Synthesis and Exchange Reactions of 5-Alkyl-2-oxo-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylic Acids

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An improved preparation of 2-oxo-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylic acid **3**, a potent inhibitor of dihydroorotase is presented. *Trans*-5-alkyl-2-oxo-6-thioxohexahydropyrimidine-4-carboxylic acids **12a-c** were synthesised *via* the thiation of the *p*-methoxybenzyl esters of 5-alkyldihydroorotic acids with Lawesson's reagent followed by subsequent de-protection. The corresponding *cis* isomers were prepared by reduction of 5-alkyl-6-thioxoorotic acids with zinc in acetic acid. The stability and exchange reactions of **12a-c** under physiological conditions were investigated by ultra-violet and ^{1}H nmr spectroscopy. The attempted synthesis of **16**, a fused cyclopentyl derivative of **3** is also presented.

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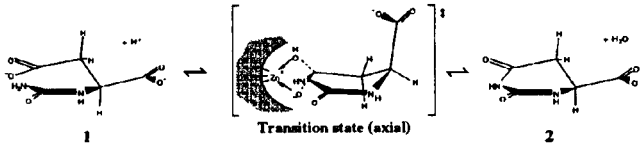
Introduction.

The *de novo* pyrimidine biosynthetic pathway has been proposed as an excellent target against which to develop anti-malarial drugs [1]. The malarial parasite, is unable to salvage pyrimidine bases or nucleosides and so must synthesise these *via* the *de novo* pathway [2]. Humans on the other hand are able to utilise both the *de novo* and salvage pathways for their pyrimidine requirements. Thus, inhibitors of the *de novo* pyrimidine pathway could be effective anti-malarials, as blockage of the pathway by these inhibitors would result in parasite death, while humans could continue to obtain their pyrimidine requirements *via* the alternative salvage pathway.

Dihydroorotase catalyses the third reaction of *de novo* pyrimidine biosynthesis, the reversible cyclisation of carbamyl aspartate **1** to dihydroorotate **2** (Figure 1). The rate of cyclisation of **1** is maximal at acidic *p*H (4.0-6.0), whilst ring cleavage is predominant under alkaline conditions (*p*H 9.0-12.0). Under physiological conditions (*p*H = 7.4) at equilibrium, the molar ratio **1**:**2** is 16.6:1 in solution, indicating that either substrate or product analogues may be effective inhibitors of dihydroorotase [3].



1 2

Figure 1. The reaction catalysed by DHOase

The catalytic mechanism of dihydroorotase is similar to that of a zinc protease, with the active site zinc stabilising the transition state of the reaction by formation of a co-ordination complex with the two oxygens at C-6 of the dihydropyrimidine ring (Figure 1) [3]. Inhibitor studies also suggest that the 4-carboxyl group is in the axial orientation in the transition state [4].

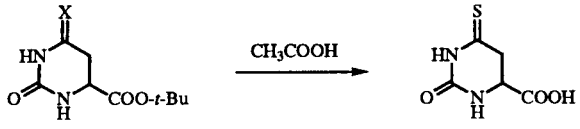
Adams *et al.* have synthesised a number of *cis* and *trans*-4-carboxy-3,4,5,6-tetrahydropyrimidin-2(1*H*)-ones incorporating either a mercaptomethyl, hydroxymethyl or carboxyl at C-6 [5]. Of these, only the *cis*-6-mercaptomethyl analogue inhibited dihydroorotase, suggesting that inhibition is primarily a result of co-ordination of the thiolate to the active site zinc. We have recently synthesised several potent inhibitors of dihydroorotase which induce blockade of *de novo* pyrimidine biosynthesis in human CCRF-CEM leukemia cells growing in culture [6]. One of these, 2-oxo-6-thioxohexahydropyrimidine-4-carboxylic acid **3**, as the free acid or methyl ester pro-drug, also induces a large accumulation of **1** in the malarial parasite, *Plasmodium falciparum* treated with **3**, indicating inhibition of dihydroorotase. Although compound **3** is an effective inhibitor of dihydroorotase (K_i 6.5 μM *c.f.* K_m of 19 μM for **2** at *p*H 7.4 [6]) it suffers from several drawbacks including its lability under aqueous conditions and its current mode of synthesis which results in material of only moderate purity [7]. We wish to report herein the synthesis of 5-alkyl-2-oxo-6-thioxohexahydropyrimidine-4-carboxylic acids, which have been prepared in order to overcome the problems associated with **3**.

Results and Discussion.

Two approaches have previously been reported for the preparation of **3**. One of these, the direct reduction of 6-thioxoorotic acid with zinc in acetic acid afforded **3** in only 65% purity along with 8% of **2** [8]. Similarly, alkaline hydrolysis of the methyl ester of **3** resulted in a significant

loss of sulphur, yielding a preparation of **3** which was contaminated with large quantities of **2** [7]. To overcome the problem of hydrolysis encountered above, the *t*-butyl group was used to protect the carboxylic acid functionality in 2 prior to thiation. *t*-Butyl dihydroorotate **4** was synthesised from **2** according to the method of Suzuki *et al.* (Scheme 1) [9]. Thiation of **4** employing Lawesson's reagent afforded the 6-thioxo derivative **5** in good yield (63%). Removal of the protecting group with anhydrous trifluroacetic acid afforded **3**, analytically pure, and in almost quantitative yield (93%). Utilising the *p*-methoxyphenylmethyl ester of **2** in place of the *t*-butyl ester proved equally effective for the preparation of **3**. Interestingly, the *t*-butyldimethylsilyl ester of **2** underwent simultaneous thiation and de-silylation with Lawesson's reagent to afford **3** directly, albeit in moderate yield and purity (55% and 62%, respectively).

Scheme 1

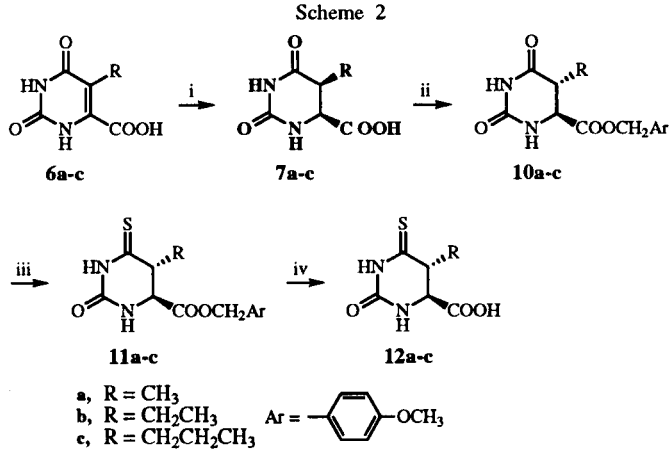


4, X = O
5, X = S

3

Scheme 2 outlines the synthetic sequences employed in the preparation of 5-alkyl-2-oxo-6-thioxohexahydropyrimidine-4-carboxylic acids. The 5-alkyl orotic acids **6a-c** utilised as starting materials were prepared according to the general procedure of Laursen *et al.* [10]. Reduction of these employing zinc dust in acetic acid at reflux afforded the corresponding 5-alkyldihydroorotic acids **7a-c** in good yield. Analysis by ^{1}H nmr indicated that these were formed as a mixture of *cis* and *trans* isomers (typically, *cis:trans* 3:1). The typical coupling constant of 6.0-6.6 Hz observed betweeen H-4 and H-5 ($J_{4,5}$) indicated

Scheme 2



6a-c **7a-c** **10a-c**

11a-c **12a-c**

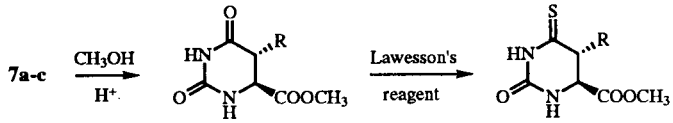
a, R = CH_3
b, R = CH_2CH_3
c, R = $CH_2CH_2CH_3$

i: Zn/HOAc, reflux; ii: *p*-methoxy benxylchloride/Cs_2CO_3/DMF; iii: P_2S_5
iv: CF_3COOH

that the 4-carboxyl and 5-alkyl substituent were in the *cis* configuration for each of the major isomers formed, with the $J_{4,5}$ for the minor isomer (2.8-3.1 Hz) being in accord with a *trans* disposition for these substituents [11].

The 5-alkyl dihydroorotic acids were readily converted to their corresponding methyl esters **8a-c** *via* Fischer-Speier esterification (Scheme 3). Methylation significantly affected the distribution of isomers, with the thermodynamically favoured *trans* configuration now contributing significantly to the isomeric distribution (*e.g. trans: cis* 4:1 for **8c**). Incorporation of sulphur at C-6 was readily accomplished employing Lawesson's reagent to afford predominately the *trans*-5-alkyl-6-thioxodihydroorotate methyl ester prodrugs **9a-c** as yellow crystalline solids in good yield (typically 60%).

Scheme 3



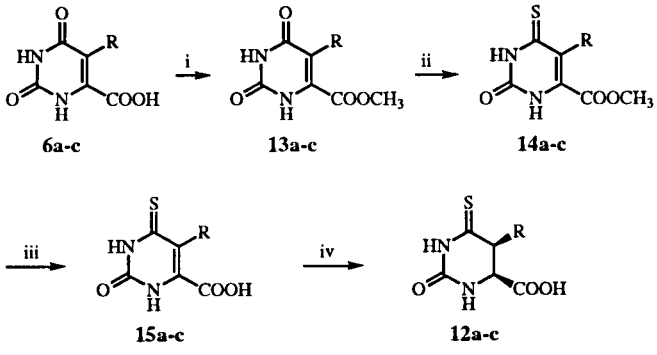
8a-c **9a-c**

a, R = CH_3
b, R = CH_2CH_3
c, R = $CH_2CH_2CH_3$

Preparation of the 5-alkyl-6-thioxodihydroorotic acids required suitable protection of the carboxylic acid functionality prior to thiation. Preparation of the *t*-butyl esters of **7a-c** proved unsuccessful, presumably due to steric constraints imposed at C-5. Reaction of *p*-methoxybenzyl chloride with the 5-alkyldihydroorotic acids **7a-c**, however proved successful, affording the corresponding *p*-methoxybenzyl esters **10a-c** in reasonable yield (50-60%). The resulting product was typically obtained as an approximately equimolar mixture of *cis* and *trans* isomers. Thiation at C-6 proved troublesome, with Lawesson's reagent typically giving very low yields (5%) of the desired 6-thioxo species **11a-c.** Resort was made to phosphorus pentasulfide as a thiating reagent, which under vigorous conditions (reflux in dioxane) increased the yields of the 6-thioxo species **11a-c** to 20-25%. Analysis by ^{1}H nmr indicated that thiation had significantly affected the distribution of stereoisomers, with approximately 85% of each of the 6-thioxo esters **11a-c** now in the *trans* configuration. Removal of the *p*-methoxy protecting group with anhydrous trifluoroacetic acid afforded the 5-alkyl-6-thioxodihydroorotic acids **12a-c** as pale yellow solids in high purity and good yield. Removal of the protecting group did not affect the stereoisomeric distribution.

Preparation of the 5-alkyl-6-thioxodihydroorotic acids **12a-c** enriched in the *cis* isomer was conveniently achieved by reduction of the corresponding 5-alkyl-6-thioxoorotic acids **15a-c** with zinc in acetic acid at 60° for 1 hour. The requisite acids **15a-c,** were readily prepared from the corresponding 5-alkylorotic acids **6a-c** employing the reaction

Scheme 4



sequence outlined in Scheme 4. In this way good yields (70-80%) of the 5-alkyl-6-thioxodihydroorotic acids **12a-c** were obtained with excellent stereoselectivities (*cis:trans* 9:1). Extended reaction times for the reduction, resulted in a gradual equilibration of the isomers, ultimately resulting in an equilibrium mixture comprising, *trans:cis* ~ 85:15.

Hydrolytic Stability of 6-Thioxodihydroorotic Acids.

Compound **3** decomposes spontaneously in aqueous solution resulting in the quantitative formation of **2** [12]. Consequently, the decompostion of **3** can conveniently be followed by monitoring the decrease in the λ_{max} of **3** at 280 nm. The loss of absorbance at 280 nm for **3** in 0.1 *M* phosphate buffer (*p*H 7.2) at 37° was found to be pseudo first order, with **3** exhibiting a half life of 6.6 hours under these conditions. The half lives for the 5 alkyl analogues **12a-c** were similarly determined, and are tabulated in Table I. It can be seen that a dramatic increase in hydrolytic stability is observed with increasing steric demand at C-5, with both the 5-ethyl **12b** and 5-propyl **12c** analogues showing no detectable decomposition over the 12 hour incubation time.

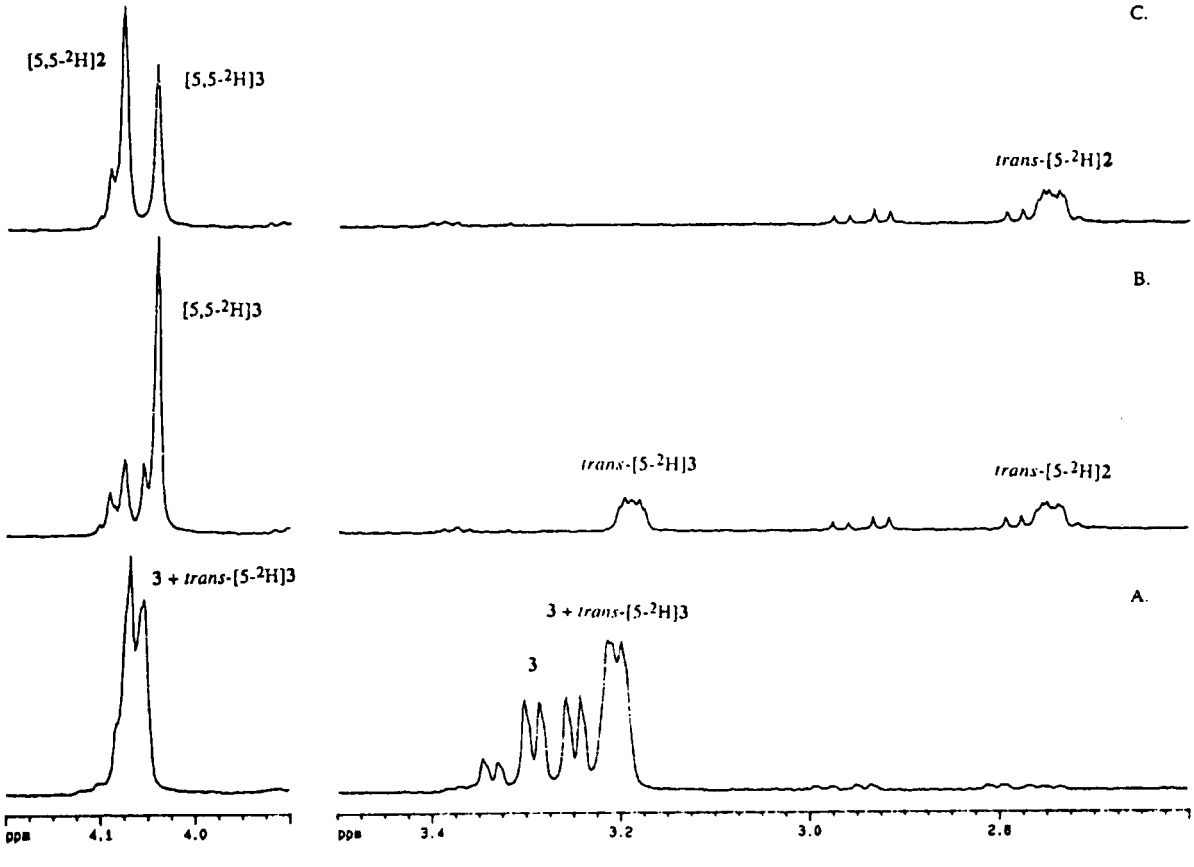
Table I

Reaction Half-times for the Hydrolysis of 5-Alkyl-6-thioxodihydroorotic Acids

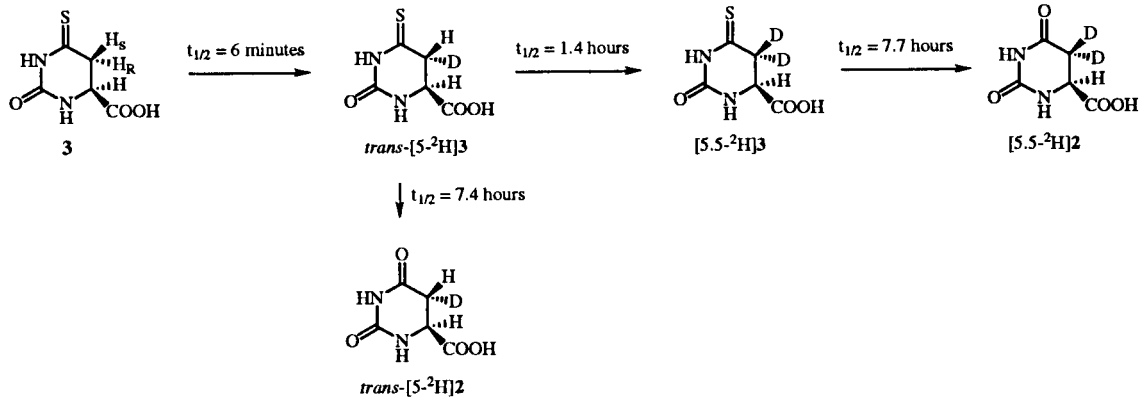
Compound	**3**	**12a**	**12b**	**12c**
Half-time (hours)	6.6	10.6	>100	>100

Figure 2.

^1H NMR Spectra of **3** in 9.5 m*M* Sodium Phosphate Buffer *p*H 7.2, 3.7° at **A** 0.1 hours, **B** 3 hours and **C** 10 hours.



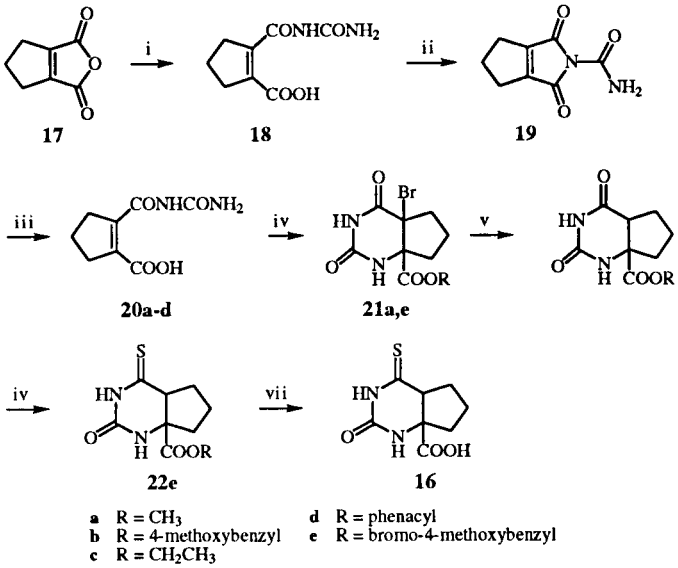
Scheme 5



The kinetics and mechanism of decomposition were further investigated by ^{1}H nmr spectroscopy. The decomposition of 3 was followed by ^{1}H nmr in phosphate buffered deuterium oxide (*p*H 7.2) at 38°. Spectra were recorded every 6 minutes over a period of 12 hours. Initially, the ^{1}H nmr spectrum of **3** exhibited two sets of double doublets at 3.23 ppm and 3.34 ppm, assigned to the *5-pro-S* and *S-pro-R* protons respectively [13], and an apparent triplet for H-4 at 4.08 ppm (Figure 2). During the course of incubation, the signal for the *S-pro-S* proton rapidly collapsed into a broadened doublet centered at 3.23 ppm, while the signal for the *5-pro-S* proton disappeared. Simultaneously, the signal for H-4 collapsed into a doublet. The ^{1}H nmr data is consistent with the new compound formed being *trans*-[5-^{2}H]**3**, arising *via* stereoselective abstraction of the *5-pro-R* proton from **3** followed by deuteration at C-5 on the opposite face to the carboxylate at C-4 (Scheme 5). Keys and Johnston have demonstrated a similar stereoselective abstraction and deuteration in deuterium oxide of the *S-pro-R* proton in **2**, the reaction having an apparent half time of 128 hours [13]. The corresponding half-time for **3** was found to be 6 minutes, presumably reflecting the ease of enolisation due to the relatively weak C=S bond. *Trans*-[5-^{2}H]**3** gradually decomposed with an apparent half life of 1.4 hours, with the doublet for H-4 collapsing into a singlet, and the signal for H-5 disappearing (Figure 2). The new compound formed in this reaction is proposed to be [5,5-^{2}H]**3**, being formed by slow abstraction of the H-5 proton from *trans*-[5-^{2}H]3 followed by subsequent deuteration (Scheme 5). A similar effect has also been observed for **2** [13]. Compound [5,5-^{2}H]**3** finally undergoes a slow hydrolysis to [5,5-^{2}H]**2** with a half-time for decomposition of 7.7 hours. This half-time correlates well with the observed reaction half-time of 6.7 hours for the decomposition of **3** into **2** as determined *via* uv spectroscopy, with the increased half-time probably a reflection of a solvent or secondary deuterium isotope effect. The final ^{1}H nmr spectrum for the decomposition of **3** recorded after 200 hours, indicated that the reaction mixture was composed largely (84%) of [5,5-^{2}H]**2** along with a smaller amount of *trans*-[5-^{2}H]**2** (16%) [14] arising from a slow ($t_{1/2}$ = 7.4 hours) competing hydrolysis of *trans*-[5-^{2}H]**3** (Scheme 5).

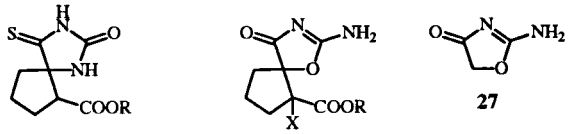
In the case of the *cis*-5-alkyl-6-thioxodihydroorotic acids **12a-c** a similar rapid removal of the H-5 proton and deuteration exclusively *trans* to the C-4 carboxylate, thereby maintaining the *cis* configuration in the deuterated product, is observed by ^{1}H nmr in phosphate-buffered deuterium oxide. This was indicated by the rapid decay of the signal for H-5, and the collapse of the doublet for H-4 into a slightly broadened singlet. Half-times for the

Scheme 6



a R = CH_3 **d** R = phenacyl
b R = 4-methoxybenzyl **e** R = bromo-4-methoxybenzyl
c R = CH_2CH_3

i: NH_2CONH_2/acitic acid; ii:: Acetic anhydride; iii: ROH/$ZnCl_2$ iv: Br_2/DMF/pyrmidne; v: Bu_3SnH/catalyst $_{88}$; vi: CF_3COOH



exchange reaction are of the order of 4-6 minutes. For the *trans* isomers of **12a-c**, a slow deuterium exchange is observed, indicated by slow decay of the H-5 signal. It was anticipated that the intermediate in the exchange reactions would be identical for the *cis* and *trans* isomers, with deuterium incorporation, ultimately resulting in the same product for either isomer. However, deuterium incorporation in the *trans* isomers resulted in exclusive retention of the *trans* configuration. This finding would indicate that proton abstraction at C-5 in both the *cis* and *trans* isomers (presumably with H-5 in the axial configuration) is followed by an extremely rapid deuteration at a rate significantly greater than that for a conformational change which would equilibrate the respective intermediates.

Attempted Synthesis of 2-Oxo-4-thioxo-1,3-diazabicyclo [4.3.0]nonane-9-carboxylic acid **16**.

The transition state for the reaction catalysed by dihydroorotase is thought to require the axial disposition of the 4-carboxylate of dihydroorotic acid (Figure 1) [4]. Consequently, the potential inhibitor **16** (Scheme 6) was designed to constrain the carboxylic acid moiety in the axial orientation while at the same time maintaining favourable hydrolytic stability and any potential hydrophobic interactions with the active site of dihydroorotase.

O'Murchu has reported a simple and high yielding preparation of 5-bromodihydroorotic acid *via* the reaction of bromine with maleuric acid [15]. Extension of this useful reaction to the synthesis of **16** would provide a covenient route to a relatively inaccessible compound. The proposed synthesis of **16** is outlined in Scheme 6. Incubation of the anhydride of cyclopentenyl-1,2-dicarboxylic acid **17** with urea in acetic acid at 50° according to the method of Tawney *et al.* [16] afforded the cyclopentyl acid **18** in good yield (78%). Formation of the crystalline imide **19** was readily accomplished by treatment of **18** with acetic anhydride at 90° in excellent yield (85%). The imide **19** was smoothly converted into a variety of esters **20a-d** (Scheme 6) by reaction with the appropriate alcohol utilising zinc chloride as a Lewis acid catalyst. Due to the success of the *p*-methoxybenzyl group as a carboxyl protecting agent in the synthesis of **12a-c**, the *p*-methoxybenzyl ester **20b** was employed for subsequent transformations. Reaction of **20b** with 2 equivalents of bromine in dimethylformamide/pyridine afforded a single compound which on the basis of O'Murchu's findings should have the structure **21e**. Removal of the aliphatic bromine was conveniently accomplished employing tributyltin hydride. This debrominated adduct was treated with Lawesson's reagent to afford a thiated adduct in 50-55% yield, with this compound expected to have the structure **22e**. However the product isolated was not the desired derivative **16**, but the hydantoin **23**. Structural confirmation was obtained *via* an

Table II

Bond Lengths (Å) for **23**

Br(1)-C(12)	1.897(5)	S(1) C(1)	1.639(6)
O(1)-C(2)	1.323(7)	0(2)-C(8)	1.349(6)
O(2)-C(9)	1.449(6)	Q(3)-C(8)	1.193(6)
O(4)-C(13)	1.355(6)	0(4) C(16)	1.407(7)
N(1)-C(1)	1.321(6)	N(1) C(2)	1.320(7)
N(2)-C(2)	1.326(6)	N(2) C(3)	1.461(6)
C(1)-C(3)	1.514(7)	C(3) C(4)	1.538(7)
C(3)-C(7)	1.542(7)	C(4) C(5)	1.498(8)
C(5)-C(6)	1.513(8)	C(6)-C(7)	1.522(7)
C(7)-C(8)	1.489(7)	C(9)-C(10)	1.495(7)
C(10)-C(11)	1.369(7)	C(10)-C(15)	1.371(8)
C(11)-C(12)	1.375(7)	C(12)-C(13)	1.386(7)
C(13)-C(14)	1.383(7)	C(14)-C(15)	1.385(7)

Table III

Bond Angles (°) for **23**

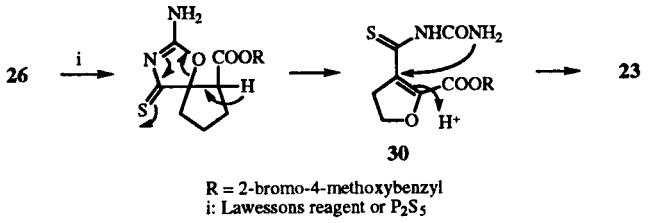
C(8)-O(2)-C(9)	115.1(4)	C(13)-O(4)-C(16)	117.9(5)
C(1)-N(1)-C(2)	107.7(5)	C(2)-N(2)-C(3)	105.9(4)
S(1)-C(1)-N(1)	128.4(4)	S(1)-C(1)C(3)	122.4(4)
N(1)-C(1)-C(33	109.1(4)	O(1)-C(2)-N(1)	126.4(5)
O(1)-C(2)-N(2)	117.7(5)	N(1)-C(2)-N(2)	115.8(5)
N(2)-C(3)-C(1)	101.3(4)	N(2)-C(3)-C(4)	109.3(4)
N(2)-C(3)-C(7)	109.2(4)	C(1)-C(3)-C(43	114.0(5)
C(1)-C(3)-C(7)	117.1(4)	C(4)-C(3)-C(7)	105.6(4)
C(3)-C(4)-C(5)	106.5(5)	C(4)-C(5)-C(6)	105.2(5)
C(5)-C(6)-C(7)	103.2(5)	C(3)-C(7)-C(6)	103.7(4)
C(3)-C(7)-C(8)	115.0(4)	C(6)-C(7)-C(8)	115.1(5)
O(2)-C(8)-O(3)	123.4(5)	O(2)-C(8)-C(7)	111.3(5)
O(3)-C(8)-C(7)	125.3(5)	O(2)-C(9)-C(10)	109.4(5)
C(9)-C(10)-C(11)	119.3(5)	C(9)-C(10)-C(15)	122.8(5)
C(11)-C(10)-C(15)	117.8(5)	C(10)-C(11)-C(12)	120.6(5)
Br(1)-C(12)-C(11)	119.6(4)	Br(1)-C(12)-C(13)	118.5(4)
C(11)-C(12)-C(13)	121.9(5)	O(4)-C(13)-C(12)	117.6(5)
O(4)-C(13)-C(14)	124.7(5)	C(12)-C(13)-C(14)	117.7(5)
C(13)-C(14)-C(15)	119.5(5)	C(10)-C(15)-C(14)	122.5(5)

X-ray crystal structure determination of **23**, which gave suitable crystals for an X-ray analysis (Figure 3). Treatment of **23** with anhydrous trifluroacetic acid cleanly cleaved the 2-bromo-4-methoxybenzyl protecting group to afford the 4-thioxohydantoin **24** in excellent yield (84%).

It was assumed that the adduct obtained *via* bromocyclisation of **20b** would also be an hydantoin, but this was found not to be the case [17]. Rather the adduct arising from the bromocyclisation of **20b** was determined to be the spiro 2-aminooxazolone **25**, with the debrominated product obtained upon reduction with tributyltin hydride having structure **26**. Evidence for an 2-aminooxazol one, in the absence of a crystal structure, is best gained by a comparison of the spectroscopic properties of the methyl ester adducts **27** and **28** and 5,5-dimethyl-2-amino-4(5*H*)oxazolone **29**. As can be seen from Table IV, excellent agreement is found between the compounds, strongly supporting structure **24** for the bromocyclisation adduct.

A tentative mechanism for the novel thiation/rearrangement of **26** with either Lawesson's reagent or phosphorus pentasulfide is shown in Scheme 7. Thiation of **26** to a

Scheme 7



R = 2-bromo-4-methoxybenzyl
i: Lawessons reagent or P_2S_5

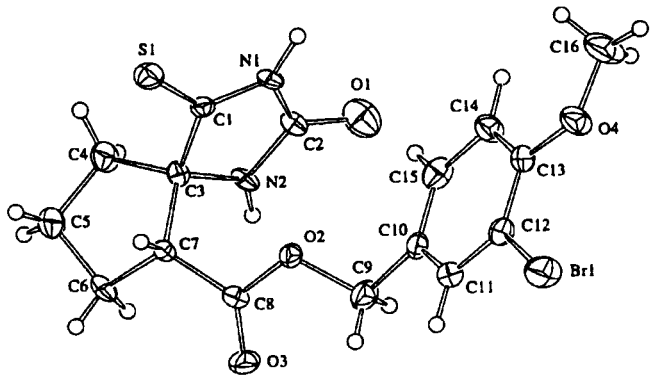


Figure 3. ORTEP plot (30% thermal ellipsoids) with atom scheme.

EXPERIMENTAL

Melting points were determined thermoelectrically on a Reichert hot stage apparatus and are uncorrected. Infrared spectra were recorded on a Perkin Elmer Infrared Spectrophotometer model 783 as mulls in nujol. The ^1H nmr spectra were acquired on either a Bruker 200 or Varian XL400 spectrometer operating at 200 and 400 MHz respectively. The spectra were measured in DMSO-d_6 unless otherwise stated. Each signal was described in terms of chemical shift as parts per million (ppm) downfield from tetramethylsilane as the internal standard (δ 0.00). Signals were described as s (singlet), d (doublet), t (triplet), q (quartet) and m (multiplet). Ultraviolet absorbance spectra were acquired using a Hewlett Packard 8452A diode array spectrophotometer. Microanalyses (C, H, N) were performed by the Australian National University Analytical Services, Canberra.

Reactions were followed by tlc on Merck F254 silica gel plates. Merck silica gel (70-230 mesh) was used for column chromatography. All solvents and reagents were obtained from commercial sources and purified before use if necessary.

NMR Time Course Reactions.

Compounds, **3** and **12a-c** were dissolved in phosphate buffered deuterium oxide (1.5 m*M* potassium dihydrogen phosphate, 8 m*M* disodium hydrogen phosphate; *p*H 7.2) at 37° to give 10 mmole solutions along with sodium 3-(trimethylsilyl)-2,2,3,3-tetradeuteropropionate (~0.2 mg) as internal reference. Spectra were acquired after a 6 minute delay at 1.8 minute intervals for ~40 minutes and at 16 minute intervals for the following 10 hours. The sample was subsequently incubated at 37° for 200 hours before acquisition of the final spectrum. An interval of 5 seconds was allowed between each acquisition to ensure full relaxation of protons. Half times for the decay and increase of signals were calculated by non-linear regression of data to first-order exponential decrease and increase equations as appropriate, using a compiled version of the non-linear regression program, DNRP53, written in basic by Duggleby [19]. Half times of signals which increased and subsequently decreased with time were also fitted to the appropriate rate equation [19].

X-Ray Crystallography.

A crystal was mounted on a glass fibre with cyanoacrylate resin and cell constants were determined by a least-squares fit to the setting angles of 25 independent reflections collected on an Enraf Nonius CAD-4F four-circle diffractometer employing graphite monochromated MoKα radiation.

The crystal data for **23** follows: Formula $C_{16}H_{17}BrN_2O_4S$; *M* 262.33, orthorhombic, space group Pbca, *a* 19.298(4), *b* 7.321(2),

Table IV

Comparative Spectroscopic Data for **27-29**

Entry	uv: (methanol) λ max	^1H nmr: (DMSO-d_6) δ	ir: ν (cm^{-1})
27	220 (log ε = 4.22))	8.35 (1H), 8.64 (1H)	3332, 3328, 1743 (C=O), 1663 (C=N)
28	218 (1og ε = 4.22)	8.32 (1H), 8.34 (1H)	3327, 3250, 1743 (C=O), 1663 (C=N)
29	215 (log ε = 4.35)	8.31 (2H)	3340, 3260, 1742 (C=O), 1641 (C=N)

thioxooxazolone, followed by ring opening to **30**, which is promoted by the presence of a relatively acidic proton and readily enolised C=S bond, followed by ring closure to the favoured 5 membered hydantoin, completes the proposed decomposition sequence for **26**.

In summary, the successful synthesis of *cis* and *trans* 5-alkyl-2-oxo-6-thioxohexahydropyrimidine-4-carboxylic acids has been accomplished. A dramatic increase in the stability of these compounds with respect to hydrolysis was observed with increasing size of the 5-alkyl substituent. The *cis* and *trans* isomers were found to readily undergo exchange at C-5, but were found not to interconvert at physiological *p*H, thus allowing the inhibition of the individual isomers against dihydroorotase to be determined. The biological evaluation of these isomers is in progress and will be reported subsequently.

c 24.328(6) Å, V 3437(1) Å^3, Z 8, D_c 1.597 g mol^{-1}, μ(MoKα) 25.43 cm^{-1}, λ(MoKα) 0.71069 Å, F(000) 1680 electrons, θ_{max} 25.0°, T_{max} 0.755, T_{min} 0 505, N 3467, N_o 1912 (I > 2.5 σ(I)), N_{var} 220, max shift 0.02σ) R* 0.049, R_w 0.043 and w = $1/(\sigma^2(F_o))$, $\Delta\rho_{max}$ 0.68, $\Delta\rho_{min}$ -0.76 e Å^{-3}.

Data reduction and application of Lorentz, polarisation and analytical absorption corrections were carried out using teXsan [20]. The structure was solved by direct methods using SHELXS-86 [21] and refined using full-matrix least-squares methods with teXsan [20]. Hydrogen atoms were included at calculated sites with isotropic thermal parameters based on that of the riding atom and the non-hydrogen atoms were refined anisotropically. Neutral atom scattering factors were taken from International Tables [22]. Anomalous dispersion effects were included in F_c [23]; the values for Δf' and Δf'' were those of Creagh and McAuley [24]. The values for the mass attenuation coefficients are those of Creagh and Hubbell [25]. All other calculations were performed using the teXsan [20] crystallographic software package of Molecular Structure Corporation. An ORTEP [26] plot of **23** is shown in Figure 3 and selected bond lengths and angles are given in Tables II and III respectively.

1,1-Dimethylethyl 2-Oxo-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **5**.

To a solution of **4** [9] (200 mg, 0.93 mmole) in dry tetrahydrofuran (15 ml) under nitrogen was added Lawesson's reagent (228 mg, 0.55 mmole). The reaction mixture was stirred at room temperature for 24 hours. Solvent was removed under vacuum, to give after column chromatography (dichloromethane/hexane 1:1, dichloromethane, dichloromethane/ethyl acetate (1:1)) 163 mg (76%) of **5** as a bright yellow solid, mp 180-181°; ir: ν 3243 (NH), 3121 (NH), 1746 (C=O), 1721 (C=O), 1156 (C=S) cm^{-1}; ^{1}H nmr: δ 1.38 (s, 9H, CH_3), 3.07 (dd, 1H, J = 1.9, 17.2 Hz, CH_2), 3.21 (dd, 1H, J = 7.1, 17.2 Hz, CH_2), 4.03 (ddd, 1H, J = 1.9, 5.1, 7.1 Hz, *CH*), 8.17 (brs, 1H, N*H*), 11.82 (brs, 1H, N*H*).

Anal. Calcd. for $C_9H_{14}N_2O_3S$: C, 46.96; H, 6.09; N, 12.17. Found: C, 47.30; H, 6.22; N, 11.96.

2-Oxo-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylic Acid **3**.

To **5** (50 mg, 0.22 mmole) was added trifluoroacetic acid (1 ml) and the mixture stirred at room temperature for 20 minutes. Solvent was removed under vacuum and the residue slurried with ethyl acetate/ether (4 ml, 1:1) for 1 minute. The resulting precipitate was collected by filtration and washed with a little ether to yield 35 mg (93%) of **3** as a bright yellow solid, mp 240-242°; ir: ν 3233 (NH), 3180 (NH), 1723 (C=O), 1687 (C=O) cm^{-1}; uv: (water): λ max 280 nm (ε 17,000); ^{1}H nmr: δ 3.11 (dd, 1H, J = 2.5, 17.4 Hz, CH_2), 3.21 (dd, 1H, J = 7.1, 17.4 Hz, CH_2), 4.05 (ddd, 1H, J = 2.5, 4.6, 7.1 Hz, *CH*), 8.14 (brs, 1H, N*H*), 11.80 (brs, 1H, NH), 13.2 (brs, 1H, COO*H*); ^{13}C nmr: δ 41.7, 49.6, 149.8, 172.2, 203.2.

Anal. Calcd. for $C_5H_6N_2O_3S$: C, 34.48; H, 3.45; N, 16.09. Found: C, 34.62; H, 3.51; N, 15.86.

General Procedure for the Preparation of **7a, 7b** and **7c**.

The 5-alkylorotic acid **6** (32 mmoles) was dissolved in acetic acid (250 ml, 17 *M*) under nitrogen, and stirred at 65° for 1 hour, after which time excess zinc powder (8.0 g, 122.4 mmoles) was added. Heating and stirring were continued for a further 1.5 hours, when further zinc (3.0 g, 46 mmoles) was added. The mixture was heated at reflux for 45 minutes, filtered hot, and solvent removed under vacuum. The residue was treated with water (50 ml), hydrochloric acid (500 μl, 10 *M*) and the precipitate collected and dried to give **7a, 7b** and **7c** in yields of 47%, 69% and 52% respectively.

5-Methyl-2,6-dioxo-1,2,3,6-hexahydropyrimidine-4-carboxylic Acid **7a**.

This compound was obtained as a mixture of *cis* and *trans* isomers, (*cis:trans*, 95:5), mp 241-243° (lit [27], 238-240°).

5-Ethyl-2,6-dioxo-1,2,3,6-hexahydropyrimidine-4-carboxylic Acid **7b**.

This compound was obtained as a mixture of *cis* and *trans* isomers, (*cis:trans*, 3:1), mp 236-241° (water); ir: ν 3234 (NH), 1739 (C=O), 1720 (C=O), 1660 (C=O) cm^{-1}; *cis*-**7b**: ^{1}H nmr: δ 0.94 (dd, 3H, J = 6.1, 7.4 Hz, CH_3), 1.16-1.24 (m, 1H, CH_2CH_3), 1.80-1.90 (m, 1H, CH_2CH_3), 2.70 (dt, J = 6.6, 8.2 Hz, H-5), 3.97 (dd, J = 3.9, 6.6 Hz, H-4), 7.78 (brs, 1H, NH), 9.99 (brs, 1H, NH), 13.14 (brs, 1H, COO*H*); *trans*-**7b** ^{1}H nmr: δ 0.92 (t, 3H, J = 7.7 Hz, CH_3), 1.59-1.66 (m, 2H, CH_2), 2.46 (m, 1H, H-5), 3.82 (dd, 1H, J = 1.6, 4.3 Hz, H-4), 7.75 (brs, 1H, N*H*), 10.05 (brs, 1H, N*H*), 13.14 (brs, 1H, COO*H*).

Anal. Calcd. for $C_7H_{10}N_2O_4$: C, 45.16; H, 5.38; N, 15.05. Found: C, 45.22; H, 5.46; N, 14.83.

2,6-Dioxo-5-propyl-1,2,3,6-hexahydropyrimidine-4-carboxylic Acid **7c**.

This compound was obtained as a mixture of *cis* and *trans* isomers, (*cis:trans*, 7:3), mp 225-228° (water); ir: ν 3238 (NH), 1739 (C=O), 1720 (C=O), 1662 (C=O) cm^{-1}; *cis*-**7c** ^{1}H nmr: δ 0.88 (t, J = 7.6 Hz, CH_3), 1.32-1.43 (m, 2H, CH_2CH_3), 1.57 (m, 2H, $CH_2CH_2CH_3$), 2.76 (ddd, J = 6.3, 6.6, 7.7 Hz, H-5), 3.96 (dd, 1H, J = 3.9, 6.3 Hz, H-4), 7.77 (brs, 1H, N*H*), 10.00 (brs, 1H, N*H*), 13.13 (brs, 1H, COO*H*); *trans*-**7c** ^{1}H nmr: δ 0.86 (t, 3H, 7.5 Hz, CH_3), 1.11-1.20 (m, 1H, $CH_2CH_2CH_3$), 1.30-1.43 (m, 2H, CH_2CH_3), 1.70-1.80 (m, 1H, $CH_2CH_2CH_3$), 2.58 (dt, 1H, J = 0.8, 3.7 Hz, H-5), 3.80 (dd, 1H, J = 0.8, 2.2 Hz, H-4), 7.77 (brs, 1H, N*H*), 10.05 (brs, 1H, N*H*), 13.13 (brs, 1H, COO*H*).

Anal. Calcd. for $C_8H_{12}N_2O_4$: C, 48.00; H, 6.00; N, 14.00. Found: C, 48.16; H, 6.13; N, 13.77.

General Procedure for the Preparation of **8a, 8b** and **8c**.

To a suspension of **7** (2.9 mmoles) in methanol (20 ml) was added a few drops of concentrated sulphuric acid. The solution was heated at reflux for 2 hours and cooled overnight. The resulting precipitate was collected by filtration, washed with water and dried to give **8a, 8b** and **8c** in yields of 50%, 54% and 51% respectively.

Methyl 5-Methyl-2,6-dioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **8a**.

This compound was obtained as white needles as a mixture of *cis* and *trans* isomers, (*cis:trans* 8:1), *cis*-**8a** ^{1}H nmr: δ 0.99 (d, 3H, J = 7.1 Hz, $CHCH_3$), 2.96 (dq, 1H, J = 6.7, 7.1 Hz, H-5), 3.65 (s, 3H, OCH_3), 4.09 (dd, 1H, J = 4.0, 6.7 Hz, H-4), 7.86 (brs, 1H, N*H*), 10.10 (brs, 1H, N*H*).

Anal. Calcd. for $C_7H_{10}N_2O_4$: C, 45.16; H, 5.38; N, 15.05. Found: C, 45.26; H, 5.46; N, 15.00.

Methyl 5-Ethyl-2,6-dioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **8b**.

This compound was obtained as a white powder as a mixture of *cis* and *trans* isomers, (*cis:trans*, 1:1), mp 163-165°; *cis*-**8b** ^{1}H nmr:

δ 0.95 (t, 3H, J = 7.4 Hz, CH_2CH_3), 1.07-1.15 (m, 1H, CH_2CH_3), 1.78-1.84 (m, 1H, CH_2CH_3), 2.76 (dt, 1H, J = 6.5, 7.1 Hz, H-5), 3.65 (s, 3H, OCH_3), 4.12 (dd, 1H, J = 2.0, 6.5 Hz, H-4), 7.87 (brs, 1H, N*H*), 10.08 (brs, 1H, N*H*); *trans*-**8b** ^{1}H nmr: δ 0.94 (t, 3H, J = 7.4 Hz, CH_2CH_3), 1.63 (dq, 1H, J = 10.7, 7.4 Hz, CH_2CH_3), 2.54 (dt, 1H, J = 2.2, 10.7 Hz, H-5), 3.67 (s, 3H, OCH_3), 3.99 (dd, 1H, J = 2.2, 2.2 Hz, H-4), 7.88 (brs, 1H, N*H*), 10.13 (brs, 1H, N*H*).

Anal. Calcd. for $C_8H_{12}N_2O_4$: C, 48.00; H, 6.00; N, 14.00. Found: C, 48.32; H, 6.16; N, 13.74.

Methyl 5-Propyl-2,6-dioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **8c**.

This compound was obtained as a white powder as a mixture of *cis* and *trans* isomers, (*cis:trans,* 1:4), mp 165-167°; *trans*-**8c** ^{1}H nmr: δ 0.90 (t, 3H, J = 7.2 Hz, CH_2CH_3), 1.08-1.16 (m, 1H, $CH_2CH_2CH_3$), 1.32-1.45 (m, 2H, $CH_2CH_2CH_3$), 1.55-1.63 (m, 1H, $CH_2CH_2CH_3$), 2.62 (dt, 1H, J = 2.9, 7.1, Hz, H-5), 3.68 (s, 3H, OCH_3), 3.98 (dd, 1H, J = 2.9, 5.9 Hz, H-4), 7.88 (brs, 1H, N*H*), 10.14 (brs, 1H, N*H*).

Anal. Calcd. for $C_9H_{14}N_2O_4$: C, 50.47; H, 6.54; N, 13.08. Found: C, 50.63; H, 6.75; N, 12.81.

General Procedure for the Preparation of **9a, 9b,** and **9c**.

To a solution of **8** (1.33 mmoles) in dry tetrahydrofuran (20 ml) was added Lawesson's reagent (325 mg, 0.78 mmole) and the mixture stirred at room temperature for 36 hours. Solvent was removed under vacuum and the residue purified *via* column chromatography eluting with dichloromethane followed by dichloromethane/ethyl acetate (3:1) to give **9a, 9b** and **9c** in yields of 61, 69 and 62% respectively.

Methyl 5-Methyl-2-oxo-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **9a**.

This compound was obtained as a yellow solid consisting of a mixture of *cis* and *trans* isomers, (*cis:trans,* 2:3), mp 175-177°; ir: ν 3224 (NH), 3115 (NH), 1744 (C=O), 1711 (C=O) cm^{-1}; *cis*-**9a** ^{1}H nmr: δ 1.26 (d, 3H, J = 7.2 Hz, $CHCH_3$), 3.29 (dq, 1H, J = 3.7, 7.2 Hz, H-5), 3.68 (s, 3H, OCH_3), 4.24 (dd, 1H, J = 3.7, 6.8 Hz, H-4), 7.39 (brs, 1H, N*H*), 11.15 (brs, 1H, N*H*); *trans*-**9a** ^{1}H nmr: δ 1.37 (d, 3H, J = 7.2 Hz, $CHCH_3$), 3.23 (dq, 1H, J = 1.9, 7.2 Hz, H-5), 3.64 (s, 3H, OCH_3), 3.75 (dd, 1H, J = 1.9, 4.3 Hz, H-4), 7.87 (brs, 1H, N*H*), 11.46 (brs, 1H, N*H*).

Anal. Calcd. for $C_7H_{10}N_2O_3S$; C, 41.58; H, 4.95; N, 13.86. Found: C, 41.65; H, 5.06; N, 13.59.

Methyl 5-Ethyl-2-oxo-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **9b**.

This compound was obtained as a pale yellow powder as a mixture of *cis* and *trans* isomers (*cis:trans* 1:4), mp 195-197°; ir: ν 3223 (NH), 3109 (NH), 1750 (C=O), 1709 (C=O), 1160 (C=S) cm^{-1}; *cis*-**9b** ^{1}H nmr: δ 0.98 (t, 3H, J = 7.3 Hz, CH_2CH_3), 1.60-1.75 (m, 2H, CH_2CH_3), 2.81 (m, 1H, H-5), 3.69 (s, 3H, OCH_3), 4.30 (dd, 1H, J = 3.4, 5.4 Hz, H-4), 8.18 (brs, 1H, N*H*), 11.79 (brs, 1H, N*H*); *trans*-**9b** ^{1}H nmr: δ 1.07 (t, 3H, J = 7.3 Hz, CH_2CH_3), 1.70 (dq, 2H, J 7.3, 7.3 Hz, CH_2CH_3), 3.01 (dd, 1H, J = 1, 7.3 Hz, H-5), 3.66 (s, 3H, OCH_3), 4.06 (dd, 1H, J = 1.0, 4.4 Hz, H-4), 8.23 (brs, 1H, N*H*), 11.82 (brs, 1H, N*H*).

Anal. Calcd. for $C_8H_{12}N_2O_3S$; C, 44.44; H, 5.55; N, 12.96. Found: C, 44.64; H, 5.79; N, 12.73.

Methyl 5-Propyl-2-oxo-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **9c**.

This compound was obtained as a pale yellow powder as a mixture of *cis* and *trans* isomers, (*cis:trans,* 1:5), 187-189°; ir: ν 3227 (NH), 3109 (NH), 1751 (C=O), 1714 (C=O), 1160 (C=S) cm^{-1}; *cis*-**9c** ^{1}H nmr: δ 0.83 (t, 3H, J = 7.2 Hz, CH_2CH_3), 1.32-1.82 (m, 4H, $CH_2CH_2CH_3$), 2.97 (m, 1H, H-5), 3.64 (s, 3H, OCH_3), 4.26 (dd, 1H, J = 2.1, 4.5 Hz, H-4), 7.52 (brs, 1H, N*H*), 11.32 (brs, 1H, N*H*); *trans*-**9c** ^{1}H nmr: δ 0.87 (t, 3H, J = 7.2 Hz, CH_2CH_3), 1.32-1.82 (m, 4H, $CH_2CH_2CH_3$), 3.14 (dt, 1H, J = 1.3, 7.4 Hz, H-5), 3.68 (s, 3H, OCH_3), 3.82 (dd, 1H, J = 1.3, 4.6 Hz, H-4), 7.87 (brs, 1H, NH), 11.37 (brs, 1H, N*H*).

Anal. Calcd. for $C_9H_{14}N_2O_3S$: C, 46.96; H, 6.09; N, 12.17. Found: C, 47.15; H, 6.15; N, 11.91.

General Procedure for the Preparation of **10a, 10b** and **10c**.

To a suspension of **7** (14.5 mmoles) in dimethylformamide (25 ml) was added *p*-methoxybenzyl chloride (2.27 g, 14.5 mmoles) and triethylamine (1.45 g, 14.40 mmoles). The solution was stirred for 48 hours at room temperature and the solvent then removed under high vacuum at 45-50°. The residue was treated with water (50 ml) for 10 minutes and the resulting precipitate collected by filtration to give **10a, 10b** and **10c** in yields of 45, 50 and 53% respectively.

p-Methoxybenzyl 5-Methyl-2,6-dioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **10a**.

This compound was obtained as a white solid as a mixture of *cis* and *trans* isomers, (*cis:trans,* 6:1), mp 175-177°; ir: ν 3260 (NH), 1715 (C=O), 840, 810 cm^{-1}; *cis*-**10a** ^{1}H nmr: δ 0.94 (d, 3H, J = 7.1 Hz, $CHCH_3$), 2.93 (dq, 1H, J = 6.1, 7.1 Hz, H-5), 3.74 (s, 3H, OCH_3), 4.08 (dd, 1H, J = 3.1, 6.1 Hz, H-4), 5.03-5.09 (m, 2H, OCH_2Ar), 6.91 (d, 2H, J = 8.6 Hz, Ar-*H*), 7.28 (d, 2H, J = 8.6 Hz, Ar-*H*), 7.87 (brs, 1H, N*H*), 10.10 (brs, 1H, N*H*).

Anal. Calcd. for $C_{14}H_{16}N_2O_5$: C, 57.53; H, 5.48; N, 9.59. Found: C, 57.69; H, 5.66; N, 9.43.

p-Methoxybenzyl 5-Ethyl-2,6-dioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **10b**.

This compound was obtained as an off white solid as a mixture of *cis* and *trans* isomers, (*cis:trans,* 3: 2), mp 156-158°; ir: ν 3200 (NH), 1720 (C=O), 815 cm^{-1}; *cis*-**10b** ^{1}H nmr: δ 0.93 (dd, 3H, J = 6.1, 7.7 Hz, CH_2CH_3), 1.15-1.26 (m, 1H, CH_2CH_3), 1.74-1.81 (m, 1H, CH_2CH_3) 2.77 (dt, 1H, J = 6.3, 6.1, 6.4 Hz, H-5), 3.74 (s, 3H, OCH_3), 4.11 (dd, 1H, J = 2.0, 6.3 Hz, H-4), 5.03-5.10 (m, 2H, OCH_2Ar), 6.85-6.92 (m, 2H, Ar-*H*), 7.20-7.29 (m, 2H, Ar-*H*), 7.89 (brs, 1H, N*H*), 10.08 (brs, 1H, N*H*).

Anal. Calcd. for $C_{15}H_{18}N_2O_5$: C, 58.82; H, 5.88; N, 9.15. Found: C, 58.99; H, 6.03; N, 8.98.

p-Methoxybenzyl 2,6-Dioxo-5-propyl-1,2,3,6-hexahydropyrimidine-4-carboxylate **10c**.

This compound was obtained as an off white solid as a mixture of *cis* and *trans* isomers, (*cis:trans,* 1:1), mp 160-164°; ir: ν 3310 (NH), 1700 (C=O), 840 cm^{-1}; *cis*-**10c** ^{1}H nmr: δ 0.86 (t, 3H, J = 7.3 Hz, CH_2CH_3), 1.16-1.70 (m, 4H, $CH_2CH_2CH_3$), 2.81 (dt, 1H, J = 6.4, 7.6 Hz, H-5), 3.74 (s, 3H, OCH_3), 4.09 (dd, 1H, J = 4.0, 6.4 Hz, H-4), 5.02 (d, 1H, J = 12.0 Hz, OCH_2Ar), 5.11 (d, 1H, J = 12.0 Hz, OCH_2Ar), 6.87-6.92 (m, 2H, Ar-*H*), 7.26-7.30 (m, 2H, Ar-*H*), 7.88 (brs, 1H, N*H*), 10.08 (brs, 1H, N*H*); *trans*-**10c** ^{1}H nmr: δ 0.77 (t, 3H, J = 7.3 Hz, CH_2CH_3), 1.16-1.70 (m, 4H, $CH_2CH_2CH_3$), 2.58 (dt, 1H, J = 0.8, 7.3 Hz, H-5), 3.74 (s, 3H, OCH_3), 3.97 (dd, 1H, J = 0.8, 2.8 Hz, H-4), 5.08 (s, 2H, OCH_2Ar), 6.87-6.92 (m, 2H, Ar-*H*), 7.26-7.30 (m, 2H, Ar-*H*), 7.88 (brs, 1H, N*H*), 10.12 (brs, 1H, N*H*).

Anal. Calcd. for $C_{16}H_{20}N_2O_5$: C, 60.00; H, 6.25; N, 8.75. Found: C, 60.17; H, 6.37; N, 8.71.

General Procedure for the Preparation of **11a, 11b** and **11c**.

To a suspension of **10** (5.13 mmoles) in anhydrous dioxane (15 ml) was added phosphorus pentasulfide (0.6 g, 2.7 mmoles). The solution was heated at reflux for 2 hours and the solvent removed under vacuum. The residue was purified *via* column chromatography, eluting sequentially with dichloromethane/hexane (9:1, v/v), dichloromethane, and finally dichloromethane/ethyl acetate (4:1, v/v). The yellow band eluting with dichloromethane/ethyl acetate was evaporated, and the residue recrystallised from ethyl acetate to give **11a, 11b** and **11c** in yields of 18, 21 and 16% respectively.

p-Methoxybenzyl 5-Methyl-2-oxo-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **11a**.

This compound was obtained as yellow crystals as a mixture of *cis* and *trans* isomers, (*cis:trans*, 5:1), mp 154-156°; ir: ν 3242 (NH), 3197 (NH), 1733 (C=O), 1693 (C=O) cm^{-1}; *cis*-**11a** ^1H nmr: δ 1.13 (d, 3H, J = 7.1 Hz, $CHCH_3$), 3.13 (dq, 1H, J = 6.0, 7.1 Hz, H-5), 3.75 (s, 3H, OCH_3), 4.27 (dd, 1H, J = 3.3, 6.0 Hz, H-4), 5.07 (s, 2H, OCH_2Ar), 6.92 (d, 2H, J = 8.6 Hz, Ar-*H*), 7.29 (d, 2H, J = 8.6 Hz, Ar-*H*), 8.19 (brs, 1H, N*H*), 11.80 (brs, 1H, N*H*).

Anal. Calcd. for $C_{14}H_{16}N_2O_4S$: C, 54.55; H, 5.19; N, 9.09. Found: C, 54.74; H, 5.32; N, 8.93.

p-Methoxybenzyl 5-Ethyl-2-oxo-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **11b**.

This compound was obtained as a yellow powder as a mixture of *cis* and *trans* isomers, (*cis:trans* 2:5); mp 159-163°; ir: ν 3220 (NH), 1710 (C=O), 820 cm^{-1}; *cis*-**11b** ^1H nmr: δ 0.93 (t, 3H, J = 7.4 Hz, CH_2CH_3), 1.15 (m, 1H, CH_2CH_3), 2.04 (m, 1H, CH_2CH_3), 2.91 (dt, 1H, J = 5.9, 7.4 Hz, H-5), 3.75 (s, 3H, OCH_3), 4.30 (dd, 1H, J = 3.4, 5.9 Hz, H-4), 5.07 (s, 2H, CH_2Ar), 6.92 (d, 2H, J =8.8 Hz, Ar-*H*), 7.30 (d, 2H, J = 8.8 Hz, Ar-*H*), 8.20 (brs, 1H, N*H*), 11.78 (brs, 1H, N*H*); *trans*-**11b** ^1H nmr: δ 0.98 (t, 3H, J = 7.2 Hz, CH_2CH_3), 1.63-1.78 (m, 2H, CH_2CH_3), 3.02 (dt, 1H, J = 1.5, 7.4 Hz, H-5), 3.74 (s, 3H, OCH_3), 4.07 (dd, 1H, J = 1.5, 4.6 Hz, H-4), 5.07-5.14 (m, 2H, OCH_2Ar), 6.91 (d, 2H, J = 8.3 Hz, Ar-*H*), 7.28 (d, 2H, J = 8.3 Hz, Ar-*H*), 8.20 (brs, 1H, N*H*), 11.82 (brs, 1H, N*H*).

Anal. Calcd. for $C_{15}H_{18}N_2O_4S$: C, 55.90; H, 5.59; N, 8.70. Found: C, 55.63; H, 5.73; N, 8.54.

p-Methoxybenzyl 2-Oxo-5-propyl-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylate **11c**.

This compound was obtained as a yellow powder as a mixture of *cis* and *trans* isomers, (*cis:trans* 15:85), mp 163-165°; ir: ν 3370 (NH), 3170 (NH), 1740 (C=O), 1700 (C=O), 805 cm^{-1}; *trans*-**11c** ^1H nmr: δ 0.88 (t, 3H, J = 7.3 Hz, CH_2CH_3), 1.33-1.51 (m, 2H, $CH_2CH_2CH_3$), 1.54-1.71 (m, 2H, $CH_2CH_2CH_3$), 3.12 (dt, 1H, J = 1.6, 7.2 Hz, H-5), 3.75 (s, 3H, OCH_3), 4.04 (dd, 1H, J = 1.6, 5.0 Hz, H-4), 5.09 (s, 2H, OCH_2Ar), 6.91 (d, 2H, J = 8.8 Hz, Ar-*H*), 7.28 (d, 2H, J = 8.8 Hz, Ar-*H*), 8.24 (brs, 1H, N*H*), 11.82 (brs, 1H, N*H*).

Anal. Calcd. for $C_{16}H_{20}N_2O_4S$: C, 57.14; H, 5.95; N, 8.33. Found: C, 57.34; H, 5.93; N, 8.30.

General Procedure for the Preparation of **12a, 12b** and **12c.**

Method 1.

To **11** (0.20 mmole) was added trifluoroacetic acid (1 ml) and the mixture stirred at room temperature for 30 minutes. Solvent was removed and the residue slurried with ethyl acetate/ether (4 ml, 1:1) for 5 minutes. The resulting precipitate was collected by filtration and washed with ether to give **12a, 12b** and **12c** in yields of 48, 62 and 42% respectively.

5-Methyl-2-oxo-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylic Acid **12a**.

This compound was obtained as a yellow powder as a mixture of *cis* and *trans* isomers, (*cis:trans* 3:17), mp 206-209°; ir: ν 3346 (NH), 3250 (NH), 1741 (C=O), 1670 (C=O), 1157 (C=S) cm^{-1}; uv: (methanol): λ max 282 nm (ε 10,038), *cis*-**12a** ^1H nmr: δ 1.21 (d, 3H, J = 6.8 Hz, $CHCH_3$), 3.11 (dq, 1H, J = 5.8, 6.8 Hz, H-5), 4.14 (dd, 1H, J = 2.8, 5.8 Hz, H-4), 8.03 (brs, 1H, N*H*), 11.75 (brs, 1H, N*H*), 13.15 (brs, 1H, COO*H*); *trans*-**12b** ^1H nmr: δ 1.34 (d, 3H, J = 7.2 Hz, $CHCH_3$), 3.23 (dq, 1H, J = 2.0, 7.2 Hz, H-5), 3.84 (dd, 1H, J = 2.0, 4.4 Hz, H-4), 8.16 (brs, 1H, N*H*), 11.78 (brs, 1H, N*H*), 13.15 (brs, 1H, COO*H*).

Anal. Calcd. for $C_6H_8N_2O_3S$: C, 38.30; H, 4.26; N, 14.89. Found: C, 38.45; H, 4.49; N, 14.63.

5-Ethyl-2-oxo-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylic Acid **12b**.

This compound was obtained as a yellow powder as a mixture of *cis* and *trans* isomers, (*cis:trans* 1:5), mp 170-172°; *cis*-**12b**: ^1H nmr: δ 0.96 (t, 3H, J = 7.3 Hz, CH_2CH_3), 1.61-1.90 (m, 2H, CH_2CH_3), 2.93 (dt, 1H, J = 5.7, 7.8 Hz, H-5), 4.09 (dd, 1H, J = 2.5, 5.7 Hz, H-4), 7.65 (brs, 1H, N*H*), 11.78 (brs, 1H, N*H*), 13.10 (brs, 1H, COO*H*); *trans*-**12b**, ^1H nmr: δ 1.02 (t, 3H, J = 7.3 Hz, CH_2CH_3), 1.55-1.71 (m, 2H, CH_2CH_3), 3.01 (dt, 1H, J = 1.5, 7.2 Hz, H-5), 3.88 (dd, 1H, J = 1.5, 4.9 Hz, H-4), 8.16 (brs, 1H, N*H*), 11.78 (brs, 1H, N*H*), 13.10 (brs, 1H, COO*H*).

Anal. Calcd. for $C_7H_{10}N_2O_3S$: C, 41.58; H, 4.95; N, 13.86. Found: C, 41.69; H, 5.22; N, 13.80.

2-Oxo-5-propyl-6-thioxo-1,2,3,6-hexahydropyrimidine-4-carboxylic Acid **12c**.

Preparation from **11c** according to the method described for **3** afforded 42% of the titled compound **12c** as a yellow powder as a mixture of *cis* and *trans* isomers, (*cis:trans* 1:5), mp 173-175°; ir: ν 3342 (NH), 3250 (NH), 1744 (C=O), 1671 (C=O), 1155 (C=S) cm^{-1}; uv: (methanol): δ max 282 nm (ε = 12,331); *cis*-**12c** ^1H nmr: δ 0.88 (t, 3H, J = 6.8 Hz, CH_3), 1.25-1.61 (m, 3H, $CH_2CH_2CH_3$), 1.85-1.93 (m, 1H, $CH_2CH_2CH_3$), 2.99 (dt, 1H, J = 5.8, 6.1 Hz, H-5), 4.17 (dd, 1H, J = 3.0, 5.8 Hz, H-4), 8.00 (brs, 1H, N*H*), 11.72 (brs, 1H, N*H*), 13.10 (brs, 1H, N*H*); *trans*-**12c** ^1H nmr: δ 0.91 (t, 3H, J = 7.2 Hz, CH_2CH_3), 1.35-1.51 (m, 2H, $CH_2CH_2CH_3$), 1.58-1.68 (2H, m, $CH_2CH_2CH_3$), 3.12 (dt, 1H, J = 1.4, 7.2 Hz, H-5), 3.85 (dd, 1H, J = 1.4, 4.8 Hz, H-4), 8.10 (brs, 1H, N*H*), 11.76 (brs, 1H, N*H*), 13.10 (brs, 1H, COO*H*).

Anal. Calcd. for $C_8H_{12}N_2O_3S$: C, 44.44; H, 5.56; N, 12.96. Found: C, 44.61; H, 5.73; N, 12.79.

Method 2.

To a suspension of **15** (1.61 mmoles) in de-oxygenated acetic acid (45 ml) was added zinc dust (200 mg, 3.17 mmoles) and the mixture heated under nitrogen at 60° with stirring until reaction was complete (~ 1-2 hours). The bright yellow solution was allowed to cool, filtered and the filtrate reduced in volume to 15 ml at 20-30°. The concentrate was treated with a solution of oxalic acid (145 mg, 1.61 mmoles) in acetic acid (5 ml). The mixture was stirred at room temperature for 15 minutes, filtered and the filtrate lyophilised to afford **12a, 12b** and **12c** in yields of 83, 78 and 77%

respectively. Analysis by ^{1}H nmr indicated that the compounds were all composed largely (85-90%) of the *cis* isomer.

2,6-Dioxo-5-propyl-1,2,3,6-tetrahydropyrimidine-4-carboxylic Acid **6c**.

Preparation from 2-(2,5-dioxo-4-imidazolidnylidene)pentanoic acid according to the method of Laursen *et. al.* [10] afforded after recrystallisation from water, the titled compound **6c** (64%) as a white solid, mp 269-271°; ^{1}H nmr: δ 0.83 (t, 3H, J = 7.4 Hz, CH_3), 1.34-1.40 (m, 2H, CH_2CH_3), 2.37 (t, 2H, J = 7.5 Hz, $CH_2CH_2CH_3$), 10.64 (brs, 1H, N*H*), 11.28 (brs, 1H, N*H*), 13.13 (brs, 1H, COO*H*).

Anal. Calcd. for $C_8H_{10}N_2O_4$: C, 48.48; H, 5.05; N, 14.14. Found: C, 48.59; H, 4.93; N, 14.06.

General Procedure for the Preparation of **13a, 13b** and **13c**.

To a suspension of **6** (38.0 mmoles) in dimethylformamide (300 ml) was added cesium carbonate (5.2 g, 16.0 mmoles) and iodomethane (4.64 g, 38.0 mmoles). The solution was stirred overnight and the solvent removed under high vacuum (0.1 mm Hg) at 45-50°. The residue was treated with water (50-100 ml), and the product collected by filtration to give after recrystallisation from methanol **13a, 13b** and **13c** in yields of 66, 20 and 33% respectively.

Methyl 5-Methyl-2,6-dioxo-1,2,3,6-tetrahydropyrimidine-4-carboxylate **13a**.

This compound was obtained as a white solid, mp 253-255°; ^{1}H nmr: δ 1.91 (s, 3H, CCH_3), 3.82 (s, 3H, OCH_3), 10.75 (brs, 1H, N*H*), 11.39 (brs, 1H, N*H*).

Anal. Calcd. for $C_7H_8N_2O_4$: C, 45.65; H, 4.35; N, 15.22. Found: C, 45.31; H, 4.26; N, 15.23.

Methyl 5-Ethyl-2,6-dioxo-1,2,3,6-tetrahydropyrimidine4-carboxylate **13b**.

This compound was obtained as white crystals, mp 215-217°; ir: ν 1720 (C=O), 1650 (C=O); ^{1}H nmr: δ 0.96 (t, 3H, J = 7.3 Hz, CH_2CH_3), 2.37, (q, 2H, J = 7.3 Hz, CH_2CH_3), 3.83 (s, 3H, OCH_3), 10.81 (brs, 1H, N*H*), 11.37 (brs, 1H, N*H*).

Anal. Calcd. for $C_8H_{10}N_2O_4$: C, 48.48; H, 5.05; N, 14.14. Found: C, 48.19; H, 4.82; N, 14.06.

Methyl 2,6-Dioxo-5-propyl-1,2,3,6-tetrahydropyrimidine-4-carboxylate **13c**.

This compound was obtained as white crystals, mp 193-195°; ir: ν 3490, 3350, 1710 (C=O), 1650 (C=O); ^{1}H nmr: δ 0.83 (t, 3H, J = 7.3 Hz, CH_2CH_3), 1.33-1.40 (m, 2H, CH_2CH_3), 2.33, (t, 2H, J = 7.3 Hz, $CH_2CH_2CH_3$), 3.82 (s, 3H, OCH_3), 10.83 (brs, 1H, N*H*), 11.36 (brs, 1H, N*H*).

Anal. Calcd. for $C_9H_{12}N_2O_4$: C, 50.94; H, 5.66; N, 13.21. Found: C, 50.79; H, 5.61; N, 13.11.

General Procedure for the Preparation of **14a, 14b** and **14c**.

To a-suspension of **13** (26.1 mmoles) in dioxane (125 ml) was added phosphorus pentasulfide (1.7 g, 10.7 mmoles). The mixture was heated at reflux for 24 hours. Solvent removed under vacuum and the residue treated with dilute hydrochloric acid (0.1 *M*, 100 ml) for 10 minutes. The product was collected to give after recrystallisation from methanol **14a, 14b** and **14c** in yields of 69, 79 and 85% respectively.

Methyl 5-Methyl-2-oxo-6-thioxo-1,2,3,6-tetrahydropyrimidine-4-carboxylate **14a**.

This compound was obtained as orange needles, mp 209-211°; ir: ν 3240 (NH), 1710 (C=O), 1070 (C=S); ^{1}H nmr: δ 2.10 (s, 3H, CCH_3), 3.85 (s, 3H, OCH_3), 11.58 (brs, 1H, N*H*), 12.84 (brs, 1H, N*H*).

Anal. Calcd. for $C_7H_8N_2O_3S$: C, 42.00; H, 4.00; N, 14.00. Found: C, 41.69; H, 3.81; N, 13.93.

Methyl 5-Ethyl-2-oxo-6-thioxo-1,2,3,6-tetrahydropyrimidine-4-carboxylate **14b**.

This compound was obtained as an orange powder, mp 212-215°; ir: ν 1741 (C=O), 1704 (C=O) cm^{-1}; ^{1}H nmr: δ 1.00 (t, 3H, J = 7.2 Hz, CH_2CH_3), 2.64, (q, 2H, J = 7.2 Hz, CH_2CH_3), 3.85 (s, 3H, OCH_3), 11.59 (brs, 1H, N*H*), 12.80 (brs, 1H, N*H*).

Anal. Calcd. for $C_8H_{10}N_2O_3S$: C, 44.86; H, 4.67; N, 13.08. Found: C, 44.72; H, 4.65; N, 12.99.

Methyl 2-Oxo-5-propyl-6-thioxo-1,2,3,6-tetrahydropyrimidine-4-carboxylate **14c**.

This compound was obtained as an orange powder, mp 189-191°; ir: ν 1745 (C=O), 1710 (C=O), 1140 (C=S) cm^{-1}; ^{1}H nmr: δ 0.84 (t, 3H, J = 7.3 Hz, CH_2CH_3), 1.40-1.46 (m, 2H, CH_2CH_3), 2.59 (t, 2H, J = 7.3 Hz, $CH_2CH_2CH_3$), 3.84 (s, 3H, OCH_3), 11.60 (brs, 1H, N*H*), 12.79 (brs, 1H, N*H*).

Anal. Calcd. for $C_9H_{12}N_2O_3S$: C, 47.37; H, 5.26; N, 12.28. Found: C, 47.26; H, 5.13; N, 12.03.

General Procedure for the Preparation of **15a, 15b** and **15c**.

To a suspension of **14** (15.0 mmoles) in methanol (40 ml) was added water (20 ml) and potassium hydroxide solution (20 ml, 1*M*). The mixture was stirred at room temperature for 1 hour. The mixture was acidified to *p*H 2 with hydrochloric acid (1*M*). Most of the methanol was removed under vacuum and the resulting orange solid collected and washed with water. Recrystallisation from dimethylformamide/ethanol afforded **15a, 15b** and **15c** in yields of 65, 86 and 56% respectively.

5-Methyl-2-oxo-6-thioxo-1,2,3,6-tetrahydropyrimidine-4-carboxylic Acid **15a**.

This compound was obtained as an orange powder, mp 275°; ir: ν 3380 (NH), 3200 (NH), 1665 (C=O); ^{1}H nmr: δ 2.11 (s, 3H, CCH_3), 11.44 (brs, 1H, N*H*), 12.70 (brs, 1H, N*H*), 13.82 (brs, 1H, COO*H*).

Anal. Calcd. for $C_6H_6N_2O_3S$: C, 38.71; H, 3.23; N, 15.05. Found: C, 38.51; H, 3.10; N, 14.87.

5-Ethyl-2-oxo-6-thioxo-1,2,3,6-tetrahydropyrimidine-4-carboxylic Acid **15b**.

This compound was obtained as an orange powder, mp > 275°; ir: ν 3200 (NH), 1710 (C=O), 1680 (C=O) cm^{-1}; uv: (methanol): λ max 348 nm (ε 10,430); ^{1}H nmr: δ 1.01 (t, 3H, J = 7.1 Hz, CH_2CH_3), 2.67, (q, 2H, J = 7.1 Hz, CH_2CH_3), 11.52 (brs, 1H, N*H*), 12.70 (brs, 1H, N*H*), 13.68 (brs, 1H, COO*H*).

Anal. Calcd. for $C_7H_8N_2O_3S$: C, 42.00; H, 4.00; N, 14.00. Found: C, 41.83; H, 3.91; N, 13.81.

2-Oxo-5-propyl-6-thioxo-1,2,3,6-tetrahydropyrimidine-4-carboxylic Acid **15c**.

This compound was obtained as an orange powder, mp 266-268°; ir: ν 3174 (NH), 1705 (C=O), 1599 (C=C), 1151 (C=S) cm^{-1}; ^{1}H nmr: δ 0.83 (t, 3H, J = 7.3 Hz, CH_2CH_3), 1.41-1.47 (m, 2H, CH_2CH_3), 2.63 (t, 2H, J = 7.3 Hz,

$CH_2CH_2CH_3$), 11.46 (brs, 1H, N*H*), 12.63 (brs, 1H, N*H*), 13.81 (brs, 1H, COO*H*).

Anal. Calcd. for $C_8H_{10}N_2O_3S$: C, 44.86; H, 4.67; N, 13.08. Found: C, 44.63; H, 4.60; N, 12.99.

2-[(Aminocarbonyl)aminocarbonyl]-1-cyclopentenecarboxylic Acid **18**.

To a solution of 1-cyclopentene-1,2-dicarboxylic anhydride **17** (100 mg, 0.72 mmole) in glacial acetic acid (500 µl) was added urea (43 mg, 0.72 mmole) and the mixture heated with stirring at 50° for 16 hours. The mixture was allowed to stand at room temperature overnight and the resulting crystals collected by filtration and washed with ether (1 ml) to yield 112 mg (78%) of the titled compound **18** as a white solid, mp 220°; ir: ν 3390 (NH), 3205 (NH), 3150 (NH), 1697 (C=O) cm^{-1}; ^1H nmr: δ 1.86 (m, 2H, $CH_2CH_2CH_2$), 2.56 (t, 2H, J = 7.7 Hz, $CH_2CH_2CH_2$), 2.66 (t, 2H, J = 7.7 Hz, $CH_2CH_2CH_2$), 7.23 (brs, 1H, N*H*), 7.63 (brs, 1H, N*H*), 10.41 (brs, 1H, N*H*), 12.60 (brs, 1H, COO*H*).

Anal. Calcd. for $C_8H_{10}N_2O_4$: C, 48.48; H, 5.05; N, 14.14. Found: C, 48.40; H, 5.25; N, 14.11.

1-Aminocarbonyl-1-aza-2,8-dioxobicyclo [3.3.0]-3-octene **19**.

A suspension of **18** (1 g, 5.05 mmoles) in acetic anhydride (3 ml) was heated at 95° with stirring for 30 minutes after which time **18** had entered solution. The mixture was cooled to room temperature and the resulting solid collected by filtration and washed with acetone (2 ml) to yield 0.77 g (85%) of the titled compound **19** as fine needles, mp 183°; ir: ν 3424 (NH), 3254 (NH), 1790 (C=O), 1731 (C=O) cm^{-1}; ^1H nmr: δ 2.35 (m, 2H, $CH_2CH_2CH_2$), 2.59 (t, 4H, J = 7.7 Hz, $CH_2CH_2CH_2$), 7.25 (brs, 1H, N*H*), 7.61 (brs, 1H, N*H*).

Anal. Calcd. for $C_8H_8N_2O_3$: C, 53.33; H, 4.44; N, 15.55. Found: C, 52.99; H 4.34; N, 15.09.

General Procedure for the Preparation of **20a, 20b, 20c** and **20d**.

To a suspension of **19** (750 mg, 4.17 mmoles) in the alcohol (2 ml) was added zinc chloride (10 mg) and the mixture stirred at 70° for 1 hour. Afer cooling the suspension was diluted with ether (2 ml) and the precipitate collected by filtration and washed with ether to afford the following esters.

Methyl 2-[(Aminocarbonyl)aminocarbonyl]-1-cyclopentenoate **20a**.

Employing methanol as the alcohol afforded 93% of **20a** as a white solid, mp 192°; ir: ν 3370 (NH), 3308 (NH), 3233 (NH), 1717 (C=O), 1632 cm^{-1}; ^1H nmr: δ 1.89 (m, 2H, $CH_2CH_2CH_2$), 2.59 (t, 2H, J = 7.6 Hz, $CH_2CH_2CH_2$), 2.68 (t, 2H, J = 7.6 Hz, $CH_2CH_2CH_2$), 3.64 (s, 3H, OCH_3), 7.27 (brs, 1H, N*H*), 7.60 (brs, 1H, N*H*), 10.34 (brs, 1H, N*H*).

Anal. Calcd. for $C_9H_{12}N_2O_4$: C, 50.94; H, 5.66; N, 13.21. Found: C, 50.89; H 5.84; N, 13.00.

4'-Methoxyphenylmethyl 2-[(Aminocarbonyl)aminocarbonyl]-1-cyclopentenoate **20b**.

Employing *p*-methoxybenzyl alcohol afforded 79% of the titled compound **20b** as a white solid after recrystallisation from acetone, mp 170-171°, ir: ν 3284 (NH), 3329 (NH), 3240 (NH), 1710 (C=O), 1663 cm^{-1}; ^1H nmr: δ 1.88 (m, 2H, $CH_2CH_2CH_2$), 2.59 (t, 2H, J = 7.4 Hz, $CH_2CH_2CH_2$), 2.68 (t, 2H, J = 7.4 Hz, $CH_2CH_2CH_2$), 3.74 (s, 3H, OCH_3), 5.05 (s, 2H, OCH_2), 6.89 (d, 2H, J = 8.6 Hz, Ar*H*), 7.26 (brs, 1H, N*H*), 7.27 (d, 2H, J = 8.6 Hz, Ar*H*), 7.57 (brs, 1H, N*H*), 10.38 (brs, 1H, N*H*).

Anal. Calcd. for $C_{16}H_{18}N_2O_5$: C, 60.38; H, 5.66; N, 8.81. Found: C, 60.23; H 5.63; N, 8.76.

Ethyl 2-[(Aminocarbonyl)aminocarbonyl]-1-cyclopentenoate **20c**.

Employing ethanol afforded 86% of the titled compound **20c** as a white solid after recrystallisation from ethanol, mp 165°, ir: ν 3412 (NH), 3319 (NH), 3235 (NH), 1721 (C=O), 1683 (C=O), 1637 (C=C) cm^{-1}; ^1H nmr: δ 1.17 (t, 3H, J = 7.2 Hz, CH_2CH_3), 1.79 (m, 2H, $CH_2CH_2CH_2$), 2.35-2.75 (m, 4H, J = 7.6 Hz, $CH_2CH_2CH_2$), 3.99 (q, 2H, J = 7.2 Hz, CH_2CH_3), 7.15 (brs, 1H, N*H*), 7.50 (brs, 1H, N*H*), 10.35 (brs, 1H, N*H*).

Anal. Calcd. for $C_{10}H_{14}N_2O_4$: C, 53.10; H, 6.19; N, 12.39. Found: C, 53.25; H 6.23; N, 12.45.

2'-Phenyl-2'-oxoethyl 2-[(Aminocarbonyl)aminocarbonyl]-1-cyclopentenoate **20d**.

Employing phenacyl alcohol afforded 69% of the titled compound **20d** as a white solid, mp 213-214°; ir: ν 3382 (NH), 3325 (NH), 3219 (NH), 1722 (C=O), 1692, 1635 cm^{-1}; ^1H nmr: δ 1.91-1.99 (m, 2H, $CH_2CH_2CH_2$), 2.65-2.75 (m, 4H, J = 7.6 Hz, $CH_2CH_2CH_2$), 5.54 (s, 2H, CH_2), 7.25 (brs, 1H, N*H*), 7.55 (t, 2H, J = 7.4 Hz, Ar-*H*), 7.58 (brs, 1H, N*H*), 7.69 (t, 1H, J = 7.4 Hz, Ar-*H*), 7.95 (d, 2H, J = 7.4 Hz, Ar-*H*), 10.37 (brs, 1H, N*H*).

Anal. Calcd. for $C_{16}H_{16}N_2O_5$: C, 60.76; H, 5.06; N, 8.86. Found: C, 60.91; H, 5.12; N, 8.76.

2'-Bromo-4'-methoxyphenylmethyl 2-Amino-6-bromo-1-oxa-4-oxo-3-azaspiro[4.4]non-2-ene-6-carboxylate **25**.

To a solution of **20b** (500 mg, 1.57 mmoles) in dry dimethyl formamide (2 ml) was added pyridine (261 mg, 3.30 mmoles) followed by bromine (502 mg, 3.14 mmoles). The mixture was stirred at room temperature for 24 hours, upon which time water (10 ml) was added over 5 minutes. The resulting precipitate was collected and washed with water to afford after recrystallisation from ethyl acetate/hexane to yield 665 mg (89%) of the titled compound **25** as a white solid, mp 212-214° dec; ir: ν 3205 (NH), 3190 (NH), 1740 (C=O), 1680 (C=O) cm^{-1}; ^1H nmr: δ 1.95-2.64 (m, 6H, CH_2), 3.85 (s, 3H, OCH_3), 5.02 (d, 1H, J = 12.4 Hz, OCH_2), 5.14 (d, 1H, J = 12.4 Hz, OCH_2), 7.11 (d, 1H, J = 8.6 Hz, Ar-*H*), 7.34 (dd, 1H, J = 2.1, 8.5 Hz, Ar-*H*), 7.58 (d, 1H, J = 2.1 Hz, Ar-*H*), 8.34 (brs, 1H, NH_2), 8.62 (brs, 1H, NH_2).

Anal. Calcd. for $C_{16}H_{16}Br_2N_2O_5$: C, 40.34; H, 3.36; N, 5.88. Found: C, 40.51; H 3.29; N, 5.80.

Methyl 2-Amino-6-bromo-1-oxa-4-oxo-3-azaspiro [4.4]non-2-ene-6-carboxylate **27**.

To a solution of **20a** (500 mg, 2.36 mmoles) in dimethylformamide (2 ml) was added pyridine (198 mg, 2.50 mmoles) followed by bromine (377 mg, 2.36 mmoles). The mixture was stirred at room temperature for 24 hours, upon which time water (10 ml) was added over 5 minutes. The resulting precipitate was collected and washed with water to afford after recrystallisation from methanol, 582 mg (86%) of the titled compound **27** as a white solid, mp 226° dec; ir: ν 3332 (NH), 3328 (NH), 1742 (C=O), 1663 cm^{-1}; uv (methanol): λ max 220 nm (ε = 16,775); ^1H nmr: δ 1.87-2.43 (m, 5H, CH_2), 2.58 (dt, 1H, J = 9.4, 14.8 Hz, CH_2), 3.66 (s, 3H, OCH_3), 8.35 (brs, 1H, N*H*), 8.64 (brs, 1H, N*H*).

Anal. Calcd. for $C_9H_{11}BrN_2O_4$: C, 37.13; H, 3.78; N, 9.62. Found: C, 36.83 H, 3.81; N, 9.55.

General Procedure for the Preparation of **26** and **28**.

To a solution of **25** or **27** (3.4 g, 7.14 mmoles) in THF (80 ml) under an atmosphere of nitrogen was added 1,1'-azobis-(cyclohexanecarbonitrile) (200 mg, 0.82 mmole) followed by tributyltin hydride (2.08 g, 7.14 mmoles). The solution was stirred at 40° overnight. Solvent was removed under vacuum and the residue crystallised from ethyl acetate to yield **26** and **28** in yields of 69 and 62% respectively.

2'-Bromo-4'-methoxyphenylmethyl 2-Amino-1-oxa-4-oxo-3-azaspiro[4.4]non-2-ene-6-carboxylate **26**.

This sample was obtained as a white solid. Recrystallisation from isopropanol afforded an analytical sample, mp 206°; ir: ν 3276 (NH), 3200 (NH), 1706 (C=O), 1600 cm^{-1}; ^1H nmr: δ 1.70-2.10 (m, 6H, CH_2), 3.27 (t, 1H, J =9.1 Hz, CH), 3.85 (s, 3H, OCH_3), 4.87 (d, 1H, J = 12.4 Hz, OCH_2), 5.02 (d, 1H, J = 12.4 Hz, OCH_2), 7.08 (d, 1H, J = 8.5 Hz, Ar-*H*), 7.30 (dd, 1H, J = 1.9, 8.5 Hz, Ar-*H*), 7.54 (d, 1H, J = 1.9 Hz, Ar-*H*), 8.31 (brs, 1H, NH_2), 8.54 (brs, 1H, NH_2).

Anal. Calcd. for $C_{16}H_{17}BrN_2O_5$: C, 48.36; H, 4.28; N, 7.05. Found: C, 48.19; H, 4.32; N, 6.92.

Methyl 2-Amino-1-oxa-4-oxo-3-azaspiro[4.4]non-2-ene-6-carboxylate **28**.

This compound was obtained as a white solid, mp 182-191°; ir: ν 3327 (NH), 3250 (NH), 1743 (C=O), 1663 cm^{-1}; uv: (methanol): λ max 218 nm (ε 16,600); ^1H nmr: δ 1.71-2.06 (m, 6H, CH_2), 3.20 (t, J = 9.1 Hz, CH), 3.53 (s, 3H, OCH_3), 8.32 (brs, 1H, NH_2), 8.34 (brs, 1H, NH_2).

Anal. Calcd. for $C_9H_{12}N_2O_4$: C, 50.94; H, 5.66; N, 13.21. Found: C, 51.08; H, 5.75; N, 13.10.

2'-Bromo-4'-methoxyphenylmethyl 2-Oxo-6-thioxo-1,3-diazaspiro[4.4] nonane-6-carboxylate **23**.

To a solution of **26** (100 mg, 0.25 mmole) in tetrahydrofuran (5 ml) was added Lawesson's reagent (62 mg, 0.15 mmoles) and the mixture stirred at 50° for 3 days. Solvent was removed under vacuum and the residue purified *via* column chromatography eluting with dichloromethane followed by dichloromethane/ethyl acetate (4:1) to yield, after recrystallisation from ethyl acetate, 54 mg (52%) of the titled compound **23** as a yellow solid, mp 210°; ir ν 3313 (NH), 1736 (C=O), 1673 (C=O) cm^{-1}; uv: (methanol): λ max 296 nm (ε 20,560); ^1H nmr: δ 1.81-2.30 (m, 6H, CH_2), 3.52 (t, 1H, J = 9.3 Hz, CH), 3.84 (s, 3H, OCH_3), 4.87 (d, 1H, J = 12.5 Hz, OCH_2), 5.00 (d, 1H, J = 12.5 Hz, OCH_2), 7.07 (d, 1H, J = 8.5 Hz, Ar-*H*), 7.28 (dd, 1H, J = 2.0, 8.5 Hz, Ar-*H*), 7.53 (d, 1H, J = 2.0 Hz, Ar-*H*), 9.01 (brs, 1H, NH_2), 9.18 (brs, 1H, NH_2).

Anal. Calcd. for $C_{16}H_{17}BrN_2O_4S$: C, 46.49; H, 4.12; N, 6.78. Found: C, 46.68; H, 4.31; N, 6.45.

2-Oxo-6-thioxo-1,3-diazaspiro[4.4]nonane-6-carboxylic Acid **24**.

To **23** (50 mg, 0.12 mmole) was added trifluoroacetic acid (2 ml), and the mixture stirred at room temperature for 1 hour. Solvent was removed under vacuum and the residue slurried with dichloromethane/hexane (2:1, 5 ml). The mixture was allowed to stand at 4° overnight and the precipitate collected and air dried to yield 22 mg (84%) of **24** as a yellow solid, mp 156-158°; ir: ν 3397 (NH), 1723 (C=O) cm^{-1}; ^1H nmr: δ 1.75-2.20 (m, 6H, CH_2), 3.39 (t, 1H, J = 9.8 Hz, CH), 9.05 (brs, 1H, NH_2), 9.16 (brs, 1H, NH_2), 11.15 (brs, 1H, COOH).

Anal. Calcd. for $C_8H_{10}N_2O_3S$: C, 44.86; H, 4.67; N, 13.08. Found: C, 44.69; H, 4.51; N, 12.81.

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