[CONTRIBUTION FROM THE LEDERLE LABORATORIES DIVISION OF AMERICAN CYANAMID CO.]

$\Delta^{5,7}$-STEROIDS. V (1). 7-DEHYDROCHOLESTERYL MERCAPTAN

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In a further extension of the NBS[1] (Wohl-Zeigler) method (1) for the preparation of $\Delta^{5,7}$-steroids, we have succeeded in preparing 7-dehydrocholesteryl mercaptan (provitamin D_3 mercaptan) (VI) from cholesteryl mercaptan (I).[2]

Cholesteryl mercaptan (I), prepared by the method of King, Dodson, and Subluskey (2), was converted in the usual manner to the previously unreported thiobenzoate (III). Treatment of III in petroleum ether and carbon tetrachloride with NBS, with subsequent dehydrobromination of the intermediate bromo-compound (IV) by *s*-collidine in xylene gave the $\Delta^{5,7}$-thiobenzoate (V). In one experiment, the bromo-intermediate, 7α(?)-bromocholesteryl thiobenzoate (IV),[3] was isolated and characterized. On dehydrobromination it also gave the expected thiobenzoate (V). Hydrolysis with either alcoholic potash or potassium carbonate in ethanol gave the desired 7-dehydrocholesteryl mercaptan (VI). The mercaptan

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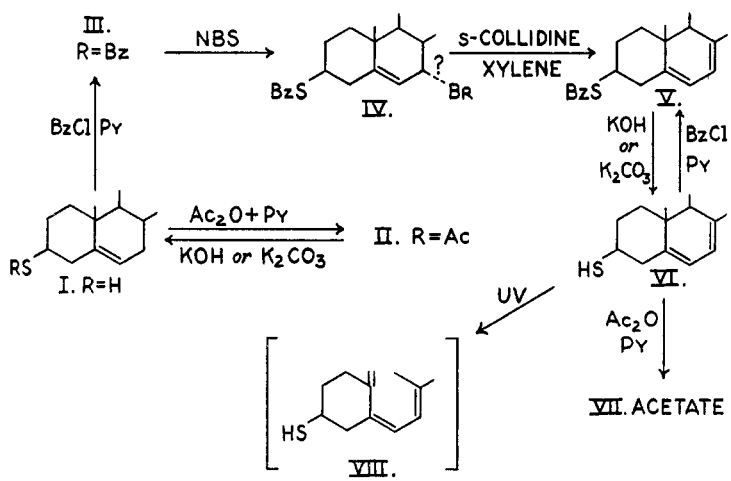

[1] NBS = N-Bromosuccinimide.

[2] While this manuscript was in the process of preparation for publication, there appeared an article by Strating and Backer, *Rec. trav. chim.*, **69**, 909 (1950), on the preparation of 7-dehydrocholesteryl mercaptan along similar lines. Their method of synthesis is as follows: A. Cholesteryl rhodanide $\xrightarrow{\text{NBS}}$ 7-bromocholesteryl rhodanide $\xrightarrow{\text{Collidine}}$ 7-dehydrocholesteryl rhodanide $\xrightarrow{LiAlH_4}$ 7-dehydrocholesteryl mercaptan; B. 7-Dehydrocholesteryl rhodanide $\xrightarrow{NaOC_2H_5}$ 7-dehydrocholesteryl disulfide $\xrightarrow{LiAlH_4}$ 7-dehydrocholesteryl mercaptan.

[3] There is insufficient evidence at present to indicate whether or not allylic bromination with NBS of Δ^5-steroids at the C-7 position is stereospecific. The assignment of the α-configuration at the C-7 position follows that of Fieser, *Experientia*, **6**, 312 (1950).

(VI) was further characterized by the preparation of its acetate (VII); on benzoylation of VI the benzoate (V) produced was identical in all respects with the material prepared directly from cholesteryl thiobenzoate (III) by the NBS method.

In one experiment the above transformation was carried out on cholesteryl thioacetate (II) rather than on the thiobenzoate (III). Difficulty was encountered in isolating a pure product, and, consequently, the utility of this ester in this preparation was not further investigated. A similar result has been recorded by Strating and Backer (*loc. cit.*) who also were unable to isolate pure 7-dehydrocholesteryl thioacetate (VII). These investigators made no mention of the possibility of utilizing the thiobenzoate (III), which was highly successful in our hands. Strating and Backer abandoned thioesters in favor of the rhodanide. Their decision was based in part on the anticipation of encountering difficulty in the hydrolysis of $\Delta^{5,7}$-thioesters. Strating and Backer reported that they did not "succeed" in isolating pure cholesteryl mercaptan (I) from the thioacetate (II). In our experience, hydrolysis of cholesteryl thioacetate (II) with potassium carbonate in ethanol gave an 88% yield of pure mercaptan (I); with alcoholic potash an 86% yield was obtained. In both hydrolyses traces of cholesteryl disulfide were isolated. The cholesteryl mercaptan (I) was identical with that prepared by King, Dodson, and Subluskey's method (2), in which alkaline conditions prevailed.

The thiol group at the C-3 position in 7-dehydrocholesteryl mercaptan (VI) has been assigned the β-configuration in accordance with the work of Ralls, Dodson, and Riegel (3), who have presented evidence for assigning the β-configuration to this group in cholesteryl mercaptan (I).

It was interesting to observe that neither cholesteryl mercaptan[4] (I) nor 7-dehydrocholesteryl mercaptan (VI) gave a sparingly soluble digitonide with digitonin in 90% alcohol. This was also true for dehydroisoandrosteryl mercaptan (4).

In Figure I are presented the infrared absorption curves of compounds I, II, III, V, VI, and VII.

In Table I are presented the physical properties of cholesteryl mercaptan (I) and 7-dehydrocholesteryl mercaptan (VI), and their esters. Also included for comparison purposes are the ultraviolet absorption constants of cholesteryl benzoate and dehydroisoandrosteryl benzoate.

An examination of the ultraviolet spectra in Table I reveals the bathochromic effect of a C-3 sulfur atom. This effect with other pertinent observations has been discussed elsewhere (4). Also, it is apparently true that in the absorption spectra of $\Delta^{5,7}$-steroids [free steroid, acetate, and benzoate (1b), methyl ether (1d), chloride and bromide (1a)] that the relative order of magnitude (ϵ) of the principal maxima are $\epsilon_{282-285} > \epsilon_{271-274} > \epsilon_{293-297}$.[5] 7-Dehydrocholesteryl mer-

[4] Strating and Backer (*loc. cit.*) have made a similar observation with cholesteryl mercaptan.

[5] It will be noted that *p*-nitrobenzoates and 3,5-dinitrobenzoates are not included in

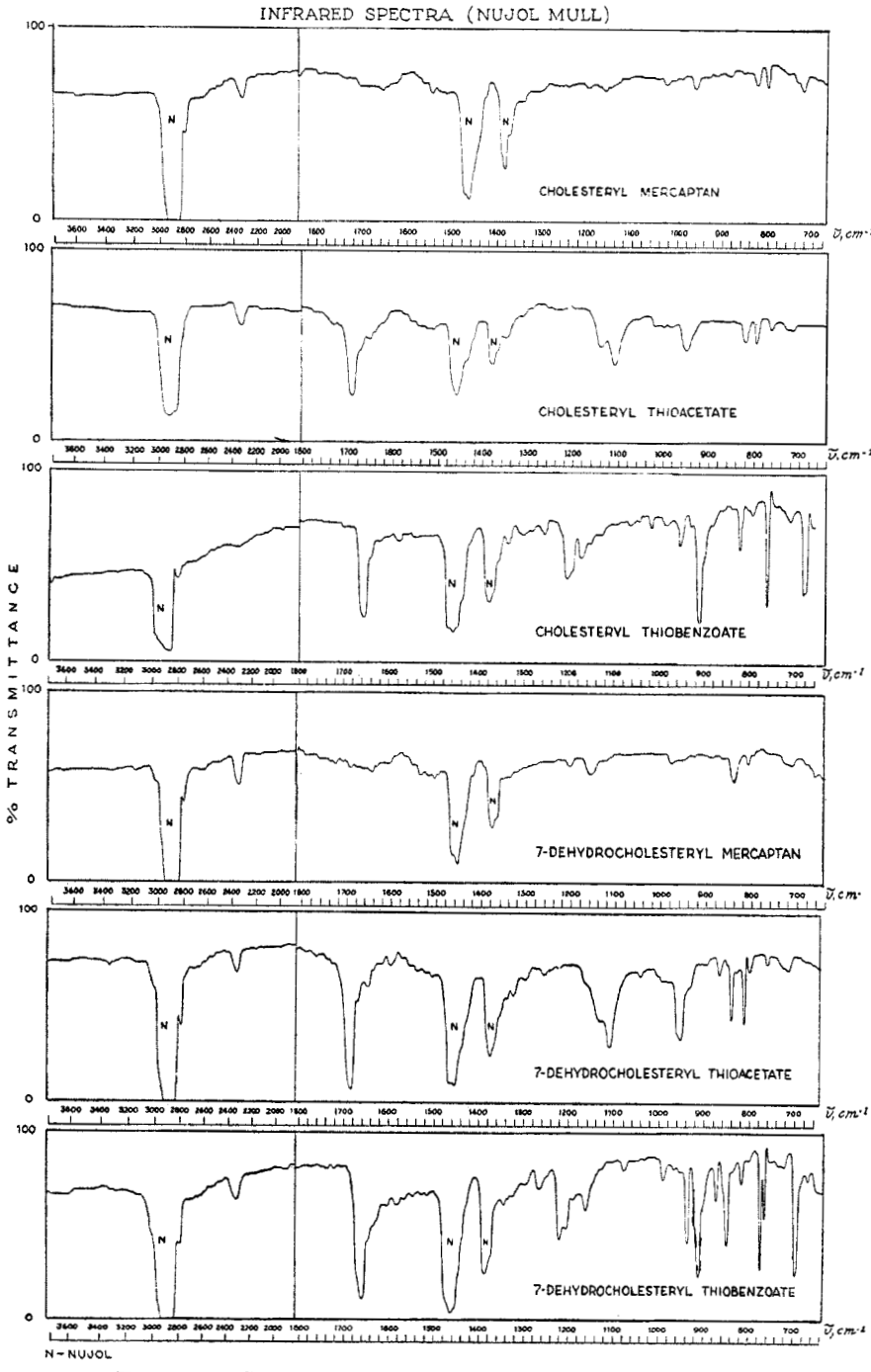

FIG. 1. $\Delta^{5,7}$-STEROIDS V. 7-DEHYDROCHOLESTERYL MERCAPTAN

TABLE I

PHYSICAL PROPERTIES

COMPOUND	M.P., C°.	$[\alpha]^{\circ}_{D}$	$[\alpha]^{\circ}_{Hg}$	α_{Hg}/α_{D}	U.V. ABSORPTION MAXIMA WITH MOLECULAR EXTINCTION COEFFICIENTS (1% CA)
Cholesteryl mercaptan[a]	96–99[b] 100–102 98–100	−23.1 −22.6 −22.7	−26.4 −26.8 −26.8	1.14 1.19 1.18	
Cholesteryl disulfide[a]	144–145	−42.7	−51.0	1.20	
Cholesteryl thioacetate	122.5–124.5	−49.6	−59.5	1.20	$\epsilon_{232.5\text{-}234} = 4630$
Cholesteryl thiobenzoate	160–162, 188	−27.2	−31.7	1.17	$\epsilon_{238\text{-}239} = 11{,}500$, $\epsilon_{269\text{-}271} = 8350$
7-Bromocholesteryl thiobenzoate	153–157.5	−139.2	−159.8	1.15	
7-Dehydrocholesteryl mercaptan	135–137	−90.9	−119.9	1.32	$\epsilon_{273} = 12{,}800$, $\epsilon_{283} = 13{,}550$, $\epsilon_{295} = 7800$ ($E^{1\%}_{1cm} = 318$, 336, 194 resp.)
7-Dehydrocholesteryl thioacetate	110–112, 137	−33.0	−43.7	1.37	$\epsilon_{231\text{-}232} = 5490$ (inflection), $\epsilon_{264\text{-}265.6} = 9000$ (inflection), $\epsilon_{273.5} = 12{,}800$, $\epsilon_{284} = 13{,}800$, $\epsilon_{295.5} = 7950$
7-Dehydrocholesteryl thiobenzoate	163–165, 210	+11.2	+16.2	1.45	$\epsilon_{239} = 13{,}000$, $\epsilon_{274} = 21{,}400$, $\epsilon_{284} = 20{,}750$, $\epsilon_{295} = 12{,}400$
Cholesteryl benzoate	—	—	—	—	$\epsilon_{229.5} = 15{,}300$, $\epsilon_{273} = 970$, $\epsilon_{280} = 760$
Dehydroisoandrosteryl benzoate	—	—	—	—	$\epsilon_{229} = 14{,}300$, $\epsilon_{272.5} = 990$, $\epsilon_{280} = 790$

[a] King, Dodson, and Subluskey (2), mercaptan, m.p. 97.0–97.5°, $[\alpha]^{25}_{D}$ −26.6° ($CHCl_3$); disulfide, m.p. 141–143°. Wagner-Jauregg and Lennartz, *Ber.*, **74**, 27 (1941), mercaptan, m.p. 99.5°, $[\alpha]_{D}$ −23.85°; disulfide, m.p. 144.5°.

[b] The variance in melting point from three different preparations apparently is not significant; *cf.* optical rotatory powers.

captan (VI) and its acetate (VII) conform to this rule. However, this is not so with the benzoate (V), where the order of magnitude is $\epsilon_{274} > \epsilon_{284} > \epsilon_{295}$. This may be understood when it is realized that cholesteryl thiobenzoate has a relatively strong maximum at 269–271 mμ (ϵ = 8350). Consequently, a more or less additive effect from this group may be expected in the spectrum of a $\Delta^{5,7}$-thiobenzoate. In the spectrum of cholesteryl benzoate there are two subsidiary maxima at 273 and 280 mμ (*cf.* spectrum of dehydroisoandrosteryl benzoate, Table I), but their magnitudes, ϵ = 790, 760 resp., are very low, and the ultimate effect on the spectrum of $\Delta^{5,7}$-benzoate is relatively small (1b).

In Table I, the rotatory dispersion (α_{Hg}/α_D) values are given. It will be noticed that for cholesteryl mercaptan and derivatives the value is 1.14–1.20, to be compared with the value of 1.48–1.52 for dehydroisoandrosteryl mercaptan and its benzoate (4), and the value of 1.33–1.45 for 7-dehydrocholesteryl mercaptan and derivatives. The utility of rotatory dispersion values other than for more rigorous characterization is, at present, of questionable value due to the lack of extensive data.[6]

7-Dehydrocholesteryl mercaptan (VI) in ether solution was irradiated with an ultraviolet lamp (Hanovia) in the usual manner (continuous flow system). A sample of 7-dehydrocholesterol was similarly irradiated for control purposes.

The samples were assayed by a modified AOAC chick bone ash test (5) in which the distal portion of the middle toe of one foot was used in place of the tibia for the ash determination. The results indicated a conversion of approximately 25% of the 7-dehydrocholesterol to Vitamin D_3. On the basis of this conversion figure the irradiated mercaptan was inactive at levels of 150 units and 5 mg. per kilogram of diet. U.S.P. reference cod liver oil was also used as a positive control, and gave the expected response.

The assumption has been made that the irradiation of 7-dehydrocholesteryl mercaptan (VI) gave, among other products, Vitamin D_3 mercaptan (VIII). The only evidence for this is that the irradiation products of both 7-dehydrocholesterol and 7-dehydrocholesteryl mercaptan (VI) gave approximately the same ultraviolet absorption curve. It may be concluded that irradiation of 7-dehydrocholesteryl mercaptan gave a product devoid of vitamin activity.

EXPERIMENTAL

Melting points. All m.p.'s are uncorrected, and were determined with uncalibrated Anschütz thermometers (total immersion). When a compound has a cloudy melt, the clearing point is given after the m.p., *e.g.* 160–162°, 188°.

Ultraviolet absorption spectra. All spectra were determined with a Beckman quartz spectrophotometer (mfg'd by the National Technical Laboratories, Pasadena, California), and were determined in 1% chloroform-absolute alcohol (1% CA), *i.e.*, the weighed sample

this generalization, *e.g.* 7-dehydrocholesteryl *p*-nitrobenzoate ($\epsilon_{271} > \epsilon_{282}$), 7-dehydrocholesteryl 3,5-dinitrobenzoate ($\epsilon_{271} > \epsilon_{282} > \epsilon_{293}$) (1b). Here, also, the ester grouping has a pronounced effect on the spectrum of the $\Delta^{5,7}$-steroid.

[6] In a comparison of the rotations of C-20 epimers in the allopregnane and isoallopregnane series (19 compounds), Fieser and Fieser, *Natural Products Related to Phenanthrene*, 3rd Edition, Reinhold Publishing Corp., N. Y., 1949, p. 434, have noted that $[\alpha]_D$ is 19% lower than $[\alpha]_{5461}$.

was dissolved in 1 ml. of chloroform and this solution was rapidly diluted to 100 ml. with commercial absolute alcohol.

Infrared absorption spectra. All spectra were determined with a Perkin-Elmer instrument converted to a double beam spectrophotometer.

Optical rotations. The weighed sample was dissolved in chloroform to make 2 ml. of solution, and the observed rotation was determined in a 1-dm. semi-micro polarimeter tube.

Cholesteryl thioacetate (*Δ^5-cholestene-3β-thiol acetate*) (II). Cholesteryl mercaptan (2), (4.6 g.) m.p. 96–99°, $[\alpha]_D^{29}$ −23.1°, $[\alpha]_{Hg}^{29}$ −26.4°, α_{Hg}/α_D = 1.14 (α_D^{29} −0.21°, α_{Hg}^{29} −0.24°; 18.2 mg.), in 30 ml. of pyridine was acetylated at room temperature in the usual manner with 10 ml. of acetic anhydride. Recrystallization from acetone to constant m.p. gave 3.8 g. of pure acetate, m.p. 122.5–124.5°;[7] $[\alpha]_D^{30}$ −49.6°, $[\alpha]_{Hg}^{30}$ −59.5°, α_{Hg}/α_D = 1.20 (α_D^{30} −0.65°, α_{Hg}^{30} −0.78°, 26.2 mg.), $[M]_D$ −220°; $\lambda_{max}^{1\%_{CA}}$ 232.5–234 mμ, ϵ = 4630.

Anal. Calc'd for $C_{29}H_{48}OS$ (444.73): C, 78.31; H, 10.88; S, 7.21.

Found: C, 78.61; H, 10.94; S, 7.65.

Hydrolysis of cholesteryl thioacetate. A. A mixture of 0.46 g. of cholesteryl thioacetate (II), 0.30 g. potassium carbonate, and 45 ml. of alcohol was refluxed for 4 hours, cooled, and treated with 1 ml. of glacial acetic acid. After dilution with water, the product was worked up in ether. The extract was washed 3 times with water and dried with magnesium sulfate. The ether solution was concentrated with simultaneous addition of acetone, until the latter had displaced all of the ether. The acetone solution was concentrated with addition of methanol. This gave 0.42 g. of crude mercaptan, m.p. 93–96°. Recrystallization from acetone gave 6 mg. of a product (probably disulfide), m.p. 139–141.5°. From the mother liquor by concentration with addition of methanol there was obtained 0.37 g. of pure cholesteryl mercaptan, m.p. 100–102°, $[\alpha]_D^{29}$ −22.6°, $[\alpha]_{Hg}^{29}$ −26.8°, α_{Hg}/α_D = 1.19 (α_D^{29} −0.27°, α_{Hg}^{29} −0.32°, 23.9 mg.). Yield 88%.

B. A mixture of 0.46 g. of cholesteryl thioacetate (II), 0.30 g. of potassium hydroxide, and 45 ml. of alcohol was refluxed for 0.5 hour. After dilution with water, 1 ml. of glacial acetic acid was added, and the product was worked up in ether in the usual manner. The ether solution was concentrated with simultaneous addition of methanol, and this gave 0.33 g. of a product, m.p. below 90°. Two recrystallizations from acetone gave 3 mg., m.p. 144–145°, of cholesteryl disulfide (mixed m.p. with authentic sample gave no depression of the melting point). The mother liquors by triangular recrystallization from acetone-methanol gave three fractions of mercaptan, wt. 0.23 g., m.p. 97–98°, wt. 0.10 g., m.p. 98–100°, and wt. 0.03 g., m.p. 97.5–99.5°. Total wt. 0.36 g., yield 86%.

The three fractions were combined and recrystallized once more from acetone-methanol, wt. 0.30 g., m.p. 98–100°. $[\alpha]_D^{29}$ −22.7°, $[\alpha]_{Hg}^{29}$ −26.8°, α_{Hg}/α_D = 1.18 (α_D^{29} −0.33°, α_{Hg}^{29} −0.39°; 29.1 mg.).

Cholesteryl thiobenzoate (*Δ^5-cholestene-3β-thiol benzoate*) (III). Cholesteryl mercaptan (I) (3 g.) in 25 ml. of pyridine was treated dropwise with 2 ml. of benzoyl chloride (ice-cooling). The mixture was allowed to stand at room-temperature for about 67 hours. It was then poured into ice-water and the crude product was washed successively with water, cold dilute hydrochloric acid, water, and methanol; wt. 3.69 g., m.p. 158–160°, 182.5°. Recrystallization from acetone, and ethyl acetate-alcohol to constant melting point gave 3.17 g. of the pure benzoate, m.p. 160–162°, 188° $[\alpha]_D^{29}$ −27.3°, $[\alpha]_{Hg}^{29}$ −31.7°, α_{Hg}/α_D = 1.16 (35.95 mg., α_D^{29} −0.49°, α_{Hg}^{29} −0.57°), $[M]_D$ −138°; $\lambda_I^{1\%_{CA}}$ 238–239, 269–271 mμ, $\epsilon_{238-239}$ = 11,500, $\epsilon_{269-271}$ = 8350.

Anal. Calc'd for $C_{34}H_{50}OS$ (506.80): C, 80.57; H, 9.94; S, 6.63.

Found: C, 80.71; H, 10.10; S, 6.50.

7-Dehydrocholesteryl thiobenzoate (*$\Delta^{5,7}$-cholestadiene-3β-thiol acetate*). (V) *A.* Cholesteryl thiobenzoate (III) (5.26 g., 0.0105 *M*) was dissolved in 85 ml. of petroleum ether

[7] Rosenberg and Turnbull, Jr., U. S. Patent 2,375,874 (May 15, 1945), m.p. 102–104°. Strating and Backer, *loc. cit.*, m.p. 124–125°, $[\alpha]_D^{20.5}$ −52.1° ($CHCl_3$).

(b.p. 64–66°, purified with conc'd sulfuric acid and potassium permanganate), and 15 ml. of carbon tetrachloride. NBS (2.24 g., 0.0126 *M*) was added, and the mixture was refluxed and irradiated for 3.5 minutes by the heat and light of a photospot lamp (General Electric Co., RSP-2). One ml. of *s*-collidine was added, and the suspension was cooled immediately to 20° in an ice-bath. The succinimide was removed and washed with cold carbon tetrachloride.

The filtrate was evaporated *in vacuo;* near the end of the distillation 75 ml. of xylene was added, and the distillation was continued for a short time after the xylene began to distill. The xylene solution was brought to reflux temperature (140°) and 0.75 ml. of *s*-collidine was added. The mixture was refluxed for 8 minutes, cooled, and extracted twice with cold 15% acetic acid. The xylene solution was then washed three times with water, dried with magnesium sulfate, treated with Norit, and filtered. Evaporation *in vacuo* gave a partly crystalline residue, which on treatment with ethyl acetate and a little acetone became crystalline. The mixture was cooled and the crude product was collected, wt. 2.5 g., m.p. 150–153°, 185°. Three recrystallizations from ethyl acetate gave 1.4 g., m.p. 163–165°, 204° (yield 26.6%).

Anal. Calc'd for $C_{34}H_{48}OS$ (504.78): C, 80.89; H, 9.58; S, 6.35.
Found: C, 81.11; H, 9.99; S, 6.40.

B. In another run with 15.18 g. of cholesteryl thiobenzoate (III), 200 ml. of petroleum ether (b.p. 64–66°), 6.42 g. of NBS, 6.6 ml. of *s*-collidine, and 100 ml. of xylene gave 3.46 g. of the product, m.p. 160–162.5°, 202°.

C. 7α(?)-Bromocholesteryl thiobenzoate (0.57 g.) (IV) in 40 ml. of xylene was treated with 1 ml. of *s*-collidine, and was refluxed for 15 minutes. The mixture was cooled, and filtered. Evaporation *in vacuo* gave a semi-solid residue which on crystallization from ethyl acetate-ethyl alcohol gave white needles, wt. 0.35 g., m.p. 155–159°, 191°. Recrystallization from ethyl acetate-ethyl alcohol, and ethyl acetate-acetone gave 0.23 g., m.p. 159.5–161.5°, 206°. Yield 46.5%.

From the mother liquor there was obtained 80 mg. of a material, m.p. 131–132.5°, 155°, $\lambda^{1\%CA}_{max}$ 245, 272 mμ, ϵ_{245} = 29,800, ϵ_{272} = 13,700, which consisted principally of the $\Delta^{4,6}$-thiobenzoate. No attempt was made to isolate pure $\Delta^{4,6}$-cholestadienyl thiobenzoate.

D. 7-Dehydrocholesteryl mercaptan (VI) (0.55 g.) in 10 ml. of pyridine was treated with 0.3 ml. of benzoyl chloride (ice-cooling), and was allowed to stand at room temperature for 48 hours. The reaction mixture was poured into ice-acetic acid, and the crude product was washed with methanol. Two recrystallizations from ethyl acetate gave 0.30 g., m.p. 163–165°, 210°. $\lambda^{1\%CA}_{max}$ 239, 274, 284, and 295 mμ, ϵ_{239} = 13,000, ϵ_{274} = 21,400, ϵ_{284} = 20,750, ϵ_{295} = 12,400. $[\alpha]^{30}_{D}$ +11.2°, $[\alpha]^{30}_{Hg}$ +16.2°, α_{Hg}/α_D = 1.45 (α^{30}_{D} +0.11°, α^{30}_{Hg} +0.16°; 19.7 mg.). $[M]_D$ +56°.

Anal. Found: C, 81.01; H, 9.68; S, 6.24.

7α(?)-Bromocholesteryl thiobenzoate [*7α(?)-bromo-Δ^5-cholestene-3β-thiol benzoate*] (IV). Cholesteryl thiobenzoate (III) (7.59 g., 0.015 *M*) was mixed with 3.21 g. (0.018 *M*) of NBS in 190 ml. of petroleum ether (b.p. 64–66°, purified with conc'd sulfuric acid and potassium permanganate), and the mixture was refluxed and irradiated for 4 minutes by the heat and light of 2 photospot lamps (General Electric Co., RSP-2). The suspension was cooled to about 30°, and the succinimide was removed, wt. 1.8 g. (calc'd 1.8 g.). The filtrate was concentrated on the steam-bath and cooled. The crystals so obtained melted below 100° (crystals were solvated). Triangular recrystallization from petroleum ether (b.p. 64–66°) gave two fractions of needles, m.p. 139–140.5°d. with much previous softening, strong positive Beilstein test for halogen, and m.p. 143–148°d.

The two fractions were combined and recrystallized twice from acetone, wt. 1.1 g., m.p. 153–155°d. (with previous softening). One further recrystallization from acetone gave m.p. 153–157.5°d. (sample placed in melting bath at 150°, m.p. appeared to be dependent on manner performed). $[\alpha]^{32}_{D}$ −139.2°, $[\alpha]^{32}_{Hg}$ −159.8°, α_{Hg}/α_D = 1.15 (α^{32}_{D} −1.35°, α^{32}_{Hg} −1.55°, 19.4 mg.), $[M]_D$ −814°.

Anal. Calc'd for $C_{34}H_{49}BrOS$ (585.71): C, 69.72; H, 8.43; Br, 13.64; S, 5.47.
Found: C, 69.50; H, 8.30; Br, 13.72; S, 5.83.

7-Dehydrocholesteryl mercaptan (*$\Delta^{5,7}$-cholestadiene-3β-thiol*) (VI). *A.* 7-Dehydrocholesteryl thiobenzoate (V) (1.3 g.) was refluxed for 3 hours (nitrogen atmosphere) with 0.45 g. of potassium carbonate in 90 ml. of alcohol. The solution was cooled, diluted with water, and filtered. The crude product (0.85 g., m.p. 130–135°) was recrystallized from acetone to give 0.75 g., m.p. 135–137°. Further recrystallization did not raise the melting point. $\lambda^{1\%CA}_{max}$ 273, 283, and 295 mμ, ϵ_{273} = 12,800, ϵ_{283} = 13,550, and ϵ_{295} = 7800. $E^{1\%}_{1cm}$ = 318, 336, 194 resp. $[\alpha]^{30}_{D}$ −90.9°, $[\alpha]^{30}_{Hg}$ −119.9°, α_{Hg}/α_D = 1.32 (α^{30}_{D} −1.23°, α^{30}_{Hg} −1.62°, 27.05 mg.). $[M]_D$ −364°.[8]

Anal. Calc'd for $C_{27}H_{44}S$ (400.68): C, 80.93; H, 11.07; S, 8.00.
Found: C, 80.70; H, 11.27; S, 8.07.

In another hydrolysis of this type with 0.3 g. of thiobenzoate, 0.20 g. of potassium carbonate, and 75 ml. of alcohol (2 hours reflux) there was obtained 0.20 g. of mercaptan, m.p. 136–138°. Yield 84%.

B. A mixture of 3.46 g. of 7-dehydrocholesteryl thiobenzoate, 1.0 g. of potassium hydroxide, and 75 ml. of alcohol was refluxed for 30 minutes in a nitrogen atmosphere. Two ml. of glacial acetic acid was added, and the mixture was cooled in an ice-acetone bath. The crude product, wt. 2.85 g., m.p. 133.5–135°, was recrystallized three times from benzene-acetone and once from ethyl acetate-methanol. This gave 1.3 g., m.p. 136.5–137.5°.

7-Dehydrocholesteryl thioacetate (*$\Delta^{5,7}$-cholestadiene-3β-thiol acetate*) (VII). *A.* 7-Dehydrocholesteryl mercaptan (VI) (0.20 g.) in 5 ml. of pyridine was treated with ice-cooling with 0.3 ml. of acetic anhydride. After 18 hours at room temperature the mixture was poured into ice-acetic acid and the crude product was collected. Three recrystallizations from acetone-methanol gave 90 mg., m.p. 110–112°, 137°. $\lambda^{1\%CA}_{max}$ 231–232 (inflection), 264–265.5 (inflection), 273.5, 284, and 295.5 mμ, $\epsilon_{231-232}$ = 5490, $\epsilon_{264-265.5}$ = 9000, $\epsilon_{273.5}$ = 12,800, ϵ_{284} = 13,800, and $\epsilon_{295.5}$ = 7950. $[\alpha]^{30}_{D}$ −33.0°, $[\alpha]^{30}_{Hg}$ −43.7°, α_{Hg}/α_D = 1.33 (α^{30}_{D} −0.43°, α^{30}_{Hg} −0.57°; 26.1 mg.). $[M]_D$ −146°.

Anal. Calc'd for $C_{29}H_{46}OS$ (442.72): C, 78.67; H, 10.47; S, 7.24.
Found: C, 78.63; H, 10.32; S, 7.32.

B. 7-Dehydrocholesteryl mercaptan (VI) (0.5 g.) in 15 ml. of acetic anhydride was refluxed in a nitrogen atmosphere for 40 minutes. The excess anhydride was removed by distillation *in vacuo*, and the residue was crystallized from ethyl acetate-methanol; yield, 0.48 g., m.p. 109–110°, 133°. Four recrystallizations from acetone-methanol gave the pure acetate, m.p. 111–112°, 135°.

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SUMMARY

1. 7-Dehydrocholesteryl mercaptan (provitamin D_3 mercaptan) has been prepared from cholesteryl thiobenzoate by allylic bromination with NBS, with subsequent elimination of hydrogen bromide, and hydrolysis.

[8] Strating and Backer, *loc. cit.*, *Method A.*, m.p. (*in vacuo*) sinter at 129°, clear melt at 135° (at 115° a slight alteration was observed); $[\alpha]^{19}_{D}$ −71.1° ($CHCl_3$), λ_{max} 272.5, 283, and 295 mμ, $E^{1\%}_{1cm}$ = 270, 296, and 173 resp. (solvent not stated). *Method B.*, m.p. (*in vacuo*) 132–135°; at 108° a slight alteration was observed; $[\alpha]^{17.5}_{D}$ −62.2° ($CHCl_3$), λ_{max} 272.5, 283, and 295 mμ, $E^{1\%}_{1cm}$ = 290, 307, and 179 resp. (solvent not stated).

2. The mercaptan was characterized by the formation of its acetate and benzoate.

3. Irradiation of 7-dehydrocholesteryl mercaptan gave a product devoid of Vitamin D activity.

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REFERENCES

(1) (a) I. BERNSTEIN, SAX, AND SUBBAROW, *J. Org. Chem.*, **13**, 837 (1948); (b) II. BERNSTEIN, BINOVI, DORFMAN, SAX, AND SUBBAROW, *J. Org. Chem.*, **14**, 433 (1949); (c) III. BERNSTEIN, OLESON, RITTER, AND SAX, *J. Am. Chem. Soc.*, **71**, 2576 (1949); (d) IV. BERNSTEIN AND SAX, *J. Am. Chem. Soc.*, **73**, 846 (1951).

(2) KING, DODSON, AND SUBLUSKEY, *J. Am. Chem. Soc.*, **70**, 1176 (1948).

(3) RALLS, DODSON, AND RIEGEL, *J. Am. Chem. Soc.*, **71**, 3320 (1949).

(4) BERNSTEIN AND SAX, *J. Org. Chem.*, **16**, preceding article.

(5) EVANS AND ST. JOHN, *J. Assoc. Offic. Agr. Chemists*, **27**, 283 (1944).