC, m.p. and $^1$H NMR spectrum) with authentic samples. 59

### 3.5.3 General procedure for the synthesis of C-4 unsubstituted DHPMs

A solution of oxazinane 15 (5.74 mmol), ethyl acetoacetate 16 (5.74 mmol) and urea/N-methyl urea 17 (5.74 mmol) in anhydrous acetonitrile (30-40 ml) containing trifluoroacetic acid (0.5 ml) was refluxed till the reaction was completed (TLC). The reaction mixture was basified with cold aqueous sodium carbonate solution and extracted with chloroform (3×50 ml). The extract was washed with cold water (2×50 ml) and dried.
(anhydrous sodium sulfate). Solvent was removed and the residue was crystallized from methanol. The characteristic data of compounds is given below.

**5-Ethoxycarbonyl-6-methyl-3,4-dihydropyrimidin-2(1H)-one (2m)**

Colorless solid. Rf: 0.6 (ethyl acetate:hexane/80:20). Yield: 62%. m.p. 256-257°C (methanol). IR (KBr): $\nu_{\text{max}}$ 1705, 1730 cm$^{-1}$. $^1$H (300 MHz, DMSO-$d_6$, 25°C): $\delta$ 1.15 (t, 3H, $J$ 7.2 Hz, ester-CH$_3$), 2.12 (s, 3H, C6-CH$_3$), 3.86 (s, 2H, C4-CH$_2$), 4.02 (q, 2H, $J$ 7.2 Hz, ester-CH$_2$), 6.97 (br, 1H, D$_2$O exchangeable, NH), 8.80 (br, 1H, D$_2$O exchangeable, NH). $^{13}$C NMR (75 MHz, DMSO-$d_6$, 25°C): $\delta$ 14.3, 17.4, 40.6, 59.1, 94.4, 148.7, 152.8 and 165.4. Anal. Calcd. for C$_8$H$_{12}$N$_2$O$_3$: C, 52.17; H, 6.52; N, 15.22; Found: C, 51.90; H, 6.30; N, 15.00. MS: $m/z$ 185 (M$^+$+1).

**5-Ethoxycarbonyl-1,6-dimethyl-3,4-dihydropyrimidin-2(1H)-one (2n)**

Colorless solid. Rf: 0.5 (ethyl acetate:hexane/80:20). Yield: 80%. m.p. 160-162°C (methanol). IR (KBr): $\nu_{\text{max}}$ 1640, 1710 cm$^{-1}$. $^1$H NMR (300 MHz, CDCl$_3$, 25°C): $\delta$ 1.28 (t, 3H, $J$ 7.2 Hz, ester-CH$_3$), 2.26 (s, 3H, C6-CH$_3$), 2.93 (s, 3H, N1-CH$_3$), 4.06 (d, 2H, $J$ 1.2 Hz, C4-CH$_2$), 4.18 (q, 2H, $J$ 7.2 Hz, ester-CH$_2$), 7.94 (br, 1H, D$_2$O exchangeable, NH). $^{13}$C NMR (75 MHz, CDCl$_3$, 25°C): $\delta$ 14.3, 18.0, 34.0, 48.6, 59.8, 95.4, 147.5, 153.1 and 165.7. Anal. Calcd. for C$_9$H$_{14}$N$_2$O$_3$: C, 54.54; H, 7.07; N, 14.14; Found: C, 54.48; H, 7.01; N, 14.05. MS: $m/z$ 221 (M$^+$+23).

### 3.5.4 General procedure for the oxidation of 3,4-dihydropyrimidin-2(1H)-ones

A solution of appropriate DHPM 1a-n (1.97 mmol) in DCM was stirred with PCC (5.90 mmol) till the reaction was completed (16-25 h, TLC). The reaction mixture was filtered over a Celite bed to remove any suspended particles and the residue obtained after removal of the solvent was flash chromatographed to obtain corresponding 1,2-dihydropyrimidin-2(1H)-one derivatives 3a-n. The characteristic data is given below.

**5-Ethoxycarbonyl-6-methyl-4-phenylpyrimidin-2(1H)-one (3a)**

White crystalline solid. Rf: 0.2 (ethyl acetate:hexane/80:20). Yield: 63%. m.p. 175-178°C (dichloromethane/hexane). IR (KBr): $\nu_{\text{max}}$ 1459, 1601, 1650, 1729, 3430 cm$^{-1}$. $^1$H NMR (400 MHz, CDCl$_3$, 25°C): $\delta$ 0.93 (t, 3H, $J$ 7.1 Hz, ester-CH$_3$), 2.62 (s, 3H, C6-CH$_3$), 4.05 (q, 2H, $J$ 7.1 Hz, ester-CH$_2$), 7.46 (m, 3H, ArH), 7.59 (m, 2H, ArH), 13.67 (br, 1H, D$_2$O...
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.

exchangeable, NH). $^{13}$C NMR (100 MHz, CDCl$_3$, 25°C): δ 13.4, 61.6, 111.5, 128.0, 128.4, 130.8, 158.3 and 166.1. Anal. Calcd. for C$_{14}$H$_{14}$N$_2$O$_3$: C, 65.12; H, 5.43; N, 10.85; Found: C, 65.42; H, 5.59; N, 11.01. MS: $m/z$ 281 (M$^+$+23).

5-Ethoxycarbonyl-1,6-dimethyl-4-phenylpyrimidin-2(1H)-one (3b)
White crystalline solid. Rf: 0.5 (ethyl acetate). Yield: 60%. m.p. 116-118°C (dichloromethane/hexane). IR (KBr): $\nu_{\text{max}}$ 1286, 1426, 1590, 1640, 1725, 3323 cm$^{-1}$. $^1$H NMR (300 MHz, CDCl$_3$, 25°C): δ 0.87 (t, 3H, J 6.9 Hz, ester-CH$_3$), 2.55 (s, 3H, C6-CH$_3$), 3.65 (s, 3H, N1-CH$_3$), 4.00 (q, 2H, J 7.2 Hz, ester-CH$_2$), 7.42 (m, 3H, ArH), 7.58 (m, 2H, ArH). $^{13}$C NMR (75 MHz, CDCl$_3$, 25°C): δ 13.4, 61.6, 111.5, 128.0, 128.4, 130.8, 158.3 and 166.1. Anal. Calcd. for C$_{14}$H$_{14}$N$_2$O$_3$: C, 65.12; H, 5.43; N, 10.85; Found: C, 65.42; H, 5.59; N, 11.01. MS: $m/z$ 281 (M$^+$+23).

5-Methoxycarbonyl-6-methyl-4-phenylpyrimidin-2(1H)-one (3c)
White crystalline solid. Rf: 0.2 (ethyl acetate). Yield: 71%. m.p. 195-197°C (dichloromethane/hexane). IR (KBr): $\nu_{\text{max}}$ 1428, 1596, 1642, 1731, 3331 cm$^{-1}$. $^1$H NMR (300 MHz, CDCl$_3$, 25°C): δ 2.61 (s, 3H, C6-CH$_3$), 3.58 (s, 3H, OCH$_3$), 7.48 (m, 3H, ArH), 7.60 (m, 2H, ArH), 13.5 (br, 1H, D$_2$O exchangeable, NH). $^{13}$C NMR (75 MHz, CDCl$_3$, 25°C): δ 52.3, 111.1, 128.0, 128.4, 131.0, 137.6, 142.6, 158.2 and 166.6. Anal. Calcd. for C$_{13}$H$_{12}$N$_2$O$_3$: C, 63.93; H, 4.92; N, 11.48; Found: C, 63.60; H, 5.20; N, 11.74. MS: $m/z$ 267 (M$^+$+23).

5-Ethoxycarbonyl-6-propyl-4-phenylpyrimidin-2(1H)-one (3d)
White crystalline solid. Rf: 0.6 (ethyl acetate). Yield: 70%. m.p. 143-145°C (dichloromethane/hexane). IR (KBr): $\nu_{\text{max}}$ 1209, 1430, 1610, 1720, 3430 cm$^{-1}$. $^1$H NMR (300 MHz, CDCl$_3$, 25°C): δ 0.93 (t, 3H, J 7.2 Hz, ester-CH$_3$), 1.03 (t, 3H, J 7.2 Hz, CH$_3$), 1.82 (m, 2H, CH$_2$), 2.85 (t, 2H, J 7.2 Hz, CH$_2$), 4.03 (q, 2H, J 7.2 Hz, ester-CH$_2$), 7.43 (m, 3H, ArH), 7.59 (m, 2H, ArH). $^{13}$C NMR (75 MHz, CDCl$_3$, 25°C): δ 13.6, 14.0, 22.4, 29.6, 61.5, 111.3, 126.5, 127.9, 128.5, 130.6, 158.2 and 166.2. Anal. Calcd. for C$_{16}$H$_{18}$N$_2$O$_3$: C, 67.13; H, 6.29; N, 9.79; Found: C, 66.90; H, 6.42; N, 10.10. MS: $m/z$ 309 (M$^+$+23).
5-Ethoxycarbonyl-6-methyl-4-(4-methoxyphenyl)pyrimidin-2(1H)-one (3e)
White crystalline solid. Rf: 0.3 (ethyl acetate). Yield: 60%. m.p. 153-155°C (dichloromethane/hexane). IR (KBr): \( \nu_{\text{max}} \) 1514, 1663, 1714, 3421 cm\(^{-1}\). \(^1\)H NMR (400 MHz, CDCl\(_3\), 25°C): \( \delta \) 1.05 (t, 3H, \( J \) 7.2 Hz, ester-CH\(_3\)), 2.58 (s, 3H, C6-CH\(_3\)), 3.86 (s, 3H, OCH\(_3\)), 4.12 (q, 2H, \( J \) 7.2 Hz, ester-CH\(_2\)), 6.94 (d, 2H, \( J \) 8.6 Hz, ArH), 7.61 (d, 2H, \( J \) 8.4 Hz, ArH), 13.76 (br, 1H, D\(_2\)O exchangeable, NH). \(^{13}\)C NMR (100 MHz, CDCl\(_3\), 25°C): \( \delta \) 13.7, 55.4, 61.6, 100.0, 113.9, 130.1, 141.7, 153.1 and 166.6. Anal. Calcd. for C\(_{15}\)H\(_{16}\)N\(_2\)O\(_4\): C, 62.50; H, 5.55; N, 9.72; Found: C, 62.71; H, 5.68; N, 9.50. MS: \( m/z \) 311 (M\(^+\)+23).

5-Ethoxycarbonyl-6-methyl-4-(3,4,5-trimethoxyphenyl)pyrimidin-2(1H)-one (3f)
White crystalline solid. Rf: 0.4 (ethyl acetate). Yield: 60%. m.p. 160-162°C (dichloromethane/hexane). IR (KBr): \( \nu_{\text{max}} \) 1240, 1655, 1720, 3422 cm\(^{-1}\). \(^1\)H NMR (300 MHz, CDCl\(_3\), 25°C): \( \delta \) 0.96 (t, 3H, \( J \) 7.2 Hz, ester-CH\(_3\)), 2.52 (s, 3H, C6-CH\(_3\)), 3.82 (s, 9H, 3×OCH\(_3\)), 4.03 (q, 2H, \( J \) 7.2 Hz, ester-CH\(_2\)), 6.80 (s, 2H, ArH). \(^{13}\)C NMR (75 MHz, CDCl\(_3\), 25°C): \( \delta \) 13.6, 56.2, 60.9, 61.7, 105.4, 111.6, 128.3, 140.4, 153.1, 158.2, 158.8 and 166.4. Anal. Calcd. for C\(_{17}\)H\(_{20}\)N\(_2\)O\(_6\): C, 58.62; H, 5.74; N, 8.04; Found: C, 58.89; H, 5.41; N, 8.10. MS: \( m/z \) 371 (M\(^+\)+23).

5-Ethoxycarbonyl-1,6-dimethyl-4-(4-nitrophenyl)pyrimidin-2(1H)-one (3g)
White crystalline solid. Rf: 0.5 (ethyl acetate). Yield: 60%. m.p. 128-130°C (dichloromethane/hexane). IR (KBr): \( \nu_{\text{max}} \) 1680, 1690, 3420 cm\(^{-1}\). \(^1\)H NMR (300 MHz, CDCl\(_3\), 25°C): \( \delta \) 0.93 (t, 3H, \( J \) 7.2 Hz, ester-CH\(_3\)), 2.61 (s, 3H, C6-CH\(_3\)), 3.69 (s, 3H, N1-CH\(_3\)), 4.03 (q, 2H, \( J \) 7.2 Hz, ester-CH\(_2\)), 7.75 (d, 2H, \( J \) 8.7 Hz, ArH), 8.27 (d, 2H, \( J \) 8.7 Hz, ArH). \(^{13}\)C NMR (75 MHz, CDCl\(_3\), 25°C): \( \delta \) 13.4, 18.2, 33.3, 61.9, 111.1, 123.4, 128.8, 144.3, 148.7, 155.2, 160.1, 166.0 and 169.05. Anal. Calcd. for C\(_{15}\)H\(_{18}\)N\(_3\)O\(_5\): C, 56.78; H, 4.73; N, 13.25; Found: C, 56.40; H, 4.59; N, 13.27. MS: \( m/z \) 340 (M\(^+\)+23).
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.

5-Isopropoxycarbonyl-6-methyl-4-(3-nitrophenyl)pyrimidin-2(1H)-one (3h)

Light yellowish solid. Rf: 0.5 (ethyl acetate). Yield: 55%. m.p. 203-205°C (dichloromethane/hexane). IR (KBr): ν_max 1220, 1600, 1673, 3399 cm⁻¹. ¹H NMR (300 MHz, CDCl₃, 25°C): δ 1.08 (d, 6H, J 6.3 Hz, 2×CH₃), 2.67 (s, 3H, C6-CH₃), 5.03 (m, 1H, CH), 7.64 (t, 1H, J 7.8 Hz, ArH), 7.79 (d, 1H, J 8.7 Hz, ArH), 8.35 (d, 1H, J 7.8 Hz, ArH), 8.47 (s, 1H, ArH), 13.75 (br, 1H, D₂O exchangeable, NH). ¹³C NMR (75 MHz, CDCl₃, 25°C): δ 18.3, 21.2, 70.2, 111.6, 123.2, 125.1, 129.4, 134.0, 147.9, 158.0 and 164.6. Anal. Calcd. for C₁₅H₁₅N₃O₅: C, 56.78; H, 4.73; N, 13.25; Found: C, 56.41; H, 5.10; N, 13.20. MS: m/z 340 (M⁺+23).

5-Ethoxycarbonyl-4,6-dimethylpyrimidin-2(1H)-one (3i)

White crystalline solid. Rf: 0.2 (ethyl acetate). Yield: 69%. m.p. 134-136°C (dichloromethane/hexane). IR (KBr): ν_max 1450, 1561, 1662 cm⁻¹. ¹H NMR (400 MHz, CDCl₃, 25°C): δ 1.39 (t, 3H, J 7.2 Hz, ester-CH₃), 2.57 (br, 6H, C₄-CH₃ & C₆-CH₃), 4.36 (q, 2H, J 7.0 Hz, ester-CH₂), 13.58 (br, 1H, D₂O exchangeable, NH). ¹³C NMR (100 MHz, CDCl₃, 25°C): δ 14.2, 61.6, 111.3, 158.1 and 165.3. Anal. Calcd. for C₉H₁₂N₂O₃: C, 55.10; H, 6.12; N, 14.29; Found: C, 54.87; H, 6.42; N, 14.03. MS: m/z 219 (M⁺+23).

5-Methoxycarbonyl-4,6-dimethylpyrimidine-2(1H)-one (3j)

White crystalline solid. Rf: 0.3 (ethyl acetate). Yield: 65%. m.p. 160-162°C (dichloromethane/hexane). IR (KBr): ν_max 1550, 1710 cm⁻¹. ¹H NMR (300 MHz, CDCl₃, 25°C): δ 2.56 (br, 6H, C₄-CH₃ & C₆-CH₃), 3.89 (s, 3H, OCH₃), 13.53 (br, 1H, D₂O exchangeable, NH). ¹³C NMR (75 MHz, CDCl₃, 25°C): δ 20.7, 52.2, 110.9, 158.1 and 165.3. Anal. Calcd. for C₈H₁₀N₂O₃: C, 52.75; H, 5.49; N, 15.39; Found: C, 52.38; H, 5.43; N, 15.75. MS: m/z 205 (M⁺+23).

5-Ethoxycarbonyl-6-phenyl-4-methylpyrimidin-2(1H)-one (3k)

White crystalline solid. Rf: 0.4 (ethyl acetate). Yield: 71%. m.p. 183-185°C (dichloromethane/hexane). IR (KBr): ν_max 1209, 1431, 1650, 1730, 2818, 3431 cm⁻¹. ¹H NMR (300 MHz, CDCl₃, 25°C): δ 0.93 (t, 3H, J 7.2 Hz, ester-CH₃), 2.62 (s, 3H, C₄-CH₃), 4.05 (q, 2H, J 7.2 Hz, ester-CH₂), 7.46 (m, 3H, ArH), 7.60 (m, 2H, ArH), 13.63 (br, 1H, D₂O exchangeable, NH). ¹³C NMR (75 MHz, CDCl₃, 25°C): δ 13.3, 61.5, 111.4, 128.1, 130.7, 158.3 and 166.0.
Anal. Calcd. for C$_{14}$H$_{14}$N$_2$O$_3$: C, 65.12; H, 5.43; N, 10.85; Found: C, 65.09; H, 5.77; N, 11.03. MS: $m/z$ 281 (M$^+$+23).

**5-Ethoxycarbonyl-6-methyl-4-pentylpyrimidin-2(1H)-one (3l)**

Brownish solid. Rf: 0.4 (ethyl acetate). Yield: 62%. m.p. 110-112°C (dichloromethane/hexane). IR (KBr): $\nu_{\text{max}}$ 1247, 1601, 1654 cm$^{-1}$. $^1$H NMR (400 MHz, CDCl$_3$, 25°C): $\delta$ 0.94 (t, 3H, $J$ 7.3 Hz, ester-CH$_2$), 1.31 (t, 3H, $J$ 7.1 Hz, CH$_3$), 1.41 (m, 2H, CH$_2$), 1.70 (m, 4H, 2×CH$_2$), 2.66 (s, 3H, C6-CH$_3$), 2.97 (t, 2H, $J$ 7.5 Hz, CH$_2$), 4.29 (q, 2H, $J$ 7.1 Hz, ester-CH$_2$), 13.51 (br, 1H, D$_2$O exchangeable, NH).

$^{13}$C NMR (100 MHz, CDCl$_3$, 25°C): $\delta$ 13.8, 19.2, 22.2, 24.8, 39.0, 62.0, 107.8, 158.3 and 163.6. Anal. Calcd. for C$_{13}$H$_{20}$N$_2$O$_3$: C, 61.90; H, 7.94; N, 11.10; Found: C, 61.50; H, 7.60; N, 10.80. MS: $m/z$ 253 (M$^+$+1).

**5-Ethoxycarbonyl-6-methylpyrimidin-2(1H)-one (3m)**

Off-white solid. Rf: 0.5 (ethyl acetate:hexane/80:20). Yield: 79%. m.p. 210-211°C (methanol). IR (KBr): $\nu_{\text{max}}$ 1675, 1680 cm$^{-1}$. $^1$H NMR (400 MHz, DMSO-d$_6$, 25°C): $\delta$ 1.28 (t, 3H, $J$ 7.12 Hz, ester-CH$_3$), 2.54 (s, 3H, C6-CH$_3$), 4.23 (q, 2H, $J$ 7.0 Hz, ester-CH$_2$), 8.74 (s, 1H, C4-H), 12.46 (br, 1H, D$_2$O exchangeable, NH). $^{13}$C NMR (100 MHz, DMSO-d$_6$, 25°C): $\delta$ 13.8, 19.2, 22.2, 24.8, 39.0, 62.0, 107.8, 158.3 and 163.6. Anal. Calcd. for C$_8$H$_{10}$N$_2$O$_3$: C, 52.75; H, 5.49; N, 15.38; Found: C, 52.35; H, 5.21; N, 15.77. MS: $m/z$ 183 (M$^+$+1).

**5-Ethoxycarbonyl-1,6-dimethylpyrimidin-2(1H)-one (3n)**

Off-white solid. Rf: 0.5 (ethyl acetate:hexane/70:30). Yield: 63%. m.p. 212-213°C (dichloromethane). IR (KBr): $\nu_{\text{max}}$ 1680, 1690 cm$^{-1}$. $^1$H NMR (300 MHz, CDCl$_3$, 25°C): $\delta$ 1.30 (t, 3H, $J$ 7.2 Hz, ester-CH$_3$), 2.63 (s, 3H, C6-CH$_3$), 3.55 (s, 3H, N-CH$_3$), 4.25 (q, 2H, $J$ 7.2 Hz, ester-CH$_2$), 8.35 (s, 1H, C4-H). $^{13}$C NMR (75 MHz, CDCl$_3$, 25°C): $\delta$ 14.1, 26.0, 39.0, 61.1, 108.1, 152.6, 154.9, 163.1 and 176.5. Anal. Calcd. for C$_9$H$_{12}$N$_2$O$_3$: C, 55.10; H, 6.12; N, 14.28; Found: C, 54.75; H, 6.01; N, 14.14. MS: $m/z$ 219 (M$^+$+23).

### 3.5.5 General procedure for the synthesis of 5-alkoxycarbonyl-6-methyl-2-chloropyrimidines

A solution of compound 3 (10 mmol) in phosphorous oxychloride (10 ml) was heated under reflux (105°C) for 30 minutes. The resulting reaction mixture was distilled under reduced pressure to remove excess of phosphorous oxychloride and last traces of POCl$_3$ were removed through azeotropic distillation with dry benzene and crude product.
was purified by column chromatography to afford pure product 4. The characteristic data of compounds 4a-c is given below.

### 5-Ethoxycarbonyl-6-methyl-4-phenyl-2-chloropyrimidine (4a)

Viscous liquid. Rf: 0.6 (ethyl acetate:hexane/10:90). Yield: 91%. IR (KBr): \(\nu_{\text{max}}\) 715, 1655, 1740, 3245 cm\(^{-1}\). \(^1\)H NMR (300 MHz, CDCl\(_3\), 25\(^\circ\)C): \(\delta\) 1.07 (t, 3H, \(J\) 7.2 Hz, ester-CH\(_3\)), 2.61 (s, 3H, C6-CH\(_3\)), 4.20 (q, 2H, \(J\) 7.2 Hz, ester-CH\(_2\)), 7.47 (m, 3H, ArH), 7.65 (m, 2H, ArH).

\(^{13}\)C NMR (75 MHz, CDCl\(_3\), 25\(^\circ\)C): \(\delta\) 13.5, 22.4, 62.1, 124.3, 128.3, 128.5, 130.6, 136.2, 160.4, 166.1, 166.9 and 168.5. Anal. Calcd. for C\(_{14}\)H\(_{13}\)N\(_2\)O\(_2\)Cl: C, 60.76; H, 4.70; N, 10.13; Found: C, 60.36; H, 4.35; N, 10.02. MS: \(m/z\) 276 (M\(^+\)).

### 5-Methoxycarbonyl-6-methyl-4-phenyl-2-chloropyrimidine (4b)

Viscous liquid. Rf: 0.7 (ethyl acetate:hexane/25:75). Yield: 93%. IR (KBr): \(\nu_{\text{max}}\) 665, 1610, 1710, 3245 cm\(^{-1}\). \(^1\)H NMR (300 MHz, CDCl\(_3\), 25\(^\circ\)C): \(\delta\) 2.61 (s, 3H, C6-CH\(_3\)), 3.58 (s, 3H, ester-CH\(_3\)), 7.48 (m, 3H, ArH), 7.65 (m, 2H, ArH).

\(^{13}\)C NMR (75 MHz, CDCl\(_3\), 25\(^\circ\)C): \(\delta\) 22.4, 52.7, 124.0, 128.2, 128.6, 130.8, 136.0, 160.5, 166.0, 167.5 and 168.5. Anal. Calcd. for C\(_{13}\)H\(_{11}\)N\(_2\)O\(_2\)Cl: C, 59.43; H, 4.19; N, 10.67; Found: C, 59.23; H, 4.30; N, 10.30. MS: \(m/z\) 263 (M\(^+\)+1).

### 5-Ethoxycarbonyl-6-methyl-2-aminopyrimidine (4c)

Viscous liquid. Rf: 0.6 (ethyl acetate:hexane/25:75). Yield: 85%. IR (KBr): \(\nu_{\text{max}}\) 650, 1650, 1750 cm\(^{-1}\). \(^1\)H (300 MHz, CDCl\(_3\), 25\(^\circ\)C): \(\delta\) 1.44 (t, 3H, \(J\) 7.2 Hz, ester-CH\(_3\)), 2.84 (s, 3H, C6-CH\(_3\)), 4.42 (q, 2H, \(J\) 7.2 Hz, ester-CH\(_2\)), 9.02 (s, 1H, C4-H).

\(^{13}\)C NMR (75 MHz, CDCl\(_3\), 25\(^\circ\)C): \(\delta\) 14.0, 24.6, 63.6, 122.0, 162.0, 162.8, 164.7 and 172.5. Anal. Calcd. for C\(_8\)H\(_9\)N\(_2\)O\(_2\)Cl: C, 47.88; H, 4.49; N, 13.96; Found: C, 47.60; H, 4.10; N, 14.10. MS: \(m/z\) 200 (M\(^+\)).

#### 3.5.6 General procedure for the synthesis of 5-ethoxycarbonyl-6-methyl-2-aminopyrimidines

A solution of compound 4a-b (1.9 mmol) in anhydrous THF was treated with THF saturated with ammonia gas (evolved by using standard assembly by warming 30% aqueous ammonia solution and evolved ammonia was dried using KOH trap) at room temperature for 45 minutes and then stirred reaction mixture at room temperature till reaction get completed (TLC, 1 h). The resulting reaction mixture was distilled under...
reduced pressure to remove excess THF and crude product was purified through crystallization to isolate compounds 14a-b. The characteristic data is presented below.

**5-Ethoxycarbonyl-6-methyl-4-phenyl-2-aminopyrimidine (14a)**

White crystalline solid. Rf: 0.7 (ethyl acetate:hexane/80:20). Yield: 80%. m.p. 290-292°C (methanol). IR (KBr): \( \nu_{\text{max}} \) 1276, 1550, 1649, 1700, 3177, 3409 cm\(^{-1}\). \(^1\)H NMR (300 MHz, CDCl\(_3\), 25°C): \( \delta \) 0.94 (t, 3H, J 7.2 Hz, ester-CH\(_3\)), 2.48 (s, 3H, C6-CH\(_3\)), 4.05 (q, 2H, J 7.2 Hz, ester-CH\(_2\)), 5.70 (br, 2H, D\(_2\)O exchangeable, NH\(_2\)), 7.42 (m, 3H, ArH), 7.51 (m, 2H, ArH). \(^1^3\)C NMR (75 MHz, CDCl\(_3\), 25°C): \( \delta \) 13.4, 22.6, 61.1, 116.4, 127.7, 128.3, 129.5, 138.6, 161.8, 166.5, 167.5 and 168.4. Anal. Calcd. for C\(_{14}\)H\(_{15}\)N\(_3\)O\(_2\): C, 65.37; H, 5.84; N, 16.34; Found: C, 65.20; H, 5.50; N, 15.96. MS: \( m/z \) 258 (M\(^+\)+1).

**5-Ethoxycarbonyl-6-methyl-2-aminopyrimidine (14b)**

White crystalline solid. Rf: 0.6 (ethyl acetate:hexane/60:40). Yield: 75%. m.p. 250-253°C (methanol). IR (KBr): \( \nu_{\text{max}} \) 1260, 1450, 1700, 1730, 3300 cm\(^{-1}\). \(^1\)H NMR (300 MHz, DMSO-\(d_6\), 25°C): \( \delta \) 1.26 (t, 3H, J 6.9 Hz, ester-CH\(_3\)), 2.49 (s, 3H, C6-CH\(_3\)), 4.19 (q, 2H, J 6.9 Hz, ester-CH\(_2\)), 7.34 (br, 2H, D\(_2\)O exchangeable, NH\(_2\)), 8.61 (s, 1H, C4-H). \(^1^3\)C NMR (75 MHz, DMSO-\(d_6\), 25°C): \( \delta \) 14.1, 24.1, 59.9, 111.4, 160.8, 163.7, 164.8 and 169.6. Anal. Calcd. for C\(_8\)H\(_{11}\)N\(_3\)O\(_2\): C, 53.04; H, 6.07; N, 23.20; Found: C, 53.32; H, 5.86; N, 22.90. MS: \( m/z \) 182 (M\(^+\)+1).

**3.5.7 General procedure for the synthesis of C-2 amine substituted 5-alkoxycarbonyl-6-methyl pyrimidines**

To a solution of appropriate 4 (1.9 mmol) in absolute ethanol (20 ml), an amine (2.85 mmol) was added and reaction heated at 80°C until completion (2 h, TLC). Ethanol was removed under reduced pressure and the crude product was purified by column chromatography to isolate corresponding 14c-n. The characteristic data of 14c-n is presented below.

**5-Ethoxycarbonyl-6-methyl-4-phenyl-2-benzylaminopyrimidine (14c)**

Colorless solid. Rf: 0.7 (ethyl acetate:hexane/20:80). Yield: 92%. m.p. 70-72°C (dichloromethane/hexane). IR (KBr): \( \nu_{\text{max}} \) 1255, 1558, 1715, 3126, 3258 cm\(^{-1}\). \(^1\)H NMR (300 MHz, CDCl\(_3\), 25°C): \( \delta \) 0.95 (t, 3H, J 7.2 Hz, ester-CH\(_3\)), 2.49 (s, 3H, C6-CH\(_3\)), 4.05 (q, 2H, J 7.2 Hz, ester-CH\(_2\)), 4.71 (d, 2H, J 6.0 Hz, CH\(_2\)NH), 5.63 (br, 1H, D\(_2\)O exchangeable, NH), 7.34 (m, 8H, ArH), 7.53 (m, 2H,
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\[ \text{ArH}. \]

$^{13}$C NMR (75 MHz, CDCl$_3$, 25°C): $\delta$ 13.5, 22.9, 45.3, 61.0, 115.5, 127.2, 127.5, 127.9, 128.2, 128.5, 129.4, 138.9, 161.0 and 168.8. Anal. Calcd. for C$_{21}$H$_{21}$N$_3$O$_2$: C, 72.62; H, 6.05; N, 12.10; Found: C, 72.30; H, 5.83; N, 11.92. MS: $m/z$ 348 (M$^+$+1).

5-Methoxycarbonyl-6-methyl-4-phenyl-2-benzylaminopyrimidine (14d)

Brownish solid. Rf: 0.7 (ethyl acetate:hexane/30:70). Yield: 85%. m.p. 103-105°C (dichloromethane/hexane). IR (KBr): $\nu_{\text{max}}$ 1249, 1558, 1721, 3127, 3261 cm$^{-1}$. $^1$H NMR (300 MHz, CDCl$_3$, 25°C): $\delta$ 2.47 (s, 3H, C$\_6$-CH$_3$), 3.58 (s, 3H, ester-CH$_3$), 4.71 (d, 2H, J$^{6.0}$ Hz, CH$_2$NH), 5.68 (br, 1H, D$_2$O exchangeable, NH), 7.35 (m, 8H, ArH), 7.54 (m, 2H, ArH). $^{13}$C NMR (75 MHz, CDCl$_3$, 25°C): $\delta$ 22.9, 45.3, 51.9, 115.2, 127.2, 127.5, 127.9, 128.2, 128.5, 129.5, 138.8, 138.9, 161.1 and 169.4. Anal. Calcd. for C$_{20}$H$_{19}$N$_3$O$_2$: C, 72.07; H, 5.70; N, 12.61; Found: C, 72.23; H, 5.90; N, 12.30. MS: $m/z$ 334 (M$^+$+1).

5-Ethoxycarbonyl-6-methyl-2-benzylaminopyrimidine (14e)

Colorless solid. Rf: 0.8 (ethyl acetate:hexane/20:80). Yield: 82%. m.p. 93-95°C (dichloromethane/hexane). IR (KBr): $\nu_{\text{max}}$ 1225, 1545, 1720, 3115, 3200 cm$^{-1}$. $^1$H NMR (300 MHz, CDCl$_3$, 25°C): $\delta$ 1.36 (t, 3H, J$^{7.2}$ Hz, ester-CH$_3$), 2.66 (s, 3H, C$\_6$-CH$_3$), 4.31 (q, 2H, J$^{7.2}$ Hz, ester-CH$_2$), 4.70 (d, 2H, J$^{6.0}$ Hz, CH$_2$NH), 5.94 (br, 1H, D$_2$O exchangeable, NH), 7.29 (m, 5H, ArH), 8.85 (s, 1H, C$\_4$-H). $^{13}$C NMR (75 MHz, CDCl$_3$, 25°C): $\delta$ 14.3, 24.7, 45.3, 60.4, 113.2, 127.4, 127.5, 128.6, 138.4, 161.1, 162.2 and 165.4. Anal. Calcd. for C$_{15}$H$_{17}$N$_3$O$_2$: C, 66.42; H, 6.27; N, 15.50; Found: C, 66.70; H, 6.01; N, 15.18. MS: $m/z$ 272 (M$^+$+1).

5-Ethoxycarbonyl-6-methyl-4-phenyl-2-butylaminopyrimidine (14f)

Viscous liquid. Rf: 0.8 (ethyl acetate:hexane/40:60). Yield: 97%. IR (KBr): $\nu_{\text{max}}$ 720, 1382, 1560, 1730, 3310 cm$^{-1}$. $^1$H NMR (300 MHz, CDCl$_3$, 25°C): $\delta$ 0.94 (m, 6H, CH$_2$ & ester-CH$_2$), 1.42 (m, 2H, CH$_2$), 1.59 (m, 2H, CH$_2$), 2.48 (s, 3H, C$\_6$-CH$_3$), 3.49 (m, 2H, CH$_2$NH), 4.04 (q, 2H, J$^{7.2}$ Hz, ester-CH$_2$), 5.31 (br, 1H, D$_2$O exchangeable, NH), 7.40 (m, 3H, ArH), 7.53 (m, 2H, ArH). $^{13}$C NMR (75 MHz, CDCl$_3$, 25°C): $\delta$ 13.5, 13.7, 19.9, 23.0, 31.7, 41.0, 60.9, 114.9, 127.8, 128.1, 129.3, 139.2, 161.2, 166.1 and 168.9. Anal. Calcd. for C$_{18}$H$_{23}$N$_3$O$_2$: C, 69.00; H, 7.35; N, 13.42; Found: C, 68.82; H, 6.99; N, 13.11. MS: $m/z$ 336 (M$^+$+23).
5-Ethoxycarbonyl-6-methyl-4-phenyl-2-(3-hydroxypropylamino)pyrimidine (14g)
Viscous liquid. Rf: 0.3 (ethyl acetate:hexane/30:70). Yield: 75%. IR (KBr): \( \nu_{\text{max}} \) 726, 1261, 1597, 1717, 2925, 3431 cm\(^{-1}\). \(^1\)H NMR (300 MHz, CDCl\(_3\), 25\(^\circ\)C): \( \delta \) 0.94 (t, 3H, \( J \) 7.2 Hz, ester-CH\(_3\)), 1.74 (m, 2H, CH\(_2\)NH), 2.50 (s, 3H, C6-CH\(_3\)), 3.63 (m, 4H, (CH\(_2\))\(_2\)OH), 4.04 (q, 2H, \( J \) 7.2 Hz, ester-CH\(_2\)), 5.59 (br, 1H, D\(_2\)O exchangeable, NH), 7.43 (m, 3H, ArH), 7.52 (m, 2H, ArH). \(^{13}\)C NMR (75 MHz, CDCl\(_3\), 25\(^\circ\)C): \( \delta \) 13.5, 22.8, 33.1, 37.3, 58.3, 61.1, 127.7, 128.3, 129.6, 138.7, 161.7 and 164.0. Anal. Calcd. for C\(_{17}\)H\(_{21}\)N\(_3\)O\(_3\): C, 64.76; H, 6.67; N, 13.33; Found: C, 64.40; H, 6.30; N, 12.99. MS: \( m/z \) 338 (M\(^+\)+23).

5-Ethoxycarbonyl-6-methyl-4-phenyl-2-isopropylamino pyrimidine (14h)
Viscous liquid. Rf: 0.9 (ethyl acetate:hexane/20:80). Yield: 87%. IR (KBr): \( \nu_{\text{max}} \) 1382, 1256, 1590, 1705, 3250 cm\(^{-1}\). \(^1\)H NMR (300 MHz, CDCl\(_3\), 25\(^\circ\)C): \( \delta \) 0.94 (t, 3H, \( J \) 7.2 Hz, ester-CH\(_3\)), 1.26 (d, 6H, \( J \) 6.6 Hz, 2×CH\(_3\)), 2.47 (s, 3H, C6-CH\(_3\)), 4.04 (q, 2H, \( J \) 7.2 Hz, ester-CH\(_2\)), 4.27 (m, 1H, CH), 5.40 (br, 1H, D\(_2\)O exchangeable, NH), 7.41 (m, 3H, ArH), 7.53 (m, 2H, ArH). \(^{13}\)C NMR (75 MHz, CDCl\(_3\), 25\(^\circ\)C): \( \delta \) 13.4, 22.7, 22.9, 42.7, 60.9, 114.8, 127.8, 128.1, 129.3, 139.2, 160.4, 166.1, 167.2 and 168.8. Anal. Calcd. for C\(_{17}\)H\(_{21}\)N\(_3\)O\(_2\): C, 68.23; H, 7.02; N, 14.05; Found: C, 68.10; H, 6.85; N, 13.70. MS: \( m/z \) 300 (M\(^+\)+1).

5-Ethoxycarbonyl-6-methyl-4-phenyl-2-(2-hydroxyphenylamino)pyrimidine (14i)
Yellowish solid. Rf: 0.5 (ethyl acetate:hexane/30:70). Yield: 85%. m.p. 145-147\(^\circ\)C (dichloromethane/hexane). IR (KBr): \( \nu_{\text{max}} \) 737, 1258, 1562, 1715, 3361 cm\(^{-1}\). \(^1\)H NMR (300 MHz, CDCl\(_3\), 25\(^\circ\)C): \( \delta \) 0.98 (t, 3H, \( J \) 7.2 Hz, ester-CH\(_3\)), 2.57 (s, 3H, C6-CH\(_3\)), 4.09 (q, 2H, \( J \) 7.2 Hz, ester-CH\(_2\)), 6.87 (m, 1H, ArH), 7.06 (m, 3H, ArH), 7.27 (br, 1H, D\(_2\)O exchangeable, NH), 7.45 (m, 3H, ArH), 7.56 (m, 2H, ArH), 10.23 (br, 1H, D\(_2\)O exchangeable, OH). \(^{13}\)C NMR (75 MHz, CDCl\(_3\), 25\(^\circ\)C): \( \delta \) 13.5, 22.8, 61.3, 115.5, 116.6, 121.7, 121.8, 127.9, 128.3, 129.6, 131.7, 138.6, 151.8, 158.8, 166.0, 167.3 and 168.6. Anal. Calcd. for C\(_{20}\)H\(_{19}\)N\(_3\)O\(_3\): C, 68.77; H, 5.44; N, 12.03; Found: C, 68.42; H, 5.14; N, 11.82. MS: \( m/z \) 372 (M\(^+\)+23).
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5-Ethoxycarbonyl-6-methyl-4-phenyl-2-(4-hydroxyphenylamino)pyrimidine (14j)
Viscous liquid. Rf: 0.3 (ethyl acetate:hexane/30:70).
Yield: 94%. IR (KBr): v_max 730, 1250, 1560, 1710, 3365 cm^{-1}. \(^1\)H NMR (300 MHz, CDCl\(_3\), 25°C): δ 0.98 (t, 3H, J 7.2 Hz, ester-CH\(_3\)), 1.65 (br, 1H, D\(_2\)O exchangeable, OH), 2.54 (s, 3H, C6-CH\(_3\)), 4.09 (q, 2H, J 7.2 Hz, ester-CH\(_2\)), 6.80 (m, 2H, ArH), 7.20 (br, 1H, D\(_2\)O exchangeable, NH), 7.48 (m, 7H, ArH).

\(^{13}\)C NMR (75 MHz, CDCl\(_3\), 25°C): δ 13.5, 22.7, 61.5, 120.1, 122.2, 126.0, 127.2, 127.8, 128.5, 130.2, 135.0, 137.6, 148.6, 158.6 and 167.6. Anal. Calcd. for C\(_{20}\)H\(_{19}\)N\(_3\)O\(_3\): C, 68.77; H, 5.44; N, 12.03; Found: C, 68.90; H, 5.30; N, 11.90. MS: m/z 372 (M\(^++\)+23).

5-Ethoxycarbonyl-6-methyl-4-phenyl-2-(3-aminophenylamino)pyrimidine (14k)
Viscous liquid. Rf: 0.3 (ethyl acetate:hexane/20:80). Yield: 75%. IR (KBr): v_max 735, 1250, 1550, 1705, 3550 cm\(^{-1}\). \(^1\)H NMR (300 MHz, CDCl\(_3\), 25°C): δ 0.98 (t, 3H, J 7.2 Hz, ester-CH\(_3\)), 2.56 (s, 3H, C6-CH\(_3\)), 4.09 (q, 2H, J 7.2 Hz, ester-CH\(_2\)), 6.39 (m, 1H, ArH), 7.07 (m, 2H, ArH), 7.32 (br, 1H, D\(_2\)O exchangeable, NH), 7.43 (m, 4H, ArH), 7.60 (m, 2H, ArH).

\(^{13}\)C NMR (75 MHz, CDCl\(_3\), 25°C): δ 13.5, 22.9, 61.2, 105.9, 109.7, 109.8, 117.0, 128.0, 128.3, 129.6, 138.6, 140.1, 146.8, 158.6, 165.8, 167.1 and 168.4. Anal. Calcd. for C\(_{20}\)H\(_{20}\)N\(_4\)O\(_2\): C, 68.97; H, 5.75; N, 16.09; Found: C, 68.93; H, 5.40; N, 15.93. MS: m/z 349 (M\(^++\)+1).

5-Ethoxycarbonyl-6-methyl-4-phenyl-2-[(1H-Indol-3-yl)ethylamino]pyrimidine (14l)
IR (KBr): v_max 697, 1261, 1556, 1693, 3259, 3364 cm\(^{-1}\). \(^1\)H NMR (300 MHz, CDCl\(_3\), 25°C): δ 0.94 (t, 3H, J 7.2 Hz, ester-CH\(_3\)), 2.48 (s, 3H, C6-CH\(_3\)), 3.08 (t, 2H, J 6.9 Hz, CH\(_2\)), 3.84 (q, 2H, J 6.9 Hz, CH\(_2\)NH), 4.04 (q, 2H, J 7.2 Hz, ester-CH\(_2\)), 5.45 (br, 1H, D\(_2\)O exchangeable, NH), 7.17 (m, 4H, ArH), 7.37 (m, 3H, ArH), 7.53 (m, 2H, ArH), 7.67 (m, 1H, ArH), 8.04 (br, 1H, D\(_2\)O exchangeable, NH).

\(^{13}\)C NMR (75 MHz, CDCl\(_3\), 25°C): δ 13.5, 22.9, 25.4, 41.5, 61.0, 111.1, 113.0, 115.1, 118.9, 119.3, 122.0, 127.3, 127.9, 128.2, 129.3, 136.3, 139.1, 162.1, 166.1, 168.9 and
172.5. Anal. Calcd. for C_{24}H_{24}N_{4}O_{2}: C, 72.00; H, 6.00; N, 14.00; Found: C, 71.84; H, 5.84; N, 13.83. MS: m/z 423 (M^+23).

5-Ethoxycarbonyl-6-methyl-4-phenyl-2-(piperidin-1-yl)pyrimidine (14m)

Yellowish solid. Rf: 0.5 (ethyl acetate:hexane/10:90). Yield: 68%. m.p. 80-82°C (dichloromethane/hexane). IR (KBr): ν_{max} 700, 1558, 1718, 2852, 2932, 3412 cm^{-1}. ^1H NMR (300 MHz, CDCl₃, 25°C): δ 0.93 (t, 3H, J 7.2 Hz, ester-CH₃), 1.64 (m, 6H, 3×CH₂), 2.48 (s, 3H, C₆-CH₃), 3.88 (t, 4H, J 4.8 Hz, 2×NCH₂), 4.02 (q, 2H, J 7.2 Hz, ester-CH₂), 7.38 (m, 3H, ArH), 7.57 (m, 2H, ArH). ^13C NMR (75 MHz, CDCl₃, 25°C): δ 13.4, 23.1, 24.7, 25.7, 44.5, 60.7, 113.1, 128.0, 128.2, 129.1, 139.7, 160.1, 165.6, 166.8 and 169.1. Anal. Calcd. for C_{19}H_{23}N_{3}O_{2}: C, 70.15; H, 7.08; N, 12.92; Found: C, 69.91; H, 6.95; N, 12.63. MS: m/z 326 (M^+1).

5-Ethoxycarbonyl-6-methyl-4-phenyl-2-morpholinopyrimidine (14n)

Yellowish solid. Rf: 0.3 (ethyl acetate:hexane/10:90). Yield: 70%. m.p. 120-122°C (dichloromethane/hexane). IR (KBr): ν_{max} 770, 1517, 1718, 2972 cm^{-1}. ^1H NMR (300 MHz, CDCl₃, 25°C): δ 0.95 (t, 3H, J 7.2 Hz, ester-CH₃), 2.49 (s, 3H, C₆-CH₃), 3.75 (t, 4H, J 4.5 Hz, 2×NCH₂), 3.92 (t, 4H, J 4.5 Hz, 2×OCH₂), 4.05 (q, 2H, J 7.2 Hz, ester-CH₂), 7.40 (m, 3H, ArH), 7.56 (m, 2H, ArH). ^13C NMR (75 MHz, CDCl₃, 25°C): δ 13.4, 23.1, 24.7, 44.0, 60.9, 66.8, 114.4, 128.0, 129.3, 139.2, 160.1, 165.6, 166.9 and 168.9. Anal. Calcd. for C_{18}H_{21}N_{3}O_{3}: C, 66.06; H, 6.42; N, 12.84; Found: C, 65.72; H, 6.21; N, 12.53. MS: m/z 328 (M^+1).

5-Ethoxycarbonyl-6-methyl-4-phenyl-2-ethoxypyrimidine (14o)

Compound 4a (1.9 mmol) was treated with ethanol at 80°C for 2 h and after completion of reaction (TLC) solvent was removed under reduced pressure. Purification of the residue by column chromatography using hexane and ethylacetate as eluents resulted in the isolation of compound 14o as a viscous liquid. Rf: 0.3 (ethyl acetate:hexane/10:90). Yield: 87%. IR (KBr): ν_{max} 1150, 1410, 1700, 1730, 3300 cm^{-1}. ^1H NMR (300 MHz, CDCl₃, 25°C): δ 1.03 (t, 3H, J 6.9 Hz, ester-CH₃), 1.45 (t, 3H, J 6.9 Hz, CH₃), 2.57 (s, 3H, C₆-CH₃), 4.15 (q, 2H, J 7.2 Hz, ester-CH₂), 4.50 (q, 2H, J 7.2 Hz, OCH₂), 7.44 (m, 3H, ArH), 7.63 (m, 2H, ArH). ^13C NMR (75 MHz, CDCl₃, 25°C): δ 13.4, 14.3, 22.6, 61.4, 63.5, 119.6, 128.1, 128.9, 129.9, 137.7, 163.9, 166.3, 168.2 and
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168.5. Anal. Calcd. for C_{16}H_{18}N_{2}O_{3}: C, 67.13; H, 6.29; N, 9.79; Found: C, 66.82; H, 6.10; N, 9.45. MS: m/z 309 (M^+23).

3.6 References


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