158. *The Application of the Method of Molecular Rotation Differences to Steroids. Part IV. Optical Anomalies.*

By D. H. R. Barton and J. D. Cox.

The correctness of the assumptions that the molecular rotations of steroidal substances are, for practical purposes, independent of concentration and temperature, has been checked by experiment. Special attention has been paid to the validity in steroids of the Principle of Optical Superposition and of the Rule of Shift, and many cases of disobedience of these generalisations, usually ascribed to " vicinal action ", have been examined. The variation of vicinal action with distance has been studied for several series of substituents. It has been found that steroidal molecules may exhibit optical anomalies if two easily polarised groups are present, and that, for covalently unsaturated substances, there is an approximate correlation between the magnitude of the optical anomaly ($\Delta\Delta$ value; see below) and the position of maximum absorption of the substituent groups in the far ultra-violet. This is, however, only one aspect of the problem, and the optical anomalies observed seem to depend also upon the capacity of the substituent groups for distortion of the framework of the molecule. It is possible, from these studies, to make a fairly reliable estimate of the maximum value of the optical anomaly in a particular steroid molecule.

Parts I, II, and III * (Barton, *J.*, 1945, 813; 1946, 512, 1116) have depended on the assumptions that the molecular rotations of steroids were, for practical purposes, concentration- and temperature-independent and that the additivity of molecular rotations was not invalidated by " vicinal action ". This paper examines these three postulates.

In the experimental section many recordings of specific rotations over wide ranges of concentration in chloroform solution for the Na_D line will be found. In no case was the variation in rotation sufficient to render valueless the application of the Method of Molecular Rotation Differences. All the experience that has been acquired has also shown that over the temperature range of 15° to 25° within which recordings were made, the rotations of steroids show very little, if any, variation, and the temperature independence of molecular rotations in such compounds seems to be well proven.

A much more important cause of uncertainty is the failure of some compounds to obey, within the experimental error, the principle of Optical Superposition and the quantitative form of the Rule of Shift (Freudenberg, *Ber.*, 1933, **66**, 177); a disobedience which may be attributed to " vicinal action ". There has been a very large number of studies of the validity of these generalisations, but it is not desirable to quote full references here. With regard to esters it may be said that the combination of an unsaturated alcohol with an unsaturated acid usually gives rise to an ester showing an optical anomaly;† if either of the components is saturated the anomaly is small, whilst if both are saturated it is negligible (cf. Rupe and Häfliger, *Helv. Chim. Acta*, 1940, **23**, 53). With regard to sugars there is, in general, surprisingly good agreement with Hudson's rules (Gorin, Kauzmann, and Walter, *J. Chem. Physics*, 1939, **7**, 327; cf. Kauzmann, *J. Amer. Chem. Soc.*, 1942, **64**, 1626), but, as is well known, in some cases there are serious optical anomalies the reasons for which are still not clearly understood.

In steroids the change in molecular rotation for a given chemical transformation at (say) the 3 position should be constant if the introduction of substituent groups in other parts of the molecule does not lead to vicinal action. Vicinal action and hence an optical anomaly is obviously to be anticipated in steroids which bear substituents close to the 3 position, and this is shown in practice by the data listed in Table I, which contains a few typical examples from the literature. In this table and the others in this paper Δ_1, Δ_2, and Δ_3 are the molecular rotation differences observed on acetylation, benzoylation, and oxidation respectively, of the hydroxyl attached to C_3. In calculating molecular rotations all specific rotations have been rounded off to the nearest degree. The molecular weights of the steroids examined in this paper are roughly 400 ± 100, hence a molecular rotation may be regarded as accurate to $\pm(2 \pm 0{\cdot}5)$. The difference between two molecular rotations (Δ value) will therefore be accurate to $\pm(4 \pm 1)$ and the difference between a given Δ value and a Δ value taken as a standard will be accurate to $\pm(8 \pm 2)$. In the tables the latter difference is denoted by the symbol $\Delta\Delta$ and will be clearly ≤ 10 in numerical

* In view of the very rapid growth in the literature of the steroids it may be stated that in Part I the literature reviewed is complete to the end of June 1945 whilst in Parts II and III all important references to the end of 1945 have been included.

† We have reserved the term " optical anomaly " for substances which do not show additivity of molecular rotation differences for other than trivial causes. In the latter we include anomalies caused by impurity or the incorrect assignment of structure and prefer to refer to all possible causes as leading to " molecular rotation anomalies ".

TABLE I.

Substance.	$[M]_D$*. Alcohol.	$[M]_D$*. Acetate.	Δ_1.	$\Delta\Delta_1$.	Refs.
Cholest-5-en-3(β)-ol (standard)	−154°	−188°	− 34°	—	Exptl.
4(β)-Benzoyloxycholest-5-en-3(β)-ol	−152	−329	−177	−143°	1, 2
4(β)-Butyroxycholest-5-en-3(β)-ol	−354	−468	−114	− 80	1
4(β)-Propionoxycholest-5-en-3(β)-ol	−403	−485	− 82	− 48	1
4(β)-Acetoxycholest-5-en-3(β)-ol	−395	−442	− 47	− 13	1, 2, 3, 4
4(β)-Hydroxycholest-5-en-3(β)-ol	−242	−289 †	− 47	− 13	2, 3, 5
Cholestan-3(β)-ol-2-one	+261	+324	+ 63	+ 97	6
Cholestan-3(β)-ol-4-one ‡	+ 60	− 36	− 96	− 62	6
Cholest-4-en-3(β)-ol-6-one	− 52	−226	−174	−140	7

* All molecular-rotation data in this paper are for the Na_D line and in chloroform solution unless specified to the contrary.

† The value given for the rotation of this acetate in Ref. 5 has been taken as correct. A considerably different value as well as a lower m. p. were reported earlier (Ref. 4).

‡ The assignment of configuration at C_3 is only probable and not proved.

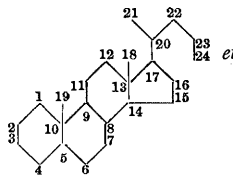
value in the absence of vicinal action. When the ΔΔ values are > 10 they may be taken as quantitative measures of the optical anomalies due to vicinal action. In Table I it may be pointed out that the series of 4(β)-acyloxy-substituents of cholesterol shows a progressive decrease in $\Delta\Delta_1$ values with decreasing bulk of the substituent. Table II, which gives ΔΔ values for the reduction of the Δ^5-bond, also illustrates the point made by Table I, it being observed that in many examples there are the expected large optical anomalies.

TABLE II.

Substance.	$[M]_D$. Unsaturated.	$[M]_D$. Saturated.	Δ.	ΔΔ.	Refs.
Cholest-5-en-3(β)-ol (standard)	−154°	+ 89°	+243°	—	Exptl.
Cholest-5-ene	−207	+ 91	+298	+55°	Exptl.
3(β)-Chloro-cholest-5-ene *	−122	+102	+224	−19	Exptl.
Cholest-5-en-3-one	− 30	+162	+192	−51	Exptl.
Cholest-5-en-3(β)-yl acetate	−188	+ 60	+248	+ 5	Exptl.
Cholest-5-en-3(β)-yl benzoate	− 74	+ 94	+168	−75	Exptl.

* The designation 3(β) has the true stereochemical significance; see Shoppee (*J.*, 1946, 1138, 1147).

It is generally conceded that the forces responsible for vicinal action must decrease with increase of distance (Kauzmann, Walter, and Eyring, *Chem. Reviews*, 1940, **26**, 339), and in the sugar field Freudenberg and Kuhn (*Ber.*, 1931, **64**, 719) consider that they are negligible after transmission through two saturated carbon atoms. So far there have been no systematic studies to test this assumption with optically active molecules containing a variety of chromophores, although steroid molecules would seem particularly suited to this purpose. The correctness of the view that vicinal action decreases with distance, for molecules bearing only the groupings $-O-H$, $-C{=}C-$, and $-C{=}O$, is proved by the data listed in Table III. It will be seen that for Δ_3 values vicinal action decreases progressively as the substituent ethylenic linkage or ketonic group is removed further from the 3 position. In both series vicinal action is negligible after transmission through three saturated carbon atoms.



Steroid skeleton.

In Table III and elsewhere in this paper comparison is made between steroids which differ from each other by the removal or addition of various saturated alkyl groups in the side chain. Such changes certainly do not cause vicinal action at the 3 position. In support of this are the data listed for androstan-3(β)-ol, 17-methylandrostan-3(β)-ol and *allo*pregnan-3(β)-ol in Table III. More striking support is the exact agreement between the Δ_3 values for cholesterol and β-sitosterol (see Part I, Table II), for Δ_3 values of this type are particularly sensitive to optical anomalies (see below). It would be possible to quote many other examples from the literature to substantiate this view.

Table IV also deals with the variation of vicinal action with distance for Δ_1 and Δ_2 values in a series of ethylenic compounds and in a series of ketones. For the acetates it will be noted that $\Delta\Delta_1$ values do seem to decrease with distance but only become negligible after transmission through at least 5 saturated carbon atoms. For the benzoates vicinal action is still strong in ergost-14-

TABLE III.

Substance.	$[M]_D$. Alcohol.	Ketone.	Δ_3.	$\Delta\Delta_3$.	Refs.
Cholestan-3(β)-ol (standard)	+ 89°	+162°	+ 73°	—	Exptl.
Cholest-5-en-3(β)-ol	−154	− 30	+124	+51°	Exptl.
Ergost-7-en-3(β)-ol	− 8	+ 88	+ 96	+23	Exptl.
Cholest-9(11)-en-3(β)-ol	+193	+277	+ 84	+11	7, 8
Ergost-8(14)-en-3(β)-ol	+ 44	+119	+ 75	+ 2	Exptl.
Ergost-14-en-3(β)-ol	+ 88	+159	+ 71	− 2	Exptl.
Androst-16-en-3(β)-ol	+ 30	+103	+ 73	± 0	9
Cholestan-3(β)-ol-6-one	− 24	+ 16	+ 40	−33	Exptl.
Cholestan-3(β)-ol-7-one	−144	− 76	+ 68	− 5	Exptl.
Androstan-3(β)-ol-17-one	+261	+325	+ 64	− 9	Exptl.
*allo*Pregnan-3(β)-ol-20-one	+305	+382	+ 77	+ 4	Exptl.
Androstan-3(β)-ol	+ 3	+ 84	+ 81	+ 8	9, 11
17-Methylandrostan-3(β)-ol	+ 17	+ 92	+ 75	+ 2	10
*allo*Pregnan-3(β)-ol	+ 57	+139	+ 82	+ 9	10
Ergosta-8(9) : 14-dien-3(β)-ol	− 76	− 4	+ 72	− 1	Exptl.

TABLE IV.

Substance.	$[M]_D$. Alcohol.	Acetate.	Benzoate.	Δ_1.	Δ_2.	$\Delta\Delta_1$.	$\Delta\Delta_2$.	Refs.
Cholestan-3(β)-ol (standard)	+ 89°	+ 60°	+ 94°	−29°	+ 5°	—	—	Exptl.
Cholest-5-en-3(β)-ol	−154	−188	− 74	−34	+80	− 5°	+75°	Exptl.
Ergost-7-en-3(β)-ol	− 8	− 18	+ 10	−10	+18	+19	+13	Exptl.
Cholest-9(11)-en-3(β)-ol	+193	+137	+201	−56	+ 8	−27	+ 3	7
Ergost-8(14)-en-3(β)-ol	+ 44	+ 4	± 0	−40	−44	−11	−49	Exptl.
Ergost-14-en-3(β)-ol	+ 88	+ 58	+116	−30	+28	− 1	+23	Exptl.
Cholestan-3(β)-ol-6-one	− 24	− 71	+ 20	−47	+44	−18	+39	Exptl.
Cholestan-3(β)-ol-7-one	−144	−163	− 86	−19	+58	+10	+53	Exptl.
Androstan-3(β)-ol-17-one	+261	+229	+272	−32	+11	− 3	+ 6	Exptl.
*allo*Pregnan-3(β)-ol-20-one	+305	+277	+319	−28	+14	+ 1	+ 9	Exptl.
Ergosta-8(9) : 14-dien-3(β)-ol	− 76	−158	− 15	−82	+61	−53	+56	Exptl.
Ergosta-7 : 22-dien-3(β)-ol 7 : 8-oxide	−182	−214	−187	−32	− 5	− 3	−10	12

en-3(β)-ol in the olefinic series, but has become negligible in the ketonic series at androstan-3(β)-ol-17-one. From the data in Table IV it will be seen that the conjugated unsaturation in ergosta-8(9) : 14-dien-3(β)-ol leads to large $\Delta\Delta$ values. It will also be perceived that while ergost-7-en-3(β)-ol shows Δ_1 and Δ_2 values which demonstrate beyond doubt the existence of optical anomalies, such anomalies are not observed in the analogous 7 : 8-oxide from ergosta-7 : 22-dien-3(β)-ol.

It has been generally taken for granted that substituents in the steroid side chain or at the 17 position are too far removed to cause vicinal action at the 3 position. The Δ_1 and Δ_2 values in the first part of Table V provide good evidence in support of this assumption for the >C=O group, the −OH group, the −C≡C− group, and even the readily polarisable −CBr–CBr− grouping as substituents. In the second part of Table V are recorded figures for substances with highly

TABLE V.

Substance.	$[M]_D$. Alcohol.	Acetate.	Benzoate.	Δ_1.	Δ_2.	$\Delta\Delta_1$.	$\Delta\Delta_2$.	Refs.
Cholest-5-en-3(β)-ol (standard)	−154°	−188°	− 74°	−34°	+ 80°	—	—	Exptl.
Androst-5-en-3(β)-ol-17-one	+ 6	− 23	+ 94	−29	+ 88	+ 5°	+ 8°	Exptl.
17(β)-Methylandrost-5-ene-3(β) : 17(α)-diol	−258	−294	−175	−36	+ 83	− 2	+ 3	Exptl.
Pregn-5-en-3(β)-ol-20-one	+ 79	+ 50	+168	−29	+ 89	+ 5	+ 9	Exptl.
Stigmasta-5 : 22-dien-3-(β)-ol	−210	−250	−134	−40	+ 76	− 6	− 4	Exptl.
Stigmasta-5 : 22-dien-3(β)-ol 22 : 23-dibromide	−132	−172	− 41	−40	+ 91	− 6	+11	Exptl.
Androst-5-ene-3(β) : 17(α)-diol 17(α)-benzoate	− 12	− 17	+ 65	− 5	+ 77	+29	− 3	Exptl.
Androst-5-ene-3(β) : 17(β)-diol 17(β)-benzoate	−445	−449	−339	− 4	+106	+30	+26	Exptl.
Androst-5-en-3(β)ol-17-one 4-phenylsemicarbazone	− 13	+ 16	+ 84	+29	+ 97	+63	+17	Exptl.

unsaturated groups attached at the 17 position. Here both acetates and benzoates show optical anomalies, the Δ_1 value for the 4-phenylsemicarbazone being remarkable. Clearly vicinal action is transmitted in these substances throughout the whole length of the molecule.

Table VI is concerned with a similar investigation, but in this case with the effect of side-chain substituents in causing optical anomalies in Δ_3 values. In androst-5-ene-3(β) : 17(α)-diol

TABLE VI.

Substance.	$[M]_D$. Alcohol.	$[M]_D$. Ketone.*	Δ_3.	$\Delta\Delta_3$.	Refs.
Cholest-5-en-3(β)-ol (standard)	−154°	+357°	+511°	—	Exptl.
Androst-5-ene-3(β) : 17(α)-diol	−183	+340	+523	+ 12°	Exptl.
17(β)-Methylandrost-5-ene-3(β) : 17(α)-diol	−258	+257	+515	+ 4	Exptl.
Androst-5-en-3(β)-ol-17-one	+ 6	+572	+566	+ 55	Exptl.
Pregn-5-en-3(β)-ol-20-one	+ 79	+631	+552	+ 41	Exptl.
Androst-5-ene-3(β) : 17(α)-diol 17(α)-benzoate	− 12	+615	+627	+116	Exptl.
Stigmasta-5 : 22-dien-3(β)-ol	−210	+258	+468	− 43	Exptl.
Stigmasta-5 : 22-dien-3(β)-ol 22 : 23-dibromide	−132	+336	+468	− 43	Exptl.
24 : 24-Diphenylchola-5 : 20(22) : 23-trien-3(β)-ol	− 10	+686	+696	+185	13

* The figures quoted in this table refer, of course, to Δ^4-3-ones.

and 17(β)-methylandrost-5-ene-3(β) : 17(α)-diol, containing covalently saturated hydroxyl groups, the Δ_3 values recorded show no significant optical anomalies, but with all the other substances listed in the table large anomalies are found. The magnitude of the $\Delta\Delta_3$ values is about the same for the $-C{=}O$ and for the $-C{\equiv}C-$ group, whilst, since there is a very large anomaly, with the highly polarisable 17(α)-benzoyloxy-substituent, it is of interest that the $\Delta\Delta_3$ value for the $-CBr{-}CBr-$ grouping is also about the same. The very highly unsaturated 24 : 24-diphenylchola-5 : 20(22) : 23-trien-3(β)-ol shows the largest $\Delta\Delta_3$ value of all the compounds examined. The substances in this table illustrate particularly well that vicinal action can, *in the appropriate compounds*, be transmitted through the whole of the steroid molecule from the side chain to ring A, a distance of over 10 A.

The effect of a further series of very highly unsaturated side-chain substituents on Δ_1 values is illustrated by the data, taken from the literature, in Table VII. It will be noted that, rather

TABLE VII.

Substance.	$[M]_D$. Alcohol.	$[M]_D$. Acetate.	Δ_1.	$\Delta\Delta_1$.	Refs.
Cholestan-3(β)-ol (standard)	+ 89°	+ 60°	− 29°	—	Exptl.
24 : 24-Diphenyl*allo*chol-23-en-3(β)-ol	+ 203	+ 183	− 20	+ 9°	14
3(β)-Hydroxy*allo*ætiochola-14 : 16-dienic acid	+1014	+1131	+117	+146	19
Cholan-3(α)-ol type	—	—	+ 83	—	15
24 : 24-Diphenylchol-23-en-3(α)-ol	+ 278	+ 344	+ 66	− 17	14
* 24 : 24-Diphenylchola-20(22) : 23-diene-3(α) : 12(α)-diol 12(α)-acetate	+1087	+1170	+ 83	± 0	16
24 : 24-Diphenylchola-20(22) : 23-dien-3(α)-ol	+ 401	+ 450	+ 49	− 34	14
* 22 : 22-Diphenylbisnorchol-20(22)-ene-3(α) : 12(α)-diol	+1718 †	+1930 †	+212	+129	17

* In the original publications these substances were listed as possessing a 12(β)-ol grouping. This has been altered here to 12(α)-ol in accordance with the view now generally accepted.

† These rotations were recorded in acetone solution.

surprisingly, large $\Delta\Delta_1$ values are not generally recorded. The one example with a very high $\Delta\Delta_1$ value is also a substance of high optical rotatory power so that its molecular rotation differences are particularly sensitive to small amounts of impurity. The optical anomaly may not, therefore, be so great as Table VII would indicate. The optical rotatory power data for 3(β)-hydroxy*allo*ætiochola-14 : 16-dienic acid are of interest because here a highly unsaturated grouping is introduced into the nucleus of the steroid molecule. In contrast with all but one of the other substances in Table VII a large optical anomaly is observed.

From the discussion given above it will be apparent that optical anomalies occur only in unsaturated steroids. It is well known that unsaturation in optically active substances is often associated with high optical rotatory power, and it is of importance therefore to establish that this high rotatory power does not automatically give rise to optical anomalies. The data of

Tables III, IV, V, and VI show that there is no such relationship. Thus derivatives of androstan-17-one and *allo*pregnan-20-one in Tables III and IV, although of high rotatory power, do not show optical anomalies, whereas derivatives of cholestan-6-one and ergost-7-ene, although of low rotatory power, do.

Many theories of optical activity concern themselves with the polarisabilities of the groups making up the optically active molecule (Kirkwood, *J. Chem. Physics*, 1937, **5**, 479). It is of interest, therefore, to see if there is any direct connection between group polarisability and the incidence of optical anomalies. The data of this paper show that major changes in the polarisability of the steroid molecule, for example the complete removal of the side chain and its replacement by saturated or relatively saturated groups of small polarisability, do not necessarily cause an optical anomaly. Differential group polarisability, defined as the difference in polarisability between an unsaturated group and that of the same group when the covalent unsaturation has been saturated with hydrogen, also bears no obvious relationship to the incidence of optical anomalies. Thus the differential polarisability of the benzoate grouping is very much greater than that of the acetate group, or of the $-C{=}C-C{=}O$ and $-C{=}C-C{=}C-$ groups, yet the optical anomalies in molecules containing the benzoate grouping are mostly comparable with those of the corresponding acetates. Moreover the $-C{=}C-C{=}O$ and $-C{=}C-C{=}C-$ groups lead to very much greater optical anomalies than the benzoate grouping as shown, for example, by the negligible $\Delta\Delta_2$ values for androstan-3(β)-ol-17-one and *allo*pregnan-3(β)-ol-20-one (Table IV) as compared with the large optical anomalies shown in the comparable $\Delta\Delta_3$ values (Table VI).

In spite of any direct correlation between polarisability and $\Delta\Delta$ values it remains true that optical anomalies only occur in steroidal molecules containing two easily polarised groups. Also the anomalies are small or negligible with groups that are covalently saturated but co-ordinatively unsaturated (see Braude, *Ann. Reports*, 1945, **42**, 117), whilst they are relatively large with groups that are covalently unsaturated. This is illustrated by the data in Table VI, which have already been discussed above. Leaving out of consideration such covalently saturated groups it will be seen from Table VIII that there is a general qualitative relationship between the incidence of optical anomalies and the position of the absorption maximum furthest in the ultra-violet which is characteristic of the whole covalent unsaturation of the grouping.

TABLE VIII.*

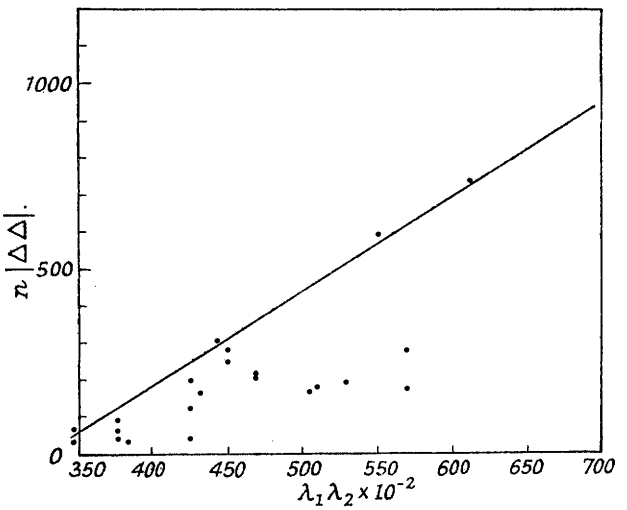
Grouping.	$\lambda_{max.}$ (mμ).	Remarks.
–C—H	<160	No optical anomalies; covalently and co-ordinatively saturated.
–C—C–	<160	
–C—Cl	<160	Doubtless no optical anomalies; covalently saturated.
–O—H	~180	No optical anomalies: covalently saturated.
–C=C–	185	Only small optical anomalies in most cases; covalently unsaturated.
–C=O	188	
–O–	190	No optical anomalies; covalently saturated.
–CO—O–	204	More liable to give optical anomalies than the –C=C– or –C=O groups; covalently unsaturated.
Ph—CO—O–	230	
–C=C—C=O	240 †	Causing large optical anomalies in most cases; covalently unsaturated.
–C=C—C=C–	248 †	
CPh_2=C–	250	Discussed in the text.
–C—Br	260	In most cases no optical anomaly; covalently saturated.
–C=C—C=C—CO_2H	~300 †	Large optical anomaly when substituted into the steroid nucleus.
CPh_2=C—C=C–	310	Discussed in the text.

* The absorption-spectra data are mostly taken from Braude, *Ann. Reports*, 1945, **42**, 105.
† The $\lambda_{max.}$ data given here refer specifically to the steroidal compounds discussed in the text.

From an empirical approach it is possible to gain some idea as to the maximum value of the optical anomaly in a steroidal molecule bearing two easily polarised groups. In the figure the product of λ_1 and λ_2, these being ultra-violet absorption maxima taken from Table VIII, is plotted against $n \times |\Delta\Delta|$ where $n \geq 3$ is the number of saturated carbon atoms between the two groups (in acyloxy-derivatives the carbon atom to which the acyloxy-group is attached is not counted) and $|\Delta\Delta|$ is the observed optical anomaly. The introduction of n into the plot takes account of the falling off of vicinal action with distance already discussed above. From the straight line the maximum value that $|\Delta\Delta|$ might be expected to attain in a given molecule may be roughly evaluated.

Besides the ease of electronic transition shown by covalently unsaturated groups, these groups must often also possess the property of causing a considerable degree of strain when introduced into saturated molecules. The amount of strain produced must depend not only on the covalently

unsaturated grouping but also upon the position into which it is substituted in the originally saturated molecule. Thus the strain produced in a ring system must usually be much greater for a pair of conjugated ethylenic linkages than for a single ethylenic linkage, and again the strain produced by a single ethylenic linkage in a ring system must generally be greater than that produced by a ketonic grouping. Furthermore the strain produced by the introduction of a pair of conjugated ethylenic linkages into one ring of the steroid nucleus must be greater than that produced by the same pair in two different rings of the nucleus. Also the strain produced by (say) a benzoate or acetate grouping attached by a single linkage to the steroidal nucleus must be relatively small. Although these concepts cannot, as yet, be placed on a quantitative basis, it will be clear that the capacity of covalently unsaturated substituents for causing strain does not necessarily vary in the same order as their ease of electronic excitation. It is suggested that this differentiation in the properties of covalently unsaturated groupings is related in a general qualitative manner to the incidence of optical anomalies. Considering, for the moment, steroid molecules which may be regarded as derived from a standard framework [usually cholesterol or cholestan-3(β)-ol] by the introduction of substituent groups not in close proximity



to each other, certain generalisations can be made regarding the incidence of optical anomalies (Barton and Cox, *Nature*, 1946, **159**, 470). Substituent groups may be divided into three classes :

(i) Those causing no anomalies in molecular rotation, *i.e.*, C–H, C–C, C–OH, –O–; groups which are not easily polarised and which do not distort the molecular framework.

(ii) Those causing major anomalies only in the presence of class (iii) substituents, *i.e.*, C–Br, C–OAc, C–OBz, C=O, C=C; groups which are more or less easily polarised but which do not cause a high degree of strain in the molecular framework.

(iii) Those causing, in general, major anomalies if another member of class (ii) or (iii) is present, *i.e.*, C=C, C=C–C=O, C=C–C=C; groups which are easily polarised and which distort the molecular framework.

Clearly there is no rigid dividing line between the groups placed in classes (ii) and (iii), and, for example, the ethylenic linkage must be placed in both of these classes, although steroids containing two ethylenic linkages, one in the side chain and the other in the steroidal nucleus, do not exhibit large optical anomalies. Moreover the amounts of distortion produced by the introduction of class (iii) substituents must be dependent on the actual positions they occupy in the molecule; it must be greater, for example, for substitution into the polycyclic ring system of the nucleus than for introduction into the long side chain (cf. Table VII).

The view that the strain produced by covalently unsaturated groups in a molecule is a very important factor in governing the incidence of optical anomalies provides a clue to the mechanism by which long distance " vicinal action " may be transmitted. This type of interaction is most probably an intramolecularly propagated effect because of its concentration independence and because there is no steady decrease in ΔΔ values with distance as might be expected for an effect transmitted through space. The introduction of covalently unsaturated groupings into a molecule leads to a flexing of valency angles and, to a lesser extent, to an alteration in bond

lengths, in the immediate environment of the substitution. This flexing of valency angles must also occur to a lesser degree for the next most proximate atomic nuclei and must be passed on through the molecule for some distance before it dies out completely. Such possibilities have been appreciated for many years, but, as far as we are aware, this is the first time that evidence has been provided that such interaction can be handed on for distances of the order of 10 A.

There are two aspects, therefore, to be considered in the discussion of optical anomalies. First, the polarisabilities of the groups which lead to easy electronic transitions and thus to induced anisotropies which make large contributions to the optical rotatory power. Secondly, the deformation of the molecular framework by the introduction of covalently unsaturated groups which provides a means by which these groups may interact.

It is suggested that the data in Table IX may provide an illustration of the two effects. Both 7-methylenecholest-5-en-3(β)-ol and ergosta-5 : 7 : 22-trien-3(β)-ol have the ring B chromophore

TABLE IX.

Substance.	$[M]_D$. Alcohol.	Acetate.	Benzoate.	Δ_1.	Δ_2.	$\Delta\Delta_1$.	$\Delta\Delta_2$.	Refs.
Cholest-5-en-3(β)-ol (standard)	−154°	−188°	− 74°	− 34°	+ 80°	—	—	Exptl.
Cholest-5-en-3(β)-ol-7-one	−416	−455	−277	− 39	+139	− 5°	+59°	Exptl.
7-Methylenecholest-5-en-3(β)-ol	−760	−783	−612	− 23	+148	+ 11	+68	18
7-Methylcholest-5-ene-3(β) : 7-diol	−125	−160	− 36	− 35	+ 89	− 1	+ 9	18
Ergosta-5 : 7 : 22-trien-3(β)-ol	−523	−413	−345	+110	+178	+144	+98	Exptl.

at equal distances from the 3 position, and both chromophores are equally polarisable. However, in the ergostane derivative where the molecule is more strained, the optical anomalies are much larger. It is significant that cholest-5-en-3(β)-ol-7-one, which must have an appreciable dipole moment, has Δ_1 and Δ_2 values identical with those of 7-methylenecholest-5-en-3(β)-ol, the obvious similarity between the two being that both distort the molecular framework to the same extent. On the other hand when the conjugation in ring B is destroyed, and the polarisability greatly reduced, as in 7-methylcholest-5-ene-3(β) : 7-diol, then the Δ_1 and Δ_2 values are in good agreement with those for cholest-5-en-3(β)-ol.

It is now possible to predict by empiricism when an optical anomaly will not occur and also when one may be expected, as shown above. Nevertheless it is an unfortunate aspect of the optical anomalies discussed in this paper that it is still *not* possible to explain the absence of an optical anomaly when, from the unsaturation of the molecule, one might be expected. This is brought out by the Δ_1 value for cholest-5-en-3(β)-ol being nearly identical with the Δ_1 value for cholestan-3(β)-ol (Table IV), and by the Δ_3 value for ergosta-8(9) : 14-dien-3(β)-ol being identical with the Δ_3 value for cholestan-3(β)-ol (Table III). Other examples will be seen from the tables. It is only by taking an overall picture of the optical anomalies in steroids that the regularities discussed in this paper become apparent.

References.

1. Petrow, Rosenheim, and Starling, *J.*, 1943, 135.
2. Spring and Swain, *J.*, 1941, 83.
3. Rosenheim and Starling, *J.*, 1937, 377.
4. Petrow, *ibid.*, p. 1077.
5. Petrow and Starling, *J.*, 1940, 60.
6. Ruzicka, Plattner, and Furrer, *Helv. Chim. Acta*, 1944, **27**, 727.
7. Wieland, Rath, and Benend, *Annalen*, 1941, **548**, 19.
8. Wieland and Benend, *Ber.*, 1942, **75**, 1708.
9. Prelog, Ruzicka, and Wieland, *Helv. Chim. Acta*, 1944, **27**, 66.
10. Ruzicka, Meister, and Prelog, *ibid.*, 1947, **30**, 867.
11. Prelog, Ruzicka, *et al.*, *ibid.*, 1945, **28**, 618.
12. Callow and Rosenheim, *J.*, 1933, 387.
13. Meystre *et al.*, *Helv. Chim. Acta*, 1946, **29**, 627.
14. Meystre and Miescher, *ibid.*, p. 33.
15. Barton, *J.*, 1946, 1116.
16. Meystre *et al.*, *Helv. Chim. Acta*, 1944, **27**, 1815.
17. Sorkin and Reichstein, *ibid.*, 1945, **28**, 875.
18. Bann, Heilbron, and Spring, *J.*, 1936, 1274.
19. Plattner, Meier, and Heusser, *Helv. Chim. Acta*, 1947, **30**, 905.

Experimental.

The substances whose rotations are listed below were all purified as carefully as possible to constant m. p. and constant rotation. All specimens were dried in a vacuum at 20° below their m. p.s, or at 120° whichever was the lower temperature. All rotations are for the Na_D line and in chloroform solution unless specified to the contrary. The measurements were made at room temperature which varied from 15° to 25°.

In order to improve the accuracy of the rotation measurements, most readings were taken using macro-tubes. Normally 1 dm. tubes were employed, but rotations marked with an asterisk were recorded using a 2 dm. macro-tube. Values obtained using a 1 dm. micro-tube have this fact indicated after each individual rotation. All values of $[\alpha]_D$ have been approximated to the nearest degree as in Parts I, II, and III. Concentrations (c) are expressed in g. per 100 ml. of solution. For the calculations the specific rotations at c. 2·00, or at the nearest concentrations to this at which measurements were made, have been taken as the most suitable. In no case, however, was there any marked variation of specific rotation with concentration (see text).

Acetylations were carried out by refluxing the substance with acetic anhydride for 30 minutes; benzoylations by the usual pyridine procedure, the reactants being left for 24 hours at room temperature to complete reaction. Alkaline hydrolyses were effected by using several equivalents of potassium hydroxide and refluxing the reactants for 30 minutes in methanolic or dioxan–methanolic solution depending upon the solubility of the ester. The use of milder conditions in all these procedures, when required, is mentioned specifically below. Chromic acid oxidations were carried out at room temperature, the reactants being left for 24 hours in acetic acid or ether–acetic acid solution depending upon solubility requirements, and slightly more than the theoretical amount of chromic acid being used.

Oppenauer oxidations were performed under the usual conditions (Barton and Jones, *J.*, 1943, 599) except that it was found advantageous to use only half the amount of acetone there specified and to carry out the refluxing for only 4 hours instead of the usual 16 hours. It was found that the use of resublimed aluminium *tert.*-butoxide is not essential, and material crystallised from benzene was employed instead. It is usually considered that the Oppenauer procedure cannot be applied for the oxidation of secondary alcohols unless the alcohol grouping is activated by the close proximity of an ethylenic linkage. Nevertheless we have experienced no difficulty in making such application as will be seen from the examples quoted below.

With some substances it has proved advantageous to ensure purity by the use of the chromatographic method (see Barton and Jones, *J.*, 1943, 602).

M. p.s are uncorrected.

Androstane Derivatives.—Androst-5-en-17-on-3(β)-yl acetate (British Schering Ltd.), recrystallised from acetone–methanol, had m. p. 172°, $[M]_D$ − 23°. The following dependence of rotation on concentration was observed :

c	14·55	7·28	5·14	2·91	2·06	1·67
$[\alpha]_D$	−8°	−7°	−7°	−7° *	−7° *	−8°

Androst-5-en-3(β)-ol-17-one (dehydro*iso*androsterone), recrystallised from methanol, had m. p. 148°, $[\alpha]_D$ + 2°(c, 5·37), + 2°(c, 2·15), $[M]_D$ + 6°.

The rotations of androstan-3(β)-ol-17-one (*iso*androsterone) and its acetate were recorded in Part III.

Androst-5-en-17-on-3(β)-yl benzoate, recrystallised from chloroform–ethyl acetate, had m. p. 257°, $[\alpha]_D$ + 24°(c, 2·22), + 23°(c, 0·89), $[M]_D$ + 94°.

Androstan-17-on-3(β)-yl benzoate, recrystallised from acetone–methanol, had m. p. 216° $[\alpha]_D$ + 69°(c, 1·44), + 68°(c, 0·58), $[M]_D$ + 272 (Found : C, 79·5; H, 8·8. $C_{26}H_{34}O_3$ requires C, 79·2; H, 8·7%).

Androst-5-en-17-on-3(β)-yl acetate 4-phenylsemicarbazone was prepared by condensation of androst-5-en-17-one-3(β)-yl acetate on the water-bath for 10 minutes with the equivalent amount of 4-phenylsemicarbazide in alcoholic solution made acid with acetic acid. Recrystallised from chloroform–methanol, it had m. p. 268—269°, $[\alpha]_D$ + 3°(c, 2·11), + 4°(c, 2·05), $[M]_D$ + 16° (Found : N, 9·3. $C_{28}H_{37}O_3N_3$ requires N, 9·1%).

Androst-5-en-3(β)-ol-17-one 4-phenylsemicarbazone was prepared by condensation of androst-5-en-17-one-3(β)-ol with 4-phenylsemicarbazide under the conditions specified above. Recrystallised from alcohol, it had m. p. 237—238° (decomp.), $[\alpha]_D$(in chloroform–alcohol, 98 : 2 parts by volume) − 4°(c, 1·76), − 2°(c, 1·50), − 2°(c, 1·08), $[M]_D$ − 13° (Found : N, 9·6. $C_{26}H_{35}O_2N_3$ requires N, 9·9%).

Androst-5-en-17-on-3(β)-yl benzoate 4-phenylsemicarbazone was prepared by benzoylation of the corresponding alcohol in pyridine solution under the usual conditions, except that the reactants were left together for 30 minutes only. Recrystallised from chloroform–methanol, it had m. p. 283—284° (decomp.) $[\alpha]_D$ + 16°(c, 1·64), + 14°(c, 0·64), $[M]_D$ + 84° (Found : C, 75·3; H, 7·5. $C_{33}H_{39}O_3N_3$ requires C, 75·4; H, 7·4%).

Androst-5-ene-3(β) : 17(α)-diol (British Schering Ltd.), recrystallised from acetone–methanol, had m. p. 180°, $[\alpha]_D$ − 61°(c, 1·50; in chloroform–alcohol, 90 : 10 parts by volume), − 61°(c, 1·00; in chloroform–alcohol, 96 : 4), − 63°(c, 0·30; in chloroform–alcohol, 98 : 2); extrapolated $[\alpha]_D$ in pure chloroform − 63°, $[M]_D$ − 183°.

17(β)-Methylandrost-5-ene-3(β) : 17(α)-diol (British Schering Ltd.), recrystallised from acetone–methanol, had m. p. 204°, $[\alpha]_D$ − 85°(c, 1·04; in chloroform–alcohol, 85 : 15 parts by volume), − 85° (c, 0·91; in chloroform–alcohol 98 : 2), $[M]_D$ − 258° (in pure chloroform by extrapolation).

17(β)-Methylandrost-5-ene-3(β) : 17(α)-diol 3(β)-acetate was prepared from the corresponding diol by acetylation with acetic anhydride in pyridine solution for 24 hours at room temperature. Recrystalliesd from acetone, it had m. p. 174°, $[\alpha]_D$ − 85°(c, 1·35), $[M]_D$ − 294°.

17(β)-Methylandrost-5-ene-3(β) : 17(α)-diol 3(β)-benzoate was prepared from the corresponding diol by treatment with benzoyl chloride in pyridine solution at room temperature for 2 hours. Recrystallised from methanol, it had m. p. 197—198°, $[\alpha]_D$ − 43° (c, 2·05; micro-tube), $[M]_D$ − 175° (Found : C 77·9; H, 8·5. $C_{27}H_{36}O_3,\frac{1}{2}H_2O$ requires C, 77·8; H, 8·9%).

Androst-5-ene-3(β) : 17(α)-diol 3(β)-acetate 17(α)-benzoate (British Schering Ltd.), recrystallised from acetone, had m. p. 175°, $[\alpha]_D$ − 4°(*c*, 3·23; micro-tube), − 4°(*c*, 2·86), − 4°(*c*, 2·33), − 4°(*c*, 2·24; micro-tube), − 3°(*c*, 1·34), $[M]_D$ − 17°.

Androst-5-ene-3(β) : 17(α)-diol 17(α)-benzoate was prepared from the corresponding 3(β)-acetate (see above) by hydrolysis with the theoretical amount of methanolic potassium hydroxide at room temperature for 24 hours. Recrystallised from acetone–methanol, it had m. p. 218°, $[\alpha]_D$ − 4°(*c*, 3·28), − 3°(*c*, 2·41), − 3°(*c*, 1·89; micro-tube), − 4°(*c* 1·64), − 3°(*c*, 0·91; micro-tube), $[M]_D$ − 12°.

Androst-5-ene-3(β) : 17(α)-*diol dibenzoate*, recrystallised from chloroform–acetone, had m. p. 217°, $[\alpha]_D$ + 13°(*c*, 1·80), + 12°(*c*, 1·55), + 13°(*c*, 0·95; micro-tube), + 13°(*c*, 0·90), $[M]_D$ + 65° (Found: C, 79·5; H, 7·8. $C_{33}H_{38}O_4$ requires C, 79·5; H, 7·7%).

Androst-5-ene-3(β) : 17(β)-diol 3(β)-acetate 17(β)-benzoate (British Schering Ltd.), recrystallised from acetone–methanol, had m. p. 129°, $[\alpha]_D$ − 103°(*c*, 2·00), − 103°(*c*, 1·05), −103°(*c*, 0·99,) $[M]_D$ − 449°.

Androst-5-ene-3(β) : 17(β)-diol 17(β)-benzoate was prepared from the corresponding 3(β)-acetate (see above) by hydrolysis with the theoretical amount of methanolic potassium hydroxide at room temperature for 24 hours. Recrystallised from aqueous methanol, it had m. p. 148·5° (after prolonged drying in a vacuum at 70° and upwards), $[\alpha]_D$ − 113°(*c*, 2·57; micro-tube), − 113°(*c*, 0·78), $[M]_D$ − 445°.

Androst-5-ene-3(β) : 17(β)-*diol dibenzoate*, recrystallised from ethyl acetate–methanol, had m. p. 152°, $[\alpha]_D$ − 68°(*c*, 1·64), $[M]_D$ − 339° (Found : C, 79·6; H, 7·9. $C_{33}H_{38}O_4$ requires C, 79·5; H, 7·7%).

Androst-4-ene-3 : 17-dione was prepared by the method of Ruzicka and Wettstein (*Helv. Chim. Acta*, 1935, **18**, 986) from androst-5-en-3(β)-ol-17-one and purified by chromatography. Recrystallised from light petroleum (b. p. 40—60°)–benzene, it had m. p. 172°, $[\alpha]_D$ + 199°(*c*, 1·98), + 201°(*c*, 1·03), + 202° (*c*, 0·52), $[M]_D$ + 572°.

Androstane-3 : 17-dione was prepared by chromic acid oxidation of androstan-3(β)-ol-17-one and purified by chromatography. Recrystallised from light petroleum (b. p. 40—60°), it had m. p. 127°, $[\alpha]_D$ + 111°(*c*, 1·22), + 113°(*c*, 1·08), $[M]_D$ + 325°.

Androst-4-en-17(α)-ol-3-one (testosterone) was prepared by alkaline hydrolysis of the corresponding propionate (British Schering Ltd.). Recrystallised from aqueous methanol, it had m. p. 150—151°, $[\alpha]_D$ + 118°(*c*, 2·29), + 119°(*c*, 1·14), + 117°(*c*, 1·08), $[M]_D$ + 340°.

Androst-4-en-3-on-17(α)-yl benzoate was prepared from androst-4-en-17(α)-ol-3-one by benzoylation. Recrystallised from chloroform–acetone, it had m. p. 193°, $[\alpha]_D$ + 156°(*c*, 1·90), + 157°(*c*, 1·54), $[M]_D$ + 615°.

17-(β)-Methylandrost-4-en-17(α)-ol-3-one (British Schering Ltd.), recrystallised from methanol, had m. p. 164°, $[\alpha]_D$ + 85°(*c*, 1·73), + 84°(*c*, 1·16), $[M]_D$ + 257°.

allo*Pregnane Derivatives*.—Pregn-5-en-20-on-3(β)-yl acetate (British Schering Ltd.), recrystallised from acetone–methanol, had m. p. 147°, $[\alpha]_D$ + 14°(*c*, 4·55), + 13°(*c*, 2·37), + 14°(*c*, 2·28), + 14°(*c*, 1·61), $[M]_D$ + 50°.

Pregn-5-en-3(β)-ol-20-one (Ciba), recrystallised from methanol, had m. p. 187—190°, $[\alpha]_D$ + 25°(*c*, 2·36), $[M]_D$ + 79°.

Pregn-5-en-20-*on*-3(β)-*yl benzoate*, recrystallised from acetone–methanol, had m. p. 197°, $[\alpha]_D$ + 40°(*c*, 0·84), $[M]_D$ + 168° (Found : C, 79·7; H, 8·7. $C_{28}H_{36}O_3$ requires C, 80·0; H, 8·6%).

*allo*Pregnan-20-on-3(β)-yl acetate was prepared by catalytic hydrogenation of pregn-5-en-20-on-3(β)-yl acetate under the same conditions as those used for the preparation of androstan-17-on-3(β)-yl acetate (see Part III). Recrystallised from acetone, it had m. p. 143·5—144·5°, $[\alpha]_D$ + 77°(*c*, 1·90), + 77°(*c*, 1·22), $[M]_D$ + 277°.

*allo*Pregnan-3(β)-ol-20-one, recrystallised from methanol, had m. p. 194—195°, $[\alpha]_D$ + 95°(*c*, 1·48), + 96°(*c*, 1·28), $[M]_D$ + 305°.

allo*Pregnan*-20-*on*-3(β)-*yl benzoate*, recrystallised from acetone, had m. p. 197—198°, $[\alpha]_D$ + 76°(*c*, 0·98), + 76°(*c*, 0·74; micro-tube), $[M]_D$ + 319° (Found : C, 79·3; H, 8·6. $C_{28}H_{38}O_3$ requires C, 79·6; H, 9·0%).

Pregn-4-ene-3 : 20-dione (progesterone) (British Schering Ltd.), recrystallised from aqueous methanol, had m. p. 128°, $[\alpha]_D$ + 201°(*c*, 4·56), + 201°(*c*, 2·28), + 202°(*c*, 1·14), $[M]_D$ + 631°.

*allo*Pregnane-3 : 20-dione was prepared by chromic acid oxidation of *allo*pregnan-3(β)-ol-20-one and purified by chromatography. Recrystallised from light petroleum (b. p. 40—60°)–benzene, it had m. p. 201°, $[\alpha]_D$ + 121°(*c*, 2·12; micro-tube), $[M]_D$ + 382°.

Cholestane and its Derivatives.—Cholest-5-ene, recrystallised from chloroform–acetone, had m. p. 92°, $[\alpha]_D$ − 55°(*c*, 2·32), − 56°(*c*, 1·16), $[M]_D$ − 207°.

Cholestane was prepared from cholest-5-ene by catalytic hydrogenation in ether–acetic acid solution using a platinum oxide catalyst. Recrystallised from chloroform–acetone, it had m. p. 80°, $[\alpha]_D$ + 24°(*c*, 2·96), + 25°(*c*, 1·48), $[M]_D$ +91°.

Cholesterol [cholest-5-en-3(β)-ol], recrystallised from ethyl acetate, had m. p. 148·5°, $[M]_D$ − 154°. The following dependence of rotation on concentration was observed :

c	28·0	25·0	20·0	15·0	10·0	8·0	6·0	4·0	3·0	2·0	1·0
$[\alpha]_D$	−39°	−39°	−39°	−39°	−39°	−39°	−39°	−40°	−40°	−40° *	−40° *

Cholesteryl acetate, recrystallised from ethyl acetate, had, m. p. 114·5°, $[M]_D$ − 188°. The following dependence of rotation on concentration was observed :

c	28·0	25·0	20·0	15·0	10·0	8·0	6·0	4·0	3·0	2·0	1·0
$[\alpha]_D$	−42°	−42°	−42°	−42°	−43°	−43°	−43°	−44°	−44°	−44° *	−44 *

Cholestan-3(β)-ol, recrystallised from ethyl acetate, had m. p. 142·5°, $[M]_D$ + 89°. The following dependence of rotation on concentration was observed :

c	10·0	8·0	6·0	4·0	3·0	2·0	1·0
$[\alpha]_D$	+23°	+23°	+23°	+23°	+23°	+23° *	+24° *

Cholestan-3(β)-yl acetate, recrystallised from ethyl acetate, had m. p. 110°, $[M]_D$ +60°. The following dependence of rotation on concentration was observed :

c	15·0	10·0	8·0	6·0	4·0	3·0	2·0	1·0
$[\alpha]_D$	+13°	+13°	+13°	+13°	+14°	+14°	+14° *	+14° *

Cholesteryl benzoate, recrystallised from acetone–methanol or ether–alcohol, had m. p. 150°, $[\alpha]_D$ − 16°(*c*, 9·19), − 15°(*c*, 4·60), − 16°(*c*, 2·30), − 14°*(*c*, 0·92), $[M]_D$ − 74°.

Cholestan-3(β)-yl benzoate, recrystallised from acetone, had m. p. 135°, $[\alpha]_D$ + 19°(*c*, 2·95), + 18°*(*c*, 1·81), $[M]_D$ + 94°.

Cholestan-6-on-3(β)-yl acetate was prepared without difficulty by following the directions of Mauthner and Suida (*Monatsh*, 1903, **24**, 654). Recrystallised from alcohol, it had m. p. 129°, $[\alpha]_D$ − 16°(*c*, 2·33), − 16°(*c*, 1·17), $[M]_D$ − 71°.

Cholestan-3(β)-ol-6-one was prepared from the acetate by hydrolysis with several equiv. of potassium carbonate in 20% aqueous methanol at the boiling point for 10 minutes; also from the acetate by acid hydrolysis using 3% aqueous concentrated hydrochloric acid in methanol solution at the boiling point for 15 minutes. Recrystallised from aqueous methanol, it had m. p. 140°, $[\alpha]_D$ − 6°(*c*, 1·69), − 6°(*c*, 1·35), − 6°(*c*, 0·98), $[M]_D$ − 24°. Preparations by both methods were identical.

Cholestan-6-on-3(β)-yl benzoate, recrystallised from ethyl acetate, had m. p. 173·5°, $[\alpha]_D$ + 4°(*c*, 3·33), + 4°(*c*, 1·66), $[M]_D$ + 20°.

Cholest-5-en-7-on-3(β)-yl acetate, recrystallised from acetone, had m. p. 159°, $[\alpha]_D$ − 103°(*c*, 6·08), − 103°(*c*, 3·59), − 103°(*c* 1·80), − 103°(*c*, 1·22), $[M]_D$ − 455°. (We are indebted to Professor E. R. H. Jones for a supply of the technical material.)

Cholest-5-en-3(β)-ol-7-one was prepared by hydrolysis of the acetate, using the mild alkaline conditions prescribed by Bergstrom and Wintersteiner (*J. Biol. Chem.*, 1941, **141**, 597). Recrystallised from methanol, it had m. p. 170°, $[\alpha]_D$ − 103°(*c*, 2·91), − 104°(*c*, 1·46), $[M]_D$ − 416°.

Cholest-5-en-7-on-3(β)-yl benzoate, recrystallised from acetone–methanol, had m. p. 159°, $[\alpha]_D$ − 54° (*c*, 3·18), − 55°(*c*, 1·59), $[M]_D$ −277°.

Cholestan-7-on-3(β)-yl acetate was prepared from cholest-5-en-7-on-3(β)-yl acetate by catalytic hydrogenation, using the procedure of Wintersteiner and Moore (*J. Amer. Chem. Soc.*, 1943, **65**, 1503). A small amount of cholestan-3(β)-yl acetate is always formed as a by-product during the hydrogenation. The cholestan-7-on-3-(β)-yl acetate was freed from this impurity by chromatography [16 fractions, the first two being cholestan-3(β)-yl acetate], or by conversion into the semicarbazone, m. p. 232° (decomp.), which furnished cholestan-3(β)-ol-7-one on acid hydrolysis with 4N-aqueous-alcoholic sulphuric acid on the water-bath (see below). The preparations by either method of purification were identical. Recrystallised from absolute alcohol, it had m. p. 149°, $[\alpha]_D$ − 36°(*c*, 6·13), − 36°(*c*, 3·07), − 37° (*c*, 1·77), − 37°(*c*, 1·53), − 36°(*c*, 0·89), $[M]_D$ − 163°.

Cholestan-3(β)-ol-7-one, recrystallised from acetone–methanol, had m. p. 168·5°, $[\alpha]_D$ − 36°(*c*, 6·01), − 36°(*c*, 2·12), − 36°(*c*, 1·84), − 36°(*c*, 1·50), − 36°(*c*, 1·06), $[M]_D$ − 144°.

Cholestan-7-on-3(β)-yl benzoate, recrystallised from acetone, had m. p. 169·5°, $[\alpha]_D$ − 17°(*c*, 3·06), − 18°(*c*, 1·60), − 18°(*c*, 1·22), $[M]_D$ − 86° (Found: C, 80·3; H, 10·0. $C_{34}H_{50}O_3$ requires C, 80·7; H, 9·8%).

Cholest-4-en-3-one had $[M]_D$ + 357° (this value is taken from Barton and Jones, *J.*, 1943, 602).

Cholest-5-en-3-one was prepared by the method of Butenandt and Schmidt-Thomé (*Ber.*, 1936, **69**, 882). Recrystallised from methanol, it had m. p. 126°, $[\alpha]_D$ − 7°(*c*, 2·42; micro-tube), − 9°(*c*, 1·03), $[M]_D$ − 30°.

Cholestan-3-one was prepared from cholestan-3-(β)-ol by chromic acid oxidation and purified by chromatography. Recrystallised from acetone, it had m. p. 127—128°, $[\alpha]_D$ + 42°(*c*, 1·93), + 42°(*c*,0·97), + 41°(*c*, 0·92), $[M]_D$ + 162°.

Cholestane-3 : 6-dione was prepared from cholestan-3(β)-ol-6-one by chromic acid oxidation and purified by chromatography. Recrystallised from light petroleum (b. p. 60—80°), it had m. p. 172°, $[\alpha]_D$ + 4°(*c*, 2·59; micro-tube), + 4°(*c*, 0·69), $[M]_D$ + 16°.

Cholestane-3 : 7-dione was prepared from cholestan-3(β)-ol-7-one by chromic acid oxidation and purified by chromatography. Recrystallised from light petroleum (b. p. 60—80°)–benzene, it had m. p. 190°, $[\alpha]_D$ − 19°(*c*, 1·84; micro-tube), − 19°(*c*, 0·91; micro-tube), $[M]_D$ − 76°.

3(β)-Chlorocholest-5-ene was prepared by the action of thionyl chloride on cholesterol. Recrystallised from ethyl acetate, it had m. p. 96°, $[\alpha]_D$ − 30°(*c*, 1·25), $[M]_D$ − 122°.

3(β)-Chlorocholestane was prepared by catalytic hydrogenation of 3(β)-chlorocholest-5-ene in acetic acid solution using a platinum oxide catalyst, and purified according to the directions of Anderson and Nabenhauer (*J. Amer. Chem. Soc.*, 1924, **46**, 1957). Recrystallised from chloroform–alcohol, it had m. p. 114—115°, $[\alpha]_D$ + 25°(*c*, 1·86), $[M]_D$ + 102°.

Ergostane Derivatives.—Ergosterol [ergosta-5 : 7 : 22-trien-3(β)-ol], recrystallised from alcohol, had m. p. 159°, $[\alpha]_D$ − 132°(*c*, 1·52), $[M]_D$ − 523°.

Ergosteryl acetate, recrystallised from chloroform–alcohol, had m. p. 175°, $[\alpha]_D$ − 92°(*c*, 1·89), − 92(*c*, 1·75), $[M]_D$ − 413°.

Ergosteryl benzoate, recrystallised from chloroform–alcohol, had m. p. 167—169°, $[\alpha]_D$ − 69°(*c*, 2·09), − 69°(*c*, 2·00), − 68°(*c*, 2·00), $[M]_D$ − 345°.

Ergost-7-en-3(β)-yl acetate was prepared from ergosteryl acetate by the method of Wieland and Benend (*Annalen*, 1943, **554**, 1). Recrystallised from chloroform–alcohol, it had m. p. 157—159°, $[\alpha]_D$ − 4°(*c*, 1·90), $[M]_D$ − 18°. This value is in satisfactory agreement with those recorded by Windaus and Langer (*ibid.*, 1933, **508**, 105) and by Reichel (*Z. physiol. Chem.*, 1934, **226**, 146), but is markedly different from that, $[\alpha]_D$ − 16°, reported recently by Wieland and Coutelle (*Annalen*, 1941, **548**, 270). There is no doubt, however, about the correctness of the rotation recorded here.

Ergost-7-en-3(β)-ol, recrystallised from chloroform–methanol, had m. p. 148°, $[\alpha]_D$ − 2°(*c*, 1·43), $[M]_D$ − 8°.

Ergost-7-en-3(β)-yl benzoate, recrystallised from chloroform–methanol, had m. p. 180·5°, $[\alpha]_D$ + 2°(*c*, 1·31), $[M]_D$ + 10°.

It will be noted that the values now found for ergost-7-en-3(β)-ol are in good agreement with those

suggested for Δ^7-unsaturated sterols as a result of literature analysis (Part I), whereas from previous workers there was a slight discrepancy. This is undoubtedly due to the relative difficulty of obtaining this compound and its derivatives in a state of purity.

Ergost-8(14)-en-3(β)-yl acetate was prepared from ergosteryl acetate by catalytic hydrogenation in ether–acetic acid solution using a platinum oxide catalyst. Recrystallised from acetone, it had m. p. 109°, $[\alpha]_D \pm 0°$(*c*, 4·26), + 1°(*c*, 2·13), $[M]_D$ + 4°.

Ergost-8(14)-en-3(β)-ol, recrystallised from acetone–methanol, had m. p. 132°, $[\alpha]_D$ + 11°(*c*, 4·01), + 11°(*c*, 2·01), + 11°(*c*, 0·80), $[M]_D$ + 44°.

Ergost-8(14)-en-3(β)-yl benzoate, recrystallised from acetone, had m. p. 116°. This m. p. is significantly lower than some of those given in the literature, but careful fractionation of two independent preparations proved their homogeneity. The rotation, however, was in complete agreement with values previously recorded; $[\alpha]_D$ − 1°(*c*, 4·42), ± 0°(*c*, 2·21), − 1°(*c*, 2·54), ± 0°*(*c*, 1·02), $[M]_D \pm 0°$.

Ergost-14-en-3(β)-yl benzoate was prepared by the method of Heilbron and Wilkinson (*J.*, 1932, 1708). Recrystallised from acetone, it had m. p. 158°, $[\alpha]_D$ + 22°(*c*, 5·49), + 22°(*c*, 2·75), + 23°(*c*, 1·37), $[M]_D$ + 116°.

Ergost-14-en-3(β)-ol, recrystallised from acetone–methanol, had m. p. 141°, $[\alpha]_D$ + 22°(*c*, 5·05), + 22°(*c*, 2·53), + 22°(*c*, 1·26), $[M]_D$ + 88°.

Ergost-14-en-3(β)-yl acetate had $[M]_D$ + 58°. [The rotation given by Heilbron and Wilkinson (*loc. cit.*) has been taken as correct. It is in good agreement with that for cholest-14-en-3(β)-yl acetate (see Part I).]

Ergosta-8(9) : 14-dien-3(β)-yl acetate was prepared by oxidation of ergost-8(14)-en-3(β)-yl acetate with selenium dioxide (Callow, *J.*, 1936, 462). The compound was most conveniently purified by chromatography eluting with light petroleum (b. p. 40—60°) (17 fractions, the first four being unchanged ergost-8(14)-en-3(β)-yl acetate). This procedure also removes most of the selenium-containing impurities which are otherwise retained very tenaciously during crystallisation. Recrystallised from acetone–methanol, it had m. p. 135—136°, $[\alpha]_D$ − 36°(*c*, 2·85), − 36°(*c*, 1·42), $[M]_D$ − 158°.

Ergosta-8(9) : 14-dien-3(β)-ol, recrystallised from acetone–methanol, had m. p. 141°, $[\alpha]_D$ − 19°(*c*, 1·47), $[M]_D$ − 76°.

Ergosta-8(9) : 14-*dien*-3(β)-*yl benzoate*, recrystallised from acetone–methanol, had m. p. 162°, $[\alpha]_D$ − 3° (*c*, 1·30), $[M]_D$ − 15° (Found: C, 83·8; H, 10·1. $C_{35}H_{50}O_2$ requires C, 83·6; H, 10·0%).

Ergost-7-*en*-3-*one* was prepared by Oppenauer oxidation of ergost-7-en-3(β)-ol. Recrystallised from methanol, it had m. p. 159°, $[\alpha]_D$ + 22°(*c*, 1·59; micro-tube), $[M]_D$ + 88° (Found: C, 83·9; H, 11·3. $C_{28}H_{46}O$ requires C, 84·4; H, 11·5%).

Ergost-8(14)-en-3-one was prepared by Oppenauer oxidation of ergost-8(14)-en-3(β)-ol. Recrystallised from chloroform–methanol, it had m. p. 129—130°, $[\alpha]_D$ + 30°(*c*, 1·91; micro-tube), + 30°(*c*, 1·32), $[M]_D$ + 119°.

Ergost-14-en-3-one was prepared by Oppenauer oxidation of ergost-14-en-3(β)-ol. Recrystallised from chloroform–methanol, it had m. p. 149—150°, $[\alpha]_D$ + 40°(*c*, 0·83), $[M]_D$ + 159°.

Ergosta-8(9) : 14-dien-3-one was prepared by Oppenauer oxidation of ergosta-8(9) : 14-dien-3(β)-ol. Recrystallised from ethyl acetate–methanol, it had m. p. 148°, $[\alpha]_D$ − 1°(*c*, 2·18; micro-tube), $[M]_D$ − 4°.

In these Oppenauer oxidations the yield of pure ketone was about 40%, but this could doubtless be improved upon by working on a larger scale and to a less exacting standard of purity.

Stigmastane Derivatives.—Stigmasta-5 : 22-dien-3(β)-yl acetate (Ciba) was recrystallised from ethyl acetate–acetone; it had m. p. 143°, $[\alpha]_D$ − 55°(*c*, 3·24), − 55°(*c*, 2·79), − 55°(*c*, 1·39), $[M]_D$ − 250°.

Stigmasta-5 : 22-dien-3(β)-ol (stigmasterol), recrystallised from acetone, had m. p. 168°, $[\alpha]_D$ − 51°(*c*, 3·56), − 51°(*c*, 2·63), − 51°(*c*, 1·78), − 51°(*c*, 1·31), $[M]_D$ −210°.

Stigmasta-5 : 22-dien-3(β)-yl benzoate, recrystallised from ethyl acetate–methanol, had m. p. 164°, $[\alpha]_D$ − 26°(*c*, 2·71), − 26°(*c*, 1·36), $[M]_D$ − 134°.

Stigmasta-5 : 22-dien-3(β)-yl acetate 22 : 23-dibromide was prepared by the method of Fernholz and Stavely (*J. Amer. Chem. Soc.*, 1939, **61**, 2956). When following the procedure of these workers it was not possible to obtain the pure acetate in satisfactory yield by recrystallisation, but shaking for a short time in ether–acetic acid solution with a platinum oxide catalyst in an atmosphere of hydrogen destroyed the contaminating stigmasta-5 : 22-dien-3(β)-yl acetate 5 : 6 : 22 : 23-tetrabromide by reduction. Recrystallisation from chloroform–methanol then furnished without difficulty the required acetate, m. p. 212° (no decomp.). Specimens not purified by partial hydrogenation melted lower and always with decomposition. $[\alpha]_D$ − 28°(*c*, 1·94), − 28°(*c*, 1·59), − 28°(*c*, 0·87), $[M]_D$ − 172° (Found: Br, 26·1. Calc. for $C_{31}H_{50}O_2Br_2$: Br, 26·1%).

Stigmasta-5 : 22-dien-3(β)-ol 22 : 23-dibromide, recrystallised from chloroform–methanol, had m. p. 208—209° (no decomp.), $[\alpha]_D$ − 23°(*c*, 1·84), − 23°(*c*, 1·11), − 23°(*c*, 0·94), $[M]_D$ − 132°.

Stigmasta-5 : 22-*dien*-3(β)-*yl benzoate* 22 : 23-*dibromide*, recrystallised from chloroform–alcohol, had m. p. 213° (no decomp.), $[\alpha]_D$ − 6°(*c*, 2·39), $[M]_D$ − 41° (Found: C, 64·5; H, 7·7. $C_{36}H_{52}O_2Br_2$ requires C, 64·0; H, 7·7%).

Stigmasta-4 : 22-dien-3-one was prepared by chromic acid oxidation of stigmasta-5 : 22-dien-3(β)-ol 5 : 6 : 22 : 23-tetrabromide in ether–acetic acid solution followed by zinc dust debromination in acetic acid solution on the water-bath, and was purified by chromatography. Recrystallised from methanol, it had m. p. 126°, $[\alpha]_D$ + 63°(*c*, 1·79), + 62°(*c*, 0·52), $[M]_D$ + 258°.

Stigmasta-4 : 22-dien-3-one 22 : 23-dibromide was prepared by Oppenauer oxidation of stigmasta-5 : 22-dien-3(β)-ol 22 : 23-dibromide and purified by chromatography. Recrystallised from acetone, it had m. p. 186—187° (no decomp.), $[\alpha]_D$ + 58°(*c*, 1·63; micro-tube), + 59°(*c*, 0·93; micro-tube), $[M]_D$ + 336°.

We thank British-Schering Ltd., Manchester, and Ciba for gifts of chemicals. One of us (D. H. R. B.) is indebted to Messrs. Imperial Chemical Industries Ltd. for an I.C.I. Fellowship during the tenure of which part of this work was carried out.

IMPERIAL COLLEGE OF SCIENCE AND TECHNOLOGY, S.W.7. [*Received, June 12th*, 1947.]