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A Concise Synthesis of Thyroxine (T_4) and 3,5,3'-Triiodo-L-thyronine (T_3)

Grzegorz M. Salamonczyk, Vibha B. Oza and Charles J. Sih*

School of Pharmacy, University of Wisconsin,
425 N. Charter St., Madison, WI 53706-1515 U.S.A.

Abstract: The mono- and di-iodo derivatives of 1-oxaspiro[2,5]bicycloocta-4,7-dien-6-one, **8** and **9**, reacted readily with 3,5-diiodo-L-tyrosine at pH 8.0 to give 3,5,3'-triiodo-L-thyronine (T_3) and thyroxine in 70% and 94% yields respectively. In turn, **8** and **9** were prepared by the sodium bismuthate oxidation of their corresponding iodinated *p*-hydroxybenzyl alcohol derivatives, **6** and **7** in 32% and 37% yields.

While the enzymatic formation of thyroxine (T_4) from the oxidative coupling of two 3,5-diiodo-L-tyrosine (DIT) residues was reported 50 years ago[1], the intimate mechanistic details of this interesting transformation remain to be clarified. Two possible mechanisms, referred to as intramolecular and intermolecular coupling, have been proposed for the formation of T_4 in the thyroid gland.[2] However, at the present time it is not definitively known whether T_4 formation *in vivo* predominantly involves the intramolecular or the intermolecular coupling mechanism.

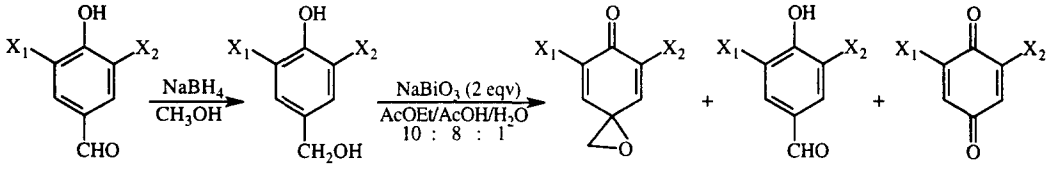
Intramolecular coupling involves oxidation of DIT to a free radical and then two DIT radicals interact to form T_4 through a quinol ether intermediate.[3] Although earlier investigators visualized that the coupling reaction involved the conversion of free DIT to free T_4, later studies showed that this process was more complicated and occurred within the thyroglobulin molecule.[4]

Intermolecular coupling was formulated from the observation that, in the presence of oxygen, DIT reacted with an active intermediate, which was formed during the oxygenation of 4-hydroxy-3,5-diiodophenylpyruvic acid (DIHPPA), to form T_4.[5] Moreover, DIHPPA also reacted with DIT residues in thyroglobulin to generate T_4.[6]

Recently, we rigorously investigated the reaction of DIHPPA with molecular oxygen and found that several unstable compounds possessing the basic spiro-4-epoxycyclohexadienone nucleus were produced.[7] Because these active species reacted readily with DIT to give T_4, we decided to investigate the chemical properties of 1-oxaspiro[2,5]bicycloocta-4,7-dien-6-one (**1**) and its iodo derivatives, **8** and **9**, with a view to utilizing them as synthons for the preparation of iodothyronines. It was reported several years ago that *p*-hydroxybenzyl alcohol, on treatment with sodium bismuthate in aqueous acetic acid, afforded the quinol epoxide **1**, as the major product in 20% yield.[8] Other derivatives of **1** were also reported,[9] but these possessed substituents in the cyclohexadienone ring and thus

were unsuitable for our use as synthetic intermediates.

By altering the reaction conditions using commercially available alcohol, **5**, we were able to improve the yield of **1** to 35%. Using this modified procedure, we successfully prepared the corresponding iodo epoxides, **8** and **9**, for the first time.



2 $X_1=X_2=H$
3 $X_1=I$; $X_2=H$
4 $X_1=X_2=I$

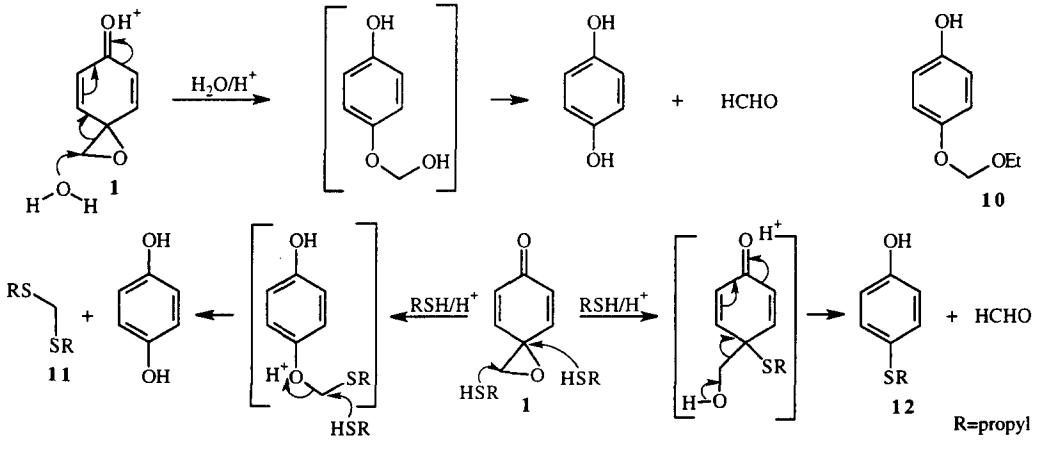
5 $X_1=X_2=H$
6 $X_1=I$; $X_2=H$
7 $X_1=X_2=I$

1 $X_1=X_2=H$ (50 min, 35%)
8 $X_1=I$; $X_2=H$ (80 min, 32%)
9 $X_1=X_2=I$ (4 h, 37%)

< 5%

~2%

Alcohols **6** and **7** were obtained by reduction of aldehydes **3**[10] and **4**[11] with an excess of sodium borohydride in methanol. Sodium bismuthate oxidations[12] of **5**, **6** and **7** afforded the desired epoxides **1**, **8** and **9**,[13] respectively, in comparable yields, accompanied by two minor side products, aldehydes **2**, **3** and **4**, and the corresponding benzoquinones; in each case, only a minor trace of unreacted alcohols was detected. As the dark colored polar byproducts (not identified) generated during the reactions adsorbed tightly onto the silica gel, they were easily separated from the reaction products by silica gel column chromatography using benzene as the mobile phase in all cases.

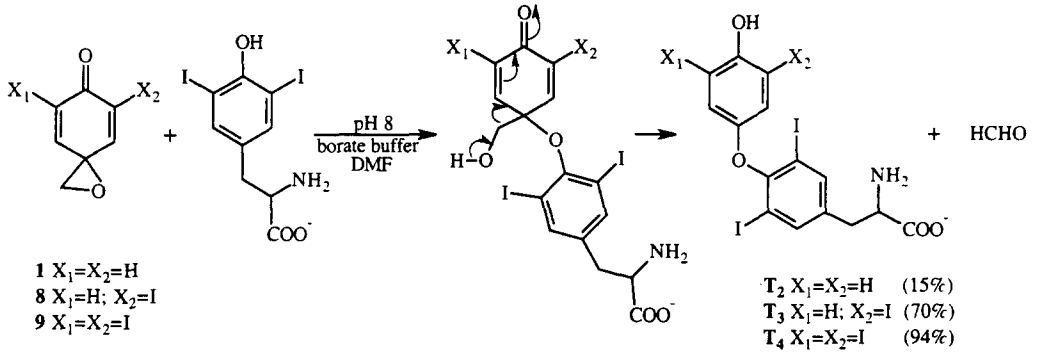


1
10
11
12

The chemical behavior of epoxides **1**, **8** and **9** towards different nucleophiles was found to be rather intriguing. Treatment of **1** with dilute aqueous hydrochloric acid resulted in the rapid hydrolysis of **1** to give formaldehyde and hydroquinone, but not via the *p*-quinol intermediate as had been proposed.[8] This mechanism of oxirane opening was confirmed by using an excess of ethanol instead of water in the presence of a catalytic

amount of TFA. The only product of this reaction was acetal **10**[14] (isolated in 91% yield), indicating that an unique epoxide ring opening reaction had occurred. It appeared that cleavage of the C–C bond was favored by the strong tendency of the cyclohexadienone to undergo aromatization. Thiolysis of **1** with 1-propanethiol, in the presence of TFA, proceeded in a similar fashion. That is, the major products of the reaction were dithioacetal **11** and hydroquinone both of which were formed in 70% yield. However, being a better nucleophile than ethanol, the thiol also reacted at the *ipso* position of **1** to yield thioether **12** (20%).

None of the epoxides (**1**, **8** and **9**) reacted with *N*-acetyl-3,5-diiodo-L-tyrosine methyl ester in the presence of TFA. Under these conditions, they all isomerized to their correponding aldehydes, **2**, **3** and **4**. On the other hand, all three epoxides reacted with unprotected DIT (2 eqv), at 24°C for 16 hrs in 0.2 M sodium borate buffer, pH 8.0, to afford their corresponding iodothyronines in modest to excellent yields. For epoxide **9**, the reaction proceeded very smoothly in high yields and only a trace of aldehyde **4** was detected.[15] Apparently, the electrophilicity at the *ipso* position in **9** is further enhanced by the iodine atoms on the cyclohexadienenone ring. This greatly facilitated S_N2 attack by DIT to furnish an unstable vinylog of a β-hydroxyketone that underwent spontaneous retro-aldolization[16] to yield T_4.



1 $X_1=X_2=H$
8 $X_1=H$; $X_2=I$
9 $X_1=X_2=I$

T_2 $X_1=X_2=H$ (15%)
T_3 $X_1=H$; $X_2=I$ (70%)
T_4 $X_1=X_2=I$ (94%)

L-Thyroxine (white powder), isolated from the reaction mixture by simple filtration, was washed with acetone and dried. However, the isolation of T_3 and T_2 required *n*-butanol extraction[17] from diluted alkaline (pH 11) reaction mixtures. All of the analytical data for T_4, T_3 and T_2 were found to be in accord with authentic samples.

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References and Notes

1. Harington, C. R.; Barger, G. *Biochem. J.* **1927**, *21*, 169-183.
2. Degroot, L. J.; Niepomniszcze, H. *Prog. in Endocrinol. and Met.* **1977**, 665-718.
3. Taurog, A.; Dorris, M.; Doerge, D. R. *Arch. Biochem. & Biophys.* **1994**, *315*. 82-89. See also: Johnson, T.; Tewkesbury, L. *Proc. Natl. Acad. Sci. USA* **1942**, *23*, 73-77.
4. Harington, C. R.; Randall, S. S. *Biochem. J.* **1929**, *23*, 373-383.
5. Hillmann, G.; Keil, B.; Tashimi, P. *Z. Naturforsch.* **1961**, *16*, 28-32. Meltzer, R. I.; Stanaback, R. J. *J. Org. Chem.*, **1961**, *26*, 1977-1979.
6. Toi, K.; Salvatore, G.; Cahnmann, H. J. *Biochim. Biophys. Acta* **1965**, *97*, 523-531.
7. Oza, V. B.; Salamonczyk, G. M.; Guo, Z. W.; Sih, C. J. Submitted to *J. Am. Chem. Soc.*
8. Adler, E.; Holmberg, K.; Ryrfors, L. O. *Acta Chem. Scand.* **1979**, *B28*, 883-887.
9. Eistert, B.; Bock, G. *Chem. Ber.* **1959**, *92*, 1247-1257. Starnes, W. H. *J. Org. Chem.* **1970**, *35*, 1974-1978.
10. Barnes, J. H.; Borrows, E. T.; Elks, J.; Hems, B. A.; Long, A. G. *J. Chem. Soc.* **1950**, 2824-2831.
11. Matsuura, T.; Cahnmann, H. J. *J. Am. Chem. Soc.* **1959**, *81*, 871-878.
12. A 0.55 M solution of the corresponding alcohol in AcOEt/AcOH/H_2O (10:8:1) at rt was used.
13. NMR (300 MHz, $CDCl_3$) data for **8** and **9**: **8**: δ ^{1}H 3.40 (AB, *J*=10.8 Hz, 2H), 6.60 (dd, *J*=10.04 1.8 Hz, 1H); 6.61 (d, *J*=10.0 Hz, 1H); 7.26 (d, *J*=1.8 Hz, 1H) ppm; δ ^{13}C 54.84, 55.62, 107.9, 130.3, 146.5, 159.7, 179.1 ppm. **9**: δ ^{1}H 3.58 (s, 2H), 7.42 (s, 2H) ppm; δ ^{13}C 54.35, 57.97, 101.1, 155.8 (2C), 174.6 ppm.
14. NMR ($CDCl_3$) of **10**: δ ^{1}H 1.11 (t, *J*=7.6 Hz, 3H), 3.72 (q, *J*=7.6 Hz, 2H), 5.16 (s, 2H), 6.85 (AB, *J*=10.9 Hz, 5H) ppm; δ ^{13}C 15.10, 64.08, 94.07, 116.0, 117.8, 150.2, 153.0 ppm.
15. Preparation of L-thyroxine: 90 mg (0.21 mmol) of 3,5-diiodo-L-tyrosine was dissolved in 10 mL of sodium borate buffer (0.2 M, pH 8). The pH of the resulting solution was then readjusted to 8.0 using 0.1 M NaOH. To this, a solution of 40 mg (0.11 mmol) of epoxide **9** in 2 mL of DMF was added. The cloudy reaction mixture was stirred at 24°C overnight. The precipitated T_4 was filtered off, washed (3 times) with 2 mL of acetone and dried *in vacuo*. Yield 77 mg (94%). The purity of the sample was found to be greater than 97% by HPLC and ^{1}H NMR analyses.
16. Sih, C. J.; Lee, S. S.; Tsong, Y. Y.; Wang, K. C. *J. Biol. Chem.* **1966**, *241*, 540-550.
17. Nishinaga, A.; Cahnmann, H. J.; Kou, H.; Matsuura, T. *Biochemistry* **1968**, *7*, 388-397.

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