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An autoxidation study of C2 substituted pyrimidine amino reductones

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Abstract—Three pyrimidine derivatives (**8a–c**), differing from isouramil and divicine at C2, have been synthesized and their autoxidation rates measured spectrophotometrically. The autoxidation rates of all five pyrimidines (**8a–c**, isouramil and divicine) were correlated with σ_p^+ values (rho=−1.28, r^2=0.949).

1. Introduction

The amino reductone structure,[1] i.e., enolamine, occurs in a number of natural products. For instance, several species of ancient organisms (fungi, cyanobacteria, and lichens) produce UV absorbing metabolites such as MAA's (mycosporine-like amino acids, Fig. 1) that are characterized by a cyclohexenone **1** or cyclohexenimine **2** chromophore conjugated with the nitrogen substituent of an amino acid or its imino alcohol and having absorption maxima ranging from 310 to 360 nm.[2–3]

MAA's can be considered as potential sunscreens as their conjugated amino enolic chromophore has both broad absorption in the UV region and antioxidant properties desirable in a sun blocker.[4] Certain pyrimidine derivatives, i.e., isouramil **8d** (6-amino-2,5-dihydroxypyrimidin-4-one) and divicine **8e** (2,6-diamino-5-hydroxypyrimidin-4-one) incorporate the amino reductone group (Scheme 2). They are found in beans as glycosides and are thought to be the causative agents in favism.[5] A synergistic cytotoxicity has been demonstrated between carboplatin and divicine on murine erythroleukemic cells.[6] Divicine also enhances in vitro and in vivo lipopolysaccharide-induced release of tumor necrosis factor (TNF).[7]

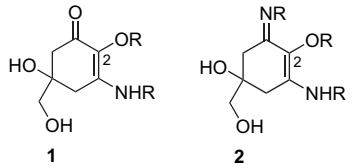

Figure 1. Structure of mycosporine-like amino acids.

A putative *cis*-enolamine **3** is thought to be the active intermediate in the formation of D-glucosamine-6-phosphate from D-fructose-6-phosphate using L-glutamine as the ammonia source, by glucosamine-6-phosphate synthase (GlmS). Because *N*-acetylglucosamine is an essential building block of both bacterial cell walls and fungal cell wall chitin, the enzyme is a potential target for antibacterial and antifungal agents.[8] Aminohexose reductones such as **4** obtained in the Maillard reaction are chiefly responsible for the development of aromas and colors during the thermal processing of foods (Fig. 2).[9]

Thus, an investigation of the redox properties of enolamines would shed light on their stability and behavior. In this paper

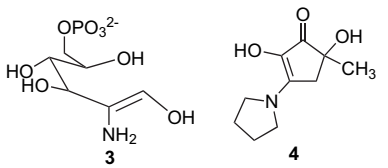

Figure 2. Naturally occurring *cis*-enoloamines.

Keywords: Amino reductones; Isouramil; Divicine; C2 pyrimidines; Autoxidation; Hammett correlation.
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we report the preparation of three pyrimidine derivatives (**8a–c**), differing from isouramil and divicine at C2, and the spectrophotometric measurement of their autoxidation rates.

2. Results and discussion

2.1. Synthesis

Pyrimidines **8a–c** were synthesized by condensing thiourea or acetamidine with 2-(tetrahydropyran-2-yloxy)-malonic acid diethyl ester (prepared in situ from tetrahydropyran-2-yloxyacetic acid ethyl ester, sodium hydride, and ethyl formate) to give **5a** and **5c**. Hydrolysis of **5a** and **5c** gave **6a** and **6c**, respectively, while treatment of **5c** with Raney Ni gave **6b**. Coupling of **6a** and **6b** with benzene diazonium chloride gave **7a** and **7b**, respectively. Methylation of **6c** followed by the same diazo coupling gave **7c**. Reduction of **7a–c** by dithionite gave **8a–c** (Scheme 1).

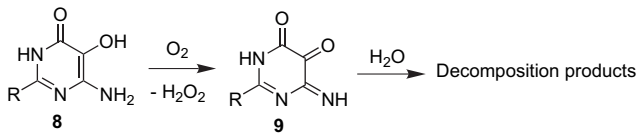
In addition to the spectroscopic and analytical data (see Section 4), the existence of an enolamine system (reductone) in compounds **8** was determined by various qualitative color tests including, ferric chloride solution,[10] 2,6-dichlorophenolindophenol solution,[11] and phosphomolybdic solution.[12]

2.2. Kinetic study of autoxidation

Compounds **8a–c**, differing from isouramil and divicine only at C2, were subjected to autoxidation under neutral conditions giving presumably H_2O_2 (Scheme 2).[5a,13–14]

The autoxidation rate was measured in air saturated buffer phosphate solutions 0.05 M at pH 7 and 25 °C. The solutions contained 1 mM EDTA to minimize catalysis of the oxidation by trace metallic cations.[5a,15] The rate of autoxidation was measured spectrophotometrically by following the decrease of the UV absorbance of the pyrimidines at their respective λ_{max} (Table 1).

All three compounds showed much slower oxidation rates compared with isouramil and divicine. This is probably the reason why there was no transient appearance of an absorption maximum around 240–255 nm, which is assumed to be due to the intermediacy of oxidized pyrimidine **9**, and was found in the autoxidation of isouramil, divicine, and related systems.[5a,14,16] The decrease in the absorbance of **8a–c**, followed a reasonably pseudo first order reaction in the pyrimidine concentration (the dissolved oxygen concentration was at least 13 times higher). However, Winterbourn et al. found that the reaction mechanism is complex and involves radical intermediates and chain reactions and that the dependency on the pyrimidine concentration is far from simple.[14,17] Therefore, we preferred to use the measured initial reaction rates for checking the quantitative dependency of the oxidation rate on the electron releasing power of the substituent in the C2 position. Our results for **8a–c** are summarized in



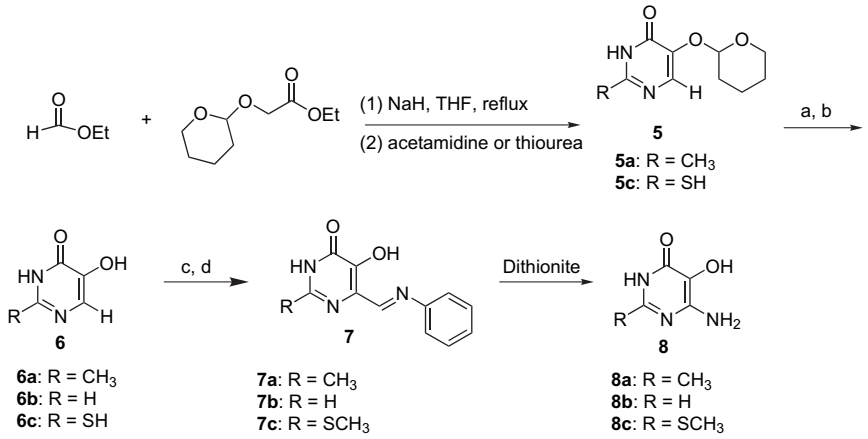
8a: R = CH_3
8b: R = H
8c: R = SCH_3
8d: R = OH (isouramil)
8e: R = NH_2 (divicine)

Scheme 2. Pyrimidine derivatives autoxidation.

Table 1. Spectral properties and initial autoxidation rates for compounds **8a–e**

Substituent R	λ_{max} nm (ε)	Initial rate μM/s	σ_p [a]	σ_p^+ [a]
H	274 (13,000)[b]	0.00087[c]	0	0
CH_3	275 (16,200)[b]	0.0015[c]	−0.17	−0.31
SCH_3	286 (10,800)[b]	0.0097[c]	0	−0.6
OH	280 (14,100)[d]	0.067[e]	−0.37	−1.6[f]
NH_2	285 (9800)[d]	0.061[e]	−0.66	−1.3

[a] Ref. 18.
[b] This work.
[c] This work; air saturated 0.05 M phosphate buffer pH=7.0, 1 mM EDTA, 25 °C, [pyrimidine]=2.4×10^{-5} M.
[d] Ref. 5a.
[e] Calculated from data in Ref. 14, see text.
[f] σ_p^+ for OH from Ref. 19.



5a: R = CH_3
5c: R = SH

6a: R = CH_3
6b: R = H
6c: R = SH

7a: R = CH_3
7b: R = H
7c: R = SCH_3

8a: R = CH_3
8b: R = H
8c: R = SCH_3

[a]H_3O^+ for **6a**, **6c**. [b]Raney Ni for **6b**. [c] PhN_2Cl for **7a** and **7b**. [d]$(MeO)_2SO_2$ then PhN_2Cl for **7c**.

Scheme 1. Synthesis of compounds **8**.

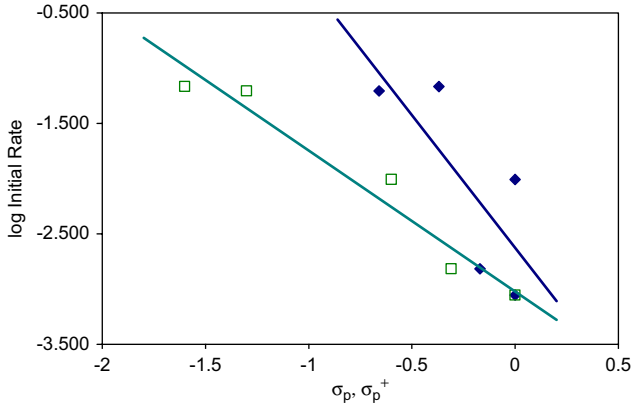
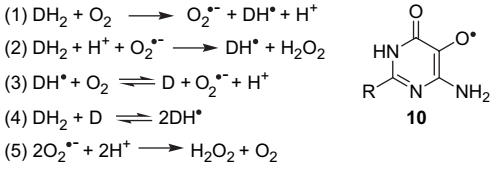


Figure 3. Hammett correlations against σ_p (◆) and σ_p^+ (□) for the autoxidation of compounds **8a–e** in air saturated solutions at pH 7.0 and 25 °C. Rho value for σ_p is −2.40, r^2=0.5868; rho value for σ_p^+ is −1.28, r^2=0.949.

Table 1. Table 1 also contains the initial rates for the reactions of isouramil and divicine (**8d** and **8e**) calculated from Winterbourn et al. data[14] (aerated 0.05 M phosphate buffer, pH 7, 23 °C, 50 mM DTPA and similar to our pyrimidine concentrations) using ΔH of 60.2 kJ/mol given by Chevion et al.[5a] A plot of the logarithmic values of the initial rates against Hammett σ_p constants did not give a reasonable correlation. However, the correlation was greatly improved when we used σ_p^+[18,19] values, rho value is −1.28 (r^2= 0.949) (Fig. 3).[20]

Winterbourn et al.[14,17] suggested the following complex chain mechanism to account for the autoxidation rates of isouramil and divicine. DH_2 stands for the reduced pyrimidine, $DH^\bullet$ for the pyrimidine radical (probably structure **10**) and D for the oxidized pyrimidine, most probably having structure **9** (Scheme 2). It is reasonable to expect the same mechanism for our compounds **8a**, **8b**, and **8c** (Scheme 3).



Scheme 3.

The observed initial reaction rates must be the result of a complex combination of the rates of the individual steps detailed in Scheme 3. Our limited data do not justify enlarging upon them, but still, the observed initial rate surely reflects the rate of the first initiation step (1) as well as the rates of the rate determining propagation steps (2) and (4). It is obvious that stabilizing the generated radical $DH^\bullet$ will enhance the rate of the above three steps. The formation of $DH^\bullet$ from DH_2 lowers the electron density on the oxygen atom and that explains the enhanced autoxidation rate with the electron releasing power of the substituent at position 2. The good correlation with σ_p^+ clearly indicates the role of resonance and partial distribution of charge in stabilizing the generated pyrimidine radical. A similar correlation was found between σ_p^+ and the one-electron reduction potential of 4-substituted phenoxyl radicals measured through their equilibrium with known redox pairs.[21,22] It is interesting to note that Xu et al.[23] checked the correlation of the first oxidation potential of substituted *N*-hydroxyacetanilides with Hammett σ^- values and found only a fair correlation (r^2=0.962). However, we found that their results correlate much better with σ^+ values (r^2=0.9943).

3. Conclusion

Several amino reductones occur naturally and are responsible for life-threatening hemolytic episodes in favism. On the other hand, amino reductones may also be useful sunscreens. Thus an understanding of their properties is desirable. We have found marked dependency of the autoxidation rate of five pyrimidine derivatives **8a–e** on the electron releasing power of the substituent at C2 and this can point a way to control the oxidation rate of future useful compounds.

4. Experimental

4.1. General procedures

All chemicals were of reagent grade and were used without further purification. ^{1}H NMR (300 MHz) spectra were recorded in $CDCl_3$. (*J* values are given in hertz). IR spectra were recorded on a Perkin–Elmer 2000 Fourier transformed infrared instrument. MS analyses were performed on Finnegan instrument (Model QDVO). All UV spectra were recorded on Unicam SP-1700. Nitrogen was bubbled for 10 min through 0.05 M phosphate buffer at pH 7 containing 1 mM EDTA. To 1.9 mL of the above buffer in a UV cell, 100 μL of freshly prepared pyrimidine solution (**8a–c**) were injected to give a final concentration of 5×10^{-5} M. The UV spectrum at the range 230–340 nm was recorded immediately. Injecting the same pyrimidine solution to air saturated buffer solution and recording the spectrum at increasing time intervals, showed a constant decrease in the UV absorption at the range 230–340 nm with no build up of a new absorption band at 240–255 nm as was found for isouramil and divicine.[5a] Kinetic measurements were performed by injecting 50 μL of the above pyrimidine solution into 2 cm^3 of the air saturated phosphate buffer thermo equilibrated at 25±1 °C, final pyrimidine concentration 2.4×10^{-5} M. The decrease of the absorbance at λ_{max} was recorded for 1–2 half-lives.

4.1.1. 2-Methyl-5-(tetrahydro-2*H*-pyran-2-yloxy)pyrimidin-4(3*H*)-one (5a).[24] To a suspension of sodium hydride (4.60 g in 55–60% paraffin oil), dry ether (50 cm^3) and dry ethyl formate (7.84 g) were added. Then (tetrahydropyran-2-yloxy)-acetic acid ethyl ester (20 g) was added dropwise under continuous stirring. After the mixture was refluxed for 2 h, acetamidine (4.3 g) was added. After the removal of ether from the reaction mixture, the remained ethanolic solution was refluxed for 4 h. Then the mixture was cooled and the volatile solvents were removed by rotory-evaporator. The residue was redissolved in water and filtered. The filtrate was acidified by acetic acid in an ice bath and the white precipitate was filtered, washed with water, and dried under reduced pressure at 100 °C. Yield (9.0 g, 58%), mp 149–151 °C; [Found: C 55.25; H 47.54; N 13.19. $C_{10}H_{16}N_2O_3$

requires C 55.59; H 7.60; N 13.20%]; δ_H (300 MHz, DMSO-d_6) 1.23–2.03 (6H, m), 2.27 (3H, s), 3.23–4.04 (2H, m), 5.47 (1H, br s), 7.60 (1H, s), 10.73 (1H, br s); ν_{max}/cm^{-1} 1670, 1610, 1385, 1310, 1205, 1190, 1120, 980, 910, 820, 775, and 740; MS(EI): *m*/*z* (%) 210 (100, M$^+$, $C_{10}H_{14}N_2O_3$), 126 (M$^+$, –C_5H_8O), 125 (M$^+$, –C_5H_9).

4.1.2. 5-Hydroxy-2-methylpyrimidin-4(3*H*)-one 6a. A few crystals of *p*-toluenesulfonic acid were added to a solution of **1a** (6.57 g) in hot methanol. After cooling the mixture in an ice bath, white crystals were formed. The crystals were filtered and dried under vacuum at 100 °C. Yield (2.5 g, 63%), mp>300 °C; δ_H (300 MHz, DMSO-d_6) 2.2 (3H, s), 7.27 (1H, s), 9.27 (1H, br s); ν_{max}/cm^{-1} 3300, 1670, 1620, 1420, 1380, 1245, 1110, 1020, 875, and 780; MS(EI): *m*/*z* (%) 126 (100, $C_5H_6N_2O_2$, M$^+$), 108 (M$^+$, –H_2O), 100 (M$^+$, −26).

4.1.3. 5-Hydroxy-2-methyl-6-phenylazo-3*H*-pyrimidin-4-one 7a. Diazotation of aniline (1.17 g) was done in hydrochloric acid (3.9 cm^3) and water (8 cm^3) by addition of sodium nitrite (0.87 g) in water (6 cm^3) at 0–5 °C. Then sodium acetate (3.1 g) was added slowly under continuous stirring, followed by the addition of a solution of **6a** (1.59 g) in 10% sodium hydroxide (10.4 cm^3). After stirring for 30 min, the reaction mixture was left for overnight at 4 °C. Then the reaction was warmed to 40 °C for 1 h and filtered. The red crystals formed were washed and dried under vacuum at 100 °C. Yield (1.61 g, 56%), mp 243–245 °C; [Found: C 57.60; H 4.22. $C_{11}H_{10}N_4O_2$ requires C 57.89; H 4.35%]; δ_H (300 MHz, DMSO-d_6) 1.95 (3H, s), 6.44–7.73 (5H, m), 11.19 (1H, br s), 11.64 (1H, br s); ν_{max}/cm^{-1} 3220, 3180, 1710, 1670, 1600, 1520, 1470, 1430, 1360, 1280, 1250, 1050, 800, 775, 715, 700, and 660; MS(EI): *m*/*z* (%) 230 (25, $C_{11}H_{10}N_4O_2$, M$^+$), 105 (100, $C_6H_5N_2^+$).

4.1.4. 6-Amino-5-hydroxy-2-methylpyrimidin-4(3*H*)-one 8a.[25] A solution of **7a** (1.61 g) in water (15 cm^3) was heated to 60–70 °C, an excess of sodium dithionite was added to the solution in batches until a bright yellow color was obtained and the solution was cooled in an ice bath. The white crystals formed were washed with water and dried under vacuum at 100 °C. Yield (0.44 g, 44%), mp>300 °C; [Found: C 42.53; H 4.98; N 29.46. $C_5H_7N_3O_2$ requires C 42.55; H 4.96; N 29.79%]; δ_H (300 MHz, DMSO-d_6) 2.13 (3H, s), 5.36 (2H, br s), 7.85 (1H, br s), 11.66 (1H, br s); ν_{max}/cm^{-1} 3420, 3320, 3160, 1600, 1440, 1380, 1280, 1210, 1020, 990, 900, 790, and 770; MS(EI): *m*/*z* (%) 141 (100, $C_5H_7N_3O_2$, M$^+$).

4.1.5. 2-Mercapto-5-(tetrahydro-pyran-2-yloxy)-3*H*-pyrimidin-4-one 5c. Identical to the procedure for the synthesis of **5a**, except for the addition of thiourea (15.29 g) instead of acetamidine. White crystals were obtained. Yield (25.0 g, 54.5%), mp>300 °C; [Found: C 47.15; H 5.38; S 14.49. $C_9H_{12}N_2O_3S$ requires C 47.37; H 5.26; S 14.00]; δ_H (300 MHz, DMSO-d_6) 0.8–1.97 (6H, m), 3.23–3.80 (2H, m), 5.27 (1H, br s), 7.16 (1H, d, J_{HNCH}=6.0 Hz), 10.8 (1H, br s), 11.3 (1H, br s); ν_{max}/cm^{-1} 3150, 3080, 1630, 1570, 1250, 1200, 1180, 1150, 1110, 1020, 980, 940, 900, 870, 810, and 670; MS(EI): *m*/*z* (%) 147 (100, M$^+$−81).

4.1.6. 5-Hydroxy-2-mercapto-3*H*-pyrimidin-4-one 6c. A suspension of **5c** (6.0 g) in 1 M H_2SO_4 (30 cm^3) was stirred for 2 h. Then the product was filtered and washed with water, methanol, and ether, and dried under vacuum at 100 °C. Yield (3.2 g, 84.4%), mp>300 °C; [Found: C 33.53; H 2.60; N 19.30; S 22.96. $C_4H_4N_2O_2S$ requires C 33.33; H 2.78; N 19.44; S 22.22%]; δ_H (300 MHz, DMSO-d_6) 6.97 (1H, d, J_{HNCH}=6.0 Hz), 9.60 (1H, br s), 10.27 (1H, br s); ν_{max}/cm^{-1} 3240, 3100, 1660, 1580, 1400, 1290, 1230, 1170, 1140, 890, 820, 760, 750, and 690; MS(EI): *m*/*z* (%) 144 (100, $C_4H_4N_2O_2S$, M$^+$).

4.1.7. 5-Hydroxy-2-methylsulfanyl-6-phenylazo-3*H*-pyrimidin-4-one 7c. Compound **6c** (2.9 g) was dissolved in a solution of sodium hydroxide (1.8 g) in water (12 cm^3) and heated to 40 °C. Dimethylsulfate (3.0 g) was added dropwise while vigorously stirring and then the mixture was cooled in an ice bath and filtered. The filtrate was acidified with concentrated hydrochloric acid and was left overnight at 4 °C. The formed crystals were filtered, successively washed with water, methanol, and ether, and dried under vacuum at 100 °C. The 2-methylthio-4,5-dihydroxypyrimidine formed (1.4 g) was subjected to the same diazotization procedure as **3a**. Yield (1.5 g, 74.0%), mp 213–215 °C; δ_H (300 MHz, DMSO-d_6) 2.73 (3H, s); 6.97–7.83 (5H, m); 10.73 (1H, br s); ν_{max}/cm^{-1} 3480, 3200, 3100, 1710, 1660, 1590, 1510, 1450, 1250, 1150, 1030, 990, 770, 750, 690, and 640; MS(EI): *m*/*z* (%) 262 (10, $C_{11}H_{10}N_4O_2S$, M$^+$), 105 (100, $C_6H_5N_2^+$), 91 ($C_6H_5N^+$), 77 ($C_6H_5^+$).

4.1.8. 6-Amino-5-hydroxy-2-(methylthio)pyrimidin-4-(3*H*)-one 8c. Identical to the procedure for the synthesis of **8a**, except for the use of **7c** (1.0 g) instead of **7a**. White crystals were obtained. Yield (0.43 g, 66%), mp 243–245 °C; [Found: C 34.27; H 4.08; N 24.21; S 19.00. $C_5H_7N_3O_2S$ requires C 34.38; H 4.05; N 24.28; S 18.50%); δ_H (300 MHz, DMSO-d_6) 2.45 (3H, s); 5.87 (2H, br s), 7.90 (1H, br s); ν_{max}/cm^{-1} 3250, 3380, 1640, 1600, 1570, 1410, 1330, 1240, 970, 830, and 760; MS(EI): *m*/*z* (%) 173 (100, $C_5H_7N_3O_2S$, M$^+$).

4.1.9. 5-Hydroxypyrimidin-4(3*H*)-one 6b. To a solution of water (76 cm^3) and concentrated aqueous ammonia (7.6 cm^3), **5c** (11.0 g) was added followed by the addition of Raney nickel (40.0 g). The mixture was refluxed for 4 h then it was cooled and filtered. All the volatile solvents were removed by rotor-evaporator and the residue was redissolved in methanol. After addition of ether, pink crystals precipitated out of the solution, and were dried under vacuum at 100 °C. Yield (2.5 g, 46%), mp 265–267 °C; δ_H (300 MHz, DMSO-d_6) 7.40 (1H, s), 7.67 (1H, s); ν_{max}/cm^{-1} 1640, 1600, 1360, 1300, 1270, 1100, 930, 880, 790, 780, and 615; MS(EI): *m*/*z* (%) 112 (100, $C_4H_4N_2O_2$, M$^+$).

4.1.10. 5-Hydroxy-6-phenylazo-3*H*-pyrimidine-4-one 7b. Identical to the procedure for the synthesis of **7a** except for the addition of **6b** (1.12 g) instead of **6a**. White crystals were obtained. Yield (1.9 g, 88%), mp 244–245 °C; δ_H (300 MHz, DMSO-d_6) 6.83–7.76 (6H, m), 11.88 (1H, br s); ν_{max}/cm^{-1} 3490, 3290, 1700, 1650, 1615, 1600, 1590, 1500, 1450, 1300, 1240, 1170, 1120, 1015, 900, 875, 750, 730, 680, 660, 640, 660, and 640; MS(EI): *m*/*z* (%) 216 (15, $C_{10}H_8N_4O_2$, M$^+$), 105 (100, $C_6H_5N_2^+$), 91 ($C_6H_5N^+$), 77 ($C_6H_5^+$).

4.1.11. 6-Amino-5-hydroxypyrimidin-4(3*H*)-one 8b. Identical to the procedure for the synthesis of **8a** except for the

addition of **7b** (1.0 g) instead of **7a**. Yield (0.45 g, 76%), mp>300 °C; [Found: C 38.02; H 3.75; N 33.35. $C_4H_5N_3O_2$ requires C 37.80; H 3.94; N 33.07%]; δ_H (300 MHz, DMSO-d_6) 5.83 (2H, br s), 7.56 (1H, s), 11.79 (1H, br s); ν_{max}/cm^{-1} 3470, 3140, 1670, 1640, 1620, 1440, 1370, 1250, 1170, 1010, 890, 810, 770, and 650; MS(EI): *m/z* (%) 127 (100, $C_4H_5N_3O_2$, M^+).

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References and notes

1. Kurata, T.; Otsuka, Y. *Adv. Exp. Med. Biol.* **1998**, *434*, 269–276.
2. Proteau, P. J.; Gerwick, W. H.; Garcia-Pichel, F.; Castenholz, R. W. *Experientia* **1993**, *49*, 825–829.
3. Gröniger, A.; Sinha, R. P.; Klisch, M.; Häder, P. D. *J. Photochem. Photobiol. B: Biol.* **2000**, *58*, 115–122.
4. (a) Bandaranayake, W. M. *Nat. Prod. Lett.* **1998**, 159–172; (b) Torres, A.; Hochberg, M.; Pergament, I.; Smoum, R.; Niddam, V.; Dembitsky, V. M.; Temina, M.; Dor, I.; Lev, O.; Srebnik, M.; Enk, C. D. *Eur. J. Biochem.* **2004**, *271*, 780–784.
5. (a) Chevion, M.; Navok, T.; Glaser, G.; Mager, J. *Eur. J. Biochem.* **1982**, *127*, 405–409; (b) McMillon, D. C.; Bolchoz, L. J. C.; Jollow, D. J. *Toxicol. Sci.* **2001**, *62*, 352–359.
6. Tonetti, M.; Sturla, L.; Bistolfi, T.; Benatti, U.; De Flora, A. *Biochem. Biophys. Res. Commun.* **1994**, *200*, 654–660.
7. Geeta, C.; Clark, I. A. *J. Immunol.* **1989**, *143*, 1290–1294.
8. Bearnet, S. L.; Blouin, C. *J. Biol. Chem.* **2000**, *275*, 135–400.
9. Hofmann, T. *J. Agric. Food Chem.* **1999**, *47*, 4763–4768.
10. (a) Singh, M.; Singh, S. S.; Sanwal, G. G. *Indian J. Exp. Biol.* **1978**, *16*, 712–714; (b) Huckel, W.; Wehrung, R. *Arch. Pharm.* **1958**, *291*, 650–663; (c) Wesp, E. F.; Brode, W. R. *J. Am. Chem. Soc.* **1934**, *56*, 1037–1042.
11. Farmer, C. J.; Abt, A. F. *Proc. Soc. Exp. Biol. Med.* **1936**, *34*, 146–150.
12. Singleton, V. L.; Rossi, J. A., Jr. *Am. J. Enol. Vitic.* **1965**, *16*, 144–158.
13. Cohen, G.; Heikkila, R. E.; MacNamee, D. *J. Biol. Chem.* **1974**, *249*, 2447–2452.
14. Winterbourn, C. C.; Cowden, W. B.; Sutton, H. *Biochem. Pharmacol.* **1989**, *38*, 611–618.
15. Munday, R. *Biochem. Pharmacol.* **1988**, *37*, 409–413.
16. Goyal, R. N.; Kumar, A.; Jain, N.; Gupta, P. *Indian J. Chem., Sect. A* **1999**, *38*, 1015–1023.
17. Winterbourn, C. C.; Munday, R. *Free Radical Res. Commun.* **1990**, *8*, 287–293.
18. Hansch, C.; Leo, A.; Taft, R. W. *Chem. Rev.* **1991**, *91*, 165–195.
19. de la Mare, P. B. D.; Newman, P. A. *Tetrahedron Lett.* **1982**, *23*, 1305–1308.
20. One referee suggested that compound **8d** is almost certainly in the 2-oxo form. However, to the best of our knowledge the structure of the most stable/common tautomer of isouramil, compound **8d**, is not yet established and it appears in the literature in various ways. We think that the similarity in the autoxidation rate of isouramil and divicine (compound **8e** which is generally believed to exist mainly as the 2-NH_2 tautomer) and especially the good fit of the reaction rate of isouramil to the Hammett correlation line that was drawn for the other four pyrimidines using sigma plus, indicate that indeed, the main tautomer of compound **8d** does have 2-OH. The similarity in the UV spectra of the five systems (Table 1) gives supporting evidence to the above claim. Furthermore, the 2-SH derivative which exists as the thiourea tautomer, showed in contrast two UV peaks, at 300 nm (17,000) and 238 nm (16,400) (unpublished results).
21. Jovanovic, S. V.; Tosic, M.; Simic, M. G. *J. Phys. Chem.* **1991**, *95*, 10824–10827.
22. Lind, J.; Shen, X.; Eriksen, T. E.; Merenyi, G. *J. Am. Chem. Soc.* **1990**, *112*, 479–482. These authors also measured the rates of the chlorine dioxide radical $ClO_2^{\bullet}$ with 4-X-substituted phenolate anions (X=H, CH_3, F, I, CH_3CO). Using their data we found a good correlation of the rate constants with σ_p (r^2=0.9512) and an even better one with σ_p^+ (r^2=0.9754).
23. Xu, F.; Deussen, H. W.; Lopez, B.; Lam, L.; Li, K. *Eur. J. Biochem.* **2001**, *268*, 4169–4176.
24. (a) Davoll, J.; Laney, D. H. *J. Chem. Soc.* **1956**, 2124–2131, triturated the reaction mixture with ether: mp 141–143 °C; (b) Tanner, E. M. *Spectrochim. Acta* **1956**, *8*, 9–18, reports IR data for various hydroxypyrimidines including **5a**.
25. Hurst, D. T. *Aust. J. Chem.* **1983**, *36*, 1285–1289, prepared the compound by Elbs persulfate oxidation followed by hydrolysis of the sulfate: mp>203 °C.