
# Improved synthesis of N1-substituted orotic acid derivatives

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## Graphical Abstract

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TETRAHEDRON LETTERS

# Improved synthesis of N1-substituted orotic acid derivatives

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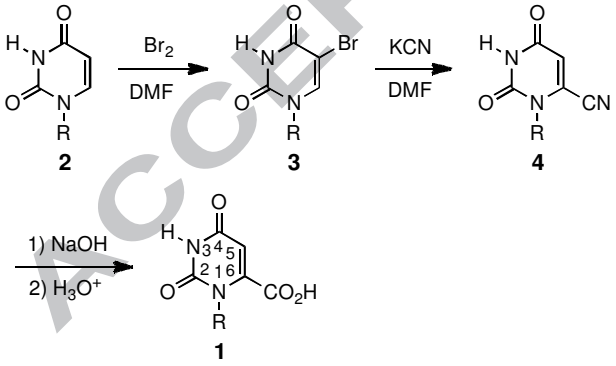
**Abstract—** An improved method for the synthesis of N1-substituted orotic acid derivatives is reported. The method involves sequential incorporation of nitrogen atoms to the pyrimidine structure from simple starting materials and thus allows the synthesis of N1-substituted orotic acid derivatives with single $^{15}N$ label at either N-1 or N-3. 

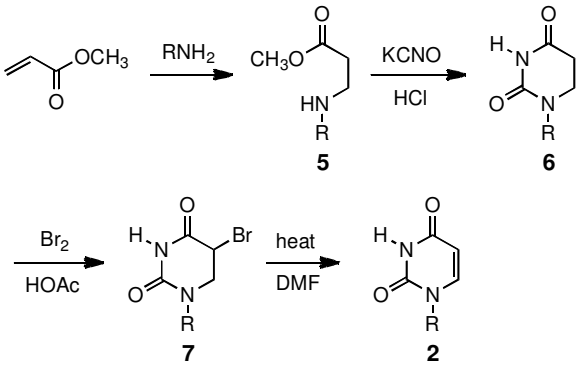
*Keywords*: Orotic acid, orotidine, 1-substituted orotic acid, 1-substituted uracil, dihydrouracil, 5-bromouracil

The decarboxylation of orotic acid and its analogues has been investigated as a model for the enzymatic decarboxylation catalyzed by orotidine-5'-monophosphate decarboxylase (ODCase).[1-12] Part of the mechanistic investigation requires N1-substituted orotic acid derivatives (structure **1** in Scheme I) as substrates. Direct alkylation of orotic acid, unfortunately, produces a mixture of disubstituted and N3-substituted orotic acid derivatives because N3 is the more reactive site.[13] N1-Substituted orotic acid derivatives are commonly synthesized from N1-substituted uracil **2** via 5-bromouracil **3** and 6-cyanouracil **4** as shown in Scheme 1.[11,14-16] In this Letter, we report a simplified synthetic method for such orotic acid derivatives from readily available 5,6-dihydrouracil derivatives.




**Scheme 1**

A few methods for the synthesis of N1-substituted uracil derivatives have been reported.[17-19] One of the methods involves sequential integration of the two nitrogen atoms from simple starting materials (as shown in Scheme 2) and thus will allow incorporation of $^{15}N$ at one position only.[17] The reported synthesis starts with the conjugated addition of a primary amine to acrylate that yields adduct **5**.[17,20] The amino group is converted to the urea group via reaction with cyanate. Cyclization as a result of the reaction of the urea group with the ester group gives the 5,6-dihydrouracil **6**. Bromination of **6** produces **7**, which undergoes thermal elimination to yield uracil **2**.




**Scheme 2**

In the process of synthesizing N1-substituted uracil derivatives following this reported synthetic route, we observed that the bromination of 5,6-dihydrouracil **6** yielded desired product **7** mixed with dibrominated product **8** as shown in Scheme 3. Further experiments of purified dibromide **8** illustrated that it underwent elimination upon heating to produce 5-bromouracil **3**, analogous to the conversion of bromide **7** to **2**. We have thus thought to shorten the synthetic route employing 5,5-dibromo-5,6-dihydrouracil **8** as a synthetic intermediate.

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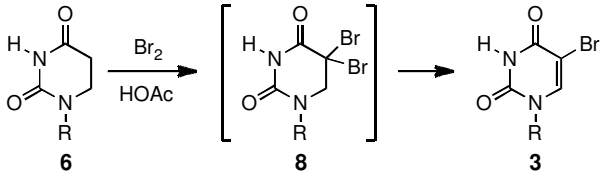


6 8 3

**Scheme 3**

The dibromination of **6** to **8** was effected by using more than two equivalents of bromine. Interestingly, it was observed that a fraction of dibromide **8** underwent elimination spontaneously to produce bromouracil **3** under the reaction conditions (acetic acid, 100 °C). We thought to further simplify the synthetic route by combining the bromination and elimination steps. The bromination reaction was thus carried out in boiling acetic acid. Under the new reaction condition, dihydrouracil **6** was converted to 5-bromouracil **3** in one step, in contrast to the three-step sequence in the original synthesis. The yields for the one-step reactions were excellent for various primary and secondary substituting groups (tertiary groups were found to be labile under the highly acidic reaction conditions as expected) tested as shown in Table 1. In the case when cyclohexyl was the substituting group, the yields for the two synthetic routes were directly compared. The direct conversion of **6a** to **3a** (R = cyclohexyl) was found to be quantitative (the yield is 89% if purified by recrystallization), whereas the reported yields for the three synthetic steps were 69% (**6a** to **7a**), 85% (**7a** to **2a**), and 75% (**2a** to **3a**), respectively, giving an overall yield of 44% from **6a** to **3a**.[14,17] Therefore, the modified one-step procedure reported here represents a significantly simplified and improved method to synthesize N1-substituted orotic acid derivatives.

Table 1. Yields for the one-step conversion of 5,6-dihydrouracil **6** to 5-bromouracil **3**[21]

| Entry | 1-Substituted 5,6-dihydrouracils (**6**) | Yield (isolated) (%) |
|---|---|---|
| 1 | **6a**, R = cyclohexyl | 100 (89)[a] |
| 2 | **6b**, R = *n*-butyl | 85 |
| 3 | **6c**, R = *n*-propyl | 88 |
| 4 | **6d**, R = isobutyl | 86 |
| 5 | **6e**, R = *sec*-butyl | 100 |

[a]Yield in parenthesis when purified by recrystallization.

It should be pointed out again that this synthetic route allows the incorporation of a single $^{15}N$ label at either N1 or N3. Such labeled compounds may aid in the mechanistic investigation of the model decarboxylation of orotic acid derivatives or the enzymatic decarboxylation catalyzed by ODCase.

## Experimental Details

All reagents were obtained from commercial sources and used without further purification. Only the experimental procedures for the conversion of dihydrouracil **6** to 5-bromouracil **3** are described here using 1-cyclohexyl-5,6-dihydrouracil as an example. The products were purified through recrystallization or column chromatography. Procedures for other synthetic steps were described in literature and were employed without modification.

*Typical experimental procedure*: 1-Cyclohexyl-5,6-dihydrouracil (1.00 g, 5.1 mmol) was dissolved in 12 mL acetic acid in a round-bottomed flask. A solution of bromine (3.24 g, 20.2 mmol) in 10 mL acetic acid was added to the reaction flask and the reaction mixture was refluxed. The reaction usually took five to six hours and was followed by thin layer chromatography (TLC). Upon completion of the reaction, the reaction mixture was evaporated to dryness under reduced pressure. The solid residue was purified by either column chromatography (10% ethyl acetate/methylene chloride as solvent) or recrystallization from ethanol to produce 5-bromo-1-cyclohexyluracil as colorless flakes; mp 220-222 °C, lit. mp 225 °C.[17]

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## References

1. Beak, P.; Siegel, B. *J. Am. Chem. Soc.* **1976**, *98*, 3601-3606.
2. Radzicka, A.; Wolfenden, R. *Science* **1995,** *267*, 90-93.
3. Lee, J. K.; Houk, K. N. *Science* **1997,** *276*, 942-945.
4. Kakanishi, M. P.; Wu, W. *Tetrahedron Lett.* **1998**, *39*, 6271–6272.
5. Feng, W. Y.; Austin, T. J.; Chew, F.; Gronert, S.; Wu, W. *Biochemistry* **2000**, *39*, 1778-1783.
6. Singleton, D. A.; Merrigan, S. R.; Kim, B. J.; Beak, P.; Phillips, L. M.; Lee, J. K. *J. Am. Chem. Soc.* **2000**, *122*, 3296–3300.
7. Wong, F. M.; Wu, W. *Bioorg. Chem.* **2006**, *34*, 99–104.
8. Yeoh, F. Y.; Cuasito, R. R.; Capule, C. C.; Wong, F. M.; Wu, W. *Biorg. Chem.* **2007**, *35*, 338–343.
9. Wong, F. W.; Capule, C. C.; Wu, W. *Org. Lett.* **2006**, *8*, 6019-6022.

10. Wong, F. M.; Capule, C. C.; Chen, D. X.; Gronert, S.; Wu, W. *Org. Lett.* **2008**, *10*, 2757-2760.
11. Lewis, C. A. Jr.; Wolfenden, R. *Biochemistry* **2009**, *48*, 8738-8745.
12. Senger, N. A.; Bliss, C. E.; Keeffe, J. R.; Gronert, S.; Wu, W. *Tetrahedron* **2013**, *69*, 5287-5292.
13. Curran, W. V.; Angier, R. B. *J. Org. Chem.* **1966**, *31*, 201–205.
14. Landesman, P. W. Ph.D. Dissertation, State University of New York, Buffalo, 1982.
15. Inoue, H.; Ueda, T. *Chem. Pharm. Bull* **1971**, *19*, 1743-1744.
16. Inoue, H.; Ueda, T. *Chem. Pharm. Bull* **1978**, *26*, 2657-2663.
17. Okano, T.; Goya, S.; Takahashi, T. *Yakugaku Zasshi* **1968**, *88*, 1112-1117.
18. Rejman, D.; Kovacˇkova´, S.; Pohl, R.; Dracˇı´nsky´, M.; Fiedler, P.; Rosenberg, I. *Tetrahedron* **2009**, *65*, 8513-8523.
19. Winckelmann, I.; Larsen, E. H. *Synthesis* **1986**, 1041-1044.
20. Padwa, A.; Hasegawa, T.; Liu, B.; Zhang, Z. *J. Org. Chem.* **2000,** *65,* 7124-7133.
21. $^1$H NMR data of **6** and **3** in $CDCl_3$:
1-*n*-Butyl-5,6-dihydrouracil **6b**. δ7.7 (bs, 1H, NH); 3.4 (t, 4H, $CH_2NCH_2$); 2.7 (t, 2H, $CH_2CO$); 1.6 (quin, 2H, $NCH_2\underline{CH_2}$), 1.3 (sext, 2H, $\underline{CH_2}CH_3$); 0.9 (t, 3H, $CH_3$).
1-*n*-Propyl-5,6-dihydrouracil **6c**. 7.5 (bs, 1H, NH); δ3.4 (m, 4H, $CH_2NCH_2$); 2.7 (t, 2H, $CH_2CO$); 1.6 (sext, 2H, $\underline{CH_2}CH_3$); 0.9 (t, 3H, $CH_3$).
1-Isobutyl-5,6-dihydrouracil **6d**. δ7.6 (s, 1H, NH); 3.4 (t, 2H, $NCH_2$ ring; 3.2 (d, 2H, $NCH_2$ side chain); 2.7 (t, 2H, $CH_2CO$); 2.0 (sept, 1H, CH); 0.9 (6H, $CH_3$ x 2).
1-*sec*-Butyl-5,6-dihydrouracil **6e**. δ8.1 (s, 1H, NH); 4.4 (1H, sext, NCH); 3.3 (2H, m, NCH2); 2.6 (t, 2H, $CH_2CO$); 1.5 (quint, 2H, $CH_3\underline{CH_2}$); 1.2 (d, 3H, $CH\underline{CH_3}$); 0.9 (t, 3H, $CH_2\underline{CH_3}$).
1-*n*-Butyl-5-bromouracil **3b**. δ8.7 (bs, 1H, NH); 7.5 (s, 1H, CH); 3.8 (t, 2H, $NCH_2$); 1.7 (quin, 2H, $NCH_2\underline{CH_2}$), 1.4 (sext, 2H, $\underline{CH_2}CH_3$); 1.0 (t, 3H, $CH_3$).
1-*n*-Propyl-5-bromouracil **3c**. δ8.5 (bs, 1H, NH); 7.5 (s, 1H, CH); 3.7 (t, 2H, $NCH_2$); 1.7 (sext, 2H, $\underline{CH_2}CH_3$); 1.0 (t, 3H, $CH_3$).
1-Isobutyl-5-bromodrouracil **3d** .δ8.5 (bs, 1H, NH); 7.5 (s, 1H, H-6); 3.6 (d, 2H, $NCH_2$); 2.1 (sept, 1H, CH); 1.0 (6H, $CH_3$ x 2).
1-*sec*-Butyl-5-bromodrouracil **3e**. δ9.0 (bs, 1H, NH); 7.5 (s, 1H, H-6); 4.6 (1H, sext, NCH); 1.7 (quint, 2H, $CH_3\underline{CH_2}$); 1.3 (d, 3H, $CH\underline{CH_3}$); 1.0 (t, 3H, $CH_2\underline{CH_3}$).

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