irradiated through uranium glass for 30 h. Fractional distillation of the reaction mixture gave, besides 0.33 g of recovered **2**, an oil, bp 36–54 °C (0.8 mm), from which was isolated by GC on column B the following products: 0.070 g of 2,3,6,7-tetramethylocta-2,6-diene (**7**), identical with material isolated in other studies;[1,4] and 0.086 g (17%) of alcohol **8**, 2,3-dimethyl-1-buten-3-ol [IR (film) 3580 cm^{-1}; NMR (CCl_4) τ 5.2 (2 H, s, br), 8.02 (1 H, s, br), 8.3 (3 H, s, br), 8.56 (6 H, s); Anal. $C_6H_{12}O$ (C, H)].

Irradiation of 3-Benzoylfuran and 3-Acetylfuran with Alkene 5. Irradiation of hexane solutions of 3-benzoylfuran (**3**)[9] with excess alkene **5** through uranium glass filters for up to 40 h gave yellow solutions from which 10–20% of diene **7** could be isolated by vacuum distillation. Chromatography of the residue on silica gel gave only ~30% of recovered **3** and high molecular weight material.

From similar experiments with **4**, using a Pyrex filter, there was isolated ~70% of recovered ketone, together with 17% of **7**, 11% of **8**, and small amounts of tarry residue.

Photolysis of 3-Formylthiophene (9) with 2,3-Dimethyl-2-Butene. Irradiation of **9** (1.4 g, 0.012 mol) with alkene **5** (20 g) in spectrograde hexane through Pyrex for 2.5 h led to complete consumption of aldehyde. Workup as described above for **2** led to the isolation of (a) alkene **12** (retention time on column B at 110°, 5.8 min.) in 46% yield [IR (film) 1080 cm^{-1}; NMR τ 2.7–3.1 (3 H, m), 3.7 (1 H, m, br), 8.0 (6 H, s, br); mass spectrum (CI) *m/e* 138 (P, 100), 133 (72), 121 (67); Anal. $C_8H_{10}S$ (C, H)], and (b) 2 + 2 ring adduct **15** (retention time 16 min) [IR (film) 1720 cm^{-1}; NMR τ 3.7 (1 H, m), 3.9 (2 H, m, br), 6.8 (1 H, s), 8.45, 8.69, 8.80, and 8.82 (3 H each, s); mass spectrum *m/e* 196 (P, 7), 181 (19), 112 (47), 84 (100); Anal. $C_{11}H_{16}OS$ (C, H)].

The photochemical reactions of aldehydes **10**, **16**, and **17** with **5** were conducted in the same manner. Spectral data on the products are given in Table I.

Photochemical Reaction of Di-2-thienyl Ketone with 2,3-Dimethyl-2-butene. A solution of ketone **20** (1.0 g) and alkene **5** (20 g) in spectrograde hexane was irradiated through a uranium glass filter for 6 h. Evaporation of solvent and excess alkene gave a yellow residue which, on trituration with 3:1 hexane–benzene, partially crystallized. Recrystallization of the solid from benzene–hexane gave 0.56 g of white prisms: mp 127–128 °C; IR (KBr) 3500 cm^{-1}; NMR ($CDCl_3$) τ 2.6–3.2 (12 H, m), 6.49 (2 H, s, br); mass spectrum *m/e* 310 (P, CI). Anal. $C_{18}H_{14}O_2S_4$ (C, H).

Acknowledgments. The author is grateful to William Landis of the National Institutes of Health for the mass spectra of the new compounds reported herein, and to the referees for suggestions.

Registry No.—**1**, 6453-99-2; **2**, 1468-83-3; **3**, 6453-98-1; **4**, 14313-09-8; **5**, 563-79-1; **6**, 63466-41-1; **7**, 18495-18-6; **8**, 10473-13-9; **9**, 498-62-4; **10**, 498-60-2; **12**, 63466-42-2; **13**, 63466-43-3; **15**, 63466-44-4; **16**, 98-03-3; **17**, 98-01-1; **18**, 63466-45-5; **19**, 63466-46-6; **20**, 704-38-1; **21**, 51248-22-7.

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Deuterium or Tritium Labeling by Ionic Hydrogenation. A Convenient Route to Specifically Labeled Dethiobiotin

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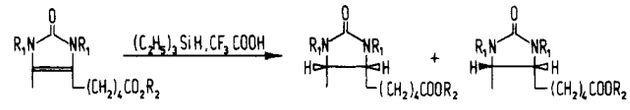
In connection with our investigation of the biosynthetic conversion of dethiobiotin to biotin, we had to synthesize specifically C-3 and/or C-4 deuterated and tritiated *cis*-dethiobiotin.[1] One convenient route was the reduction of the unsaturated precursor 4-methyl-5-(ω-carboethoxyamyl)-2-imidazolone (**1a**).[2] After a thorough investigation we found that ionic hydrogenation was the only efficient noncatalytic method to reduce the carbon–carbon double bond of the imidazolone ring.[3]

Scheme I



1a, R_1=H; R_2=C_2H_5
1b, R_1=Ac; R_2=C_2H_5

2a, R_1=H; R_2=C_2H_5
2b, R_1=Ac; R_2=C_2H_5
2c, R_1=H; R_2=H

3a, R_1=H; R_2=C_2H_5
3b, R_1=Ac; R_2=C_2H_5
3c, R_1=H; R_2=H

This paper reports our study of the scope of this reduction and observations regarding the stereoselectivity of the reaction and regioselectivity of the labeling with deuterated silanes.

We also explore the possibility of using the different combinations of hydrogenating pairs CF_3COOH, CF_3COOD, R_3SiH, and R_3SiD to invert the labeling regioselectivity or to prepare dideuterated compounds. The corresponding tritiated compound has been obtained with Et_3Si^3H. Reductions with Et_3Si^3H which have not yet been performed appear of general interest for specific tritiation of organic molecules.

Results and Discussion

Stereoselectivity. Since our goal was to obtain *cis*-dethiobiotin, we investigated the factors which might influence the course of hydrogenation and lead to the desired isomer (Table I).

1a treated with 1 equiv of Et_3SiH or Et_3SiD in CF_3COOH at 50 °C afforded in 70% yield a 1/1 mixture of *cis*- and *trans*-(*dl*) dethiobiotin ethyl ester (**2a** and **3a**) (run 1, see also Scheme I).

These isomers have been separated by TLC as *N*-diacetyl derivatives **2b** and **3b**. The cis isomer **2b** has been identified after treatment with sodium hydroxide by comparison with an authentic sample of dethiobiotin obtained by Raney nickel desulfuration of biotin.[4] The structure of the trans isomer **3b** was based on NMR and mass spectral data (see Experimental Section).[5] We also carried out, with the same hydrogenating pair, the reduction of the *N,N'*-diacetyl derivative **1b** (run 2), the double bond of which is more reactive because of the dearomatization of the imidazolone ring.[6] In this case, we observed an important variation of the stereoselectivity with a high predominance of the cis isomer (cis/trans: 95/5). The same variation in isomer ratio is observed for reductions of 3,4-dimethyl-2-imidazolone (**4a**) and its *N,N'*-diacetyl derivative **4b** (runs 6, 7).

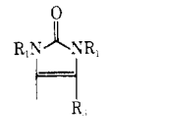
On the other hand, the variation of steric bulk of the different hydride donors tested, Et_3SiD, Ph_3SiD, Ph_2SiD_2, and Ph_3GeD, does not lead to significant variations (runs 2–5).

Since examination of molecular models shows that the acetyl groups in **1b** and **4b** do not increase significantly, with respect to **1a** and **4a**, the steric discrimination between the two faces of the imidazolone ring, our results clearly show that steric interaction between the intermediate carbenium ion and hydride donor is not the major factor governing the stereochemistry of ionic hydrogenation as previously claimed.[7a,8]

Regioselectivity. In order to select the best conditions for specific incorporation of deuterium at C-3 and/or C-4, we carried out reduction of **1a** and **1b** using hydride donors of different donating ability and steric bulk.[7a,9]

Some results are listed in Table II. In runs 1–4 the isolated dethiobiotin has incorporated, as expected, only one deuterium atom (mass spectrometry determination). There is al-

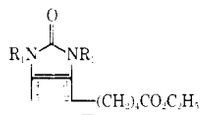
Table I. Stereoselectivity of the Reduction of the Double Bond of 2-Imidazolone Derivatives by Ionic Hydrogenation



Run no.	R_1	R_3	Registry no.	Hydride donor	Time, h	Temp, °C	% yield[c]	% cis[d]
1	H	$(CH_2)_5COOEt$	63466-47-7	Et_3SiH or Et_3SiD[a]	20	50	70	50
2	Ac	$(CH_2)_5COOEt$	27051-51-0	Et_3SiH or Et_3SiD[a]	60	25	70	95
3	Ac	$(CH_2)_5COOEt$		Ph_3SiD[b]	60	25	30	96
4	Ac	$(CH_2)_5COOEt$		Ph_2SiD_2[b]	60	25	23	94
5	Ac	$(CH_2)_5COOEt$		Ph_3GeD[b]	20	25	27	98
6	H	CH_3	1072-89-5	Et_3SiH[a]	20	50	65	50
7	Ac	CH_3	21265-71-4	Et_3SiH[a]	20	25	78	92

[a] Solvent CF_3COOH. [b] Solvent CF_3COOH + CH_2Cl_2, added for homogeneity. [c] Total yield cis + trans. [d] Accuracy on % cis isomer (determined by GLC, cf. Experimental Section) was evaluated to 1%. The reproducibility of these data, within these limits, was checked over several experiments.

Table II. Deuterium Incorporation in Dethiobiotin Obtained by Ionic Hydrogenation of 4-Methyl-5-(ω-carboethoxyamyl)-2-imidazolone



Run no.	R_1 =	Hydrogenating pair[a]	No. of D atoms on the indicated carbon[b] C-3	C-4	C-2	C-5
1	H	Et_3SiD/CF_3COOH	0.30	0.70	0	0
2	Ac	Et_3SiD/CF_3COOH	0.15	0.85	0	0
3	Ac	Ph_3SiD/CF_3COOH	0.24	0.76	0	0
4	Ac	Ph_2SiD_2/CF_3COOH	0.07	0.93	0	0
5	Ac	Ph_3GeD/CF_3COOD	0.96	0.96	0.6	1.2
8	Ac	Et_3SiH/CF_3COOD	0.79	0.10	1.2	2.1
9	H	Et_3SiD/CF_3COOD	0.96	0.96	0.8	1.9
10	Ac	Et_3SiD/CF_3COOD	0.96	0.96	0.9	1.8

[a] The experimental conditions are the same as those used for the corresponding runs listed Table I. [b] Using a strip chart recorder at low speed, we evaluated the accuracy of deuterium incorporation to 1% at C-3 and C-4 for runs 1—4. The reproducibility of these data, within these limits, was checked for runs 1 and 2 over several experiments.

ways a large predominance of the C-4 deuterated product. The regioselectivity is sensitive to the nature of the substrate (runs 1, 2) and also to the nature of silane (runs 2–4); it reaches 93% for the reduction of **1b** with Ph_2SiD_2.

It was attractive to prepare dideuterated compounds with R_3SiD in CF_3COOD or to invert the regioselectivity of the labeling with R_3SiH/CF_3COOD instead of R_3SiD/CF_3COOH. The use of deuterated solvent for this purpose has been suggested, but it has never been investigated.[7b] In CF_3COOD one cannot ignore the risk of further deuterium incorporation through olefin formation by loss of a proton α to the carbenium ion, the importance of such a competing process depending on its relative rate with respect to the trapping rate of carbenium ion by hydride donor. We carried out some experiments in CF_3COOD to ascertain this point.

Reduction of **1b** with Et_3SiH/CF_3COOD (run 8) inverts effectively the deuterium incorporation at C-3 and C-4, but unfortunately there is a fairly large deuterium incorporation at C-2 and C-5.[10]

However, this process is greatly minimized when a more reactive hydride donor like Ph_3GeD is used (run 5). This suggests that it should be possible to block this undesired reaction in some favorable cases.

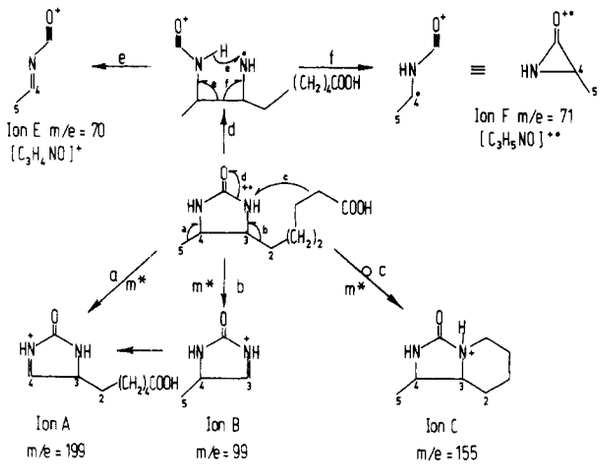
Synthesis of Tritiated Dethiobiotin. The experimental conditions described above (run 2, Tables I and II), which lead, in satisfactory yield and with excellent reproducibility, to a highly stereoselective and regioselective deuteration of **1b**, were selected to carry out the synthesis of (±)-dethiobiotin specifically tritiated at C-3 and C-4, using Et_3Si^3H prepared by LiB^3H_4 reduction of Et_3SiCl.

Since the primary kinetic isotope effect k_H/k_D measured for carbenium ion–silane hydride and deuteride transfer reaction are small,[11] we assume the same distribution of the label in deuterated and tritiated dethiobiotin prepared in the same conditions.

The use of tritiated organosilicon compounds has not yet been explored. Our finding that Et_3Si^3H of high specific activity can be readily prepared opens a valuable route for tritiation via ionic hydrogenation of olefins or more generally in carbenium ion hydride transfer reactions.

Deuterium Localization by Mass Spectrometry. Different fragmentations are found at high and low ionizing voltage. Relying on structural variation in the side chain and on deuterium labeling we could analyze the structure of the different ions and find out consistent fragmentation patterns which permit accurate localization of deuterium (Scheme II). The possibility of isotope scrambling was also ruled out by these experiments.[12] The most intense peaks in the mass spectrum of dethiobiotin at 70 eV are *m/e* 199, 155, 99. Fragmentation of the imidazolidone ring does not occur at 70 eV, but it becomes significant at low ionization potential and gives peaks at *m/e* 71 and 70 at 11 eV (ratio 9/1), the elemental composition of which was determined by high-resolution mass measurements. The molecular peak ($M^{+\cdot}$) at *m/e* 214 is not abundant and is always accompanied by peak (MH^+) at *m/e* 215 (due to chemical ionization in the source). Consequently this peak must be rejected for measuring the total deuterium content. But the ion C, in which hydrogen (or deuterium) atoms are kept intact at positions 2, 3, 4, and 5, can be used advantageously for this determination. For runs 1–4 deuterium content at C-3 and C-4 can be easily deduced from direct measurement of the peak intensities corresponding to ions C,

Scheme II



E, and F. When labeling occurs at C-2, C-3, C-4, and C-5 positions (runs 5–8), deuterium content at C-2 and C-5 is deduced from the relative heights of the peak due to ions A, B, C, E, and F.

Experimental Section

NMR spectra were recorded on a Varian HA 100 spectrometer in D_2O and chemical shifts are reported in parts per million (δ) from external HMDS. The gas chromatograph used was a GIRDEL 3000 unit. The mass spectra were obtained using a ATLAS CH_5 mass spectrometer (source at 210 °C). Scintillation counting was carried out with a Intertechnique SL_{30} spectrometer in Bray's liquor. All the results were corrected for quenching by external standard method.

Material. LiB^3H_4 (1 Ci/mmol) was obtained from C.E.A. Saclay France. All the deuterated silanes and germanes used in this study were prepared by $LiAlD_4$ reduction of the corresponding chloro compounds according to known methods.[13] No ≡SiH or ≡GeH could be detected by NMR.

The 4-methyl-5-(ω-carboethoxyamyl)-2-imidazolone (**1a**) and *N,N'*-diacetyl-4-methyl-5-(ω-carboethoxyamyl)-2-imidazolone (**1b**) were prepared in good yield according to Duschinky and Dolan.[2]

Preparation of Labeled Dethiobiotins. A typical reduction experiment was performed as follows.

Reduction of 4-Methyl-5-(ω-carboethoxyamyl)-2-imidazolone. To a mixture of 360 mg (1.5×10^{-3} mol) of **1a** in 1 mL of CF_3COOH, 180 mg (1.5×10^{-3} mol) of Et_3SiH is added gradually. The mixture is kept at 50 °C under shaking. The reduction progress is followed by NMR (disappearance of the $C(5)H_3$—C(4)= signal, singlet at δ 1.95/Me_4Si). The reduction is completed after 20 h. The excess of CF_3COOH and $Et_3SiOOCCF_3$ which has been produced is removed in vacuo. The crude material is then acetylated by two short refluxings with 5 mL of acetic anhydride, the excess of which is distilled off.

The cis/trans ratio **2b/3b** is determined on the crude acetylated mixture by GLC (SE 30 10% on Chromosorb G, WHMDS), **2b/3b** = 1/1. The preparative separation of **2b** and **3b** is performed by silica gel TLC (eluent: ethyl acetate–chloroform, 2/8).

After separation of **2b** and **3b**, saponification of each isomer with 1 N sodium hydroxide (20 °C, 2 h) afforded the corresponding dethiobiotins **2c** and **3c**.

Purification of **2c** and **3c** is carried out on a Dowex AG 50 WX_2 formate column. Dethiobiotin is eluted with 0.05 M formic acid. Total yield (**2c** + **3c**) = 70%: **2c**, mp 159 °C (lit. mp 159 °C);[4] **3c**, mp 156 °C.

The structures of **2c** and **3c** are determined by NMR and mass spectrometry. **2c:** NMR δ 1.44 (3 H, d, J = 6 Hz, CH_3CH), 4.11 (2 H, m, H_3, H_4), 2.45 (2 H, t, J = 7 Hz, $-CH_2COOH$); mass spectrum *m/e* 214 (M^+), 199, 155, 99. **3c:** NMR δ 1.54 (3 H, d, J = 5, 7 Hz, CH_3CH), 3.76 (2 H, m, H_3, H_4), 2.45 (3 H, t, J = 7 Hz, $-CH_2COOH$); mass spectrum *m/e* 214 (M^+), 199, 155, 99.

When the reduction is carried out with Et_3SiD in CF_3COOH, the deuterium distribution at C-4, and consequently at C-3, can be deduced from the integration of the signals: δ 1.43 (3 H, s, CH_3CD) and 1.44 (3 H, d, J = 6 Hz, CH_3CH). However, a better accuracy is obtained by mass spectroscopy.

[^{3}H]Triethylsilane. LiB^3H_4 (88 mg, 4×10^{-3} mol, 1 Ci/mmol) was allowed to react with Et_3SiCl (0.6 g, 4×10^{-3} mol) in 4 mL of dry triglyme under nitrogen at 50 °C. Et_3Si^3H was distilled and trapped at −70 °C in vacuo. Its chemical purity was controlled by GLC (SE 30 20% on Chromosorb Z): yield, 98%, 470 mg; 0.2 Ci/mmol.

Tritiated Dethiobiotin. Et_3Si^3H, prepared as described, was transferred into a flask containing 242 mg (0.75×10^{-3} mol) of *N,N'*-diacetyl-4-methyl-5-(ω-carboethoxyamyl)-2-imidazolone in 2 mL of CF_3COOH (freshly distilled on H_2SO_4). After 60 h at room temperature, excess Et_3Si^3H, CF_3COOH, and $Et_3SiOOCCF_3$ produced were removed in vacuo. Saponification and deacetylation of the crude residue with 1 N sodium hydroxide at 20 °C during 2 h afforded dethiobiotin. The pH of the solution was adjusted to 8 and then the solution was taken on to a Dowex AG 50 WX_2 formate column for purification. Dethiobiotin eluted with 0.05 M formic acid was obtained in 75% yield: 120 mg; mp 159 °C (198 mCi/mmol). The structure and purity were controlled by mass spectrometry and radiochromatography.

Acknowledgments. We are indebted to Dr. Pichat for helpful advice and for allowing one of us (G.G) to prepare (±)-[^{3}H]dethiobiotin in his laboratory (Service des Molecules Marquees, C.E.A. Saclay).

Registry No.—**2a**, 63526-69-2; **2a** C-3 deuterated derivative, 63466-48-8; **2a** C-4 deuterated derivative, 63466-49-9; **2a** deuterated derivative, 63466-50-2; **2b**, 63466-51-3; **2b** C-3 deuterated derivative, 63466-52-4; **2b** C-4 deuterated derivative, 63466-53-5; **2b** deuterated derivative, 63466-54-6; **2b** C-3 tritiated derivative, 63466-55-7; **2b** C-4 tritiated derivative, 63466-56-8; **2c**, 636-20-4; **3a**, 63526-70-5; **3a** C-3 deuterated derivative, 63466-57-9; **3a** C-4 deuterated derivative, 63466-58-0; **3a** deuterated derivative, 63466-59-1; **3b**, 63466-60-4; **3b** C-3 deuterated derivative, 63466-61-5; **3b** C-4 deuterated derivative, 63466-62-6; **3b** deuterated derivative, 63466-63-7; **3b** C-3 tritiated derivative, 63466-64-8; **3b** C-4 tritiated derivative, 63466-65-9; **3c**, 34879-36-2; Et_3SiT, 63466-66-0; LiB^3H_4, 23683-78-5; Et_3SiCl, 994-30-9; Et_3SiD, 1631-33-0; Ph_3SiD, 18536-60-2; Ph_2SiD_2, 17950-94-6; Ph_3GeD, 2816-42-4; Et_3SiH, 617-86-7; *cis*-3,4-dimethyl-2-imidazolinone, 63466-67-1; *trans*-3,4-dimethyl-2-imidazolinone, 63466-68-2; *cis*-3,4-dimethyl-*N,N'*-diacetylimidazolin-2-one, 63466-69-3; *trans*-3,4-dimethyl-*N,N'*-diacetylimidzolin-2-one, 63466-70-6.

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Nucleophilic Substitution with Inversion of Alcohol Configuration with the Reagent Complex Triphenylphosphine–Diethyl Azodicarboxylate–Carboxylic Acid. A Convenient Preparation of Epicholesterol

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Several methods have been reported recently for preparing epicholesterol (**1**) from cholesterol (**2**) in fair to good yield.[1,2]