To Hélène

# PREFACE

The interest surrounding the sequence and mechanisms of the various biosynthetic steps which occur during the transformation of androgens prompted the undertaking of this project involving the synthesis of potential precursors in the biosynthesis of estrogens. Another consideration was the very importance of the steroidal and modified steroidal hormones in chemotherapy and for contraceptive purposes.

## ACKNOWLEDGMENTS

The author wishes to express his sincere thanks to his research director, Professor P. Morand, for his valuable discussions and his patience and encouragement throughout the progress of this work.

The cooperation of Angela Eriksen and her excellent typing of this thesis is greatly appreciated.

A bursary from the National Research Council of Canada and fellowships from the Province of Ontario, awarded to the author, are gratefully acknowledged.

TABLE OF CONTENTS

Page no.

Page no.

# LIST OF TABLES

# LIST OF FIGURES

## ABSTRACT

A proposed intermediate, 3β, 7α, 17β, 19-tetrahydroxy-5-androstene in the biosynthesis of ring-B-unsaturated estrogens was prepared as the 7α-acetoxy derivative by the reaction of 3β, 17β, 19-triacetoxy-5-androstene with t-butylperbenzoate in acetic acid, followed by selective hydrolysis and preferential crystallization. The 7β-acetoxy epimer was also prepared.

Several approaches to the synthesis of 7α, 19-disubstituted androgens were studied and a new convenient route to these compounds was discovered. During the course of our investigations several other possible estrogen precursors were isolated and characterized.

Attempts were made to develop a synthetic route to 1α-hydroxy-$\Delta^4$-3-keto steroids.

# INTRODUCTION

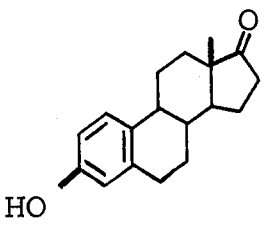
## 1. Estrogens

Estrogens are substances which are capable of producing heat (estrus) in females of various mammalian species. Although many substances with estrogenic activity have been isolated from plants, their importance has been overshadowed by those isolated from animals and those which have been chemically synthesized. In the animal body the estrogens are produced by the ovaries, testes, placenta and adrenal cortex. Since these substances are secreted by endocrine glands and are blood borne they are classified as hormones (female sex hormones). In turn these hormones belong to the chemical class of steroids. The tremendous importance of these steroidal sex hormones arises not only because of their function of stimulating the growth and development of reproductive organs in females, but also because of their growing usefulness as therapeutic agents in medicinal practice.

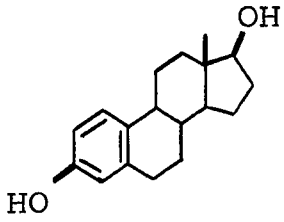
The first known steroidal hormone was isolated from the urine of pregnant women in 1929 by Doisy and coworkers[1] and also independently by Butenandt[2]. The pioneering work in the isolation and structural determination of steroidal hormones by various workers, particularly Marian, is well known and has been described in detail by Fieser and Fieser[3] and by Marrian[4]. This early work elucidated the structures of the three most important human estrogens: estrone 1, estradiol 2 and estriol 3. In 1932 the equine estrogens equilin 4 and equilenin 5 were isolated from the urine of pregnant mares[5]. The common feature amongst these steroidal estrogens is that they have eighteen carbon atoms and at least one aromatic ring.

Because of the important physiological properties[6a,b] of the estrogens, the last three decades have been witness to intense investigation into possible synthetic routes for the preparation of these hormones on a large scale. Their worth in the treatment of cancer[7a,b], circulatory diseases[8a,b] and their use in the preparation of oral contraceptives[9a,b] have already been demonstrated.

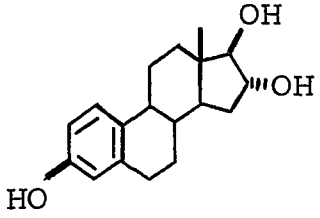
The chemical synthesis of steroidal estrogens has now been achieved and the various approaches and ultimate synthetic routes have been extensively reviewed by Morand and Lyall[10]. As well, a large number of non-steroidal and of modified steroidal compounds have been prepared and tested for estrogenic activity and some of them, such as diethylstilbestrol 6, have displayed activity comparable in potency to the naturally occurring hormones[11].



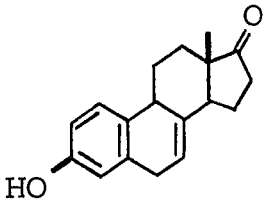

1



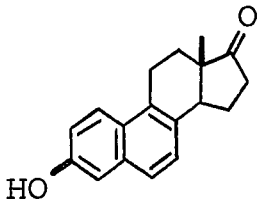

2



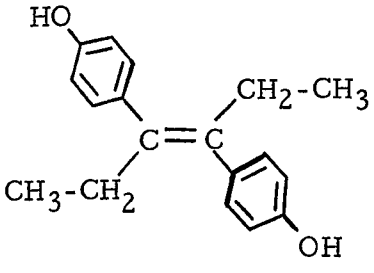

3




4




5




6

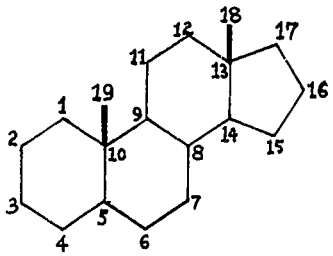
## 2. Nomenclature

The nomenclature of steroids used in this thesis is in accordance with the rules set by the International Union of Pure and Applied Chemistry (I.U.P.A.C.) in 1968[12].

Several trivial names are still used instead of the systematic names and a list of the trivial names used in this thesis is given in TABLE I.

TABLE I

| TRIVIAL NAME | SYSTEMATIC NAME |
|---|---|
| CHOLESTEROL | 5-CHOLESTEN-3β-OL |
| ESTRADIOL | 1, 3, 5(10)-ESTRATRIENE-3, 17β-DIOL |
| ESTRIOL | 1, 3, 5(10)-ESTRATRIENE-3, 16α, 17β-TRIOL |
| ESTRONE | 3-HYDROXY-1, 3, 5(10)-ESTRATRIEN-17-ONE |
| PROGESTERONE | 4-PREGNENE-3, 20-DIONE |
| TESTOSTERONE | 17β-HYDROXY-4-ANDROSTEN-3-ONE |
| EQUILIN | 3-HYDROXY-1, 3, 5(10), 7-ESTRATETRAEN-17-ONE |
| EQUILENIN | 3-HYDROXY-1, 3, 5(10), 6, 8-ESTRAPENTAEN-17-ONE |

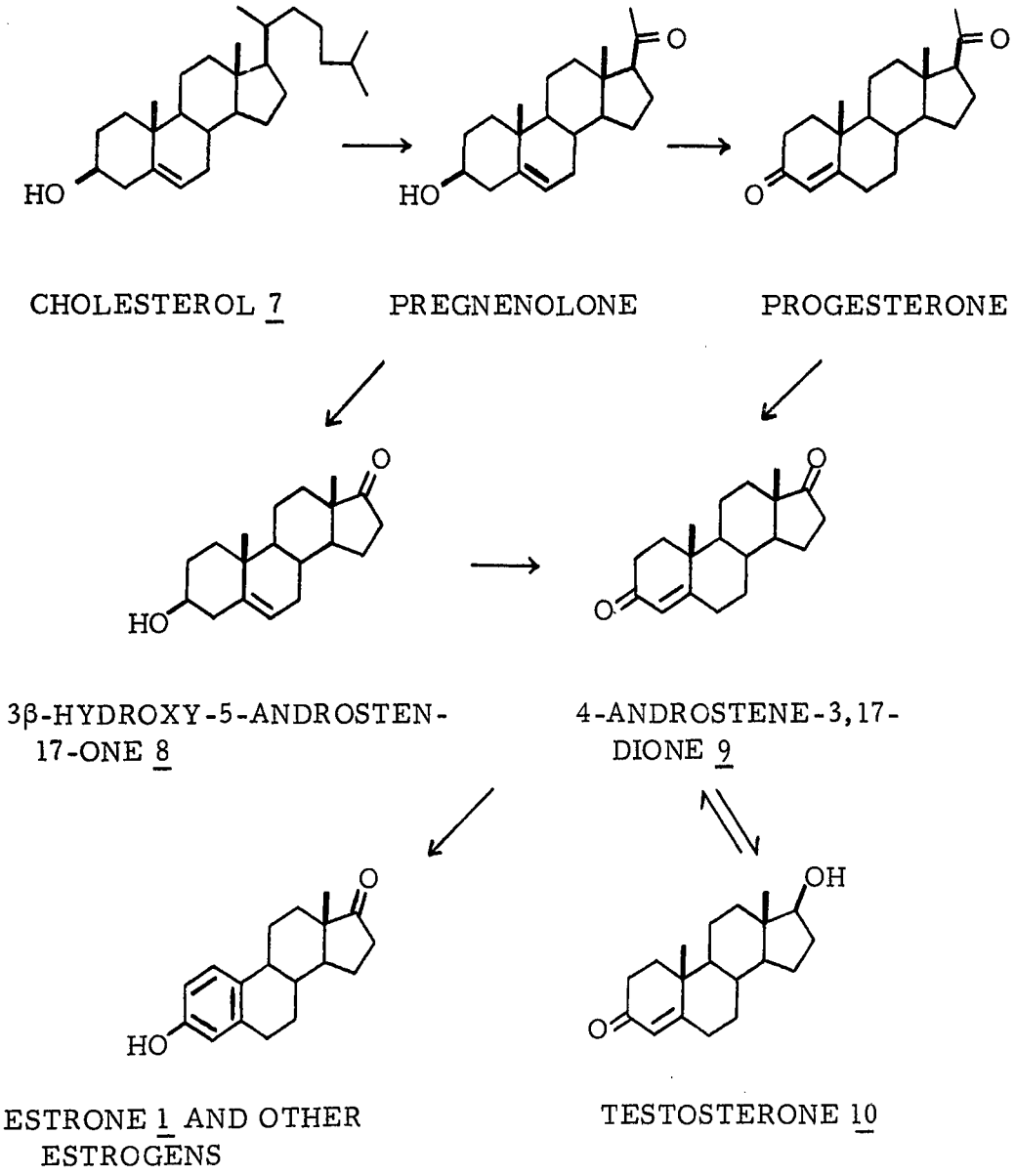


## 3. Biosynthesis of Steroidal Estrogens Saturated in the B-Ring

The biological origin of the steroidal estrogens has been the subject of a considerable amount of research over the past thirty years and a general biosynthetic pathway for these hormones has now been established. The same biosynthetic intermediates leading to the formation of cholesterol 7 are involved in the route leading to the estrogens. In 1942 Bloch and Rittenberg[13] were the first to show that acetate is the precursor for cholesterol 7. Since that time the intermediates along the pathway from acetate to cholesterol 7 have been uncovered and there are several reviews[14a,b,c,d,e] available on this subject. The important intermediates for the formation of cholesterol are illustrated in FIGURE I. Metabolic studies on cholesterol 7[15a,b,c] itself have shown that it is progressively converted into corticoids and progestagens, androgens and finally estrogens, the latter representing the final stage in the degradation of cholesterol 7. Although the metabolic studies on cholesterol 7 were performed using a variety of animal tissues both in vivo and in vitro, a general biosynthetic scheme for the formation of estrogens from cholesterol 7 may be drawn as depicted in FIGURE 2.

Acetate → Acetyl-CoA → Acetoacetyl-CoA →
3-HYDROXY-3-METHYL-GLUTARYL-CoA → MEVALONIC ACID →
ISOPENTYL PYROPHOSPHATE → GERANYL PYROPHOSPHATE
FARNESYL PYROPHOSPHATE → SQUALENE →
LANOSTEROL → ZYMOSTEROL → DESMOSTEROL →
CHOLESTEROL.

FIGURE I.




CHOLESTEROL 7

PREGNENOLONE

PROGESTERONE

3β-HYDROXY-5-ANDROSTEN-17-ONE 8

4-ANDROSTENE-3,17-DIONE 9

ESTRONE 1 AND OTHER ESTROGENS

TESTOSTERONE 10

FIGURE 2

(a) Intermediates between 4-androstene-3,17-dione 9 and estrone 1.

In 1953 Meyer[16] found that both 4-androstene-3,17-dione 9 and 3β-hydroxy-5-androsten-17-one 8 were hydroxylated at the 19-position by bovine adrenal glands and later he showed that human placenta could convert 19-hydroxy-4-androstene-3,17-dione 11 into estrone 1 and estradiol 2[17]. Using 4-androstene-3,17-dione 9 labeled with $^{14}C$ at position 4, as the substrate, Longchampt et al[18] demonstrated that human placental microsomes first hydroxylated the 19-position of the substrate and subsequently upon reincubation of the radioactive 19-hydroxy derivative, radioactive estrone 1 was isolated. Decisive experiments were later provided by Wilcox and Engel[19] when they undertook kinetic studies of the placental aromatization at early time periods and showed that 19-hydroxy-4-androstene-3,17-dione 11 was an obligatory intermediate in the conversion of 4-androstene-3,17-dione 9 to estrone 1 and estradiol 2. This result was substantiated by the fact that 19-hydroxy-4-androstene-3,17-dione 11 and 3β,19-dihydroxy-5-androsten-17-one 12 were converted to estrogens more completely than the compounds not hydroxylated in the 19-position[20]. In 1961 Morato and collaborators[21] had suggested that 19-oxo-4-androstene-3,17-dione 13 might be an intermediate in the aromatization process. They detected no 19-hydroxy-4-androstene-3,17-dione 11 following incubation of the 19-oxo-derivative while on occasion a trace of material with characteristics of the 19-aldehyde group was observed when the 19-hydroxy compound was incubated. However, the 19-oxo-compound was transformed into estrone 1 more rapidly than the 19-hydroxy compound. Under the same conditions the per cent estrone 1 formation from 4-androstene-3,17-dione 9, 19-hydroxy-4-androstene-3,17-dione 11 and 19-oxo-4-androstene-3,17-dione 13 was 30-40%, 50-60% and 100% respectively. Further evidence

for a 19-oxo-intermediate came from Axelrod et al[22] who showed that when testosterone 10 labelled with $^{14}C$ in the 19-position, was incubated with placental microsomes, the main by-product was labelled formic acid. That the formic acid produced could not be a consequence of enzymatic or non-enzymatic oxidation of formaldehyde was recently demonstrated by Skinner and Akhtar[23]. Their conclusions are based on the results of some elegant experiments involving the partial stereospecific introduction of a tritium label at C-19 in a 19-hydroxy androgen. After incubation of the tentatively assigned 19-hydroxy[(19-R)-19-$^{3}H$]-androgen they found 61.6% radioactivity in formic acid and 38.4% in water. A parallel experiment with the [(19-S)-19-$^{3}H$]-isomer gave 23.4% radioactivity in formic acid and 76.2% in water. Since the two protons in formaldehyde are chemically and biochemically equivalent, oxidation of labelled formaldehyde would give an equal distribution of radioactivity in formic acid and in water. Since this is not the case Skinner and Akhtar have suggested that "the removal of C-19 in estrogen biosynthesis occurs compulsorily at the oxidation state of a 19-aldehyde with the liberation of formic acid". In addition to this conclusion these workers have also shown the stereospecific removal of one of the C-19 hydrogen atoms in estrogen biosynthesis and they have suggested that a 19,19-dihydroxy-intermediate between the 19-hydroxy-compound and the 19-oxo-compound may be involved.

The evidence, then,for the intermediacy of 19-oxo-4-androstene-3,17-dione 13 in estrogen biosynthesis is most convincing and this compound is generally accepted as a direct precursor to estrone 1. The fact that it has never been isolated from an incubation experiment may be due to the small quantities of steroids involved and also the fast rate of reaction encountered in these systems.

Since all the above aromatizations were carried out using human placental microsomes it is of interest that Stárka and Breuer[24] were able to show that the horse placenta also yields human estrogens when certain steroidal hormones and hormone derivatives were used as substrates. They found that testosterone 10, 19-hydroxy-4-androstene-3,17-dione 11 and 19-oxo-4-androstene-3,17-dione 13 were converted to human estrogens to the extent of 20%, 24% and 33% respectively with equine placental microsomes. Although the horse placenta appears to be less efficient than the human placenta for aromatizing these particular steroids, both systems show that the efficiency with respect to the substrate used, increases in the order; 19-methyl- < 19-hydroxy- < 19-oxo-androgens. At this point one might speculate as to the possibility of a common pathway for estrogen biosynthesis in animals and man.

With the above information a more detailed scheme for the production of estrone 1 from androgens may be drawn as shown in FIGURE 3.

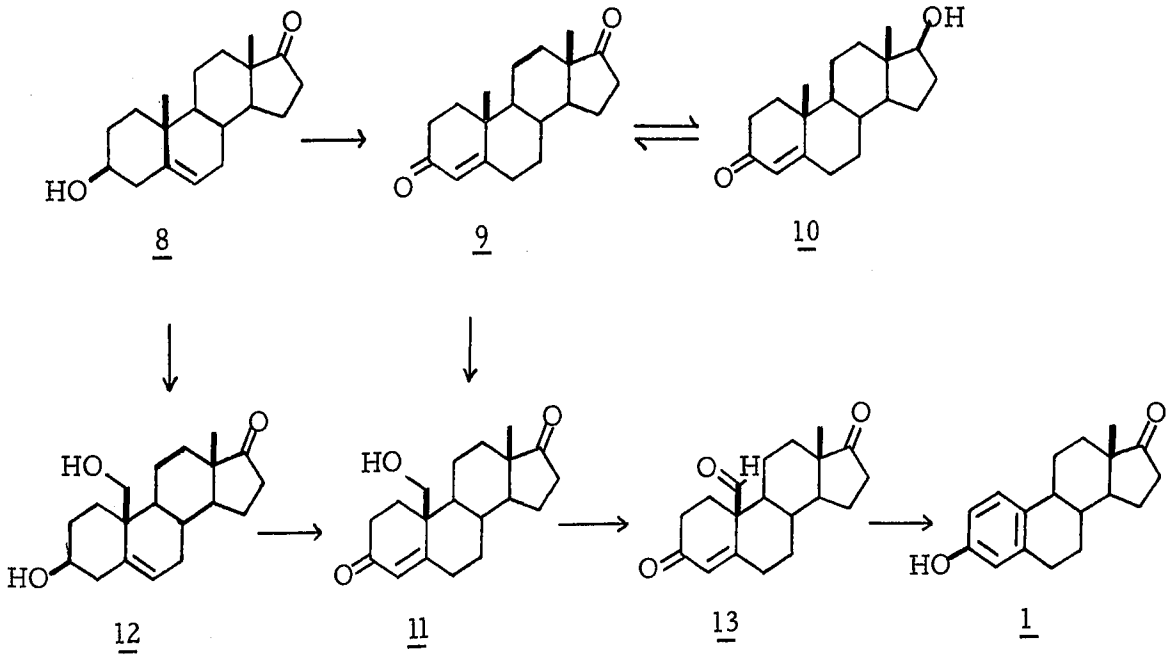



FIGURE 3

(b) Mechanism of Aromatization of the A-Ring

The placental aromatization of androgens requires NADPH and oxygen[25a,b], the usual co-factors for enzymatic hydroxylations. The hydroxylation at the 19-position has been accepted as the first step as well as the rate limiting step[19] in the aromatization process. Subsequently there remain, in chemical terms, only two operations to be effected for estrogen formation. One is the dehydrogenation at C-1 and C-2, and the other is the expulsion of the angular substituent at C-10. Whether the former is a distinct and separate step from the latter, or whether they are connected with each other is still a matter of conjecture.

Considerable progress concerning events which take place in the A-ring itself, was made by Morato et al[26] in 1962. 4-Androstene-3,17-dione 9 tritiated in the C-1 position [25% 1α-$^3$H, 75% 1β-$^3$H] was converted to estone 1 with 82% loss of tritium. When the label was predominantly reversed [93% 1α-$^3$H, 7% 1β-$^3$H] only a 15% loss of tritium occurred. Therefore the aromatization process involves loss of the 1β-hydrogen. Brodie and coworkers[27] have shown that estrone 1 formed from 1α, 2α-dideuterated 4-androstene-3,17-dione 9 retained its deuterium while the estrone 1 formed from the 2β-deuterated isomer had lost deuterium, indicating that the 2β-axial hydrogen is stereospecifically lost in the aromatization process. This result was confirmed by Fishman and Guzik[28a,b] who used tritum labelling in the 2α- and 2β-positions of 4-androstene-3,17-dione 9. Thus the overall stereochemistry of the dehydrogenation at C-1 and C-2 is β and cis. Recently Osawa and Spaeth[29] have shown the advantages of using tritium labelling in the 1α- and 2α-positions rather than in the 1β- and 2β- positions for the purpose of these studies. Tritium in the 2β-"axial" position could easily be lost by enolization of the C-3-ketone. This has been suggested as responsible for a discrepancy in the tritium distribution in testosterone reported by Brodie et al[30a] and Fishman et al[28a].

Mechanistically, the loss of the 2β-hydrogen(axial)can be explained by enolization[30b]. However, this should be a reversible process if unaccompanied by other driving forces, but Fishman et al[28b] have suggested that the hydrogen loss at C-2 is not reversible since after incubation their recovered substrates (2α- and 2β-tritiated 4-androstene-3,17-dione 9) showed no loss of tritium. This would suggest that the hydrogen loss at C-2 is concerted with or succeeds the other steps so that the driving force of aromatization precludes reversal of the enolization. Unfortunately there is some controversy on this point since Brodie et al[27] have reported isotope exchange at C-2 in their recovered substrate.

Morato et al[26] have discussed three possible pathways for the aromatization of a 19-oxo-androgen. These pathways are displayed in FIGURE 4.

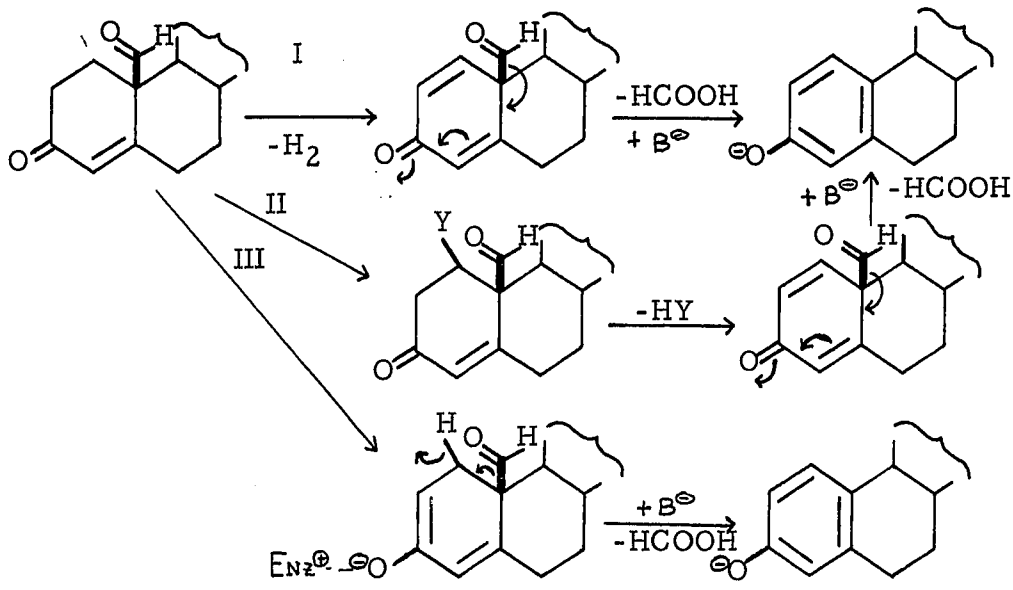
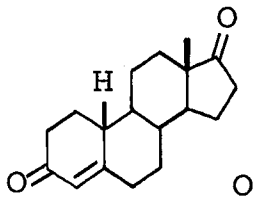
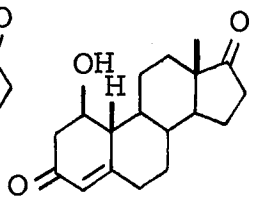



FIGURE 4.

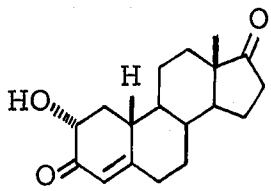
by the authors
Pathway I is not favored because the presence of a $\Delta^1$-dehydrogenase in mammalian tissue has yet to be demonstrated. Furthermore, $\Delta^1$-dehydrogenase systems such as those in microorganisms remove the 1α-hydrogen and have no requirement for NADPH and oxygen[31]. Pathway II involves the replacement of the 1β-hydrogen by a hydroxyl (or other leaving group) which can subsequently be eliminated. In fact Towsley and Brodie[32] have shown the formation of 1β-hydroxy-4-estrene-3,17-dione 15 from the incubation of 4-estrene-3,17-dione 14; however, the 1β-hydroxy compound showed no significant conversion to estrogens under conditions where there was 20% conversion of 4-estrene-3,17-dione 14. Brodie et al[27] have also found that 2α-hydroxy-4-estrene-3,-17-dione 16, 2β-hydroxy-4-estrene-3,17-dione 17 and 10β-hydroxy-4-estrene-3,17-dione 18 are poor substrates for estrone 1 formation. Earlier Gual et al[33] had shown 2β-hydroxy-4-androstene-3,17-dione 19 to be inactive as an estrogen precursor.



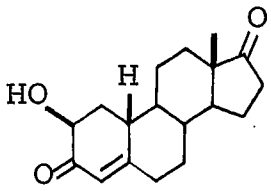

14



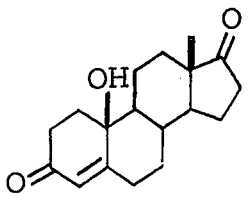

15



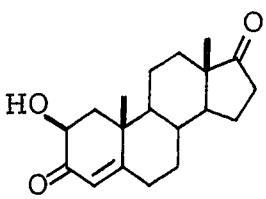

16



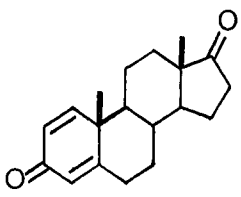

17



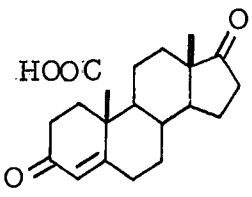

18




19




20




21

The third pathway involves removal of a hydride ion concerted with removal of the angular group at C-10. The possibility of oxidative attack on the oxygen attached to C-19 followed by abstraction of the 1β-hydride ion was ruled out by Townsley and Brodie[32] because of the fact that 4-estrene-3,17-dione 14 and 19-hydroxy-4-androstene-3,17-dione 11 have the same stereochemical and cofactor requirements for estrogen formation. These authors proposed a common mechanism for ring-A oxidation for $C_{18}$ and $C_{19}$ steroids (see FIGURE 5).

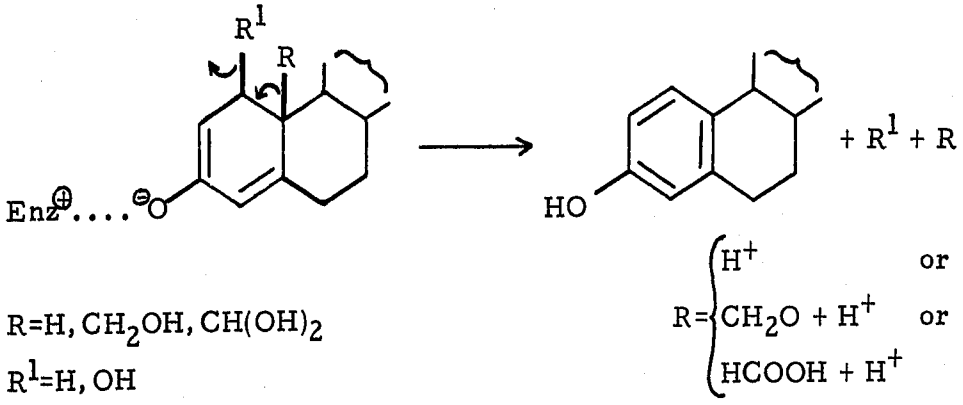
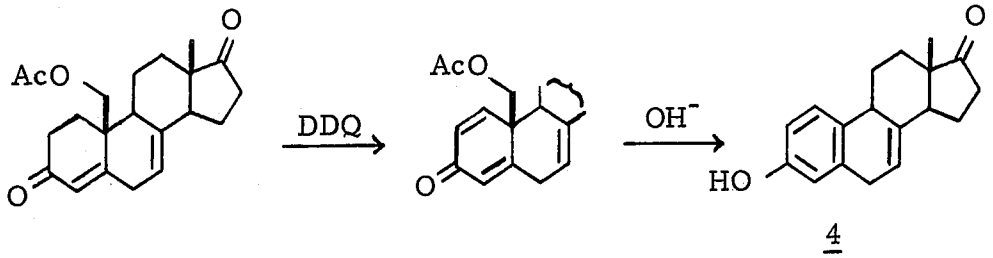



FIGURE 5

Meigs and Ryan[34] have studied the effects of oxygen and carbon monoxide on the intermediate steps of estrogen biosynthesis. Their results showed no inhibition of aromatization by carbon monoxide (5% $O_2$, 44% CO, 51% $N_2$) when 4-androstene-3,17-dione 9,19-hydroxy--4-androstene-3,17-dione 11 and 19-oxo-4-androstene-3,17-dione 13 were used as substrates. However, carbon monoxide decreased the aromatization of 4-estrene-3,17-dione 14 and 1,4-androstadiene-3,17-dione 20. Hydroxylation of the A-ring of 4-estrene-3,17-dione 14 was also inhibited by carbon monoxide to the same extent as was its

aromatization. These findings indicated the possibility that at least three mixed function oxidases might be active in the aromatization process. Earlier work[21, 33] had shown that 4-estrene-3,17-dione 14 and 1,4-androstadiene-3,17-dione 20 , each of which requires only one operation in order to be converted into estrone 1 are in fact converted less rapidly into estrone 1 than 4-androstene-3,17-dione 9 . At the same time 10β-carboxy-4-estrene-3,17-dione 21 was shown to be a poor precursor for estrone 1.

With all the information at hand it is disconcerting that the mechanism of aromatization of the A-ring is still not clear. We know that the angular group at C-10 is expelled as formic acid and that the 1β- and 2β-hydrogens are lost. The inefficiency of compounds with either unsaturation at C-1, C-2 or lacking a substituent at C-10, as precursors to estrone 1 favors a concerted mechanism and suggests a highly selective enzyme system. Finally the existence of many mixed function oxidases points out the complicated nature of this process in the biological system, a process which appears relatively simple from a purely chemical standpoint. This is evidenced by the chemical synthesis of equilin 4 by Bagli et al[35].



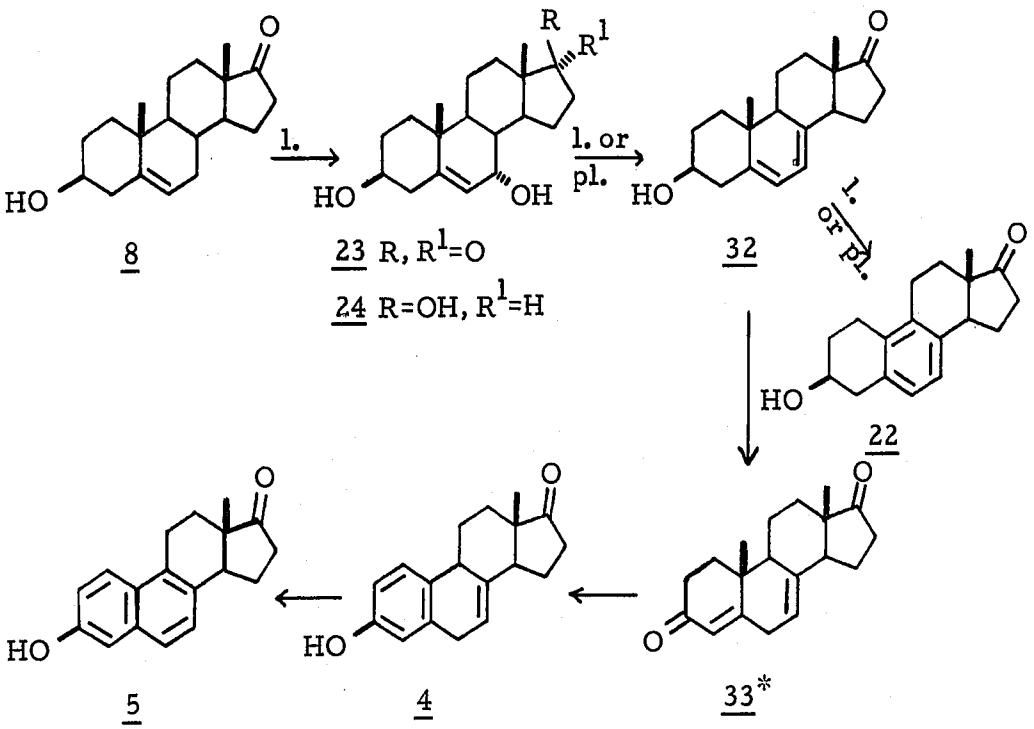

4

## 4. Biosynthesis of Steroidal Estrogens Unsaturated in the B-Ring

Phenolic (e.g. 4) and non-phenolic (e.g. 22) ring-B unsaturated estrogens have been isolated from the urine of pregnant mares[5,36a,b]. After feeding acetate-1-$^{14}$C to a pregnant mare, labelled equilin 4, equilenin 5, as well as labelled estrone 1 were obtained from the urine[37a,b]. However the equilin 4 and equilenin 5 obtained had only half the specific activity of the estrone 1 formed and it was also found that estrone 1 is not converted to ring-B unsaturated estrogens in the pregnant mare[38]. Therefore the biogenetic pathway for estrone 1 formation in the pregnant mare is not the same as that for the equine estrogens. As yet equine estrogens have not been detected in human urine. Another major estrogen isolated from equine urine is the so-called "Heard's ketone" 22[36a,b]. Stárka and Breuer[24] have studied the biogenesis of the equine estrogens from 3β-hydroxy-5-androsten-17-one 8, with preparations of equine liver and placenta. Based on their observations they have proposed the biosynthetic sequence as shown in FIGURE 6.

The formation of a 7α-hydroxy intermediate (3β, 7α-dihydroxy-5-androsten-17-one 23) from equine liver preparations is of interest since this compound had previously been isolated from human urine and was shown to be a true metabolite of 3β-hydroxy-5-androsten-17-one 8 in humans[39a,b]. This 7α-hydroxy compound has also been found in human plasma[40]. Other organs besides the liver, notably the adrenal cortex, possess steroid 7α-hydroxylating activity[41a,b]. Knuppen et al[42] showed for the first time that phenolic steroids such as estrone 1 could be hydroxylated in the 7α-position by beef adrenal brei and more recently the existence of 7α-hydroxylase activity in rat testicular glands was demonstrated[43]. Although there have been some reports[44a,b] of 7β-

hydroxylations of steroid hormones by animal tissue preparations, no 7β-hydroxy hormone derivatives have been identified in human body tissues or fluids. On the other hand several examples of 7β-hydroxylations have been observed in microbiological systems[45a,b,c,d].




l. = liver

pl. = placenta

FIGURE 6

* This compound 33 has not been detected in human placental systems.

Aromatization studies on 7-hydroxylated steroid hormones were performed by Cédard et al[46] with the following results. When 3β,-7α-dihydroxy-5-androsten-17-one 23 was incubated with human placental microsomes, 5-androstene-3β, 7α,17β-triol 24 and 7α-hydroxy-estrone 25 were produced. When 5-androstene-3β, 7β,17β-triol 26 was incubated 3β,-7β -dihydroxy-5-androsten-17-one 27, 7β-hydroxy-estrone 28 and 7β-hydroxy-estradiol 29 were formed. When the 7β-triol was perfused* with human placenta some 7-oxo-estrone 30 was also formed along with the other products. No phenolic metabolite could be detected after perfusion of 3β,17β-dihydroxy-5-androsten-7-one 31. The above results are portrayed in FIGURE 7.

A most interesting development in this field occurred when Stárka et al[48] found that human placenta could convert 5-androstene-3β,-7α,17β-triol 24 to the equine estrogens equilin 4 and equilenin 5 although under the conditions used more non-phenolic ring-B unsaturated steroids than phenolic ring-B unsaturated steroids were formed. Their results also indicated that 3β-hydroxy-5,7-androstadien-17-one 32 is a direct precursor to Heard's ketone 22. The scheme for the biogenesis of phenolic and non-phenolic ring-B unsaturated or ring-B aromatic steroids in human systems is identical to that shown in FIGURE 6, with the exception noted.

---

* Whereas incubation experiments with human placental microsomes refer to the technique developed by Ryan[25b] which involves grinding up of the placenta, centrifugation and isolation of the enzymatically active microsomal fraction, perfusion techniques use an intact placenta which is kept under conditions which simulate those in the living body. A description of the perfusion system has been given by Cédard et al[47].

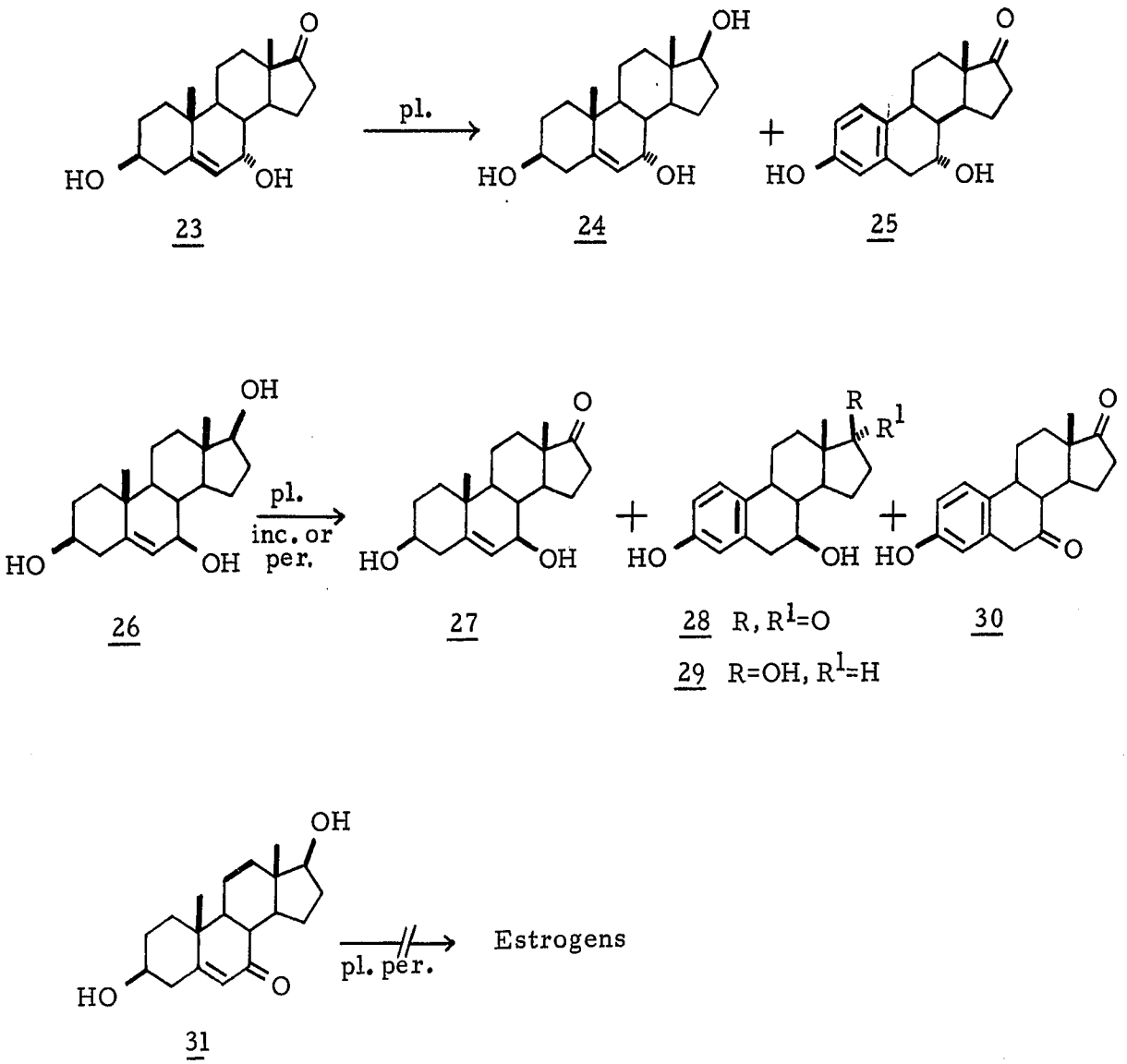

HO
OH
O
pl.
HO
OH
OH
+
HO
O
OH
23
24
25
HO
OH
OH
pl.
inc. or
per.
HO
OH
O
+
R
R1
HO
OH
+
HO
O
O
26
27
28 R, R1=O
29 R=OH, R1=H
30
OH
HO
O
pl. per.
Estrogens
31

FIGURE 7.

Aromatization studies on 7-hydroxylated steroid hormones were performed by Cédard et al[46] with the following results. When 3β,-7α-dihydroxy-5-androsten-17-one 23 was incubated with human placental microsomes, 5-androstene-3β, 7α,17β-triol 24 and 7α-hydroxy-estrone 25 were produced. When 5-androstene-3β, 7β,17β-triol 26 was incubated 3β,-7β -dihydroxy-5-androsten-17-one 27, 7β-hydroxy-estrone 28 and 7β-hydroxy-estradiol 29 were formed. When the 7β-triol was perfused* with human placenta some 7-oxo-estrone 30 was also formed along with the other products. No phenolic metabolite could be detected after perfusion of 3β,17β-dihydroxy-5-androsten-7-one 31. The above results are portrayed in FIGURE 7.

A most interesting development in this field occurred when Stárka et al[48] found that human placenta could convert 5-androstene-3β,-7α,17β-triol 24 to the equine estrogens equilin 4 and equilenin 5 although under the conditions used more non-phenolic ring-B unsaturated steroids than phenolic ring-B unsaturated steroids were formed. Their results also indicated that 3β-hydroxy-5,7-androstadien-17-one 32 is a direct precursor to Heard's ketone 22. The scheme for the biogenesis of phenolic and non-phenolic ring-B unsaturated or ring-B aromatic steroids in human systems is identical to that shown in FIGURE 6, with the exception noted.

---

* Whereas incubation experiments with human placental microsomes refer to the technique developed by Ryan[25b] which involves grinding up of the placenta, centrifugation and isolation of the enzymatically active microsomal fraction, perfusion techniques use an intact placenta which is kept under conditions which simulate those in the living body. A description of the perfusion system has been given by Cédard et al[47].

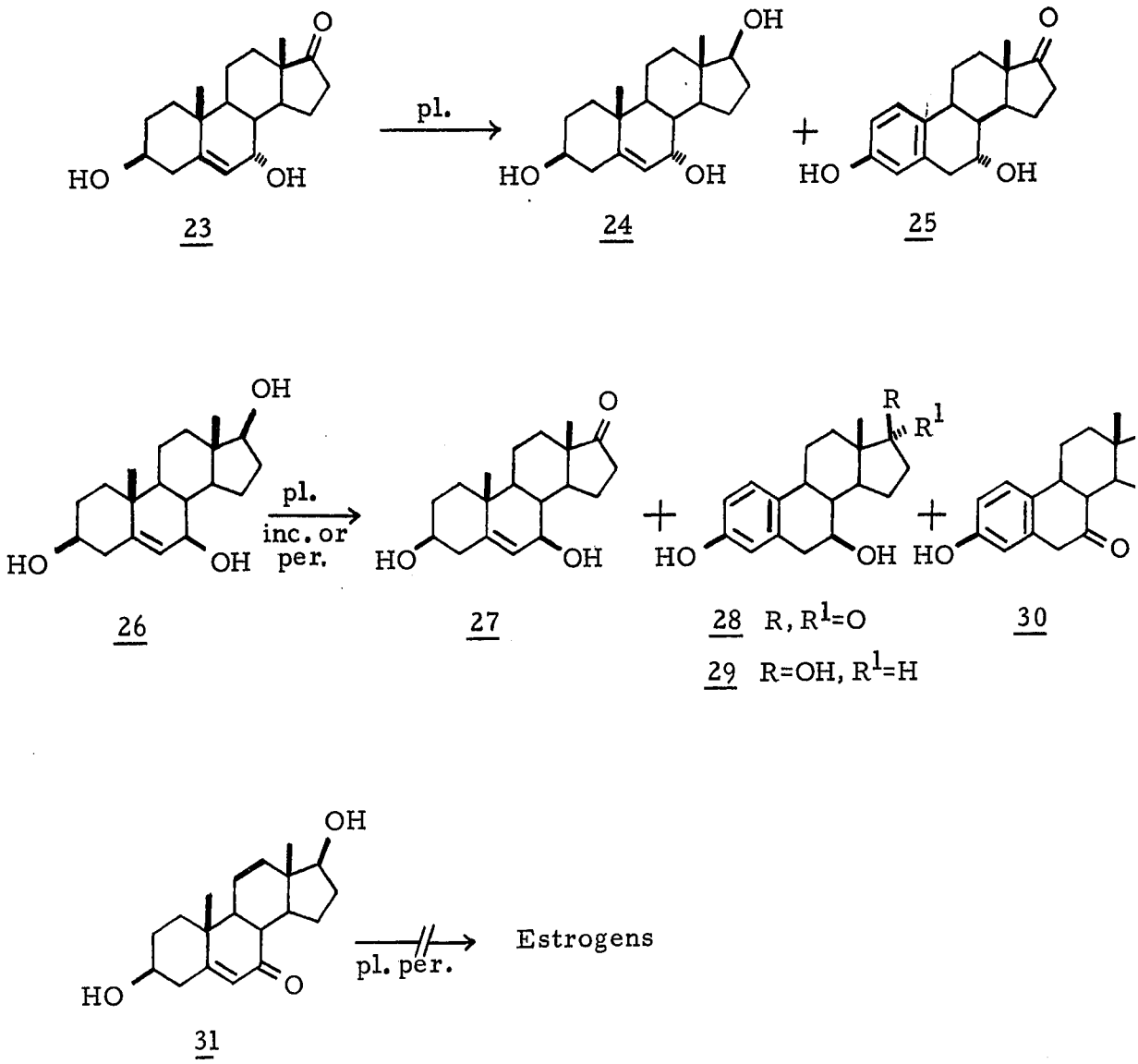

HO
OH
O
pl.
HO
OH
OH
+
HO
OH
O
23
24
25
HO
OH
OH
pl.
inc. or
per.
HO
OH
O
+
HO
R
R1
OH
+
HO
O
26
27
28 R, R1=O
29 R=OH, R1=H
30
HO
OH
O
pl. per.
Estrogens
31

FIGURE 7.

## RESULTS AND DISCUSSION

### 1. Objectives

The biochemical background which has been presented led us to speculate as to the possibility of a biosynthetic pathway for estrogen production common to all mammals. The difference in end-products, i.e. estrone 1 in man and equilin 4 in the horse, is simply a consequence of which processes are the predominant ones in the respective systems. For example, during the transformation of androgens into estrogens, there appear to be four basic processes: hydroxylations (H) ; conversion of the 3β-hydroxy-$\Delta^5$-system into a $\Delta^4$-3-keto-system (O) ; dehydrations (D) ; and aromatizations (A). With 3β-hydroxy-5-androsten-17-one 8 as precursor, a certain combination of these processes leads to estrone 1 (FIGURE 8) whereas a different combination leads to equilin 4. The sequence in which the individual processes occur may also have a bearing on the type of end-product that will be formed.

Part of FIGURE 8 is a correlation of published results (solid arrows) of the biosynthetic studies of B-ring saturated and B-ring unsaturated estrogens. The remainder of the scheme (broken arrows) is a postulated biosynthetic pathway. The 7α,19-dihydroxy intermediate (bold print), which has yet to be detected in the various systems studied emerges as a necessary link in order to have a scheme common to all mammals. It is quite possible that other intermediates*, such as 5-hydroxy and 1-hydroxy androgens, may also have a role in the biosynthesis of these estrogens but they have not been included in the general scheme presented in FIGURE 8.

---

* It is likely that because of the minute quantities of steroids involved and the rapidity of certain metabolic steps, some key intermediates may have escaped detection.

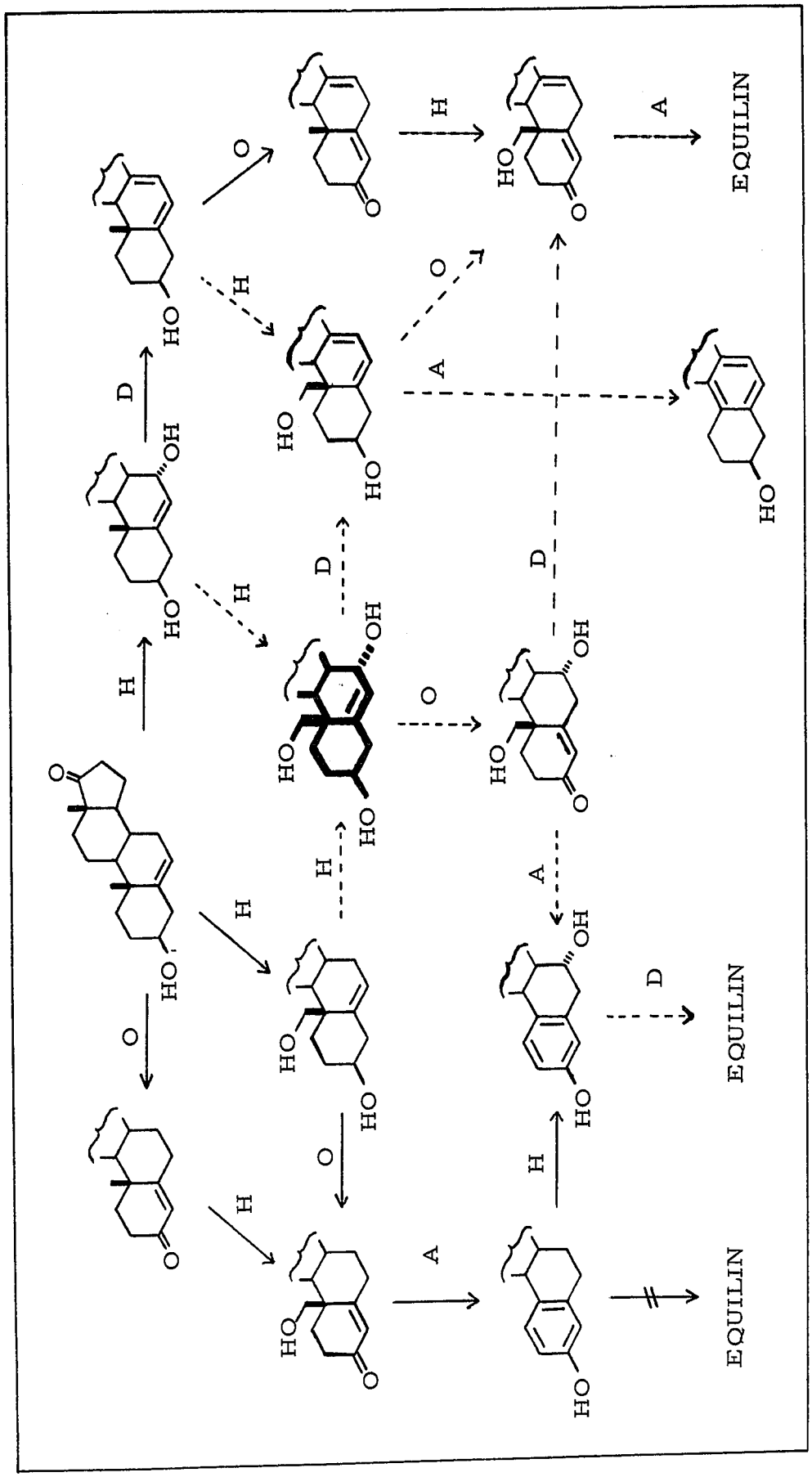



FIGURE 8.

This thesis deals with the synthesis of some possible estrogen precursors, particularly the 7α,19-dihydroxy androgens which we believe will prove to be most efficient as precursors for ring-B unsaturated estrogens.

Since we not only wished to prepare enough material for subsequent biosynthetic studies (possibly involving isotope labelling) but also for pharmacological testing, we have explored a number of synthetic approaches to these compounds with the aim of developing an efficient route in terms of yield and stereospecificity.

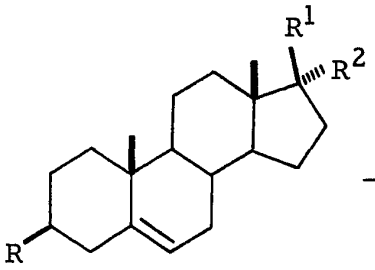
Finally, as mentioned earlier, the important applications of estrogens and related compounds in chemotherapy was an added incentive in undertaking the synthesis of potential estrogen precursors.

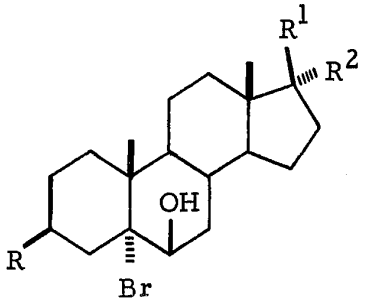
## 2. Synthesis of 7,19-Disubstituted Androgens

For this synthesis it was decided to use a substance such as 3β-acetoxy-5-androsten-17-one 34 (SCHEME 1) into which the appropriate substituents could be introduced to afford 7,19-disubstituted androgens.

The functionalization of C-19 was accomplished by a three step synthesis. Treatment of 3β-acetoxy-5-androsten-17-one 34 in dioxane with N-bromoacetamide and perchloric acid[49] yielded the bromohydrin 35. Lead tetraacetate oxidation[50] of 3β-acetoxy-5-bromo-6β-hydroxy-5α-androstan-17-one 35 gave 3β-acetoxy-5-bromo-6β,19-oxido-5α-androstan-17-one 36, which, upon reduction with zinc in ethanol[51] yielded 3β-acetoxy-19-hydroxy-5-androsten-17-one 37 in an overall yield of 18%.

In order to introduce a 7α-substituent we elected to carry out an allylic oxidation involving treatment of the $\Delta^5$-steroid with t-butyl perbenzoate and a catalytic amount of cuprous bromide. This reaction has been shown[52] to proceed without allylic rearrangement, yielding the 7α- and 7β-benzoate derivatives. Stárka[53] has applied this reaction to $\Delta^5$-steroids using acetic acid as the solvent and in his case was able to isolate a mixture of the 7α- and 7β-acetoxy derivatives in about 50% yield. Although column chromatography only partially separated the epimers, the 7α-epimer was shown to be the major product. Although it has been reported[54] that acetoxy derivatives obtained from the reaction of olefins with t-butylperbenzoate in acetic acid are a result of ester interchange, it is more likely that under these conditions a nucleophilic displacement of the benzoate group by acetic acid is involved.





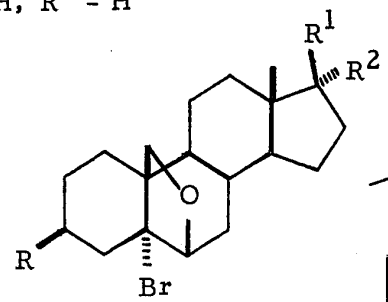
34 R = OAc, $R^1, R^2$ = O

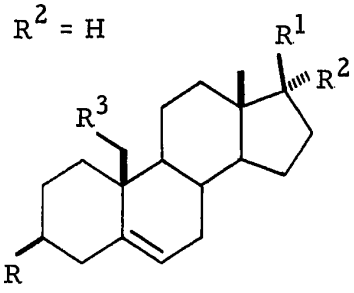
40 R = $R^1$ = OAc, $R^2$ = H

50 R = $R^1$ = OH, $R^2$ = H

35 R = OAc, $R^1, R^2$ = O

41 R = $R^1$ = OAc, $R^2$ = H





36 R=OAc, $R^1, R^2$=O

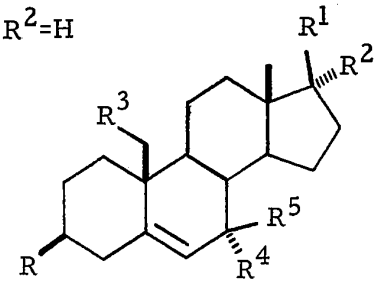
42 R=$R^1$=OAc, $R^2$=H

37 R=OAc, $R^1, R^2$=O, $R^3$=OH

38 R=$R^3$=OAc, $R^1, R^2$=O

43 R=$R^1$=OAc, $R^2$=H, $R^3$=OH

44 R=$R^1$=$R^3$=OAc, $R^2$=H



39 R=$R^3$=OAc, $R^1, R^2$=$R^4$, $R^5$=O

45 R=$R^1$=$R^3$=$R^4$=OAc, $R^2$=$R^5$=H

46 R=$R^1$=$R^3$=$R^5$=OAc, $R^2$=$R^4$=H

47 R=$R^1$=$R^3$=OH, $R^4$=OAc, $R^2$=$R^5$=H

48 R=$R^1$=$R^3$=OH, $R^5$=OAc, $R^2$=$R^4$=H

49 R=$R^1$=$R^3$=OAc, $R^2$=H, $R^4, R^5$=O

114 R=OAc, $R^3$=H, $R^1, R^2$=$R^4, R^5$=O

SCHEME 1

In applying Stárka's method, it was necessary in our case, to acetylate the 19-hydroxy group because of the free radical nature of the reaction. Thus, treatment of 3β,19-diacetoxy-5-androsten-17-one 38 (SCHEME 1) with t-butylperbenzoate and cuprous bromide in acetic acid led to the isolation of an oil consisting of the 7ξ-acetoxy derivatives in 40% yield. By nuclear magnetic resonance (n.m.r.) spectroscopic analysis this mixture was shown to contain 75% of the desired 7α-acetoxy compound* (FIGURE 9); however, all attempts to separate these epimers failed. (Similar results were obtained using t-butylperbenzoate without acetic acid. The epimeric mixture of 7-benzoates was rich in the 7α-epimer but complete separation of the epimers was not achieved).

Along with the 7ξ-acetoxy compounds we also obtained a small amount of the corresponding 7-ketone 39 whose structure was confirmed by comparison of its spectral data and by mixed melting point with 3β,19-diacetoxy-5-androstene-7,17-dione prepared by chromic acid oxidation[55] of 3β,19-diacetoxy-5-androsten-17-one 38.

When the t-butylperbenzoate reaction was performed with 3β,17β,19-triacetoxy-5-androstene 44, the product isolated in which substitution had occurred (40%) was also found to be a 3:1 mixture of the 7α- and 7β-acetoxy derivatives 45 and 46. Again, it was not possible to separate this oily mixture by repeated column chromatography.

---

* The basis for this configurational assignment is given in SECTION 3.

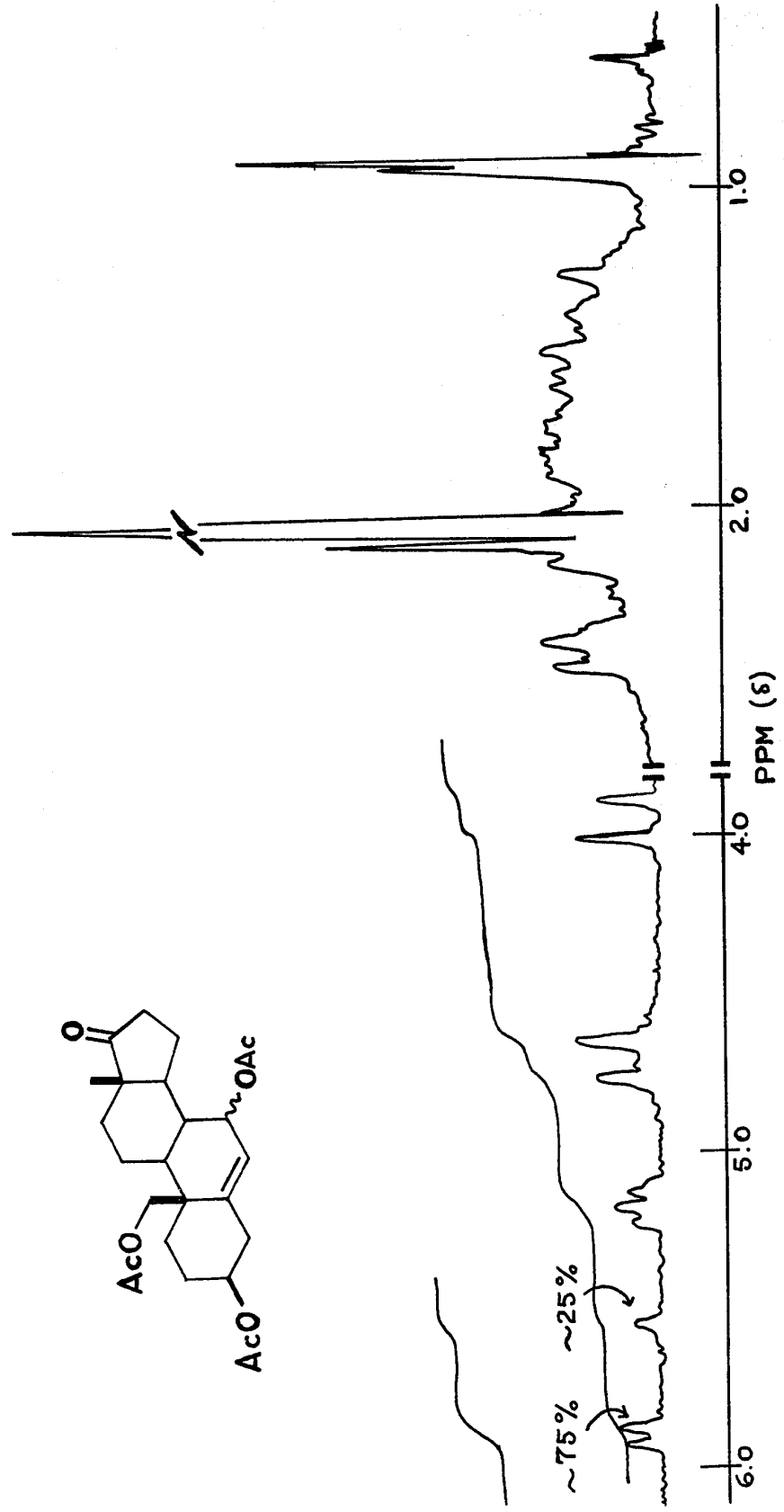
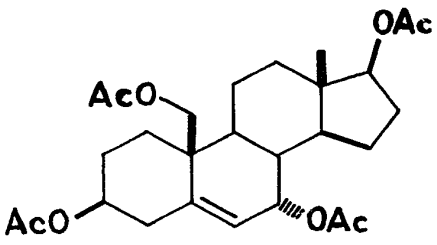



FIGURE 9.

However, by treatment of the epimeric mixture with a methanolic solution of sodium carbonate at room temperature for 24 hours, it was possible to selectively hydrolyze the 3β,17β- and 19-acetoxy groups, leaving the 7ξ-acetoxy groups intact. This result is not unexpected since the 7-position is more sterically hindered than the other three positions[56a, b]. When the resulting epimeric monoacetates were dissolved in chloroform and left standing at 5° for 24 hours, the 7α-monoacetate crystallized preferentially and in this manner we were able to obtain the desired 7α-acetoxy-3β,17β,19-trihydroxy-5-androstene 47, albeit in poor yield (14%).

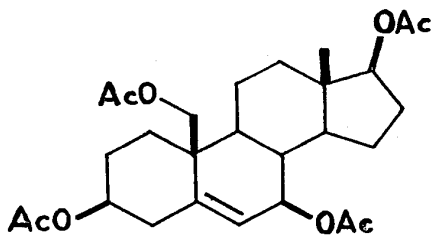
In order to confirm the configurational assignment of the 7α-epimer and for purposes of comparison with the latter in biological systems, 7β-acetoxy-3β,17β,19-trihydroxy-5-androstene 48 was prepared in the following manner. 3β,17β,19-Triacetoxy-5-androstene 44 was oxidized to the 7-keto-derivative 49 with chromic acid[55]. The ketone was reduced with sodium borohydride[56b], acetylated and subjected to selective hydrolysis, affording the 7β-monoacetate 48. Since the epimeric monoacetates 47 and 48 were insoluble in deuterated chloroform they were reacetylated to the more soluble tetraacetates 45 and 46, thus avoiding the addition of more polar solvents which tended to mask some of the signals in the n.m.r. spectra. The spectra for compounds 45 and 46 are shown in FIGURES 10 and 11.




45




FIGURE 10.




46




FIGURE 11.

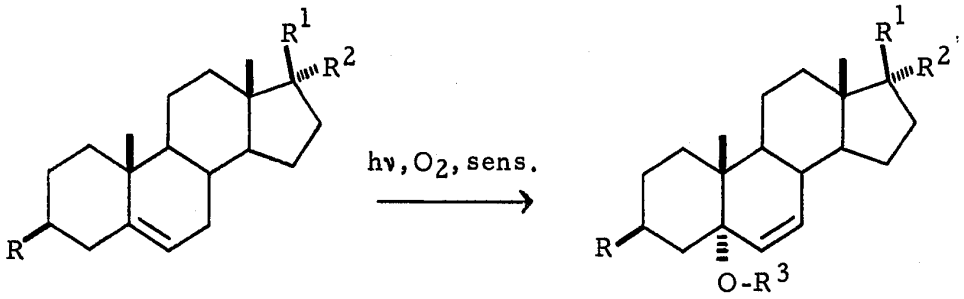
3. Identification of 7α- and 7β-Substituted-$\Delta^5$-Steroids

Since on several occasions our attempts to introduce a 7α-substituent into the steroid nucleus led to the formation of epimers, it was important that we be able to assign with certainty the configuration at C-7 in the products isolated. For this purpose some 7α- and 7β-hydroxy and -acetoxy-$\Delta^5$-steroids were prepared by known stereospecific methods. All the 7α-hydroxy compounds were obtained via a three step synthesis involving photosensitized oxygenation , followed by allylic rearrangement* and reduction. This method[57a, b, c] is discussed later in the thesis [SECTION 4(a)]. The 7β-hydroxy compounds were obtained by metal hydride reduction[56b] of the $\Delta^5$-7-keto-compounds. These reactions are outlined in SCHEME 2. The molecular rotations ($[M]_D$) and pertinent n.m.r. data for the compounds prepared are correlated in TABLE 2.

From the table it can be seen that the 7α-substituted compounds are highly levorotatory while the 7β-substituted compounds are either dextrorotatory or only slightly levorotatory. This has generally been found to be the case for 7-substituted-$\Delta^5$-steroids[61]. Consequently one may assign the configuration at C-7 with some confidence on the basis of the molecular rotation. However, epimeric mixtures cannot be analyzed for 7α- and 7β-components if neither of the epimers is available in the pure state.

---

* In all cases some unrearranged products i.e. 5α-hydroxy-$\Delta^6$-derivatives, were obtained.




51 R=OAc, $R^1=C_8H_{17}$, $R^2$=H

34 R=OAc, $R^1$, $R^2$=O

40 R=$R^1$=OAc, $R^2$=H

52 R=$R^3$=OH, $R^1=C_8H_{17}$, $R^2$=H

53 R=OAc, $R^3$=OH, $R^2$=H, $R^1=C_8H_{17}$

54 R=OH, $R^3$=$R^2$=H, $R^1=C_8H_{17}$

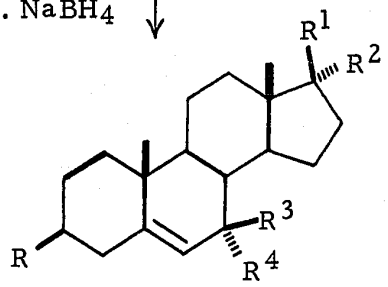
55 R=OAc, $R^3$=$R^2$=H, $R^1=C_8H_{17}$

56 $R^1$=R=OAc, $R^2$=$R^3$=H

57 R=OAc, $R^1$, $R^2$=O, $R^3$=H

1. $CrO_3$/ HOAc
2. $NaBH_4$

1. $CHCl_3$
2. NaI




58 R=$R^1$=$R^4$=OAc, $R^2$=$R^3$=H

59 R=OAc, $R^1=C_8H_{17}$, $R^2$=$R^3$=H, $R^4$=OH

60 R=OAc, $R^1=C_8H_{17}$, $R^2$=$R^4$=H, $R^3$=OH

61 R=$R^4$=OAc, $R^2$=$R^3$=H, $R^1=C_8H_{17}$

62 R=$R^3$=OAc, $R^2$=$R^4$=H, $R^1=C_8H_{17}$

63 R=OAc, $R^1$, $R^2$=O, $R^3$=H, $R^4$=OH

64 R=$R^4$=OAc, $R^1$, $R^2$=O, $R^3$=H

65 R=$R^1$=OAc, $R^2$=$R^3$=H, $R^4$=OH

66 R=$R^4$=OH, $R^2$=$R^3$=H, $R^1=C_8H_{17}$

111 R=$R^1$=OAc, $R^2$=H, $R^3$, $R^4$=O

SCHEME 2

TABLE 2

| Compound | $[M]_D$ | Ref. | ppm (δ) 6-H | ppm (δ) 7-H | $J_{6,7}$ Hz | Fig. no. |
|---|---|---|---|---|---|---|
| 3β-Acetoxy-7α-hydroxy-5-cholestene 59 | -388° | 58 | 5.60 | 3.75 | 5.4 | - |
| 3β-Acetoxy-7β-hydroxy-5-cholestene 60 | -89° | 56b | 5.30 | 3.81 | 2.0 | - |
| 3β, 7α-Diacetoxy-5-cholestene 61 | -851° | 56b, 45b, 58, 60, 69 | 5.59* | 4.97* | 5.0* | 12 |
| 3β, 7β-Diacetoxy-5-cholestene 62 | +269° | 56b | 5.23 | 5.02 | 2.0* | 13 |
| 3β-Acetoxy-7α-hydroxy-5-androsten-17-one 63 | -237° | A | 5.72 | 4.00 | 5.8 | - |
| 3β, 7α-Diacetoxy-5-androsten-17-one 64 | -694° | 45b | 5.64 | 5.10 | 5.0 | - |
| 3β, 17β-Diacetoxy-7α-hydroxy-5-androstene 65 | -454° | A | 5.61 | 3.80 | 5.0 | 14 |
| 3β, 7α, 17β-Triacetoxy-5-androstene 58 | -950° | A | 5.59 | 4.97 | 5.0 | - |

A This thesis.

* The values for $J_{6,7}$ in compounds 61 and 62 are smaller by a factor of about two than those reported by Shoppee and Newman[56b]. Professor Shoppee has informed us that the coupling constants in his paper should be halved; however, he was unable to offer any explanation for the discrepancy in the chemical shifts for the 6-H and 7-H in compound 61. Our values differ by about 0.2 and 0.4 p.p.m. respectively with those reported by the above authors.

Fortunately the nuclear magnetic resonance spectra of 7-substituted-$\Delta^5$-steroids are such that assignments of configuration as well as analyses of epimeric mixtures can be made with more certainty. This is on the basis of the chemical shift ($\delta$) of the C-6 olefinic proton and also the coupling constant ($J_{6,7}$) between the 6-H and the 7-H. TABLE 2 shows that for 7α-substituted-$\Delta^5$-steroids the signal for the 6-H generally appears at ca. 0.3 p.p.m. more downfield than in the case of 7β-substituted-$\Delta^5$-steroids. An examination of Dreiding models shows that the dihedral angle (6-H/7-H) is ca. 25° for 7α-substituted compounds and ca. 80° for the 7β-substituted compounds, which is an indication[62] that $J_{6,7}$ should be larger for the former class of compounds and this is verified in TABLE 2. The assignments of configuration for compounds in SECTION 2 were made on the basis of the methods discussed above.

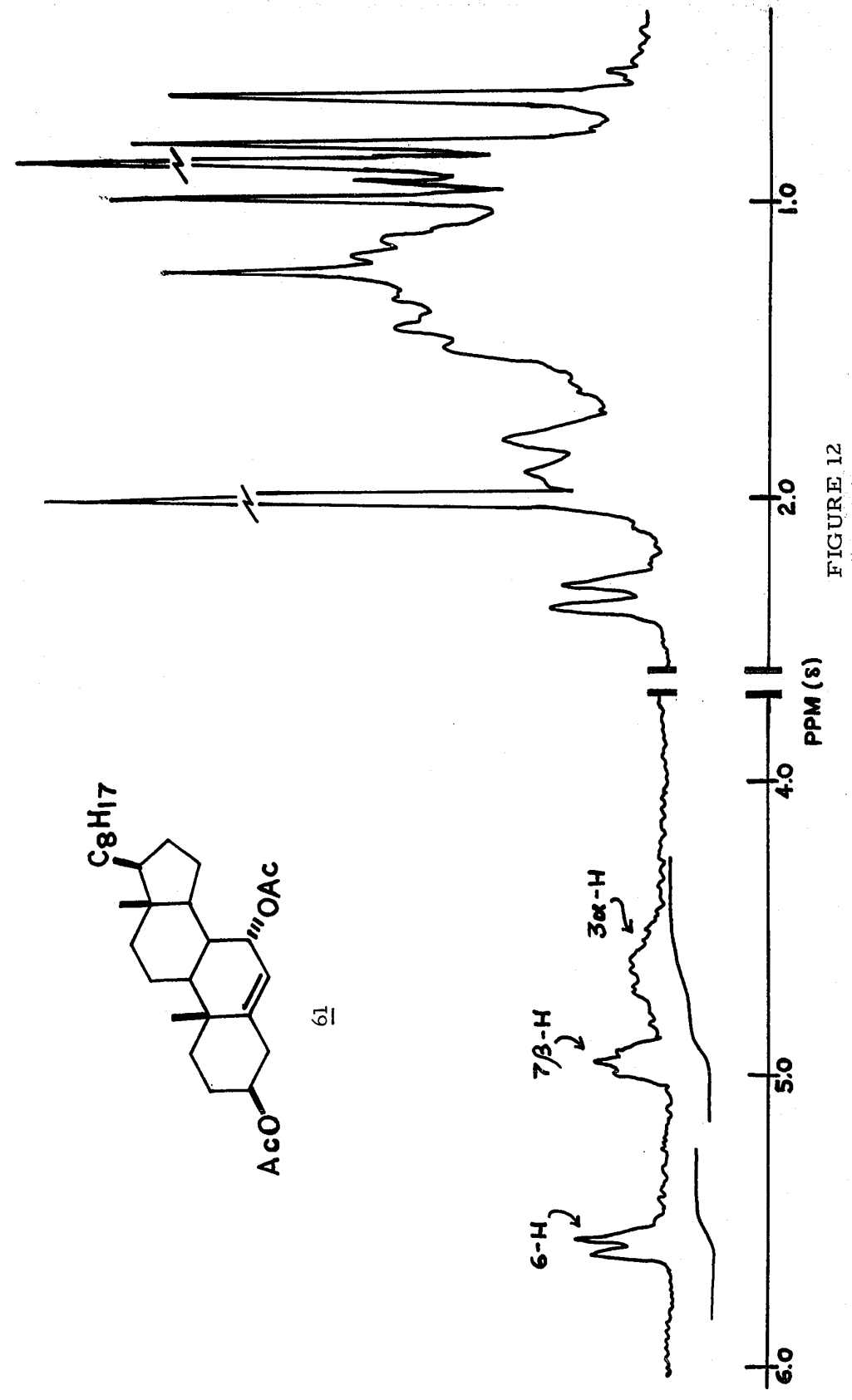
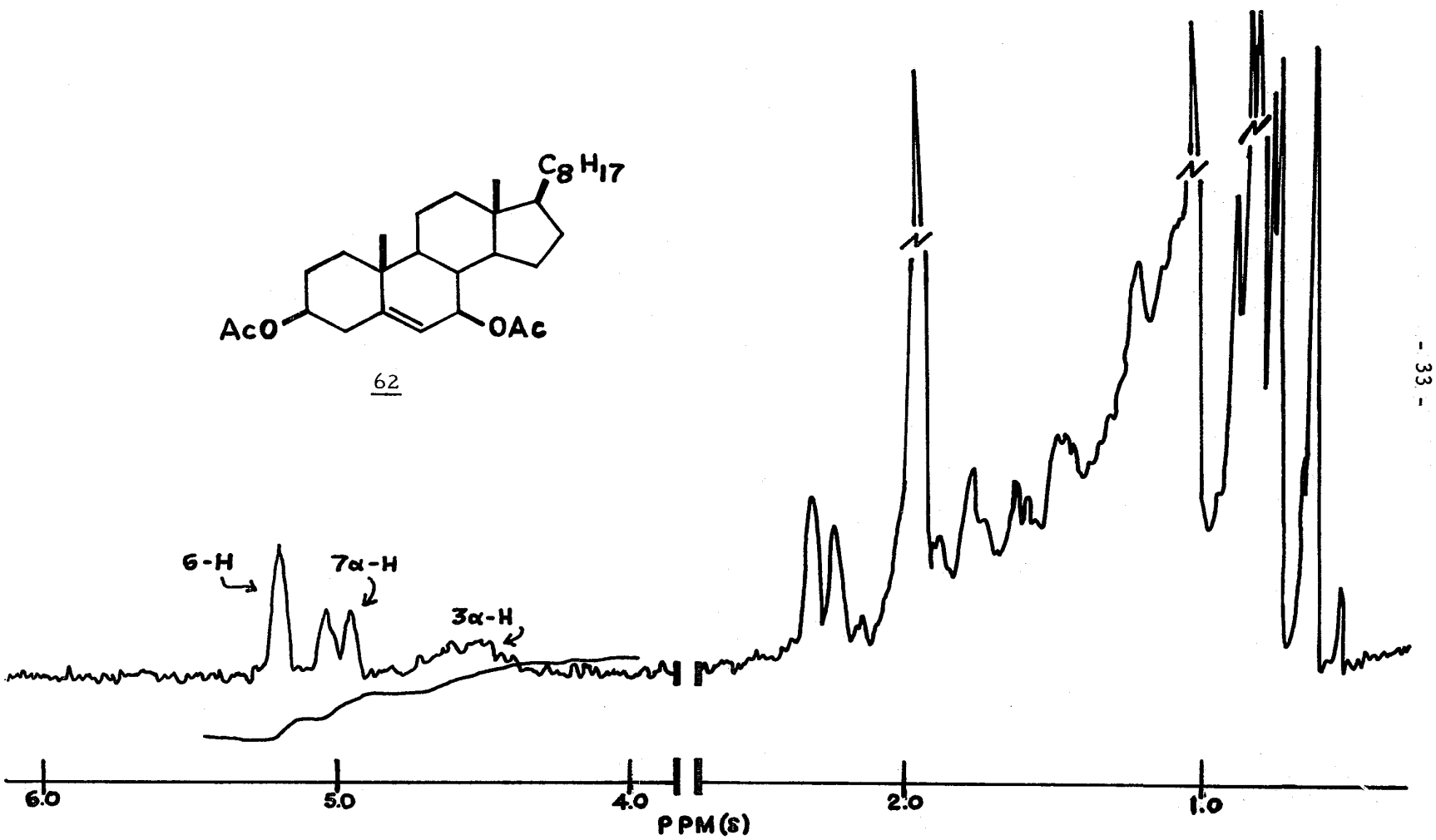
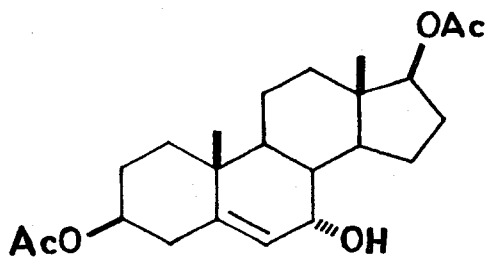



FIGURE 12




62

FIGURE 13




65




FIGURE 14

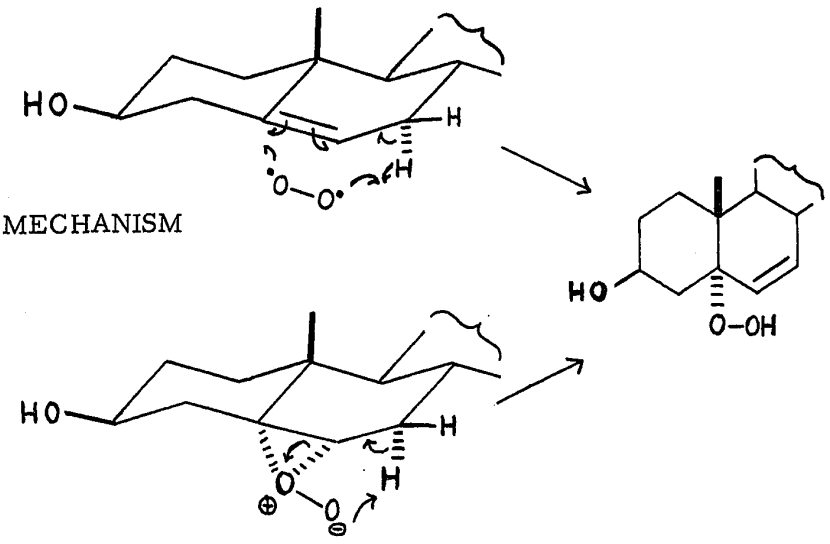
## 4. Other Approaches to the Synthesis of 7α,19-Disubstituted-$\Delta^5$-Steroids

The discovery[63] that 7-dehydrocholesterol was the provitamin $D_3$ stimulated various workers to develop an efficient synthesis of this compound. The obvious approaches of introducing into cholesterol 7 C-7 substituents which could subsequently be eliminated to yield the 5,7-diene system, have been reviewed by Fieser and Fieser[64]. In many of these approaches 7α-hydroxy-$\Delta^5$-steroids were isolated, but only as minor products. Furthermore the methods used to introduce C-7 substituents were non-stereospecific and resulted in epimeric products which, based on our own observation (SECTION 2) and those of other workers[53, 56b, 65] are often only partially separable. Thus, apart from our own work, the stereospecific introduction of 7α-hydroxy or 7α-ester groups into $\Delta^5$-steroids in good yield has at least one other important application.

### (a) Photosensitized oxygenations

When $\Delta^5$-steroids are irradiated in the presence of oxygen and a sensitizer (e.g. hematoporphyrin, methylene blue), a 5α-hydroperoxide is formed with concomitant shift of the double bond to the 6,7-position[57a]. Much work has been done to elucidate the mechanism of this reaction. Nickon and Bagli[57c] have studied the stereochemistry of the reaction by subjecting 7α- and 7β-deuterated cholesterol 7 to this photooxygenation. The product from the 7α-deuterated compound had retained only 8.5% of deuterium while that from the 7β-deuterated compound retained 95% of the deuterium label. It was concluded that the

reaction occurs stereospecifically on the α-side and since the bond which is broken and the bond which is formed are cis to each other, a cyclic mechanism has been postulated. Fenical and co-workers[66] have stated that allylic hydroperoxides are not formed via a concerted "ene" mechanism but probably through a perepoxy intermediate. However, regardless of what the transition state may be, in the case of $\Delta^5$-steroids the reaction proceeds stereospecifically on the α-face of the molecule (Figure 15).




"ENE" MECHANISM

PEREPOXIDE

FIGURE 15

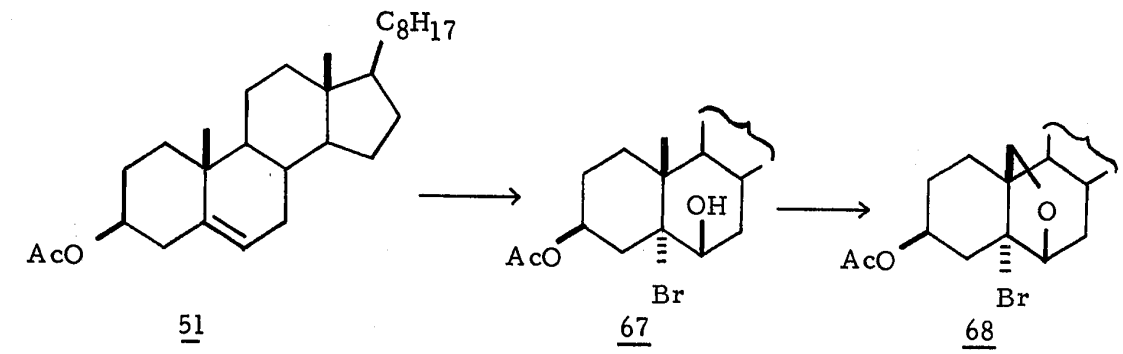
In some cases (e.g. $\Delta^4$-cholestenes) Nickon and co-workers[67a, b, c] have isolated products in which the double bond has apparently not shifted. This has been attributed to an allylic rearrangement of the initially formed hydroperoxide, thus restoring the double bond to its original position. In fact Schenck and co-workers[57b] have shown that the 5α-hydroperoxides of several $\Delta^6$-steroids (prepared by photooxygenation of $\Delta^5$-steroids) when left standing in chloroform solution, will rearrange to the 7α-hydroperoxides with concomitant shift of the double bond to the 5, 6-position. This allylic rearrangement was found[57b] to be accelerated by irradiation, benzoyl-peroxide and copper halides, although with such treatment the products isolated were the $\Delta^5$-7-keto derivatives, which were probably formed by breakdown of the 7α-hydroperoxides. Acids did not accelerate the rearrangement. Consequently Schenck visualized the rearrangement as being a free radical one. Subsequent reduction of the 7α-hydroperoxides either with Raney nickel or sodium iodide yields the 7α-hydroxy derivative.

For our purposes the above approach seemed ideal since the products have the correct stereochemistry and reasonable yields are obtained. Kaufman[68] has already shown that with 3β-acetoxy-5-cholesten-19-ol 69 , the photooxygenation reaction proceeds with the same stereospecificity as described in the above cases.

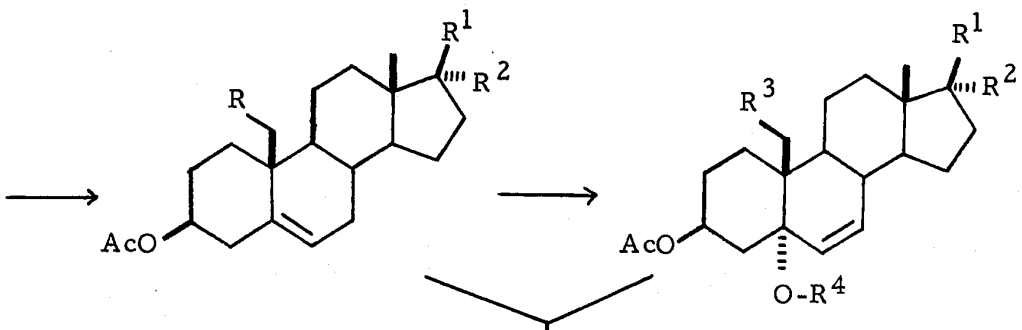
(i) Rearrangement of 3β-acetoxy-5-hydroperoxy-19-hydroxy-5α-cholest-6-ene 70

3β-Acetoxy-5-cholesten-19-ol 69 (SCHEME 3) was prepared from 3β-acetoxy-5-cholestene 51 using the same three step synthesis (51 → 67 → 68 → 69) described earlier for the androgens (SECTION 2). This compound was subjected to photosensitized oxygenation and the crude product was allowed to stand in chloroform solution[57b] for 24 hours. After reduction with sodium iodide and chromatography, two products besides the starting material were isolated: 3β-acetoxy-5,19-dihydroxy-5α-cholest-6-ene 71 (13%) and 3β-acetoxy-5-cholestene-7α,19-diol 72 (67%). The former compound 71 was identified by its n.m.r. spectrum and by mixed melting point with an authentic sample[68]. The latter compound 72 was characterized by n.m.r. analysis, elemental analysis and molecular rotation. The coupling constant ($J_{6,7}$) for this compound was 5.0 Hz (FIGURE 16) and this is consistent with the coupling constants for 7α-hydroxy compounds (TABLE 2). The molecular rotation (-348°) is also of the correct magnitude and sign.

The 7α- and 19-hydroxy groups were acetylated and the n.m.r. spectrum and molecular rotation of the resulting triacetate 73 compared with those of the triacetate 74 obtained by chromic acid oxidation[55], lithium aluminum hydride reduction[56b], and acetylation of 3β,19-diacetoxy-5-cholestene 75. The n.m.r. spectra of these triacetates 73 and 74 are shown in FIGURES 17 and 18 and it can be seen that the chemical shift for the 6-H is more downfield in compound 73 than in compound 74 and the coupling constant ($J_{6,7}$) for compound 73 is larger than that for compound 74. The molecular rotations for compounds 73 and 74 are -1085° and -45° respectively. These results are consistent with the correlations in TABLE 2 and thus confirm the structures assigned.

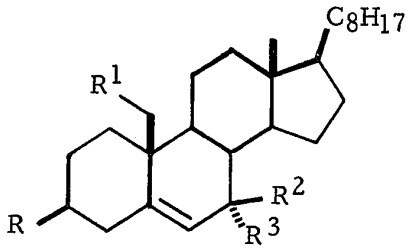



51 67 68




69 R=OH, $R^1$=$C_8H_{17}$, $R^2$=H
75 R=OAc, $R^1$=$C_8H_{17}$, $R^2$=H
37 R=OH, $R^1$, $R^2$=O
38 R=OAc, $R^1$, $R^2$=O
44 R=$R^1$=OAc, $R^2$=H

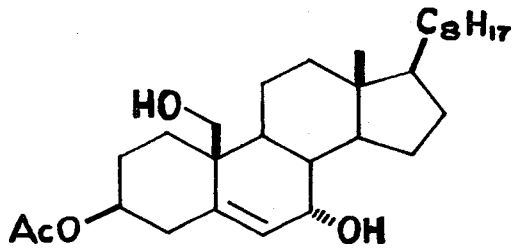
71 $R^1$=$C_8H_{17}$, $R^2$=$R^4$=H, $R^3$=OH
70 $R^1$=$C_8H_{17}$, $R^2$=H, $R^3$=$R^4$=OH
76 $R^1$, $R^2$=O, $R^3$=OH, $R^4$=H
77 $R^1$, $R^2$=O, $R^3$=OAc, $R^4$=H
78 $R^1$=$R^3$=OAc, $R^2$=$R^4$=H




72 $R^1$=$R^3$=OH, $R^2$=H, R=OAc
73 R=$R^1$=$R^3$=OAc, $R^2$=H
74 R=$R^1$=$R^2$=OAc, $R^3$=H

79 $R^1$=OH, $R^2$=H, $R^3$=OOH, R=OAc
112 R=$R^1$=OAc, $R^2$, $R^3$=O
113 R=$R^1$=$R^2$=OH, $R^3$=H

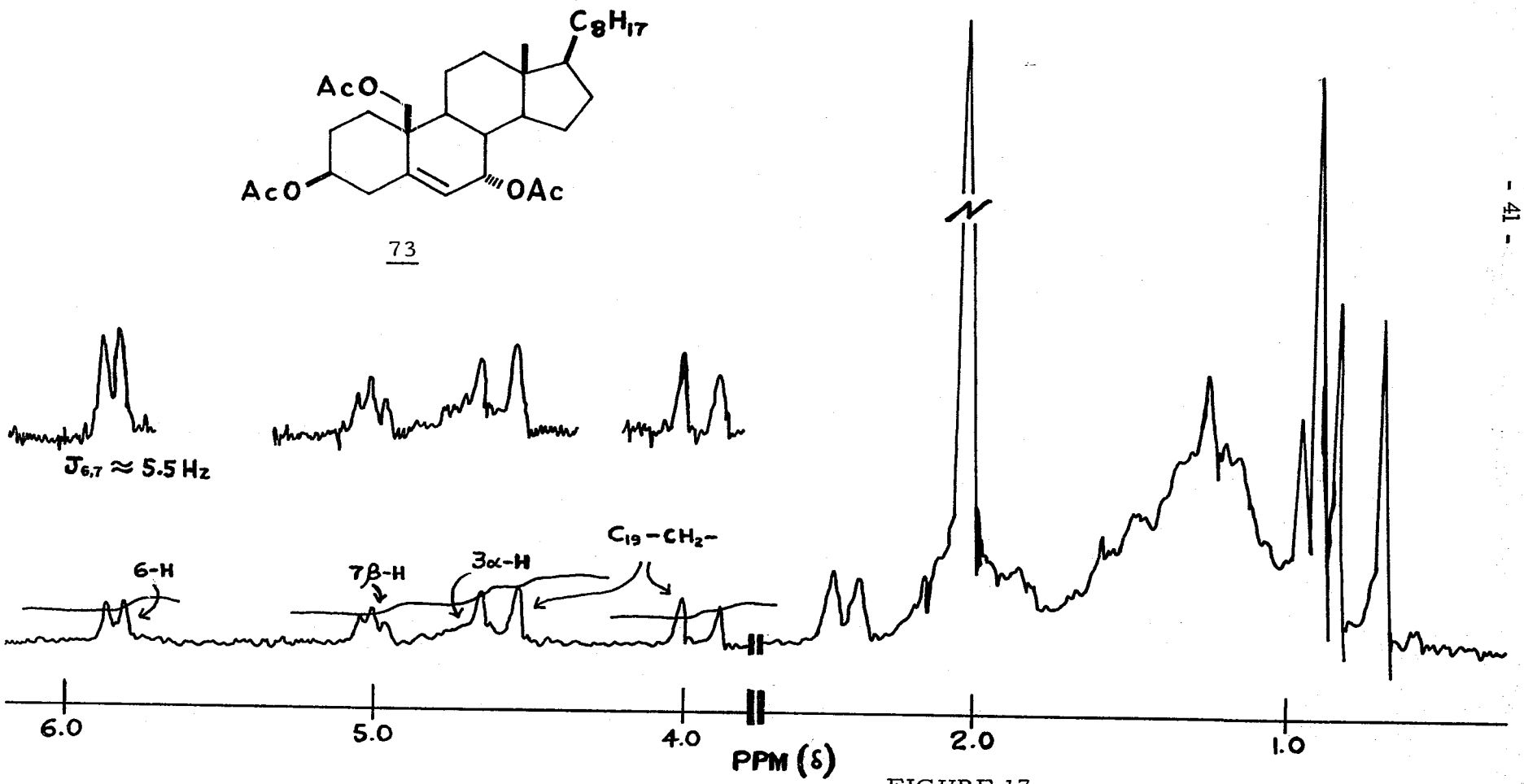
SCHEME 3




72




FIGURE 16


$C_8H_{17}$
AcO
AcO
OAc
$J_{6,7} \approx 5.5$ Hz
6-H
7β-H
3α-H
$C_{19}-CH_2-$
6.0
5.0
4.0
PPM (δ)
2.0
1.0

73

FIGURE 17

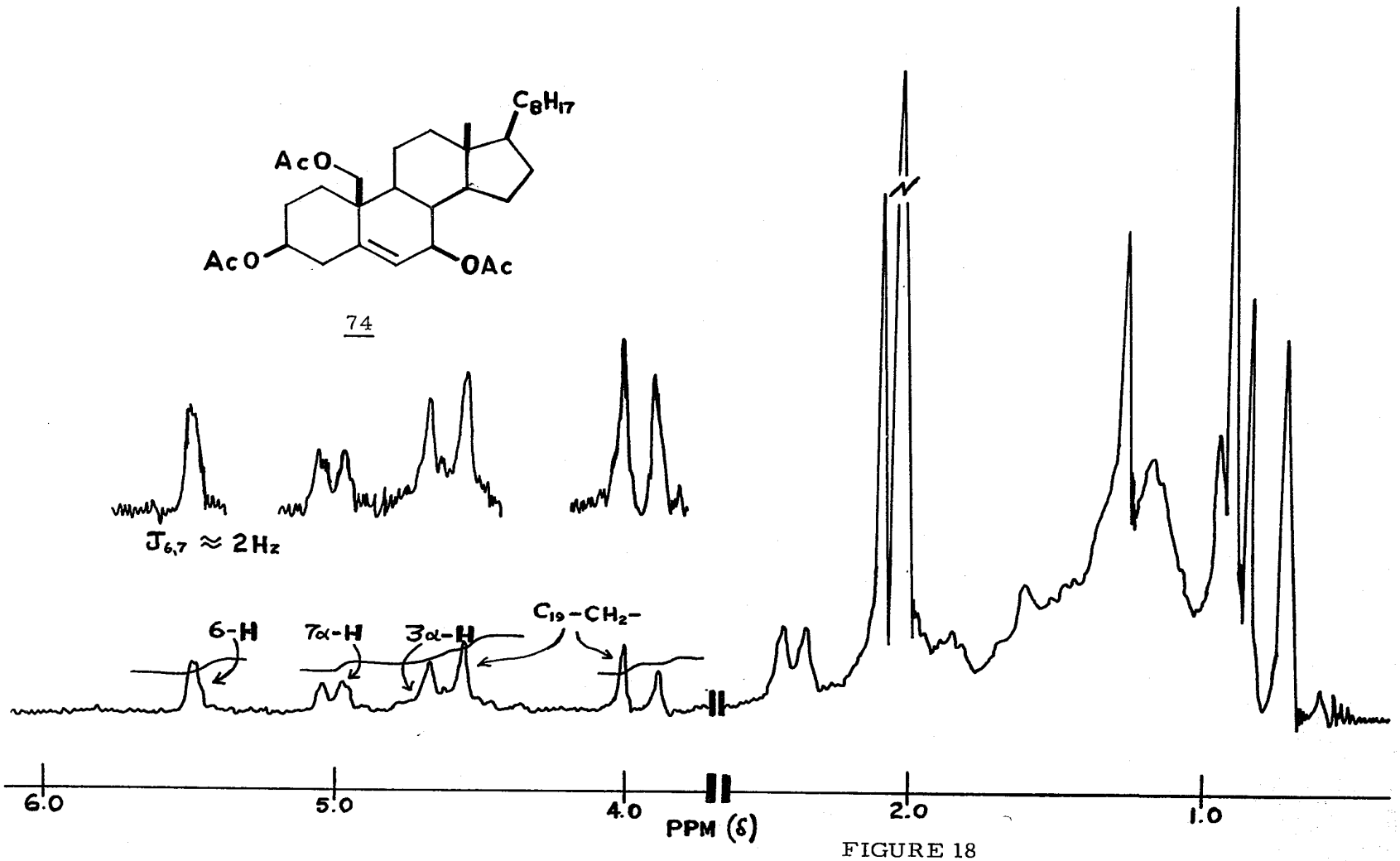

C8H17
AcO
AcO
OAc
74
J6,7 ≈ 2Hz
6-H
7α-H
3α-H
C19-CH2-
6.0
5.0
4.0
PPM (δ)
2.0
1.0

FIGURE 18

(ii) Attempted rearrangement of 5α-hydroperoxy-19-substituted-$\Delta^6$-androgens

The success encountered with the model compound 69 was encouraging and the synthetic route used for it was immediately applied to 19-substituted androgens. However, when 3β-acetoxy-19-hydroxy-5-androsten-17-one 37, 3β,19-diacetoxy-5-androsten-17-one 38 and 3β,-17β,19-triacetoxy-5-androstene 44 were photooxygenated and allowed to stand in chloroform, only the corresponding 5α-hydroxy compounds 76, 77 and 78 (SCHEME 3) were obtained after the reduction step, indicating that rearrangement had not taken place in the chloroform solution. Consequently several other solvents were substituted for chloroform, e.g. methanol, acetone and tetrahydrofuran, but with no change in the results. The addition of a few drops of HCl to the solution of hydroperoxide using chloroform, methanol and tetrahydrofuran as solvents was also ineffective. These results are partially consistent with the observations of Lythgoe and Trippett[69] who found that 3β-hydroxy-5-hydroperoxy-5α-cholest-6-ene 52* would rearrange only in chloroform and not in other solvents such as pyridine, benzene or tetrahydrofuran. In their case a trace of p-toluene-sulphonic acid in the tetrahydrofuran was also ineffective as a system for rearrangement of the 5α-hydroperoxide 52. Finally, after adding a trace of potassium cyanide to the chloroform solution of the 5α-hydroperoxide derivative of our 19-substituted androgen, there was, after a few days, evidence (t.l.c.) for a small amount of rearranged material: however, chromatography afforded only 5α-hydroxy compounds.

* SCHEME 2, p. 29.

TABLE 3

| Starting Material | Product Composition Ratio A | B | C | D |
|---|---|---|---|---|
| 3β-Acetoxy-5-cholestene 51 | 1.0 | 1.0 | 1.5 | 0.0 |
| 3β-Acetoxy-5-cholesten-19-ol 69 | 2.8 | 1.0 | 5.1 | 0.0 |
| 3β-Hydroxy-5-androsten-17-one 8 | 8.0 | 1.6 | 1.7 | 1.0 |
| 3β-Acetoxy-5-androsten-17-one 34 | 2.5 | 2.3 | 1.0 | 0.0 |
| 3β,17β-Diacetoxy-5-androstene 40 | 7.7 | 7.0 | 2.0 | 1.0 |
| 3β-Acetoxy-19-hydroxy-5-androsten-17-one 37 | 1.0 | 1.0 | 0.0 | 0.0 |
| 3β,19-Diacetoxy-5-androsten-17-one 38 | 1.5 | 1.0 | 0.0 | 0.0 |
| 3β,17β,19-Triacetoxy-5-androstene 44 | 1.5 | 1.0 | 0.0 | 0.0 |

A starting material

B $\Delta^6$-5α-hydroxy derivative

C $\Delta^5$-7α-hydroxy derivative

D $\Delta^5$-7-ketone

We have already seen that 3β-hydroxy-5-hydroperoxy-5α-cholest-6-ene 52 and the corresponding 3β-acetoxy compound 53, as well as several 5α-hydroperoxy-$\Delta^6$-androgens unsubstituted at C-19, do in fact rearrange to the 7α-hydroperoxy derivatives (SECTION 3). The failure of the 5α-hydroperoxy derivatives of C-19 functionalized androgens to rearrange was most perplexing* especially in view of the fact that 3β-acetoxy-5-hydroperoxy-19-hydroxy-5α-cholest-6-ene 70 rearranges almost completely to the 7α-hydroperoxide 79. As yet no satisfactory explanation can be given for this phenomenon but it has been generally observed by us that the rearrangements are more complete in the cases where derivatives of cholesterol 7 are used than in the case where androgens are used (TABLE 3), which perhaps implies very subtle long range effects.

(b) Peracid treatment of 3β,7α-diacetoxy-$\Delta^5$-steroids

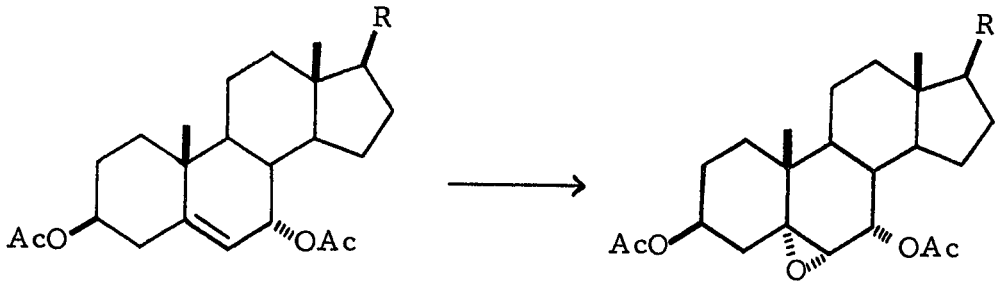
Whether or not a substituted C-19 in the case of the androgens was an influencing factor in the failure of the 5α-hydroperoxides to rearrange is still uncertain. The problem can of course be avoided by rearranging a 5α-hydroperoxide before functionalizing the C-19 position. It is known[70] that peracid treatment of $\Delta^5$-steroids leads to 5α,6α- and 5β,6β-epoxides, the latter being formed in lower yield due to sterically controlled attack of the reagent. If, by the introduction of a 7α-acetoxy group, the α-face becomes more hindered to peracid attack, then the yield of 5β,6β-epoxide might be considerably increased. This 5β,6β-epoxide could then be opened with HBr to yield a 5α-bromo-6β-hydroxy derivative which in turn could be used to functionalize C-19 by the sequence of reactions previously described.

* In a private communication (10th January, 1972), Dr. D.N. Kirk has informed us that in his laboratory the rearrangement of analogous compounds in chloroform was not a reliable reaction.

In 1936 Heilbron and co-workers[60], by epoxidation of 3β, 7α-dihydroxy-5-cholestene 66 and subsequent acetylation of the product, reported the isolation of only one epoxide with melting point 203 - 204°. That this compound was the 5α, 6α-epoxide may be rationalized on the basis of the observations of Henbest and Wilson[71], who showed that allylic alcohols are epoxidized by peracids giving products in which the epoxide and the hydroxy groups are cis to one another. The above authors have proposed that the hydroxy group exerts a promoting effect on the reaction by virtue of a hydrogen-bonded transition state. Other workers, notably Mousseron et al[72a, b], Morand et al[73] and Cross et al[74] have extended this reaction to homoallylic alcohols and have observed the same directive effect of the hydroxy group on the peracid. There are cases[74], however, where steric factors make the hydrogen-bonded transition state unfavorable, resulting in an increased amount of product in which the hydroxy group and the epoxide are trans to one another.

On the other hand, the reaction of allylic and homoallylic acetates with peracids does not involve complexing of substrate and reagent[71, 72a, b, 74]. Instead, the acetate group sterically hinders the approach of the peracid and consequently the products of epoxidation in these cases are rich in the trans acetate-epoxide. Here again other steric factors can vary considerably the ratio of cis to trans epoxides.

It seemed worthwhile to attempt the epoxidation of a $\Delta^5$-7α-acetoxy steroid and to determine whether the acetate group would have enough steric effect to overcome the steric hindrance of the 10β-methyl group which normally forces reagents to attack predominantly the α-face of the steroid molecule. When 3β, 7α-diacetoxy-5-cholestene 61 (SCHEME 4) was treated with monoperphthalic acid

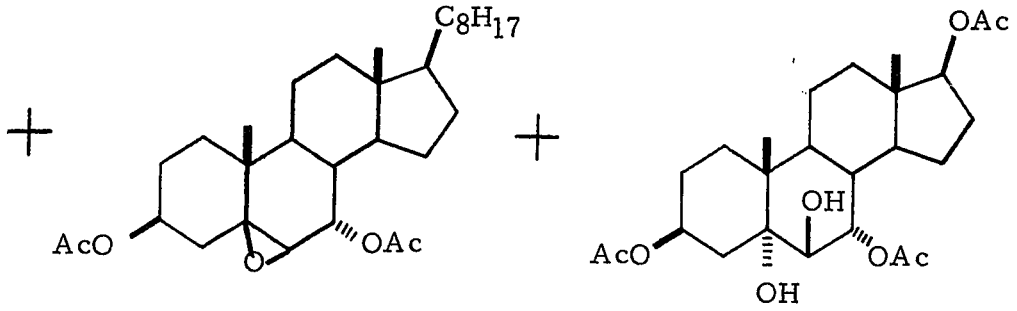



61 R=$C_8H_{17}$

58 R=OAc

80 R=$C_8H_{17}$

82 R=OAc




81

83

SCHEME 4

in ether at 5°, two epoxides 80 and 81 were isolated after chromatography, in a 2:1 ratio. The n.m.r. spectrum of the low yield epoxide 81 showed a doublet at $\delta$ 3.1 with a coupling constant ($J_{6,7}$) of ca. 3.6 Hz while the spectrum of the high-yield epoxide 80 showed a doublet at $\delta$ 3.35 with a coupling constant of ca. 4.5 Hz.

From measurements on Dreiding models Cross[75] has determined that the dihedral angle ($\phi$) subtended by the epoxidic C-H bond and the C-$H_{7\beta}$ bond is 49 ± 4° for the 5β,6β-epoxide and 28 ± 4° for the 5α,6α-epoxide and therefore the α-epoxide should have the larger coupling constant[62]. In addition Cross[75] has observed that in 5,6-epoxides the 6β-proton signal (α-epoxide) occurs at higher field than the signal for the 6α-proton (β-epoxide). However, in comparing our data with the above correlations we find that neither of the compounds has the correct values for both the coupling constant and the chemical shift to fit either a 5α,6α- or a 5β,6β-configuration. The problem of assigning the configuration of these epoxides was settled by optical rotations. It is known[76a,b,c] that 5α,6α-epoxides are more levorotatory than their 5β,6β-isomers. Unfortunately the high yield compound was the more levorotatory of the two, and is thus the undesired 5α,6α-epoxide 80*.

When 3β,7α,17β-triacetoxy-5-androstene 58 was treated with monoperphthalic acid at room temperature, 3β,7α,17β-triacetoxy-5,6α-oxido-5α-androstane 82** and 3β,7α,17β-triacetoxy-5,6β-dihydroxy-5α-androstane 83 were obtained. The latter compound is the result of acid opening of the epoxide and subsequent hydrolysis of the phthalate ester during workup with sodium bicarbonate solution.

---

* This assignment of configuration is substantiated by the fact that compound 80 has a melting point of 203-204° which is identical to that of the epoxide obtained by Heilbron et al[60]. (p. 46.)

** On the basis of the negative rotation and the n.m.r. data, this compound was assigned a 5α,6α-configuration.

(c) $\Delta^5$-7-keto-Steroids as starting materials

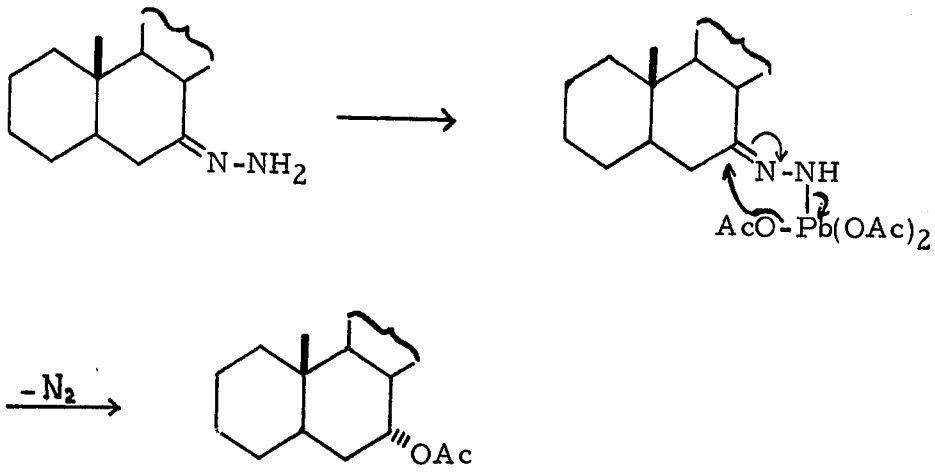
As indicated earlier, $\Delta^5$-7-keto-steroids yield 7β-hydroxy derivatives upon metal hydride reduction. Nevertheless there are indirect methods of obtaining the 7α-hydroxy-derivatives from the ketone. For example, Shoppee and co-workers[77] converted steroidal 7-ketones to the oximes, which were then reduced either by catalytic hydrogenation or by sodium in ethanol, yielding the 7α- and 7β-amino derivatives. Diazotization of these amino-compounds yields the corresponding* hydroxy compounds.

The use of lead tetraacetate for the allylicoxidation of a $\Delta^5$-steroid has been reported[65] to be non-stereospecific, yielding an epimeric mixture of 7-acetoxy-derivatives (3:2, 7β:7α) in 50% yield. Recently, Barton et al[78] have treated the hydrazone of a 7-keto B-ring-saturated steroid with lead tetraacetate at 0° in methylene dichloride have isolated the 7α-acetoxy derivative in 68% yield along with the $\Delta^7$-derivative in 20% yield. No 7β-substituted product was found. These workers have discussed two distinct pathways which could explain the products of the reaction. Pathway A (FIGURE 19) involves a cyclic intermediate which, because of its steric requirements undergoes further reaction only on the α-face of the molecule. This, however, is not consistent with the results of Debono et al[79] who obtained 3α- and 3β-acetoxy derivatives upon treating the hydrazone of a 3-keto-saturated ring-A-steroid with lead tetraacetate. The partial flexibility of the A-ring might explain the contrast in behavior between the hydrazones of

---

* The hydroxy compounds are formed with retention of configuration but in the case of the 7α-amino steroids the major products (~75%) from the diazotizations are $\Delta^7$-derivatives (elimination products) and only a small amount of 7α-hydroxy compounds was obtained.

PATHWAY A




PATHWAY B

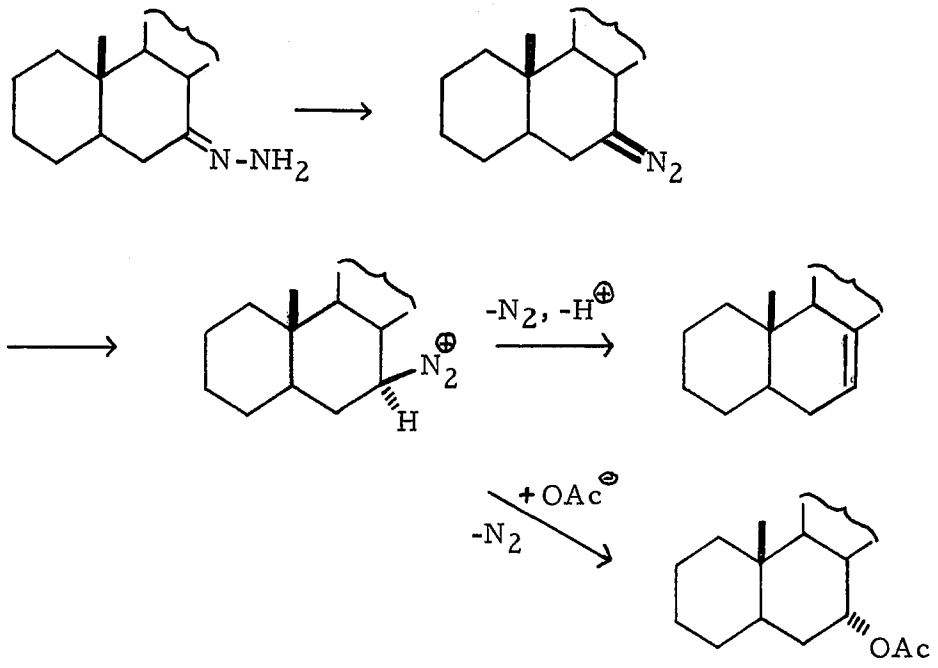
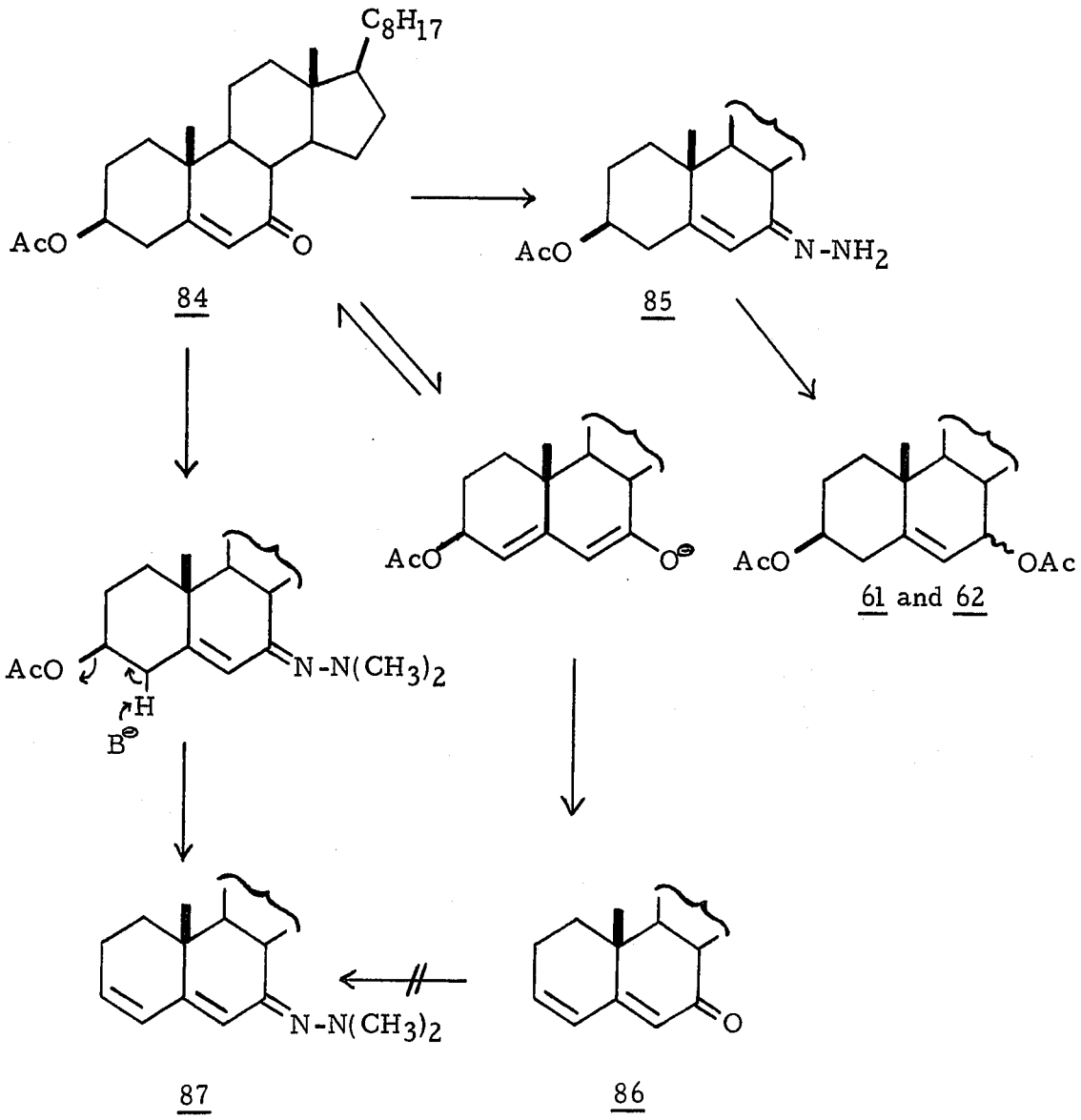



FIGURE 19

3-keto and 7-keto steroids. Pathway B involves preliminary oxidation of the hydrazone to a diazo compound. Subsequent α-protonation yields the diazonium ion which could then either lose nitrogen and a proton to yield the olefin or could be attacked by acetate ion to yield the 7α-acetoxy compound. Here again there is an inconsistency with the earlier observations of Shoppee et al[77] who found retention of configuration in the products derived from diazotization of 7β-amino B-ring-saturated steroids. Barton and co-workers[78], with reservations, favour Pathway B.

Since we wished to apply this reaction to the hydrazone of a $\Delta^5$-7-keto steroid we attempted to prepare the hydrazone of 3β-acetoxy-5-cholesten-7-one 84 (SCHEME 5) by refluxing the latter in ethanol with excess hydrazine hydrate. The yield of hydrazone 85, however, was poor ( $<$ 40%) and consequently it was decided to attempt improvement of the yield of hydrazone by a method[80] which involves the initial preparation of an N,N-dimethylhydrazone derivative followed by an exchange reaction with anhydrous hydrazine.

However, treatment of 3β-acetoxy-5-cholesten-7-one 84 with excess N,N-dimethylhydrazine in refluxing ethanol yielded products other than the expected hydrazone. Instead, the only products isolated were $\Delta^{3,5}$-compounds, the 3β-acetate group having been eliminated from the starting material 84. One of the products was identified by n.m.r. and melting point as 3,5-cholestadien-7-one 86 (SCHEME 5) and the other diene was identified by n.m.r. and elemental analysis as 3,5-cholestadien-7-N,N-dimethylhydrazone 87.


$C_8H_{17}$
AcO
O
84
AcO
N-NH$_2$
85
AcO
O$^{\ominus}$
AcO
OAc
61 and 62
AcO
N-N(CH$_3$)$_2$
H
B$^{\ominus}$
N-N(CH$_3$)$_2$
87
O
86

SCHEME 5

The results of varying the ratio of steroid to hydrazine are shown in TABLE 4. Although this reaction was not studied in any more detail it can be seen that increasing the proportion of hydrazine results in a more complete reaction. At the same time the amount of dienone 86 decreases while the proportion of hydrazone 87 increases. Also, the dienone 86, when refluxed with excess hydrazine, failed to react.

TABLE 4

| Steroid:N,N-dimethyl-hydrazine (moles) : (moles) | % Products (approx.) | | |
|---|---|---|---|
| | S.M. 84 | $\Delta^{3,5}$-7-ketone 86 | $\Delta^{3,5}$-7-hydrazone 87 |
| 1:3 | 64 | 27 | 9 |
| 1:5 | 20 | 20* | 60 |
| 1:60 | 10 | 10 | 80 |

The above results allow only a tentative proposal for the mechanism(s) involved in these reactions, the rationale of which is as follows.

The ketone 84 under the basic reaction conditions can be transformed into its enolate anion which could either revert back to starting material or could displace acetate ion** to yield the dienone 86 as shown. Since the hydrazone 87 does not form from dienone 86 it must be formed through an intermediate such as is shown in SCHEME 5. With excess hydrazine this intermediate is probably not stable and eliminates acetic acid, perhaps by a similar process as that which leads to the dienone 86.

* This value is an estimated one because a quantity of product was lost during the chromatography.

** Such eliminations have been observed when 3β-acetoxy-$\Delta^5$-7-keto steroids are treated with hot methanolic KOH[81].

The fact that there is more than one process occurring in these reactions, could account for the low yield of hydrazone 85 obtained when using hydrazine hydrate as reagent. However, any further statement on the mechanisms operative in this reaction must await additional studies, including the isolation and identification of some of the other products which are formed.

The reaction of lead tetraacetate with the hydrazone 85 resulted in formation of a 1:1 mixture (n.m.r. analysis) of epimeric 7-acetates, 61 and 62, in 85% yield. In contrast to the results of Barton et al[78] who obtained only the 7α-acetoxy derivative with a B-ring saturated steroid, our results are more in accord with those of Debono et al[79] who obtained epimeric mixtures. Our results with a compound having a conjugated double bond do not lend any weight to either of the mechanisms discussed earlier (FIGURE 19). The absence of a 6β-hydrogen in our case would lessen the steric hindrance on the β-side of the molecule allowing a cyclic intermediate (Pathway A) to undergo further reaction on both the α- and β-faces of the steroid molecule. On the other hand, the presence of a 5,6-double bond could stabilize a carbonium ion (Pathway B) which may be attacked on the re or si face. In any event, since this reaction was not stereospecific for $\Delta^5$-steroids, it was inappropriate for our purpose and no further experiments were undertaken.

(d) Rearrangement of 5α-Hydroxy-$\Delta^6$-Steroids

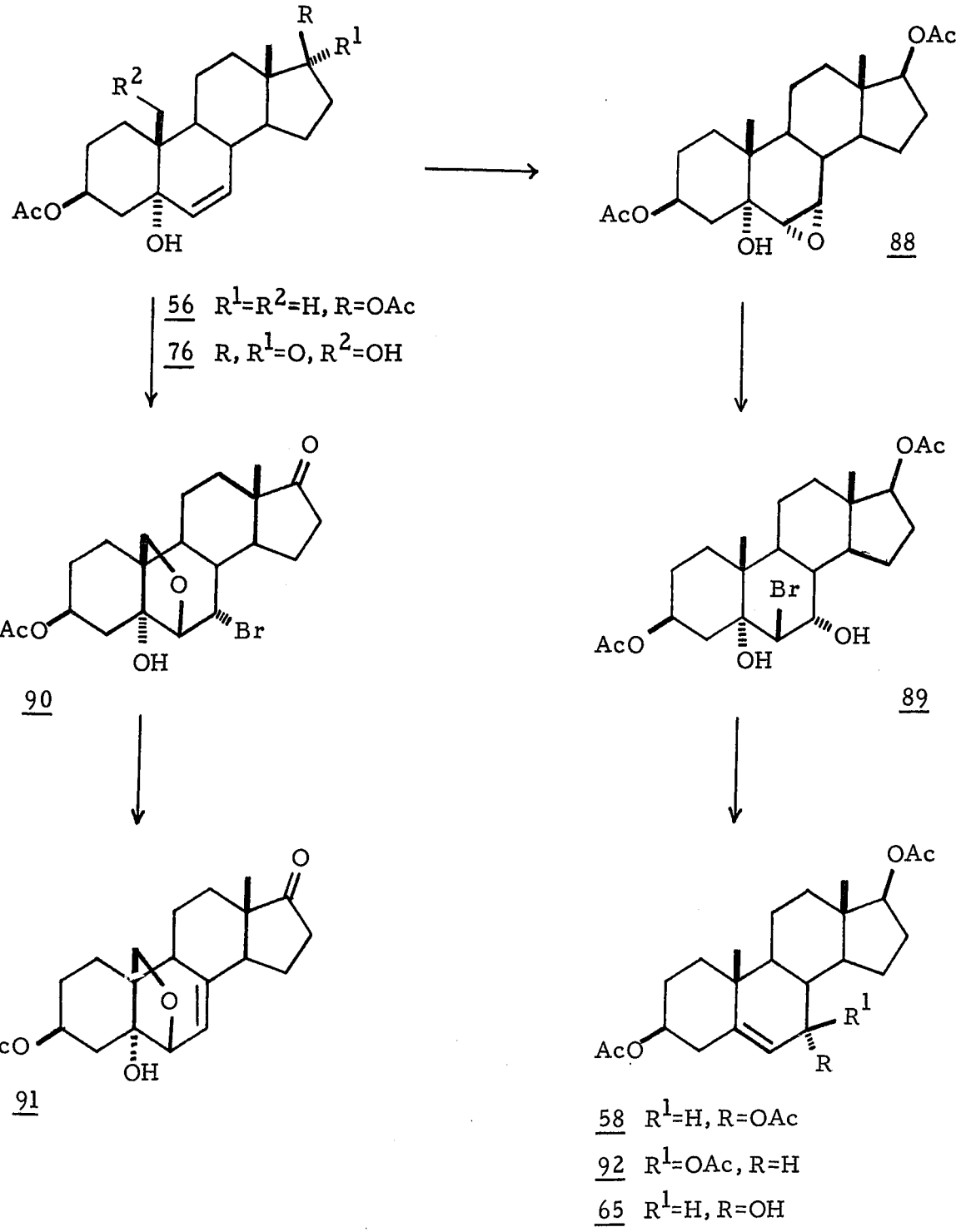
Since the photosensitized oxygenation reactions may result in the isolation of large amounts of 5α-hydroxy compounds* (TABLE 3) it seemed to be in order to attempt to transform them into their 7α-hydroxy isomers. In order to accomplish this transform ation it was decided to make use of the orienting effect of the hydroxy group on peracids [ SECTION 4 (b)]. If the peracid treatment of a 5α-hydroxy-$\Delta^6$-steroid leads exclusively to the 6α, 7α-epoxy derivative, then opening of the epoxide with HBr should yield a 5α, 7α-dihydroxy-6β-bromo-derivative, due to diaxial opening[82] of the epoxide. The bromo-derivative could then be reduced with zinc in acetic acid to yield either the starting material or the 7α-hydroxy-$\Delta^5$-isomer, hopefully the latter.

When 3β,17β-diacetoxy-5-hydroxy-5α-androst-6-ene 56 (SCHEME 6) was treated with monoperphthalic acid, only one epoxide product was isolated in a yield of 80%. This product was expected to be the 6α, 7α-isomer.

An examination of Dreiding models reveals that the dihedral angle (Ø) subtended by the C-H bonds at C-7 and C-8 in a 6α, 7α-epoxide is ca. 50°,and ca. 95° in the β-isomer. Thus in the latter case there should be virtually no coupling between the 7α-H and the 8β-H and therefore the 7α-H has adjacent coupling only with the 6α-H. (Since in our case the 5α-position is occupied by a hydroxy group, the epoxidic proton at C-6 has only one adjacent coupling proton at C-7). The signals in the n.m.r. spectrum for a 6β, 7β-epoxide of this type should thus exhibit a symmetrical pattern (analogous to that for an 11α, 12α-epoxide[83a]). The spectrum of the epoxide 88 shows a distorted multiplet in the range δ 2.80 - 3.00 which supports a 6α, 7α-configuration for the epoxide function.

---

* These compounds can also be considered as potential estrogen precursors.


R
R1
R2
AcO
OH
OAc
O
88
56 R1=R2=H, R=OAc
76 R, R1=O, R2=OH
Br
90
89
91
58 R1=H, R=OAc
92 R1=OAc, R=H
65 R1=H, R=OH

SCHEME 6

When the epoxide 88 was opened with HBr in chloroform a bromohydrin 89 was obtained. If the epoxide 88 was indeed the 6α, 7α-isomer then the bromohydrin 89 should have a 6β-bromo-5α, 7α-dihydroxy configuration, provided, of course, that diaxial opening of the epoxide had occurred. On the other hand a 6β, 7β-epoxide would yield the 5α, 6β-dihydroxy-7α-bromo isomer. It was originally thought that treatment of the bromohydrin 89 with zinc in acetic acid would provide the answer to this problem of configurational assignment since a 6β-bromo-5α, 7α-dihydroxy compound could potentially yield two products (i.e. a $\Delta^5$-7α-hydroxy and a $\Delta^6$-5α-hydroxy derivative) while a 5α, 6β-dihydroxy-7α-bromo compound could yield only one product (the $\Delta^6$-5α-hydroxy derivative). However, as will be seen later, under the reaction conditions used, $\Delta^6$-5α-hydroxy compounds rearrange to their $\Delta^5$-7α-hydroxy isomers, and therefore this approach was not valid as a means of assigning the configuration of the bromohydrin 89. It was thus decided to prepare a compound of known configuration featuring the functional groups in question. A comparison of the n.m.r. spectrum of this compound with that of the bromohydrin 89 should then resolve the problem. Such a compound was prepared in the following manner. 3β-Acetoxy-5, 19-dihydroxy-5α-androst-6-en-17-one 76 was treated with N-bromosuccinimide in t-butanol[83b]. The bromo-oxide 90 formed could either have the 6β-oxido-7α-bromo structure or the 6α-bromo-7β-oxido structure, assuming, of course, that the intermediate bromonium ion is formed by steric approach control of the reagent on the less hindered α-face of the molecule. That the former structure* is the correct one was shown by dehydrobromination of the bromo-oxide 90 with lithium carbonate and lithium chloride in N, N-dimethylformamide, which afforded an olefinic derivative 91. Dehydrobromination could not have

* An argument against the latter structure is that a 7β, 19-oxido-function forces the B-ring into a boat conformation.

been achieved if the positions of the oxygen function and the bromine atom had been reversed since this would have resulted in formation of a double bond at a bridgehead carbon[84].

By comparison of the n.m.r. spectra (FIGURE 20) of the bromohydrin 89 and the bromo-oxide 90 it was concluded that the substitution at C-6 and C-7 in the two compounds was reversed in terms of the bromine and oxygen functions. A triplet at $\delta$ 4.24 in the spectrum of the latter compound 90 due to the 7β-H is absent in the spectrum of the former compound 89. Instead the spectrum of compound 89 shows overlapping signals at ca. $\delta$ 4.0. When compound 90 is dehydrobrominated the spectrum of the resulting product 91 shows an olefinic signal (doublet) at $\delta$ 5.66 and the original triplet at $\delta$ 4.24 (compound 90 ) is no longer present. The n.m.r. signals for compounds 89, 90 and 91 in the region $\delta$ 3.0 - 6.0 p.p.m. is shown in FIGURE 20. These results indicate that the bromohydrin 89 has the 6β-bromo-7α-hydroxy configuration and this in turn rules out the possibility that the epoxide 88 is the 6β, 7β-isomer.

When the bromohydrin 89 was reduced with zinc in refluxing acetic acid (80%), two products were isolated. The minor product (~10%) was identified by n.m.r. analysis as an epimeric mixture of 3β, 7ξ, 17β-triacetoxy-5-androstenes 58 and 92. The major product (~90%) was identified (n.m.r., mixed melting point) as 3β,17β-diacetoxy-7α-hydroxy-5-androstene 65. It thus appears that transformation of $\Delta^6$-5α-hydroxy compounds into $\Delta^5$-7α-hydroxy compounds may be accomplished in good yield through the three step sequence outlined in SCHEME 6.

In order to establish the origin of the minor product in this reaction, the bromohydrin 89, the $\Delta^6$-5α-hydroxy compound 56 and the $\Delta^5$-7α-hydroxy compound 65 were treated with acetic acid (80%) for 24 hours. While the bromohydrin 89 was inert under these conditions both

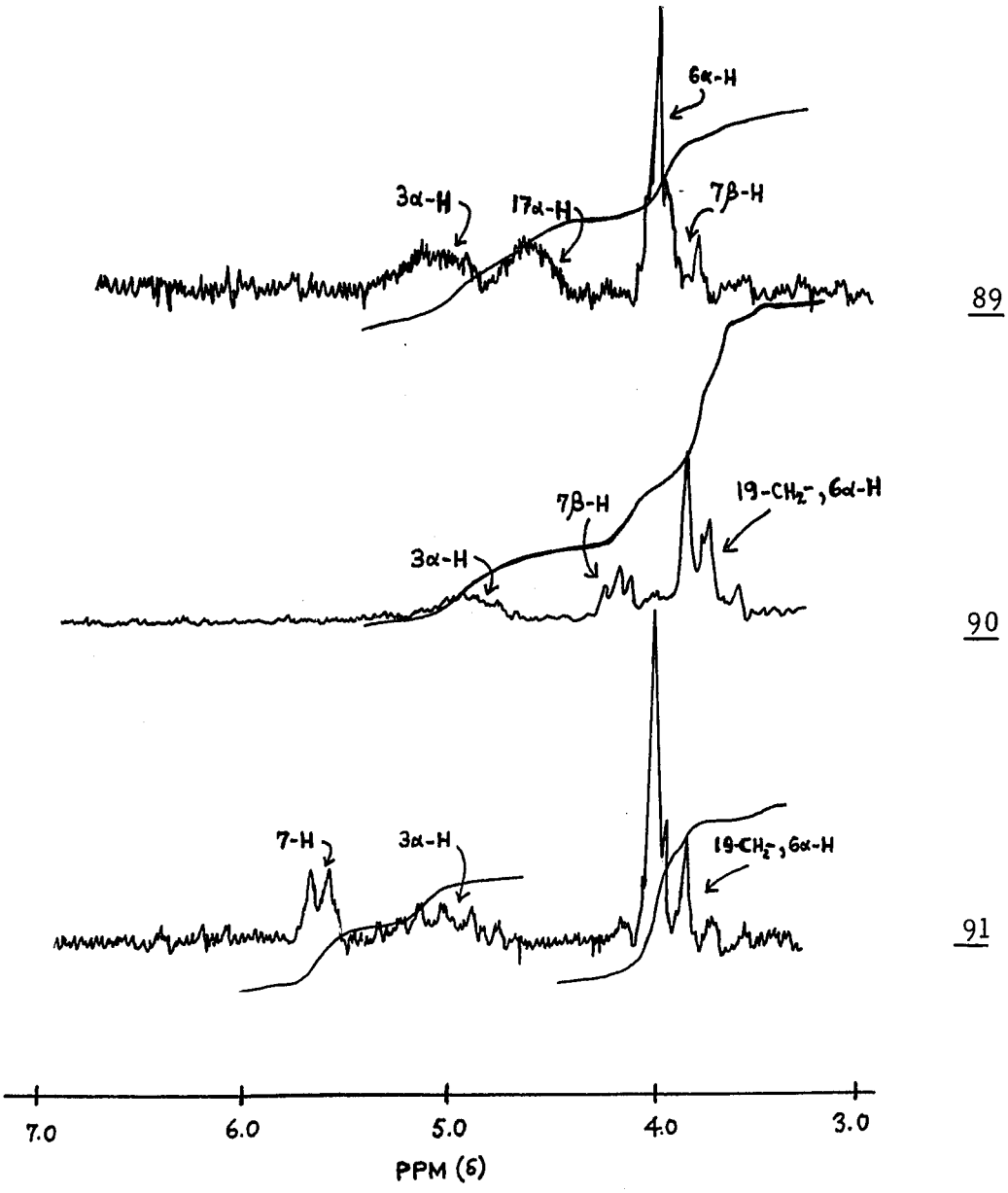

6α-H
3α-H
17α-H
7β-H
89
7β-H
19-CH₂⁻, 6α-H
3α-H
90
7-H
3α-H
19-CH₂⁻, 6α-H
91
7.0
6.0
5.0
4.0
3.0
PPM (δ)

FIGURE 20

hydroxy compounds 56 and 65 afforded 7ξ-acetoxy derivatives. Of special interest, however, was the fact that the 5α-hydroxy compound 56 had completely disappeared and there was evidence (t.l.c.) that it had given rise to some 7-hydroxy derivative. There was no evidence (t.l.c.) that the 7α-hydroxy compound 65 had been converted to the 5α-hydroxy isomer 56. Apparently acid catalyzed allylic rearrangement had occurred in the case of the 5α-hydroxy compound 56. In view of our own work and because of the potential that such mild acid allylic-alcohol rearrangements could have in synthetic chemistry, this development certainly warranted further investigation.

(e) Acid Treatment of Steroidal Allylic Alcohols

Mild acid treatment of steroidal allylic alcohols at room temperature has given indications [SECTION 4 (d)] of resulting in nucleophilic substitution reactions with little, if any, dehydration taking place. On the other hand strong acid treatment of these allylic alcohols led to dehydrated compounds. For example, 3β,5-dihydroxy-5α-cholest-6-ene 54 when treated with HCl in ether at room temperature is converted into a mixture of highly mobile (t.l.c.) compounds, which show ultra violet absorption bands characteristic of dienes and/or trienes. Schenck[85] has shown that treatment of a $\Delta^6$-3β,5α-dihydroxy steroid with HCl in chloroform leads mostly to $\Delta^{4,6}$-dienes and to $\Delta^5$-7α-chloro derivatives. A small amount of the $\Delta^{5,7}$-diene derivatives was also obtained. Such strongly acidic conditions can also lead to alkyl migration[86] as well as dehydration.

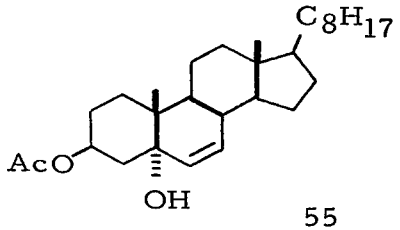
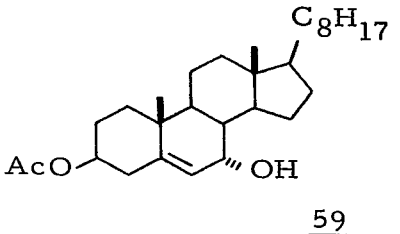
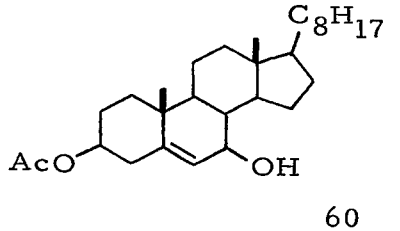
(i) Allylic rearrangement studies

Mechanisms which can be considered[87] for acid-catalyzed rearrangement of allylic alcohols fall into three categories: [a] intramolecular ($S_Ni'$); [b] unimolecular ($S_N1'$) and [c] bimolecular ($S_N2'$). TABLE 5 shows the results of mild acid treatment at room temperature of 3β-acetoxy-5-hydroxy-5α-cholest-6-ene 55. Control experiments were performed with the isomeric 3β-acetoxy-7α-hydroxy-5-cholestene 59 and 3β-acetoxy-7β-hydroxy-5-cholestene 60.

It can be seen that the 5α-hydroxy isomer 55 rearranges rapidly and after two hours has been completely converted into the corresponding 7-hydroxy and 7-acetoxy derivatives. That the 7-acetoxy derivative did not arise from a 7-hydroxy compound is clear from the control experiments which show formation of 7-acetoxy derivatives only after four and five hours respectively. After a considerable length of time ( $>$ 18 hours) all three systems exhibit similar thin layers, in all cases the amount of the unknown component A appearing to be small compared to the other products.

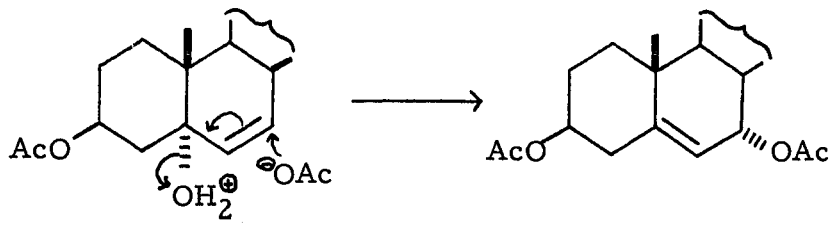
In order to determine the stereochemical course of the rearrangement, 3β-acetoxy-5-hydroxy-5α-cholest-6-ene 55 was treated with acetic acid (80%) for two hours after which the reaction was worked up and the product mixture chromatographed on SilicaR. The chromatographic separation afforded two compounds in yields of 15% and 72% which were shown (n.m.r. and mixed melting points) to be 3β, 7α-diacetoxy-5-cholestene 61 and 3β-acetoxy-7α-hydroxy-5-cholestene 59 respectively. No 7β-substituted derivative was detected. Therefore the two products appear to have arisen by stereospecific processes and thus an $S_N1'$ may be ruled out in both cases. The formation of the 7α-acetoxy derivative 61 can be adequately explained by an $S_N2'$ mechanism which is known[87] to proceed through a transition state in which the entering group and the leaving group are cis to one another (FIGURE 21).

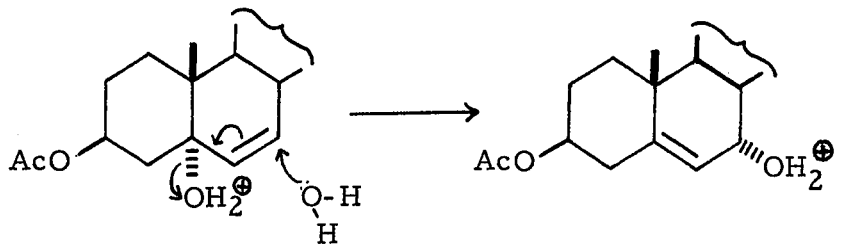
TABLE 5

| Compound | Time (hrs.) | Qualitative analysis of products by t.l.c.* | | | | | |
|---|---|---|---|---|---|---|---|
| | | 5α-OH | 7α-OH | 7β-OH | 7α-OAc | 7ξ-OAc | A |
|  55 | 0.5 | XXX | X | | X | | |
| | 1.0 | XX | XX | | X | | |
| | 2.0 | | XXXX | | X | | |
| | 3.0 | | XXXX | | X | | |
| | 18.0 | | XXXX | | X | | x |
| | 100.0 | | XXXX | | X | | x |
|  59 | 0.5 | | XXXXX | | | | |
| | 1.0 | | XXXXX | | | | |
| | 2.0 | | XXXXX | | | | |
| | 3.0 | | XXXXX | | | | |
| | 4.0 | | XXXX | | | x | |
| | 5.0 | | XXX | | | X | |
| | 18.0 | | XX | | | XX | x |
| | 100.0 | | XX | | | XXX | x |
| | | | | | | XXX | x |
|  60 | 0.5 | | | XXXXX | | | |
| | 1.0 | | | XXXXX | | | |
| | 2.0 | | | XXXXX | | | |
| | 3.0 | | | XXXXX | | | |
| | 4.0 | | | XXXXX | | x | |
| | 5.0 | | | XXXX | | X | |
| | 18.0 | | | XX | | XXX | x |
| | 100.0 | | | XX | | XXX | x |

A =unidentified compound ; * apparent relative amounts of components shown by X's. All substances exhibit an intense blue color on spraying the thin layer plates with 50% $H_2SO_4$ and estimated relative quantities were confirmed in quantitative experiments. The $R_f$ values of the various substances are given in brackets; 7α-, 7β-OH (0.46), 5α-OH (0.69), 7α, 7ξ-OAc (0.79), A (0.90). (Solvent system: ether-benzene, 1:4).

$S_N2'$




$S_Ni'$

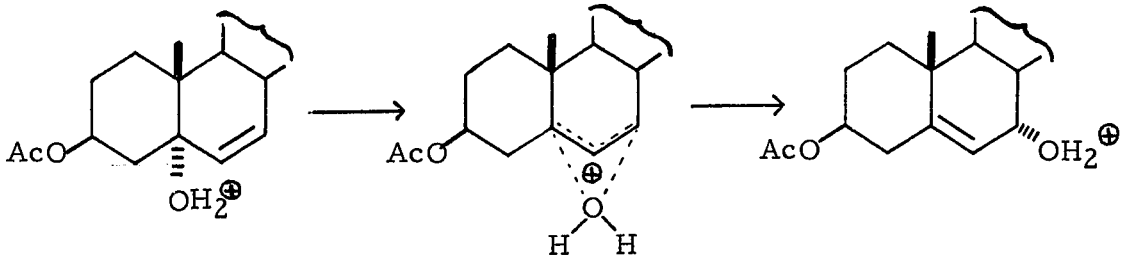



FIGURE 21

In order to determine which mechanism ($S_Ni'$ or $S_N2'$) may be operative in the formation of the 7α-hydroxy compound 59 the results of the control experiments may be invoked. Here, in contrast to the aforementioned case, the 7-acetoxy derivatives isolated in 50 - 60% yield from both control experiments after 6 hours were shown (n.m.r. analysis) to be epimeric mixtures. However, the parent 7α- and 7β-hydroxy compounds recovered were found to have retained their original configuration. These results indicate that $S_Ni$ mechanisms are operative in these systems, but the possibility also exists that $S_Ni$ and $S_N2$ processes may be operating concurrently (FIGURE 22). Similar mechanistic proposals have been made[65,88] in the case of 3β, 7α-diacetoxy-5-cholestene 61 and its 7β-isomer 62, which when separately heated in acetic acid, equilibrate to identical epimeric mixtures (3:2, 7β: 7α).

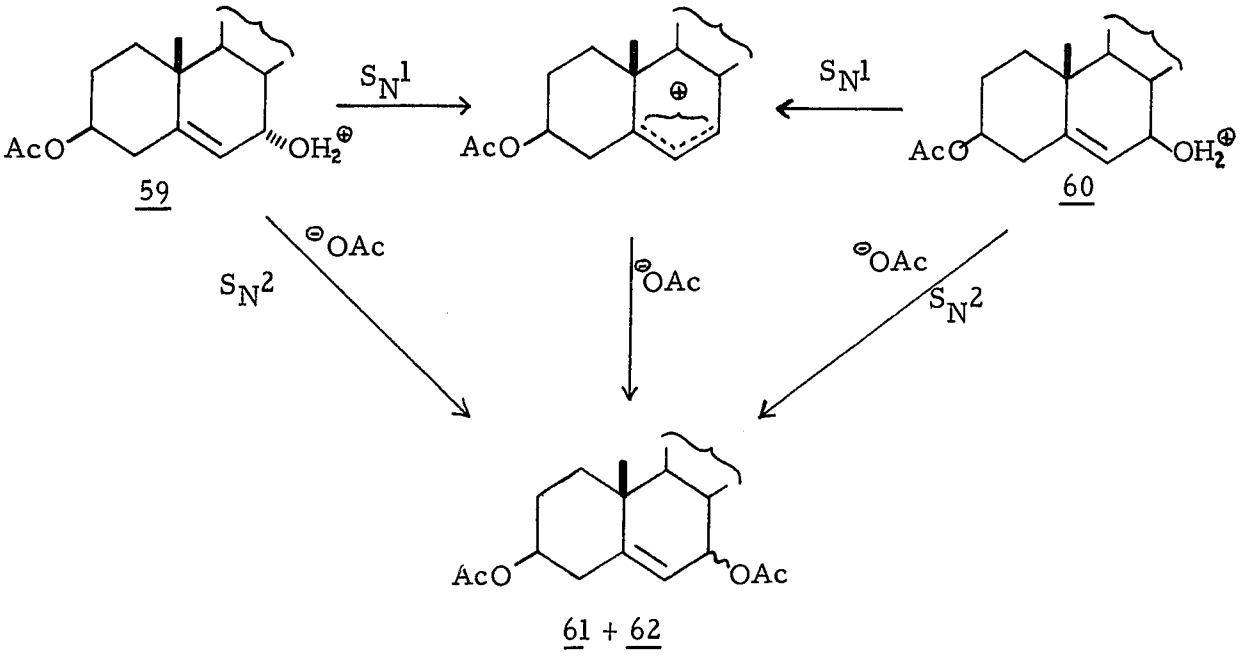



FIGURE 22

Since no epimerized 7-hydroxy compounds were detected it must be concluded that water molecules do not compete with the acetate anions as nucleophiles in this solvent system. Therefore in the case of the rearrangement of the 5α-hydroxy compound 55, since the major product is the 7α-hydroxy compound 59, an $S_N2'$ mechanism is unlikely and the operating mechanism must be an intramolecular $S_Ni'$ process. This may reflect the greater stability of the double bond in the 5, 6-position versus the 6, 7-position and also may explain why even on prolonged treatment no $\Delta^6$-5α-hydroxy compound is formed from the $\Delta^5$-7-hydroxy compounds.

The higher $R_f$ value (TABLE 5) of the unknown compound A suggested that it was a product of dehydration. In an attempt to increase its yield a solution of 3β-acetoxy-7α-hydroxy-5-cholestene 59 in acetic acid (80%) was heated on a steam bath for two hours and the crude material isolated was chromatographed. The major product from this reaction was found (u.v. and n.m.r.) to be a mixture of polyenes all of which had considerably higher $R_f$ values than the unknown compound. No compound with the same $R_f$ value as the unknown was isolated. TABLE 5 indicates that this unknown component is formed initially from the 7-hydroxy compounds 59 and 60 and may thus be a product of substitution of an acetate anion at the 5α-position of the steroid nucleus (FIGURE 23). Heating of the reaction mixture may then result in elimination of acetic acid with possible migration of alkyl groups. SCHEME 7 summarizes the experiments designed for establishing the structure of the unknown compound but the results of these studies were not conclusive.

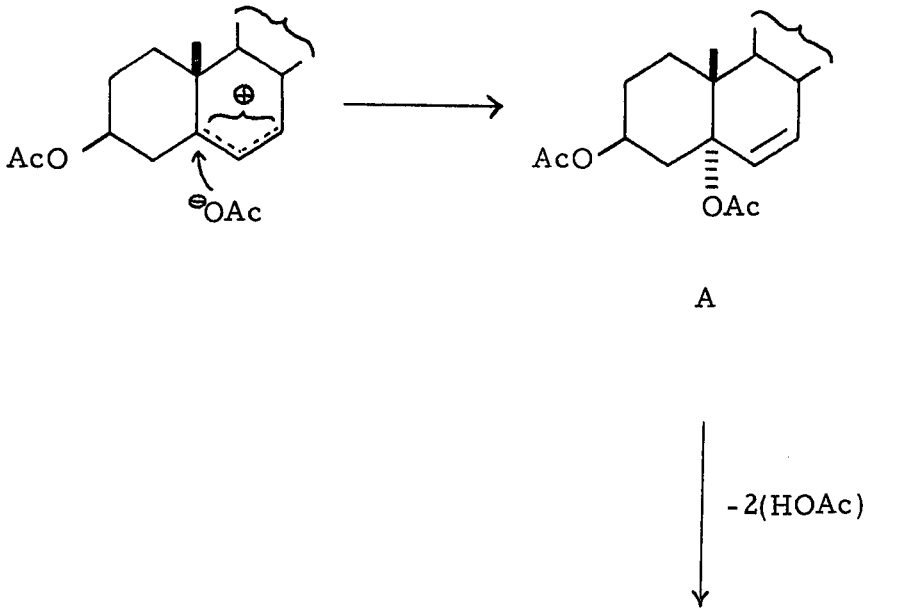
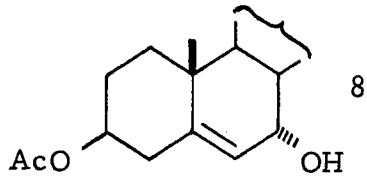


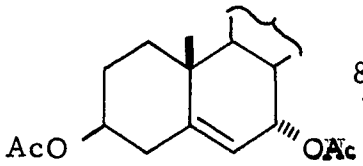

FIGURE 23




59

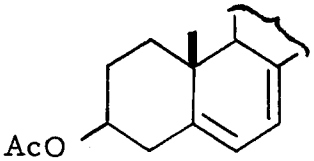
80% HOAc, Δ → polyene mixture $T^1$

80% HOAc, RT → A + 7α-OH + 7ξ-OAc



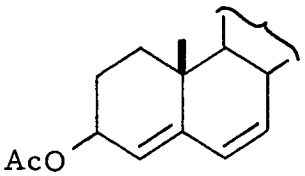

61

80% HOAc, Δ → polyene mixture $T^2$




93

80% HOAc, Δ → no reaction




94

80% HOAc, Δ → polyene mixture $T^3$

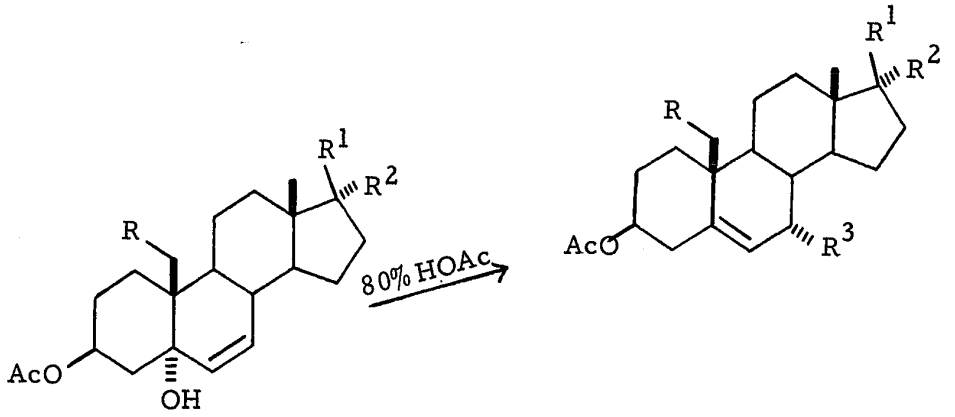
SCHEME 7

Neither of the dienes 93 or 94 have the same $R_f$ value as the unknown compound A. The u.v. spectra of the polyene mixtures $T^1$, $T^2$ and $T^3$ are almost identical and have the characteristic shape exhibited by heteroanular trienes. However none of these spectra is identical to the spectrum of either a $\Delta^{2,4,6}$- or a $\Delta^{3,5,7}$-system and their extinction coefficients (~1000) are much lower than has been observed[89] for such trienes.

Hence, as was the case with strong acids, heating of the steroidal alcohols or acetates in mild acid also results in the formation of a complex polyene mixture. More important though, is the fact that the room temperature studies have indicated that $\Delta^5$-7α-hydroxy steroids can be obtained smoothly and in good yield from their $\Delta^6$-5α-hydroxy isomers, through mild acid treatment. This technique was confirmed by the following experiments with analogous compounds in the androgen series.

The treatment of 3β-acetoxy-5-hydroxy-5α-androst-6-en-17-one 57 and its 17β-acetoxy derivative 56 (SCHEME 8) with acetic acid (80%) at room temperature for two hours resulted in the smooth formation of the $\Delta^5$-7α-acetoxy derivatives 64 and 58 (identified by n.m.r. and mixed melting points) in ca. 15% yield and the $\Delta^5$-7α-hydroxy derivatives 63 and 65 (n.m.r. and mixed melting points) in ca. 85% yield. In the case of 3β,17β-diacetoxy-5-androstene 56, the application of heat also resulted in formation of a polyene mixture which analyzed correctly for $C_{21}H_{28}O_2$. This empirical formula is consistent for a triene and the n.m.r. spectrum shows an integral value in the olefinic region $\delta$ 5.30 - 6.02 of ca. 5 protons. The u.v. spectrum was similar in pattern to those obtained for mixtures $T^1$, $T^2$ and $T^3$ but the extinction coefficient was considerably higher (~10,000) in this case.

(ii) Allylic rearrangement of 3β, 19-diacetoxy-5-hydroxy-5α-androst-6-en-17-one 77




57 R=H, $R^1$, $R^2$=O

56 R=$R^2$=H, $R^1$=OAc

77 R=OAc, $R^1$, $R^2$=O

63 R=H, $R^1$, $R^2$=O, $R^3$=OH

64 R=H, $R^3$=OAc, $R^1$, $R^2$=O

65 R=$R^2$=H, $R^1$=OAc, $R^3$=OH

58 R=$R^2$=H, $R^1$=$R^3$=OAc

95 R=$R^3$=OAc, $R^1$, $R^2$=O

96 R=OAc, $R^3$=OH, $R^1$, $R^2$=O

SCHEME 8

Of the various synthetic approaches studied for the preparation of 7α, 19-disubstituted androgens the mild acid catalyzed allylic rearrangement of $\Delta^6$-5α-hydroxy steroids has proved to be the superior method. 3β, 19-Diacetoxy-5-androsten-17-one 38 can be converted into 3β, 19-diacetoxy-5-hydroxy-5α-androst-6-en-17-one 77 in ca. 40% yield* (TABLE 3), by photosensitized oxygenation. Treatment of the 5α-alcohol 77 with acetic acid (80%) at room temperature for 1.5 hours led to the formation of 3β, 7α, 19-triacetoxy-5-androsten-17-one 95 (SCHEME 8) in 15% yield and of 3β, 19-diacetoxy-7α-hydroxy-5-androsten-17-one 96 in 74% yield. Both products were characterized by n.m.r. analysis, molecular rotations and elemental analysis.

* In all cases the starting material was recovered and recycled.

## 5. Synthesis of 1α-Hydroxy-$\Delta^4$-3-keto Steroids

In view of the fact that 1β-hydroxy-4-estrene-3,17-dione 15 is not an efficient precursor for estrone 1 [see Introduction, Section 3(b)] it was thought worthwhile to attempt the synthesis of a 1α-hydroxy-$\Delta^4$-3-keto androgen and eventually the corresponding 19-functionalized compound with the intention of determining their efficiencies as estrogen precursors, and also their biological activities. Chemically speaking the 1α-hydroxy group, being axial, is more suited for elimination with the 10β-axial group than is the 1β-hydroxy group, and consequently one can imagine an aromatization mechanism as pictured in FIGURE 24. This is similar to the mechanism proposed by Townsley and Brodie (FIGURE 5) for 1β-hydrogen elimination.

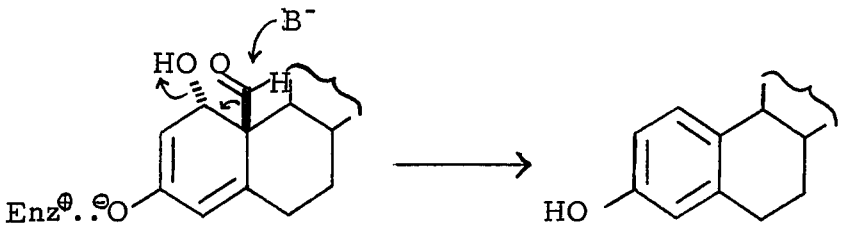



FIGURE 24

Recently a small quantity of a 1α-hydroxy corticosteroid (FIGURE 25) was isolated[90] from the interrenal glands of certain fish, and because 1-hydroxylated derivatives of $C_{21}$-steroids are relatively rare[90] and are of considerable interest to pharmacologists, the chemical synthesis of a 1α-hydroxy-$\Delta^4$-3-keto androgen could also serve as a model to study the preparation of these potentially important corticosteroids.

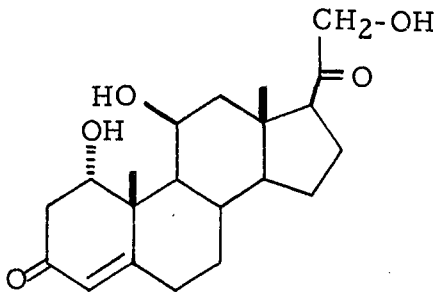
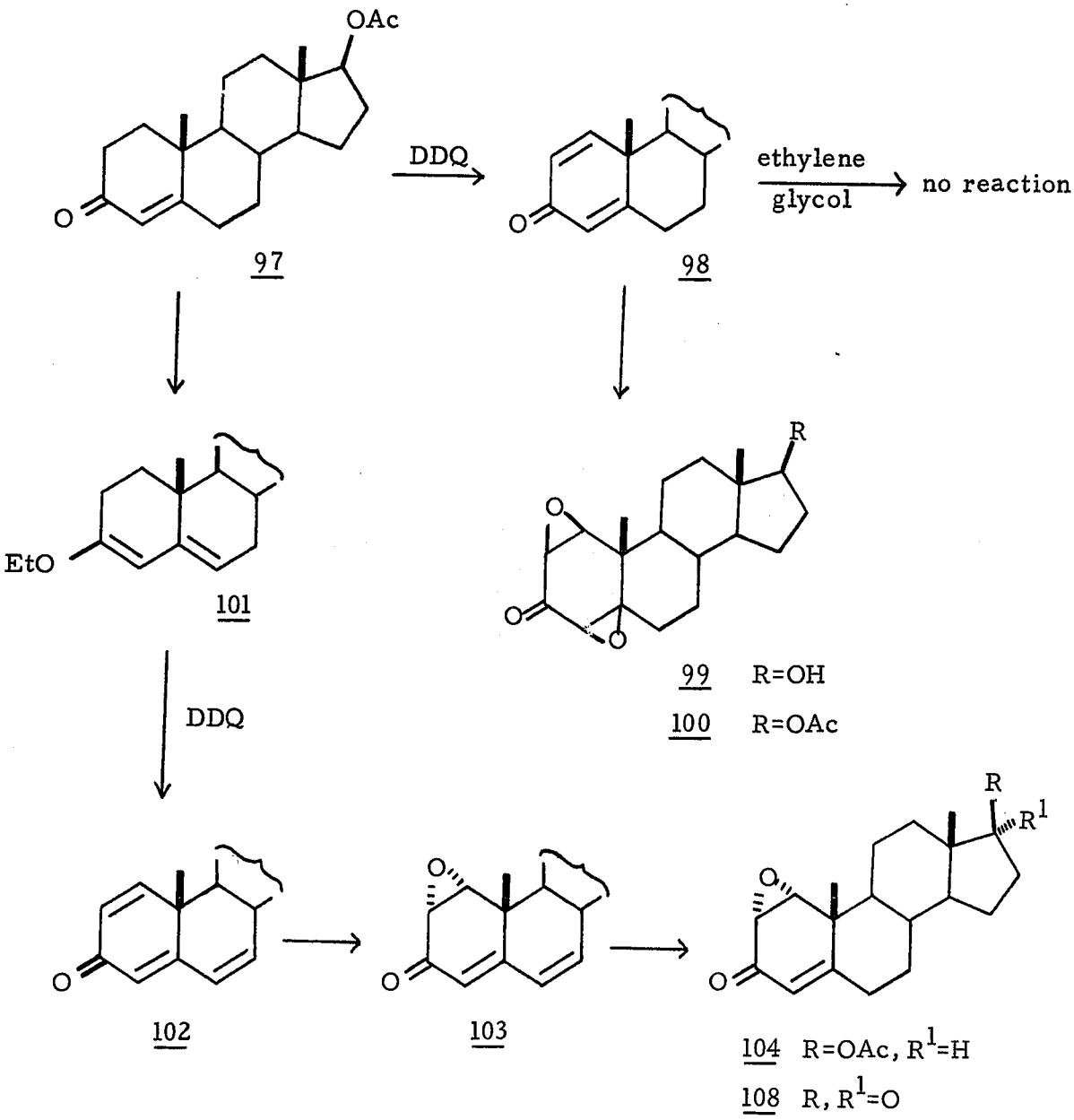



FIGURE 25

(a) $\Delta^{1,4}$-3-Keto-Steroids as Starting Materials

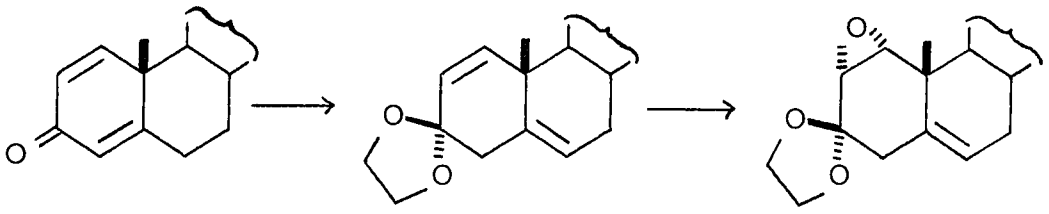
Our first attempt at introducing a 1α-hydroxy group into the A-ring of a $\Delta^4$-3-keto steroid involved treatment of 17β-acetoxy-1,4-androstadiene-3-one 98 (SCHEME 9) with hydrogen peroxide. Compound 98 was obtained by dehydrogenation of 17β-acetoxy-4-androsten-3-one 97 with 2,3-dichloro-5,6-dicyanobenzoquinone (DDQ)[91] under aprotic conditions.


OAc
DDQ
ethylene glycol
no reaction
97
98
EtO
101
R
99 R=OH
100 R=OAc
DDQ
102
103
R
R1
104 R=OAc, R1=H
108 R, R1=O

SCHEME 9

The two products 99 and 100 isolated from the peroxide reaction were both diepoxides (n.m.r.) and were related by acetylation with acetic anhydride of 99 to give 100. Compound 100 was identified by melting point, rotation and n.m.r. as 1β; 2β; 4β, 5-dioxido-17β-acetoxy-5β-androstan-3-one[92]. We had hoped that by variation of the reaction conditions, we could selectively epoxidize the 1, 2-double bond but all attempts to do this failed. Furthermore it became apparent that only β-epoxides were being formed because of steric hindrance to α-face attack. A Dreiding model of a $\Delta^{1,4}$-3-keto steroid shows that the A-ring is bent downwards (α) with respect to the plane of the rest of the molecule, making the β-side more susceptible to attack.

With the aim of removing the near chemical equivalence* of the 1, 2- and 4, 5-double bonds an attempt was made to prepare the ethylene ketal of 17β-acetoxy-1, 4-androstadien-3-one 98. Subsequent treatment of the expected product (see below) with peracid may have proven to be selective and stereospecific, but all conventional methods of preparing the ethylene ketal failed**.



* With peracid treatment it is possible to selectively epoxidize the 4, 5-double bond[96].

** It has been reported[93] that while deconjugation of the double bond in a $\Delta^4$-3-keto steroid can be chemically achieved, the same is not the case for a $\Delta^{1,4}$-3-keto steroid or for a 1α, 2α-epoxy-$\Delta^4$-3-keto steroid[100].

(b) $\Delta^{1,4,6}$-3-Keto-Steroids as Starting Materials

The problem of introducing a monoepoxide in the 1α, 2α-position of a $\Delta^{1,4}$-3-keto steroid was recently overcome by Swiss workers[94,95] who treated a $\Delta^{1,4,6}$-3-keto steroid with alkaline hydrogen peroxide at -10° and obtained the 1α, 2α-oxido derivative. (An examination of a Dreiding model of a $\Delta^{1,4,6}$-3-keto steroid shows that the A/B ring system is planar and thus makes α-face attack more favorable). Hydrogenation of the 1α, 2α-oxido-$\Delta^{4,6}$-3-keto compound over a palladium chloride-strontium carbonate catalyst afforded the 1α, 2α-$\Delta^{4}$-3-keto compound[95].

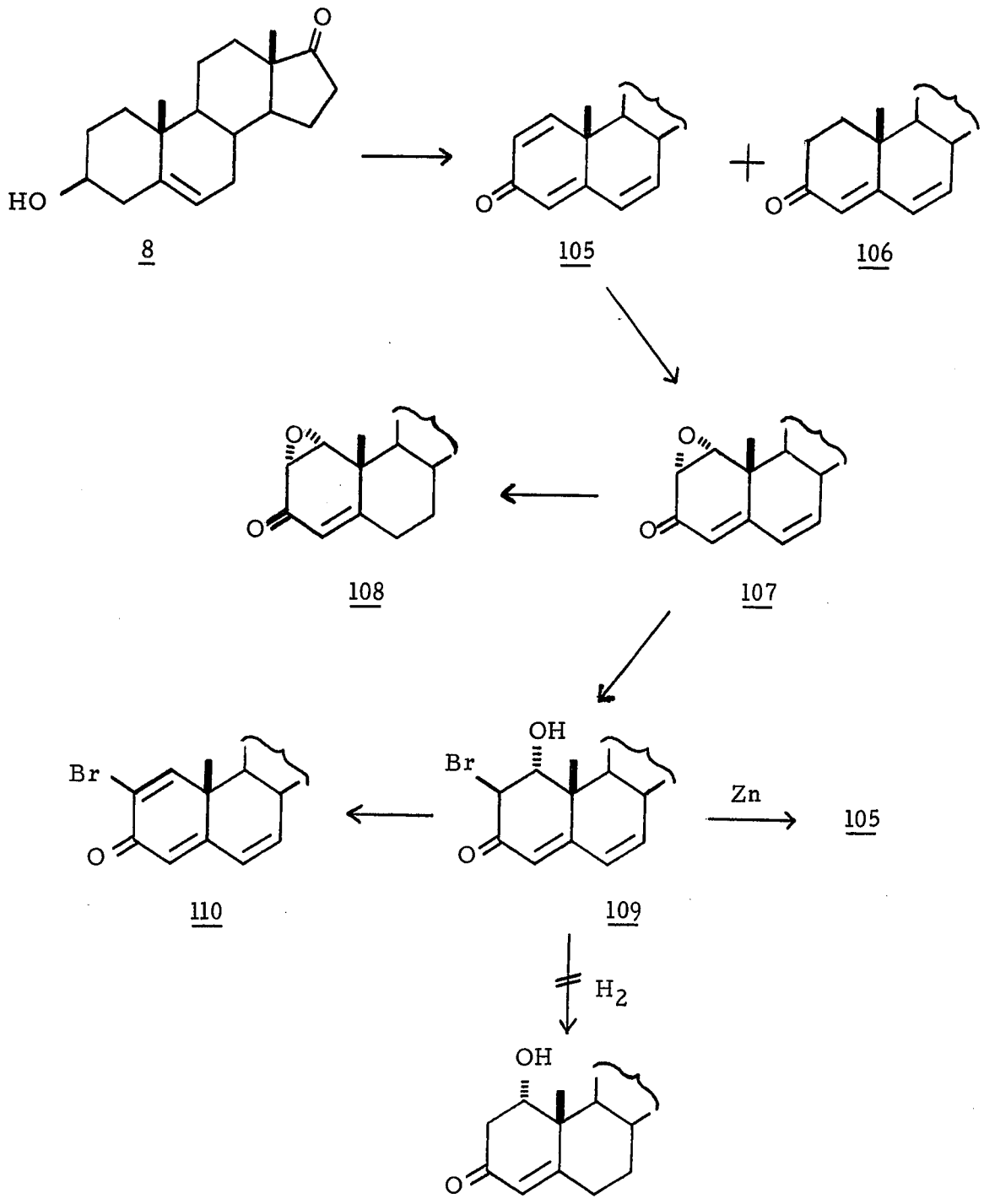
17β-Acetoxy-1, 4, 6-androstatrien-3-one 102 (SCHEME 9) was easily prepared by a method[97] involving treatment of the enol ether 101 of 17β-acetoxy-4-androsten-3-one 97 with DDQ in acetone. The triene 102 was treated with alkaline hydrogen peroxide at -10° and the resulting 1α,-2α-mono-epoxide 103 was hydrogenated in benzene with a freshly prepared catalyst (5% $PdCl_2/SrCO_3$) yielding 1α, 2α-oxido-17β-acetoxy-4-androsten-3-one 104.

Envisaging a metal hydride reduction of the epoxide to give the 1α-hydroxy derivative attempts were made to protect the carbonyl group. However, the preparation of the enol ether or the pyrrolidine enamine derivatives of compound 104 proved to be impossible, as did the preparation of the 3,3'-ethylene ketal of compound 108 (see Footnote on p. 73).

Other workers[99] have introduced a 1α-hydroxy group into a 3-keto steroid by epoxidation of the $\Delta^{1}$-3-keto compound followed by HBr opening of the epoxide to the 1α-hydroxy-2β-bromo compound and subsequent catalytic hydrogenation, which removed the bromine atom, yielding a 1α-hydroxy-3-keto compound in a yield of less than 20%.

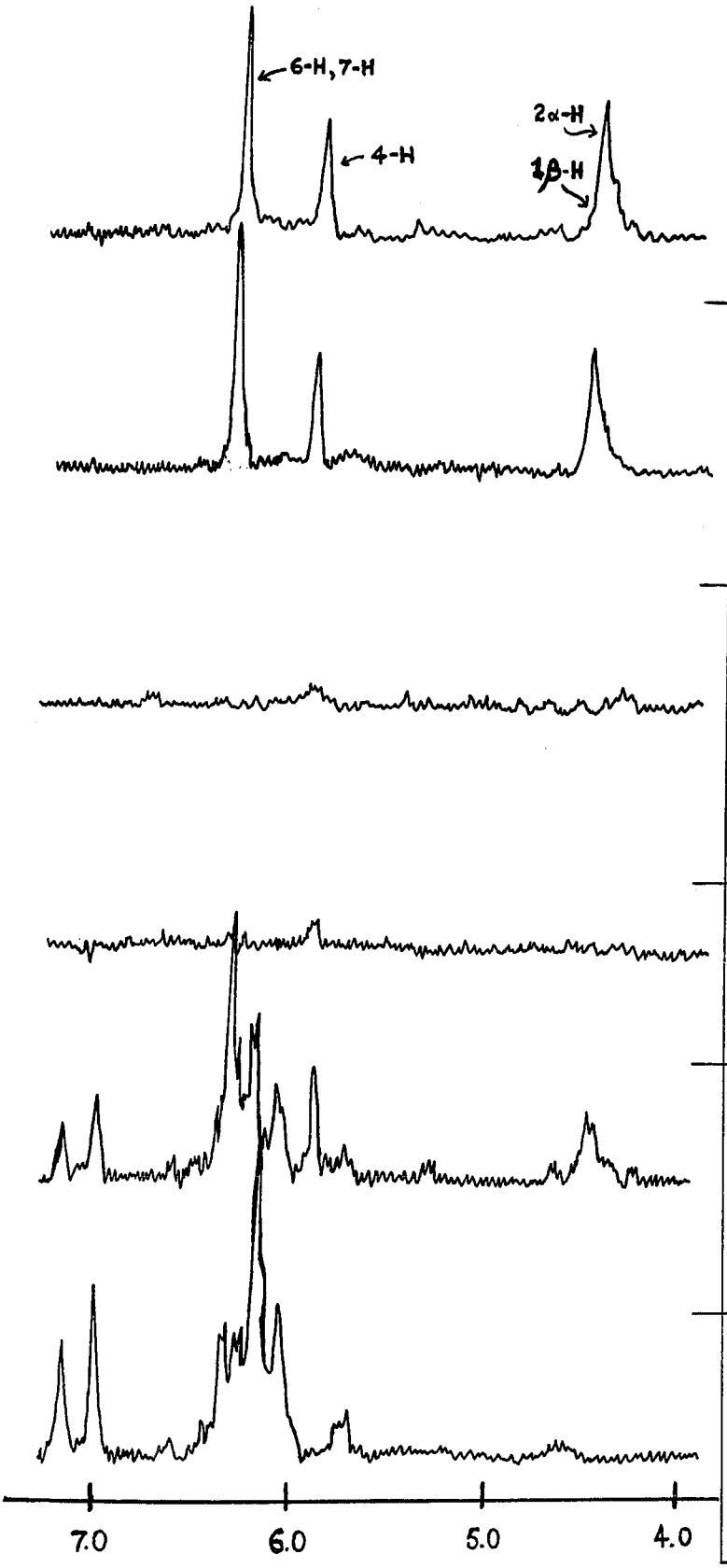
This approach was extended to a system which contained the necessary unsaturation at C-4, C-5 in the following manner. 3β-Hydroxy-5-androsten-17-one 8 (SCHEME10) was dehydrogenated with DDQ to the triene 105 in 14% yield. The triene 105 was epoxidized selectively in the 1α, 2α-position with alkaline hydrogen peroxide. The epoxide 107 when treated with HBr at room temperature gave only the known[94] compound 2-bromo-1, 4, 6-androstatriene-3, 17-dione 110 , the 1α-hydroxy group initially formed having been eliminated under the acid conditions. The elimination of the 1α-hydroxy group from a $\Delta^4$-3-keto compound has been reported[98,100] to be an extremely facile one. When the epoxide 107 was treated with cold HBr (50%) at 5°C it was possible to isolate the bromohydrin 109 . The n.m.r. spectrum of compound 109 showed signals at δ 4.40 (1β-H), 4.50 (2α-H), 6.02 (4-H) and 6.45 (6-H and 7-H). Removal of the bromine atom by catalytic hydrogenation proved to be a stumbling block. Under the conditions used it was not possible to reduce the halogen atom and the 6, 7-double bond and at the same time keep the rest of the A/B ring system intact. After 30 minutes the molecule had suffered dehydration as well as reduction of the halogen atom. The results of varying the ratio of compound to catalyst (wt.:wt.) and the reaction time are shown in FIGURE 26 and they reflect the instability of the 1α-hydroxy-$\Delta^4$-3-keto system (a "β"-hydroxy-ketone) even under the mild conditions of the reaction (benzene - 5% $PdCl_2$/ $SrCO_3$).

Attempts to reduce the bromine off with zinc powder in either ethanol or N, N-dimethylformamide resulted in triene 105 formation.




SCHEME 10

N. m. r. Spectrum 4.0-7.2 (δ)




| Ratio com. : cat. | Time (min) | Interpretation |
|---|---|---|
| | 0 | starting material |
| 1:1.5<br>1:2.5 | 30<br>20 | no reaction |
| 1:10 | 180 | reduction of all double bonds and loss of bromine atom |
| 1:4 | 45 | 4,5-double bond not fully reduced |
| 1:2.5 | 35 | dehydration and some loss of bromine atom |
| 1:1 | 55 | dehydration and complete loss of bromine |

FIGURE 26

There have been reports[94,100,101] that catalytic hydrogenation of a 1α, 2α-epoxy-$\Delta^{4,6}$-3-keto steroid results in slow hydrogenation of the epoxy group to yield a 1α-hydroxy-$\Delta^{4}$-derivative in low yield. This method may well prove to be the best route to 1α-hydroxy-$\Delta^{4}$-3-keto steroids but as was the case with the hydrogenation of our bromohydrin 109, there appears to be a fine line between selective hydrogenation and over-hydrogenation.

This summarizes the various approaches which were investigated in an attempt to synthesize the desired 1α-hydroxy steroids. It is clear that further studies will have to be initiated in other directions if this goal is to be achieved.

# EXPERIMENTAL

## 1. General

All melting points were taken on a Hoover Capillary melting point apparatus and are uncorrected. Infrared spectra were obtained from Beckman IR-8 and IR-20 instruments with chloroform as the solvent. Nuclear magnetic resonance spectra were obtained from Varian V-4302-T-60 and Varian H-100 instruments, with chloroform-d and dimethylsulphoxide-$d_6$ as the solvents and tetramethylsilane as the internal standard. Optical rotations were obtained on a Perkin-Elmer 141 automatic polarimeter with methanol and chloroform as solvents. Ultra violet spectra were obtained on a Perkin-Elmer 202 u.v.-visible spectrophotometer with ethanol and hexane as solvents.

For column chromatography an L.K.B.-3400 fraction collector was used. Silica R200-300 mesh and Woelm neutral alumina I were used as adsorbants. Benzene, ether, petroleum ether (b.p. 30-60°) and methanol were used as eluents. Silica gel G (according to Stahl) was used as adsorbant for thin layer chromatography and the plates were sprayed with 50% sulphuric acid.

Microanalyses were by Alfred Bernhardt, Mikroanalytisches Laboratorium, Elbach über Engelskirchen, West Germany, and by G. Daessle, Organic Microanalysis, Montreal, Canada.

Organic extracts were dried over anhydrous $MgSO_4$ or $Na_2SO_4$. The dilute HCl used was 3 N. The solvents were evaporated under reduced pressure unless otherwise stated.

The following abbreviations are used: i.r. (infrared); n.m.r. (nuclear magnetic resonance); u.v. (ultra violet); t.l.c. (Thin layer chromatography; m.p. (melting point); $[\alpha]_D$ (specific rotation at room temperature); pet. ether (petroleum ether); DMSO (dimethyl-sulphoxide).

2. Synthesis of 7,19-Disubstituted Androgens

(a) Synthesis of 3β,19-Diacetoxy-5-androsten-17-one 38 and Reaction with t-Butylperbenzoate

3β-Hydroxy-5-androsten-17-one 8 (20 g, 0.07 moles) was acetylated with acetic anhydride (6 ml) in pyridine (40 ml) yielding, after crystallization from acetone, pure 3β-acetoxy-5-androsten-17-one 34 (20 g); m.p. 167-169° (lit.[102], m.p. 168-170°).

A solution of the above acetate 34 (20 g, 0.06 moles) in purified dioxane (200 ml) was cooled in an ice-bath. Perchloric acid (10 ml) and N-bromoacetamide (17.2 g, 0.13 moles) were added to this solution over a period of 1 hour and after another 30 min. the reaction was stopped by the addition of 30% $Na_2SO_3$ solution (100 ml). A further 100 ml of water was added to precipitate the product which, after filtration of the mixture, was washed with water and dried in vacuo. Crystallization of the crude from ether-pet. ether 30-60 yielded 7.8 g of crude 3β-acetoxy-5-bromo-6β-hydroxy-5α-androstan-17-one 35 which still contained some starting material 34 by t.l.c. analysis.

Lead tetraacetate (12.0 g, 27 mmoles) and calcium carbonate (7 g) were added to cyclohexane (300 ml) and the mixture was refluxed for 1 hour by means of a 500 watt lamp. With continuous stirring iodine (4 g) and the crude bromohydrin 35 (3.0 g, ~7 mmoles) were added to the mixture. After refluxing again for 1 hour the mixture was filtered through Celite and the filtrate was washed with 30% $Na_2S_2O_3$ solution and water. After drying the organic layer and evaporation of the solvent 2.5 g of crude product was obtained. The above procedure was repeated and the products combined.

The crude 3β-acetoxy-5-bromo-6β,19-oxido-5α-androstan-17-one 36 (5.8 g., ~ 13 mmoles) obtained above was refluxed in ethanol (300 ml) with zinc powder (40 g) for 24 hours. After filtering the mixture through Celite and evaporation of the solvent, the residue was chromatographed on Silica R(500 g) with ether-benzene (1:20) as the eluent. The desired fraction was isolated and after crystallization from ether-pet. ether 30-60, 3β-acetoxy-19-hydroxy-5-androsten-17-one 37 (3.6 g) was obtained; m.p. 158 - 160° (lit.[51], m.p. 157-158°).

3β-Acetoxy-19-hydroxy-5-androsten-17-one 37 (1 g., 3 mmoles) was acetylated with acetic anhydride (2 ml) in pyridine (10 ml). The usual workup, followed by crystallization of the product from ether-pet. ether 30-60, afforded 900 mg of 3β,19-diacetoxy-5-androsten-17-one 38 ; m.p. 107 - 108° (lit.[50], m.p. 103-105°).

The diacetate 38 (800 mg, 2 mmoles) in acetic acid (15 ml) and cuprous bromide (0.25 g) were heated with stirring under a nitrogen atmosphere to reflux temperature. A solution of t-butyl perbenzoate* (40 ml) in acetic acid (15 ml) was added dropwise over a period of 20 minutes and the mixture was maintained at reflux temperature for an additional 20 minutes. The mixture was then poured into benzene (50 ml) and the two phases intimately mixed and then filtered. The benzene solution was separated and was washed with water, 5% $Na_2CO_3$ solution, water again, then dried and evaporated. Chromatography of the crude (830 mg) on Silica R(100 g) with ether-benzene (1:20) as eluent afforded starting material (250 mg),an oily mixture (360 mg) of the epimeric

*The t-butyl perbenzoate was prepared according to the procedure of Milas and Surgenor[103], as described below. To 90% t-butylhydroperoxide (6.0 g) at 15°C was added over a period of 1 hour with vigorous stirring, benzoyl chloride (7.0 g) and 30% KOH solution (12.0 g). This mixture was left stirring at room temperature overnight after which the organic layer was separated and washed with 5% $Na_2CO_3$ solution (50 ml) and then with water (30 ml) and dried; yield 6.4 g.

7-acetoxy derivatives: n.m.r. (100 Mc, $CDCl_3$) $\delta$ 0.90 (18-$CH_3$, doublet due to mixtur 3H), 2.0 (3β, 19, 7ξ-OAc, s, 9H), 3.85, 3.97, 4.62, 4.74 (19-$CH_2$-, q, 2H), 4.68 (3α-H, m, 1H), 5.16 (7ξ-H, m, 1H), 5.52 (6-H, d, J = 2.0 Hz), 5.87 (6-H, d, J = 5.0 Hz) and also a ketonic product X (61 mg). M.p. 160-161°; i.r. ($CHCl_3$) $\nu_{max}$ 1730 (ester C=O), 1670 $cm^{-1}$ (α, β-unsat. C=O); n.m.r. (100 Mc, $CDCl_3$) $\delta$ 0.90 (18-$CH_3$, s, 3H), 2.02 (3β, 19-OAc, s, 6H), 4.02, 4.14, 4.72, 4.84 (19-$CH_2$-, q, 2H), 4.70 (3α-H, m, 1H), 5.92 (6-H, s, 1H).

(i) Preparation of 3β,19-diacetoxy-5-androstene-7,17-dione 39

To 3β,19-diacetoxy-5-androsten-17-one 38 (800 mg, 2 mmoles), in glacial acedic acid at 51°C was added with stirring chromic anhydride (0.60 g., 6mmoles). The mixture was maintained at a temperature between 51° and 53° for 2 hours after which ethanol (5 ml) and water (10 ml) were added. The green solution was extracted with ether and the ethereal solution was washed with $NaHCO_3$ solution and water, then dried and evaporated. The crude product (500 mg) was chromatographed on SilicaR (50 g) with ether-benzene (1:20) as eluent. The chromatography afforded starting material 38 (110 mg) and 3β,19-diacetoxy-5-androstene-7,17-dione 39, m.p. 160-161°; $[\alpha]_D$ -125° (C 0.06 $CHCl_3$); the spectral data are identical to those of the ketonic product X above. Mixed m.p. (X and 39) 160-161°.

Anal. Calcd. for $C_{23}H_{30}O_6$: C, 68.63; H, 7.51.

Found: C, 67.96*; H, 7.64.

---

* Carbon analysis is not acceptable, however the spectral data confirm the structure assigned.

(b) t-Butyl perbenzoate Reaction in Absence of Acetic Acid

3β,19-Diacetoxy-5-androsten-17-one 38 (1.0 g, 2.5 mmoles) in benzene (50 ml) was refluxed with cuprous bromide (100 mg) under a nitrogen atmosphere and t-butylperbenzoate (6 ml) was added over a period of 1 hour. The usual workup and chromatography on SilicaR (80 g) with ether-benzene (1:14) afforded starting material (120 mg) and a 7ξ-benzoate mixture (343 mg). Of the epimeric mixture, the last 90 mg off the column was almost pure (n.m.r.) 7α-benzoate derivative, the original mixture having been analyzed (n.m.r.) as 16:7, 7α-:7β-benzoates. N.m.r. (60 Mc, $CDCl_3$) δ 0.98 (18-$CH_3$, s, 3H), 2.0 (3β,19-OAc, s, 6H), 3.88, 4.08, 4.66, 4.86 (19-$CH_2$-, q, 2H), 4.68 (3α-H, m, 1H), 5.40 (7ξ-H, m), 5.70 (6-H), 6.02 (6-H, d, J ≈ 5.0 Hz), 7.20 - 8.20 (  , m, 5H). The product was not crystallized.

(c) Synthesis of 3β,17β,19-Triacetoxy-5-Androstene 44 and Reaction with t-Butylperbenzoate

To 3β-hydroxy-5-androsten-17-one 8 (50 g, 0.17 moles) in tetrahydrofuran (1 l.) was added a solution of sodium borohydride (4.0 g, 0.1 mole) in water (24 ml). The mixture was stirred overnight at room temperature. Approximately half of the solvent was evaporated and water (500 ml) was added and the resulting mixture was filtered. The precipitate was dried in air and a few milligrams were crystallized from acetone-pet. ether 60-80, affording pure 3β,17β-dihydroxy-5-androstene 50 m.p. 183-184° (lit.[104], m.p. 184°). The dried precipitate (45 g) was acetylated with acetic anhydride (20 ml) in pyridine (200 ml) and usual workup yielded 3β,17β-diacetoxy-5-androstene 40 (50 g), m.p. 157-159° (lit.[104], m.p. 158-159°). To 3β,17β-diacetoxy-5-androstene 40 (50 g,

0.17 moles) in dioxane (800 ml) at 5°C was added perchloric acid (26 ml). N-Bromoacetamide (43 g., 0.31 moles) was added in the dark with stirring over a period of 1 hour after which the reaction was stopped by the addition of 30% $Na_2SO_3$ solution (100 ml) and water (400 ml). The reaction mixture was extracted twice with ether (500 ml) and the combined extracts washed with water, dried and evaporated to half volume. Pet. ether 30-60 (500 ml) was added and the solution evaporated on a steam bath to approximately half volume, whereupon crystals of 3β,17β-diacetoxy-5-bromo-6β-hydroxy-5α-androstane 41 were formed. After standing for 2 hours the mixture was filtered and the product (16.5 g) dried in vacuo. M.p. 168-170° (lit.[105], m.p. 172-174°).

To lead tetraacetate (44 g., 0.1 mole) in cyclohexane (1.1 l) was added with stirring calcium carbonate (24 g) and the mixture was refluxed for 1 hour by means of a 500 watt lamp. With continuous stirring, iodine (12.5 g) and the bromohydrin 41 (10 g., 0.02 moles), were added and the mixture refluxed as before for another hour. After filtration of the mixture through Celite the solution was washed with 30% $Na_2S_2O_3$ solution, water, dried and evaporated. The reaction and workup were performed again on 6.5 g of the bromohydrin 41 and the products combined. Yield 15 g. Crystallization of a small amount of the crude material from methanol gave pure 3β,17β-diacetoxy-5-bromo-6β,19-oxido-5α-androstane 42, m.p. 175-177° (lit.[106], m.p. 178-179°).

The 6β,19-oxide 42 (15 g) and zinc powder (125 g) in ethanol (700 ml) were vigorously stirred at reflux temperature for 24 hours. After filtration of the mixture through Celite, the solvent was evaporated yielding crude product (12 g) which was chromatographed on SilicaR (500 g) with ether-benzene (1:4) as the eluent. The chromatography afforded 3β,17β-diacetoxy-5-androsten-19-ol 43 (6.3 g), m.p. 147-148° (lit.[106], m.p. 147.5-148.5°).

The above diacetate 43 (6.0 g., 15 mmoles) was acetylated with acetic anhydride (6 ml) in pyridine (40 ml) and after the usual workup and crystallization of the crude product from ether-pet. ether, 3β,17β,19-triacetoxy-5-androstene 44 (4.7 g) was obtained. M.p. 89-91° (lit.[106], m.p. 88-89°).

The triacetate 44 (3.5 g., 8 mmoles) in acetic acid (75 ml) and cuprous bromide (1.0 g) were heated to reflux temperature with vigorous stirring under a nitrogen atmosphere. A solution of t-butyl perbenzoate (20 ml) in acetic acid (80 ml) was added to the reaction mixture dropwise over a period of 45 min and reflux temperature was maintained for an additional 30 min with constant stirring. The mixture was cooled and then poured into benzene (300 ml) and the two phases were intimately mixed and then filtered. The benzene solution was separated and washed with water, 5% $Na_2CO_3$ solution, then water again. After drying and evaporation of the solvent, 3.0 g of crude material was obtained. Chromatography of the crude product on SilicaR (230 g) with ether-benzene (1:33) afforded starting material (1.0 g) and an oil (1.39 g., 40%) which was identified (n.m.r.) as an epimeric mixture of the 7-acetoxy derivatives: n.m.r. (100 Mc., $CDCl_3$) δ 0.80 (18-$CH_3$, s, 3H), 2.00 (3β, 7ξ, 17β, 19-OAc, s, 12H), 3.88, 4.00, 4.58, 4.70 (19-$CH_2$-, q, 2H), 4.68 (3α-H, 17α-H, m, 2H), 5.03 (7β-H, t), 5.18 (7α-H, d), 5.55 (6-H, d), 5.86 (6-H, d, J = 5.0 Hz). This mixture was inseparable on repeated chromatography using SilicaR or alumina as adsorbants.

(d) 7α-Acetoxy-3β,17β,19-trihydroxy-5-androstene 47 and 7β-acetoxy-3β,17β,19-trihydroxy-5-androstene 48

To an epimeric mixture of 3β,7ξ,17β,19-tetraacetoxy-5-androstenes (800 mg) in methanol (250 ml) was added $Na_2CO_3$ (500 mg) in water (5 ml) and the mixture was stirred at room temperature for 24 hours. Dilute acetic acid was added dropwise with stirring until the pH of the solution was ca. 7 as indicated by litmus paper. Water (50ml) was added and the solution was evaporated to half volume and then extracted with ether (3 x 100 ml). After washing ($H_2O$) and drying the combined organic extracts, the ether was evaporated and the residue was dissolved in chloroform (hot) and then placed in a fridge at 4°C for 20 hours. The mixture was filtered and the precipitate was crystallized from chloroform, yielding pure 7α-acetoxy-3β,17β,19-trihydroxy-5-androstene 47 (88 mg), m.p. 165-167°; $[\alpha]_D$ -185° (C 0.21, MeOH); i.r. (salts) $\nu_{max}$ 3450 (free-OH), 3340 (bonded-OH), 1720 $cm^{-1}$ (ester C=O), n.m.r. (60 Mc, $CDCl_3$ + DMSO) $\delta$ 0.80 (18-$CH_3$, s, 3H), 2.01 (7α-OAc, s, 3H), 4.98 (7β-H, t, 1H), 5.78 (6-H, d, 1H, J = 5.0 Hz). The region between 3.0 and 4.0 masked by DMSO.

Anal. Calcd. for $C_{21}H_{32}O_5$: C, 69.20; H, 8.85.
Found : C, 69.39; H, 8.84.

The triol 47 (20 mg) was acetylated with acetic anhydride (0.5 ml) in pyridine (2.5 ml) and usual workup yielded 3β,7α,17β,19-tetraacetoxy-5-androstene 45 as an oil. $[\alpha]_D$ -161° (C 0.4, $CHCl_3$), n.m.r. (100 Mc, $CDCl_3$) $\delta$ 0.80 (18-$CH_3$, s, 3H), 2.00 (3β,7α,17β,19-OAc, s, 12H), 3.88, 4.00, 4.58, 4.70 (19-$CH_2$-, q, 2H), 4.68 (3α-H, 17α-H, m, 2H), 5.03 (7β-H, t, 1H), 5.86 (6-H, d, 1H, J = 5.5 Hz).

To 3β,17β,19-triacetoxy-5-androstene 44 (1.0 g, 2.3 mmoles) in glacial acetic acid (5.5 ml) at 51°C was added with stirring, chromic anhydride (0.75 g., 7.5 mmoles) over the period of 1 hour. The mixture was maintained at a temperature range of 51° to 53° for 2 hours and then ethanol (5 ml) and water (10 ml) were added. The solution was extracted with ether and the ethereal solution was washed with $NaHCO_3$ solution and water, then dried and evaporated. The crude product (600 mg) was chromatographed on SilicaR (50 g) with ether-benzene (1:14) as eluent. The chromatography afforded 3β,17β,19-triacetoxy-5-androsten-7-one 49 (400 mg) as an oil. $[\alpha]_D$ -107 (C 0.5, $CHCl_3$); i.r. ($CHCl_3$) $\nu_{max}$ 1730 (ester C=O), 1670 $cm^{-1}$ (α,β-unsaturated C=O); n.m.r. (60 Mc, $CDCl_3$) δ 0.80 (18-$CH_3$, s, 3H) 2.00 (3β,17β,19-OAc, s, 9H), 4.00, 4.20, 4.63, 4.83 (19-$CH_2$-, q, 2H), 5.90 (6-H, s, 1H).

Anal. Calcd. for $C_{25}H_{34}O_7$ : C, 67.24; H, 7.68.

Found : C, 67.15; H, 7.67.

The ketone 49 (400 mg, 1 mmole) in absolute ethanol (25 ml) and excess sodium borohydride were stirred at room temperature for 24 hours. Water (20 ml) was added and the mixture was extracted with chloroform (50 ml) and the organic layer was dried and evaporated. The crude product (237 mg) was acetylated with acetic anhydride (1 ml) in pyridine (10 ml) and the usual workup yielded 3β,7β,17β,19-tetraacetoxy-5-androstene 46 (220 mg) as an oil. $[\alpha]_D$ -41° (C 0.27, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 0.95 (18-$CH_3$, s, 3H), 2.04 (3β,7β,17β,19-OAc, s, shoulder, 12H), 3.78, 3.98, 4.54, 4.74 (19-$CH_2$-, q, 2H), 4.60 (3α,-17α-H, m, 2H), 5.00 (7α-H, d, 1H), 5.50 (6-H, d, 1H, J = 2.0 Hz).

To the tetraacetate 46 (200 mg) in methanol (50 ml) was added $Na_2CO_3$ (100 mg) in water (1 ml) and the mixture was stirred at room temperature for 24 hours. The solution was neutralized with dilute acetic acid and extracted with chloroform. The extract was washed with water, dried and evaporated to 20 ml and then placed in a fridge at 4°C for 24 hours. The precipitate was collected by filtration and crystallized twice from chloroform affording pure 7β-acetoxy-3β,17β,19-trihydroxy-5-androstene 48 (32 mg). M.p. 194-195°; $[\alpha]_D$ +65° (C 0.05, MeOH); n.m.r. (60 Mc, $CDCl_3$ + DMSO) δ 0.77 (18-$CH_3$, s, 3H), 1.99 (7β-OAc, s, 3H), 4.90 (7α-H, d, 1H), 5.32 (6-H, d, 1H, J ≈ 2Hz). The region between δ 3.0 and 4.0 was masked by DMSO.

Anal. Calcd. for $C_{21}H_{32}O_5$ : C, 69.20; H, 8.85.

Found : C, 69.17; H, 8.74.

3. Photosensitized Oxygenations

The oxygenations were conducted in a pyrex cylinder irradiated along its length by four 15-watt fluorescent lamps mounted about 3 inches away. Oxygen was admitted near the bottom of the cylinder at a steady rate without interruption via a fritted glass tube.

(a) Oxygenation of 3β-Acetoxy-5-cholestene 51

3β-Acetoxy-5-cholestene 51 (1.0 g, 3 mmoles) and hematoporphyrin (5 mg) in pyridine (100 ml) were irradiated for 4 days. Ether (100 ml) and charcoal were added and the mixture was shaken, filtered and the filtrate evaporated. The residue was dissolved in chloroform and allowed to stand for 2 days. After evaporation of the chloroform the residue was treated with a solution consisting of ethanol (30 ml), ether (20 ml), sodium iodide (2.5 g) and acetic acid (5 drops), for 24 hours. Water (50 ml) was added and the solution was extracted with ether. The ethereal solution was washed with 0.1 N $Na_2S_2O_3$ solution, water, and then dried and evaporated. The crude product (800 mg) was chromatographed on SilicaR (100 g) with benzene as the eluent. The first compound (200 mg) eluted was identified by t.l.c. as starting material 51. The second compound (200 mg) eluted was identified as 3β-acetoxy-5-hydroxy-5α-cholest-6-ene 55, m.p. 144-146° (lit.[58], m.p. 143°). The third compound (300 mg) eluted was identified as 3β-acetoxy-7α-hydroxy-5-cholestene 59. M.p. 139-140° (lit.[58], m.p. 139°). N.m.r. (100 Mc, $CDCl_3$) δ 0.65 (18-$CH_3$), 1.0 (19-$CH_3$), 3.75 (7β-H, m, 1H), 4.60 (3α-H, m, 1H), 5.60 (6-H, d, 1H, J = 5.4 Hz).

(i) Synthesis of 3β, 7α-diacetoxy-5-cholestene 61 and 3β, 7β-diacetoxy-5-cholestene 62

3β-Acetoxy-7α-hydroxy-5-cholestene 59 was acetylated with acetic anhydride (5 ml) in pyridine (20 ml) and usual workup afforded 3β, 7α-diacetoxy-5-cholestene 61 (300 mg). M.p. 121-122° (lit.[58], m.p. 122°). N.m.r. (100 Mc, $CDCl_3$) δ 0.64 (18-$CH_3$, s), 0.89 (19-$CH_3$), 1.98 (3β, 7α-OAc, s, 6H), 4.67 (3α-H, m, 1H), 4.97 (7β-H, t, 1H), 5.59 (6-H, d, 1H, J = 5.03 Hz).

To 3β-acetoxy-5-cholestene 51 (1 g., 2.3 mmoles) in glacial acetic acid (2 ml) at 51°C was added with stirring, chromic anhydride (0.75 g., 7.5 mmoles). The temperature was maintained between 51° and 53° for two hours. Ethanol (10 ml) and water (50 ml) were added and the green solution was extracted with ether. The ethereal solution was washed with water, $NaHCO_3$ solution, water again, and was then dried, filtered and evaporated. The crude product (800 mg) was crystallized twice from methanol yielding pure 3β-acetoxy-5-cholesten-7-one 84 (600 mg). M.p. 163-164° (lit.[55], m.p. 164°).

To 3β-acetoxy-5-cholesten-7-one 84 (500 mg., 1.1 mmoles) in ethanol (25 ml) was added a solution of sodium borohydride (20 mg) in water (1 ml) and the mixture was stirred at room temperature overnight. After the addition of water (25 ml) the mixture was filtered and the precipitate was dried in the air. Part of the product was crystallized from acetone, yielding pure 3β-acetoxy-7β-hydroxy-5-cholestene 60. M.p. 109-112° (lit.[59], m.p. 110-111°); n.m.r. (60 Mc, $CDCl_3$) δ 0.70 (18-$CH_3$, s), 2.00 (3β-OAc, s, 3H), 3.81 (7α-H, m, 1H), 4.60 (3α-H, m, 1H), 5.30 (6-H, d, 1H, J ≈ 2.0 Hz).

The rest of the product (ca. 300 mg) was acetylated with acetic anhydride (1 ml) in pyridine (5 ml). Workup as usual and crystallization of the product three times from methanol yielded pure 3β, 7β-diacetoxy-5-cholestene 62 (300 mg). M.p. 109-110° (lit.[88], m.p. 108-110°); n.m.r. (100 Mc, $CDCl_3$), δ 0.64 (18-$CH_3$, s), 1.02 (19-$CH_3$), 1.99 (3β,-7β-OAc, s, 6H), 4.60 (3α-H, m, 1H), 5.02 (7α-H, d, 1H), 5.23 (6-H, d, 1H, J ≈ 2.0 Hz).

(b) Oxygenation of 3β-Acetoxy-5-androsten-17-one 34

3β-Acetoxy-5-androsten-17-one 34 (1.0 g., 3 mmoles) was subjected to the photo-oxygenation, the mutarotation in chloroform and reduction in an analogous manner as described earlier. Chromatography on SilicaR (100 g) with ether-benzene (1:19) as eluent afforded starting material (380 mg) identified by t.l.c.; 3β-acetoxy-5-hydroxy-5α-androst-6-en-17-one 57 (350 mg). M.p. 176-178°; $[\alpha]_D$ +36° (C 0.07, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 0.92 (18-$CH_3$, s, 3H), 1.00 (19-$CH_3$, s, 3H), 2.00 (3β-OAc, s, 3H), 5.28 (3α-H, m, 1H), 5.70 (6-H, 7-H, s, 2H).

Anal. Calcd. for $C_{21}H_{28}O_4$: C, 73.22; H, 8.19.

Found: C, 72.96; H, 8.20

and 3β-acetoxy-7α-hydroxy-5-androsten-17-one 63 (150 mg); m.p. 168-169°; $[\alpha]_D$ -66° (C 0.2, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 0.90 (18-$CH_3$, s, 3H), 1.04 (19-$CH_3$, s, 3H), 4.00 (7β-H, m, 1H), 4.60 (3α-H, m, 1H), 5.72 (6-H, d, 1H, J = 5.8 Hz).

Anal. Calcd. for $C_{21}H_{28}O_4$: C, 73.22; H, 8.19.

Found : C, 73.20; H, 8.32.

(c) Oxygenation of 3β,17β-diacetoxy-5-androstene 40

As before the diacetate 40 (1.75 g., 4.7 mmoles) was subjected to the photosensitized oxygenation, the mutarotation in chloroform, reduction with sodium iodide and chromatography on SilicaR (100 g). Elution with ether-benzene (1:49) yielded starting material (503 mg) identified by t.l.c. and melting point. Further elution gave 3β,17β-diacetoxy-5-androstene-7-one 111 (65 mg); m.p. 221-223° (lit.[107], m.p. 224-226°), and 3β,17β-diacetoxy-5-hydroxy-5α-androst-6-ene 56 (450 mg). M.p. 148-149°; $[\alpha]_D$ -13° (C 0.3, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 0.89 (18-$CH_3$, s, 3H), 0.99 (19-$CH_3$, s, 3H), 4.70 (17α-H, t, 1H), 5.28 (3α-H, m, 1H), 5.62 (6-H, 7-H, s, 2H).

Anal. Calcd. for $C_{23}H_{34}O_5$ : C, 70.74; H, 8.78.

Found : C, 70.74; H, 9.13.

Further elution yielded 3β,17β-diacetoxy-7α-hydroxy-5-androstene 65 (130 mg). M.p. 170-171° ; $[\alpha]_D$ -117° (C 0.12, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 0.80 (18-$CH_3$, s, 3H), 1.00 (19-$CH_3$, s, 3H), 2.00 (3β-OAc, s, 3H), 3.80 (7β-H, m, 1H), 4.62 (17α-H, m, 1H), 5.61 (6-H, d, 1H, J = 5.0 Hz).

Anal. Calcd. for $C_{23}H_{34}O_5$ : C, 70.74; H, 8.78.

Found : C, 70.42; H, 8.62.

(d) Synthesis and Oxygenation of 3β-Acetoxy-5-cholesten-19-ol 69

Cholesterol 7 (110 g., 0.28 moles) was acetylated with acetic anhydride (40 ml) in pyridine (250 ml) yielding, after crystallization of the crude product from acetone, pure 3β-acetoxy-5-cholestene 51 (84 g., 75%). M.p. 112-113° (lit.[108], m.p. 115°).

A solution of 3β-acetoxy-5-cholestene 51 (50 g., 0.12 moles) in dioxane (500 ml) was cooled in an ice-bath and after the addition of perchloric acid (25 ml), N-bromoacetamide (45 g., 0.3 moles) was added with stirring over a period of 1 hour. After another 30 minutes the reaction was stopped by the addition of a 30% solution of $Na_2SO_3$ (250 ml). The mixture was filtered and the precipitate washed with water and dried in vacuo. Crystallization of the crude product from ether-pet. ether 30-60, yielded 3β-acetoxy-5-bromo-6β-hydroxy-5α-cholestane 67 (20 g., 33%). M. p. 171-173° (lit.[109], m.p. 172-174°).

Lead tetraacetate (26.0 g., 59 mmoles) previously dried over $P_2O_5$, and calcium carbonate (14 g) were added to cyclohexane (700 ml) and this mixture was refluxed for 1 hour by means of a 500 watt lamp and was constantly stirred. To this mixture was added freshly sublimed iodine (7.4 g) and the bromohydrin 67 (6.0 g., 11.4 mmoles) and the reaction mixture was refluxed and stirred for another hour and was then filtered through Celite. The filtrate was washed with a 30% solution of $Na_2S_2O_3$ (300 ml) and water, and then dried. After evaporation of the solvent the crude product was crystallized from acetone, yielding pure 3β-acetoxy-5-bromo-6β,19-oxido-5α-cholestane 68 (3.70 g., 62%). M.p. 152-153°, (lit.[110a,b,50], m.p. 158-159°, 154-155°, 149°). This procedure was repeated and the products combined.

The bromo-oxide 68 (7.7 g., 14.7 mmoles) and zinc powder (50 g) in 95% ethanol (400 ml) was refluxed for 24 hours. After filtering the reaction mixture through Celite, evaporation of the filtrate and crystallization of the crude material from acetone, pure 3β-acetoxy-5-cholesten-19-ol 69 (4.80 g., 73%) was obtained. M.p. 118-120°, (lit.[110a], m.p. 120-121°).

3β-Acetoxy-5-cholesten-19-ol 69 (650 mg, 1.45 mmoles) and hematoporphyrin (5 mg) in pyridine (100 ml) were irradiated for 5 days with continuous bubbling of oxygen through the solution. Ether (100 ml) and charcoal were added and the mixture shaken, filtered and the filtrate evaporated. The residue was dissolved in chloroform (100 ml) and allowed to stand in the dark for 3 days. After evaporation of the chloroform the residue was treated with a solution composed of ethanol (30 ml), ether (20 ml), sodium iodide (2.5 g) and acetic acid (5 drops), for 24 hours. After the addition of water (50 ml) the mixture was extracted with ether (100 ml) and the ethereal solution was washed with a solution of $Na_2S_2O_3$ (0.1 N) and water, then dried and evaporated.

The crude product (550 mg) was chromatographed on SilicaR (300 g). The first compound (148 mg) was eluted with benzene and was identified by t.l.c. and m.p. as starting material 69. The second compound (53 mg) was eluted with ether-benzene (1:30) and identified as 3β-acetoxy-5,19-dihydroxy-5α-cholest-6-ene 71; m.p. 163-165°, (lit.[68], m.p. 163-165°). With ether-benzene (1:13) as eluent, a third compound (269 mg) was eluted and identified as 3β-acetoxy-5-cholestene-7α,19-diol 72; m.p. 153-155°; $[\alpha]_D$ -75.6 (C 0.5, $CHCl_3$); i.r. ($CHCl_3$) $\nu_{max}$ 3630 (free-OH), 3450 (bonded-OH), 1730 $cm^{-1}$ (acetate C=O); n.m.r. (60 Mc, $CDCl_3$) δ 0.73 (18-$CH_3$), 2.01 (3β-OAc, s, 3H), 3.84 (7β-H and 19-$CH_2$-, m, 3H), 4.70 (3α-H, m, 1H), 6.00 (6-H, d, 1H, $J_{6,7}$ = 5.5 Hz).

Anal. Calcd. for $C_{29}H_{48}O_4$: C, 75.60; H, 10.50.

Found: C, 75.44; H, 10.50.

(i) Synthesis of 3β, 7α, 19-triacetoxy-5-cholestene 73 and 3β, 7β, 19-triacetoxy-5-cholestene 74

3β-Acetoxy-5-cholestene-7α, 19-diol 72 (23 mg) was acetylated with acetic anhydride (5 drops) in pyridine (3 ml). Workup as usual afforded as an oil 3β, 7α, 19-triacetoxy-5-cholestene 73 (23 mg). $[\alpha]_D$ -206° (C 1.2, $CHCl_3$); n.m.r. (100 Mc, $CDCl_3$) δ 0.70 (18-$CH_3$, s), 2.03 (3β, 7α, 19-OAc, s, 9H), 3.89, 4.01, 4.54, 4.66 (19-$CH_2$-, q, 2H), 4.65 (3α-H, m, 1H), 5.01 (7β-H, t, 1H), 5.84 (6-H, d, 1H, J = 5.0 Hz).

3β-Acetoxy-5-cholesten-19-ol 69 (1 g) was acetylated with acetic anhydride (1 ml) in pyridine (10 ml). The usual workup afforded 3β, 19-diacetoxy-5-cholestene 75 (1 g) as an oil. $[\alpha]_D$ -54° (C 0.5, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 0.70 (18-$CH_3$, s), 2.01 (3β, 19-OAc, 6H), 3.86, 4.06, 4.38, 4.58 (19-$CH_2$-, q, 2H), 4.62 (3α-H, m, 1H), 5.63 (6-H, d, 1H). Anal. Calcd. for $C_{31}H_{50}O_4$ : C, 76.50; H, 10.36.

Found : C, 76.38; H, 10.28.

To the diacetate 75 (500 mg, 1 mmole) in glacial acetic acid (4 ml) at 51°C was added with stirring, chromic anhydride (0.38 g., 3.8 mmoles). The temperature was maintained between 51° and 53° for two hours. Ethanol (5 ml) and water (20 ml) were added and the green solution was extracted with ether. The ethereal solution was washed with $NaHCO_3$ solution and water, then dried and evaporated. The residue (386 mg) was chromatographed on SilicaR (10 g) with benzene as the eluent. The chromatography afforded 3β, 19-diacetoxy-5-cholesten-7-one 112 (200 mg) as an oil. U.v. (EtOH) $\lambda_{max}$ 239 mμ, ε 12,300; i.r. ($CHCl_3$) $\nu_{max}$ 1680 (α, β-unsaturated C=O) 1730 $cm^{-1}$ (ester C=O); n.m.r. (100 Mc, $CDCl_3$) δ 0.72 (18-$CH_3$, s, 3H), 2.00 (3β, 19-OAc, s, 6H), 4.04, 4.17, 4.64, 4.76 (19-$CH_2$-, q, 2H), 4.62 (3α-H, m, 1H), 5.88 (6-H, s, 1H).

Anal. Calcd. for $C_{31}H_{48}O_5$: C, 74.38; H, 9.66

Found: C, 73.85; H, 9.60.

The ketone 112 (160 mg) in anhydrous ether (30 ml) was stirred and refluxed with lithium aluminum hydride (0.08 g) for 2 hours. The excess hydride was destroyed by the addition of methanol and the mixture was filtered and the filtrate evaporated. The residue was dissolved in ether and the solution was washed with HCl and water, dried and evaporated. The crude product (113 mg) was crystallized from acetone yielding pure 5-cholestene-3β, 7β, 19-triol 113. M. p. 158-159°; $[\alpha]_D \sim 0°$ (C 0.14, MeOH); n. m. r. (100 Mc, $CDCl_3$ + DMSO) δ 0.76 (18-$CH_3$, s) 5.58 (6-H, s, 1H); (region between δ 3.0 and 4.0 masked by DMSO).

Anal. Calcd. for $C_{27}H_{46}O_3$: C, 77.46; H, 11.08.

Found: C, 76.55*; H, 10.99.

The triol 113 (65 mg) was acetylated with acetic anhydride (5 drops) in pyridine (3 ml) and the usual workup afforded 3β, 7β, 19-triacetoxy-5-cholestene 74 (67 mg) as an oil. $[\alpha]_D$ -8.2° (C 0.83, $CHCl_3$); n. m. r. (100 Mc, $CDCl_3$) δ 0.73 (18-$CH_3$, s, 3H), 2.02, 2.08 (3β, 7β, 19-OAc, s, s, 9H), 3.89, 4.01, 4.55, 4.67 (19-$CH_2$-, q, 2H), 4.62 (3α-H, m, 1H), 5.00 (7α-H, d, 1H), 5.50 (6-H, d, 1H, J ≈ 2.0 Hz).

(e) Oxygenation of 3β-Hydroxy-5-androsten-17-one 8

3β-Hydroxy-5-androsten-17-one 8 (5.0 g) and hematoporphyrin (10 mg) in pyridine (200 ml) were oxygenated for 4 days. After the usual workup the residue was dissolved in chloroform and was allowed to stand in the dark for 24 hours. After the usual reduction with sodium iodide and acetylation of the mixture with acetic anhydride the crude product (4.5 g)

* Carbon analysis is not acceptable, however the spectral data of the acetylated derivative 74 confirm the structure assigned.

was chromatographed on SilicaR (250 g) with benzene as the eluent. The chromatography yielded 3β-acetoxy-5-androsten-17-one 34 (2.8 g), 3β-acetoxy-5-hydroxy-5α-androst-6-en-17-one 57 (550 mg), [identified as before, Experimental Section 3(b)], 3β-acetoxy-5-androstene-7,17-dione 114 (350 mg), m.p. 183-185° (lit.[111], m.p. 184-186°) and 3β,7α-diacetoxy-5-androsten-17-one 64 (600 mg). M.p. 169-170° (lit.[45b], m.p. 168-170°), n.m.r. (60 Mc, $CDCl_3$) δ 0.90 (18-$CH_3$, s, 3H), 2.00 (3β,7α-OAc, s, 6H), 4.70 (3α-H, m, 1H), 5.10 (7β-H, m, 1H), 5.64 (6-H, d, 1H, J = 5.0 Hz).

(f) Oxygenation of 3β-Acetoxy-19-hydroxy-5-androsten-17-one 37, 3β,19-diacetoxy-5-androsten-17-one 38 and 3β,17β,19-triacetoxy-5-androstene 44

3β-Acetoxy-19-hydroxy-5-androsten-17-one 37 (750 mg), 3β,19-diacetoxy-5-androsten-17-one 38 (680 mg) and 3β,17β,19-triacetoxy-5-androstene 44 (500 mg) were separately subjected to the photosensitized oxygenation reaction. Each reaction was worked up as previously described and the products dissolved in chloroform* and allowed to stand for 2 to 4 days after which they were treated with sodium iodide (2.5 g) in ethanol (25 ml) and acetic acid (5 drops). After the usual workup the residue from the reaction of compound 37 was crystallized from acetone-pet. ether (30-60) affording 3β-acetoxy-5,19-dihydroxy-5α-androst-6-en-17-one 76 (307 mg). M.p. 171-174°; $[\alpha]_D$ -70.5 (C 0.13, $CHCl_3$); i.r. (nujol) $\nu_{max}$ 3560 (-OH), 1735 ( C=O, 5-membered ring), 1724 $cm^{-1}$ ( C=O, ester), n.m.r. (60 Mc, $CDCl_3$ + DMSO) δ 0.94 (18-$CH_3$, s, 3H), 1.98 (3β-OAc, s, 3H),

* When acetone, methanol or tetrahydrofuran was used instead of chloroform the same results were obtained. Conc. HCl (3-5 drops) in the chloroform methanol or tetrahydrofuran did not aid in rearranging the 5α-hydroperoxide. (t.l.c. evidence). The addition of KCN to the chloroform aided the rearrangement but the productsobtained were $\Delta^5$-7-keto compounds (t.l.c.).

3.70 (19-$CH_2$-, m, 2H), 5.26 (3α-H, m, 1H), 5.43, 5.61, 5.68, 5.85 (6-H, 7-H, q, 2H).

Anal. Calcd. for $C_{21}H_{30}O_5$ : C, 69.58; H, 8.34.

Found : C, 69.46; H, 8.34.

The residue (400 mg) from the mother liquor was chromatographed on SilicaR (40 g) with ether-benzene (1:20) as the eluent. The first compound eluted (360 mg) was identified (t.l.c., m.p.) as starting material 37. The second compound eluted was 3β-acetoxy-5,19-dihydroxy-5α-androst-6-en-17-one 76 (40 mg) identified by t.l.c. and mixed melting point with the material previously isolated by crystallization.

The residues from the reactions of compounds 38 and 44 (650 mg and 380 mg respectively) were chromatographed on SilicaR (200 g and 130 g respectively) with ether-benzene (1:9) as the eluent in the former case and benzene as the eluent in the latter case.

In the former case the chromatography afforded starting material 38 (316 mg) identified by t.l.c. and melting point and 3β,19-diacetoxy-5-hydroxy-5α-androst-6-en-17-one 77 (210 mg). M.p. 205-208°; $[\alpha]_D$ +10° (C 0.5, $CHCl_3$); i.r. ($CHCl_3$) $\nu_{max}$ 3480 (bonded-OH), 3610 (free-OH), 1730 $cm^{-1}$ (ester C=O), n.m.r. (100 Mc, $CDCl_3$) δ 0.94 (18-$CH_3$, s, 3H), 2.03, 2.11 (3β,19-OAc, s, s, 6H), 4.03, 4.15, 4.50, 4.62 (19-$CH_2$-, q, 2H), 5.22 (3α-H, m, 1H), 5.61, 5.71, 5.79, 5.89 (6-H, 7-H, q, 2H).

Anal. Calcd. for $C_{23}H_{32}O_6$ : C, 68.29; H, 7.97.

Found : C, 68.13; H, 8.28.

In the latter case the chromatography afforded starting material (220 mg), (t.l.c., m.p.), and 3β,17β,19-triacetoxy-5-hydroxy-5α-androst-6-ene 78 (150 mg). M.p. 174.5-175.5°; $[\alpha]_D$ -45.2° (C 0.87, $CHCl_3$); n.m.r. (100 Mc, $CDCl_3$) δ 0.85 (18-$CH_3$, s, 3H), 2.01, 2.03, 2.09 (3β,17β,19-OAc, s, s, s, 9H), 3.99, 4.11, 4.45, 4.57 (19-$CH_2$-, q, 2H), 4.60 (17α-H, m, 1H), 5.19 (3α-H, m, 1H), 5.51, 5.61, 5.66, 5.76 (6-H, 7-H, q, 2H).

Anal. Calcd. for $C_{25}H_{36}O_7$ : C, 66.94; H, 8.09.

Found : C, 66.92; H, 8.11.

4. Peracid Reactions with 3β, 7α-Diacetoxy-$\Delta^5$-Steroids

(a) Reaction with 3β, 7α-diacetoxy-5-cholestene 61

3β, 7α-Diacetoxy-5-cholestene 61 (100 mg) was treated with a solution of monoperphthalic acid in ether (10 ml, 1.0 M) at 5° for 24 hours. The reaction mixture was passed through a Florisil column (50 g) with ether as the eluent. The phthalic acid-free product (100 mg) was then chromatographed on SilicaR (15 g) with benzene as the eluent. The first compound (28 mg) eluted was identified as starting material 61 by t.l.c. and m.p. The second compound (15 mg) which was not crystallizable was identified as 3β, 7α-diacetoxy-5, 6β-oxido-5β-cholestane 81. $[\alpha]_D$ +3°, (C 0.12, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 2.01, 2.07 (3β, 7α-OAc), 3.10 (6α-H, d, 1H, J = 3.6 Hz), 4.74 (3α-H, m, 1H), 5.18 (7β-H, m, 1H). The third compound (60 mg) eluted was identified as 3β, 7α-diacetoxy-5, 6α-oxido-5α-cholestane 80. M.p. 203-204°; $[\alpha]_D$ -125° (C 0.12, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 2.02, 2.10 (3β, 7α-OAc), 3.35 (6β-H, d, 1H, J = 4.5 Hz), 5.0 (7β-H, 3α-H, m, m, 2H).

Anal. Calcd. for $C_{31}H_{50}O_5$ : C, 74.06; H, 10.03.

Found : C, 74.07; H, 10.12.

(b) Reaction with 3β, 7α, 17β-triacetoxy-5-androstene 58

3β, 17β-Diacetoxy-7α-hydroxy-5-androstene 65 (820 mg) was acetylated with acetic anhydride (3 ml) in pyridine (20 ml). The usual work-up afforded an oil (830 mg) which crystallized from pentane to give pure 3β, 7α, 17β-triacetoxy-5-androstene 58. M.p. 163-164°; $[\alpha]_D$ -220° (C 0.2, $CHCl_3$); n.m.r. (100 Mc, $CDCl_3$) δ 0.79 (18-$CH_3$, s, 3H), 0.99 (19-$CH_3$, s, 3H), 4.65 (17α-H, m, 1H), 4.97 (7β-H, t, 1H), 5.59 (6-H, d, 1H, J = 5.0 Hz).

Anal. Calcd. for $C_{25}H_{36}O_6$ : C, 69.42; H, 8.39.

Found : C, 69.73; H, 8.47.

The triacetate 58 (450 mg) was treated with monoperphthalic acid in ether (40 ml, 1.0 M) at room temperature for 24 hours. The mixture was poured onto a Florisil column (200 g) and the reaction products eluted with ether. The ether solution was washed with $NaHCO_3$ solution and water, then dried and evaporated. The residue (430 mg) was chromatographed on SilicaR (50 g) with ether-benzene (1:9) as the eluent. The first compound eluted was starting material 58 (100 mg) (t.l.c. and m.p.). The second compound (80 mg) was identified as 3β,7α,17β-triacetoxy-5,6α-oxido-5α-androstane 82. M.p. 238-239°; $[\alpha]_D$ -154° (C 0.11, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 0.79 (18-$CH_3$, s, 3H), 1.13 (19-$CH_2$), 2.02, 2.06, 2.10 (3β,7α,17β-OAc, s, s, s, 9H), 3.30 (6β-H, d, 1H, J≈4.5 Hz), 4.40-5.20 (3α-H, 7β-H, 17α-H, m, 3H).

Anal. Calcd. for $C_{25}H_{36}O_7$ : C, 66.94; H, 8.09.

Found : C, 66.80; H, 7.78.

The third compound eluted (250 mg) was identified as 3β,7α,17β-triacetoxy-5,6β-dihydroxy-5α-androstane 83. M.p. 250-252°; $[\alpha]_D$ -16° (C 0.07, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 0.86 (18-$CH_3$, s, 3H), 1.20 (19-$CH_3$, s), 2.10, 2.15, 2.17 (3β,7α,17β-OAc, s, s, s, 9H), 2.80 (5α,6β-OH, b, 2H), 3.62 (6α-H, d, 1H), 4.40-5.10 (3α,7β,17α-H, m, 3H) (addition of $D_2O$ - signal at 2.80 disappears).

5. Preparation and Reactions of Hydrazones

(a) N, N-Dimethylhydrazone

A mixture of 3β-acetoxy-5-cholesten-7-one 84 (1.0 g., 2.5 mmoles) and N, N-dimethylhydrazine (1.0 ml, 13 mmoles) in absolute ethanol (25 ml) was refluxed for 20 hours. The solvent was evaporated in vacuo and the crude material was chromatographed on SilicaR (100 g) with benzene as the eluent. The second* compound eluted (500 mg) was identified as 3, 5-cholestadiene-7-N, N-dimethylhydrazone 87. M.p. 88-90°; $[\alpha]_D$ -416° (C 0.04, $CHCl_3$); i.r. ($CHCl_3$) $\nu_{max}$ 1623, w, (C=N stretch) 1604, m, $cm^{-1}$ (C=C stretch); u.v. ($CHCl_3$) $\lambda_{max}$ 277 mμ (ε 21,000) n.m.r. (60 Mc, $CDCl_3$) δ 2.42 ($-N(CH_3)_2$, s, 6H), 6.18 (3-H, 4-H, m, 2H), 6.50 (6-H, s, 1H).

Anal. Calcd. for $C_{29}H_{48}N_2$ : C, 82.01; H, 11.39; N, 6.60.

Found : C, 82.35; H, 11.38; N, 6.57.

The third compound eluted was starting material 84 (150 mg) (t.l.c. and m.p.).

The reaction was repeated using 0.6 ml of N, N-dimethylhydrazine (2 mole excess). In this case the first compound eluted (300 mg) was 3, 5-cholestadien-7-one 86. M.p. 117-118° (lit.[112], m.p. 114.5°); n.m.r. (60 Mc, $CDCl_3$) δ 5.63 (6-H, s, 1H), 6.18 (3-H, 4-H, s, 2H). The second compound eluted (100 mg) was identified (t.l.c. and m.p.) as 3, 5-cholestadiene-7-N, N-dimethylhydrazone 87. The third compound eluted (200 mg)** was identified (t.l.c. and m.p.) as starting material 84.

---

* Some of the previous fractions off the column were inadvertently thrown away. The first compound eluted (~100 mg) was identified by n.m.r. and melting point as 3, 5-cholestadien-7-one 86.

** 500 mg of compound identified as starting material had precipitated from the ethanol after the reflux period and cooling of the reaction mixture.

When the reaction was repeated with 5.0 ml of N, N-dimethyl hydrazone the compounds 86, 87 and 84 were obtained in yields of 100 mg, 800 mg, and 100 mg respectively.

(b) Hydrazine Hydrate

A mixture of 3β-acetoxy-5-cholesten-7-one 84 (1g) and hydrazine hydrate (0.5 ml) in ethanol (25 ml) was refluxed for 2 hours. The solvent was evaporated and the crude hydrazone 85 was dried for 24 hours in vacuo. N.m.r. (60 Mc, $CDCl_3$) $\delta$ 5.70 (6-H, s); starting material 84 . 6.40 (6-H, s) hydrazone 85.

(i) Reaction of hydrazone 85 with lead tetraacetate

A solution of lead tetraacetate (600 mg) in methylene chloride (20 ml) was cooled in an ice-bath to 5°. A solution of crude hydrazone 85 (650 mg) in methylene chloride (25 ml) was added dropwise to the lead tetraacetate solution over a period of 30 min. The mixture was allowed to warm up to room temperature. Water (20 ml) was added and the mixture was filtered. The organic layer was separated and washed with $NaHCO_3$ solution, water, then dried and evaporated. The crude material (650 mg) was chromatographed on SilicaR (100 g) with benzene as the eluent. The first component eluted (60 mg) was a mixture of hydrazone 85 and possibly 3,5-cholestadien-7-one 86 (n.m.r. analysis showed the signals for the 3-H, 4-H and 6-H indicative of the dienone 86 at $\delta$ 6.2 (2H) and 5.63 (1H) and a signal at $\delta$ 6.40 (1H) for the 6-H of the hydrazone 85). The second component eluted (350 mg) was a 1:1 mixture of the 7α- and 7β-acetoxy derivatives of 3β-acetoxy-5-cholestene 51. [N.m.r. analysis showed signals at $\delta$ 5.30 (1H) ($J \approx 2$Hz) and 5.62 (1H) ($J = 5.0$ Hz) indicative of the 6-H in 3β, 7β-diacetoxy-5-cholestene 62

and in 3β, 7α-diacetoxy-5-cholestene 61 respectively. Both signals had the same integral value]. The third compound eluted (220 mg) was identified (t.l.c. and m.p.) as 3β-acetoxy-5-cholesten-7-one 84, which was part of the original crude hydrazone.

6. Conversion of 3β,17β-Diacetoxy-5-hydroxy-5α-androst-6-ene 56 to 3β,17β-Diacetoxy-7α-hydroxy-5-androstene 65

3β,17β-Diacetoxy-5-hydroxy-5α-androst-6-ene 56 (1.0 g) was treated with a solution of monoperphthalic acid in ether (50 ml, 1.0 M), for 24 hours at room temperature. The reaction mixture was poured onto a Florisil column (200 g) and the products were eluted with ether. After evaporation of the ether the crude material was crystallized from ether-pet. ether (30-60), which afforded pure 3β,17β-diacetoxy-5-hydroxy-6α,7α-oxido-5α-androstane 88 (800 mg). M.p. 201.5-202°; $[\alpha]_D \sim 0°$ (C 0.13, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 0.82 (18-$CH_3$, s), 0.88 (19-$CH_3$, s), 2.02 (3β,17β-OAc, s, 6H), 2.93 (6β-H, 7β-H, d, 2H), 3.10 (-OH, b, 1H), 4.60 (17α-H, m, 1H), 5.20 (3α-H, m, 1H).

Anal. Calcd. for $C_{23}H_{34}O_6$ : C, 67.95; H, 8.43.

Found : C, 68.07; H, 8.37.

The epoxide 88 (300 mg) in chloroform (20 ml) and HBr (9 ml, 50%) were stirred at room temperature for 5 hours. The solution was washed with $NaHCO_3$ solution, water, then dried and evaporated. The residue was crystallized from ether-pet. ether (30-60) affording 3β,17β-diacetoxy-5,7α-dihydroxy-6β-bromo-5α-androstane 89 (200 mg). M.p. 167-171° ; $[\alpha]_D$ -68° (C 0.11, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$), δ 3.60 (-OH, b, 2H), 4.03 (6α-H, 7β-H, m, 2H), 4.62 (17α-H, m, 1H), 5.05 (3α-H, m, 1H).

Anal. Calcd. for $C_{23}H_{35}O_6Br$ : C, 56.67; H, 7.23; Br, 16.80

Found : C, 56.12; H, 7.08; Br, 16.51.

The bromohydrin 89 (150 mg) and zinc powder (2g) in acetic acid (10 ml) (80%) were refluxed for 3 hours. The mixture was filtered through Celite and water (10 ml) was added to the filtrate. The filtrate was extracted with ether and the ethereal solution was washed with $NaHCO_3$ solution, water, and then dried and evaporated. The residue (43 mg) was chromatographed on SilicaR with ether-benzene (1:9) as the eluent. The chromatography afforded a 3β,17ξ,17β-triacetate-mixture (18 mg), (n.m.r. analysis), and 3β,17β-diacetoxy-7α-hydroxy-5-androstene 65 (125 mg) identified by comparison of t.l.c., melting point and n.m.r. with authentic sample [Experimental Section 2(c)].

10 mg portions of the bromohydrin 89, 3β,17β-diacetoxy-5-hydroxy-5α-androst-6-ene 56 and 3β,17β-diacetoxy-7α-hydroxy-5-androstene 65 were separately treated with acetic acid (5 ml, 80%) and the reactions followed by t.l.c. The bromohydrin was unchanged after 24 hours. The t.l.c. of the reaction of the 5α-hydroxy compound 56 showed evidence of 7α-hydroxy compound 65 and 3β,7ξ,17β-triacetoxy-5-androstenes 58 and 92. The t.l.c. of the reaction of the 7α-hydroxy compound 65 showed starting material 65 and triacetates 58 and 92.

(a) Synthesis of 3β-Acetoxy-5-hydroxy-7α-bromo-6β,19-oxido-5α-androstan-17-one 90 and 3β-Acetoxy-5-hydroxy-6β,19-oxido-5α-androst-7-en-17-one 91

---

3β-Acetoxy-5,19-dihydroxy-5α-androst-6-en-17-one 76 (100 mg) and N-bromosuccinimide (160 mg) in t-butyl alcohol (5.0 ml) and water (2.0 ml) were stirred at room temperature for 0.5 hours. After the addition of $NaHSO_3$ (0.1 g) and water (10 ml) the mixture was extracted with ether and the ethereal solution was dried and evaporated. Trituration

of the residue with ether or acetone afforded crystals of 3β-acetoxy-5-hydroxy-7α-bromo-6β,19-oxido-5α-androstan-17-one 90 (80 mg). M.p. 208-209°; $[\alpha]_D$ -24.0° (C 0.1, $CHCl_3$); i.r. ($CHCl_3$) $\nu_{max}$ 3540 (-OH), 1720 $cm^{-1}$ (ester C=O, shoulder due to C=O of 5-membered ring); n.m.r. (60 Mc, $CDCl_3$) δ 0.95 (18-$CH_3$, s, 3H), 2.00 (3β-OAc, s, 3H), 3.62, 3.80, 3.84, 4.02 (19-$CH_2$-, q, 2H), 3.90 (6α-H, 1H), 4.24 (7β-H, t, 1H), 5.02 (3α-H, m, 1H).

Anal. Calcd. for $C_{21}H_{29}O_5Br$: C, 57.13; H, 6.63; Br, 18.11.

Found : C, 57.05; H, 6.64; Br, 17.86.

The above bromo compound 90 (50 mg), lithium carbonate (50 mg) and lithium chloride (25 mg) were added to N,N-dimethylformamide (5 mls) and this mixture was refluxed for 3 hours. Water (10 ml) was added and the mixture extracted with ether. The ethereal solution was dried and evaporated. Trituration of the residue with ether afforded 3β-acetoxy-5-hydroxy-6β,19-oxido-5α-androst-7-en-17-one 91 (30 mg). M.p. 212-214° ; $[\alpha]_D$ +49.6° (C 0.12, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$), δ 0.92 (18-$CH_3$, s, 3H), 2.10 (3β-OAc, s, 3H), 3.90 (6α-H, d, 1H), 4.02 (19-$CH_2$-, s, 2H), 5.06 (3α-H, m, 1H), 5.66 (7-H, d, 1H, J = 5.5 Hz).

Anal. Calcd. for $C_{21}H_{28}O_5$ : C, 69.97; H, 7.83.

Found : C, 70.04; H, 7.86.

7. Acid Treatment of Steroidal Allylic Alcohols

(a) Strong Acid

Through a solution of 3β, 5-dihydroxy-5α-cholest-6-ene 54 (160 mg) in ether was bubbled HCl gas for 1 hour. The ether solution was washed with $NaHCO_3$ solution and water, then dried and evaporated. The mixture was separated by thick layer chromatography (Silica gel G) with benzene as the eluent. Spraying part of the plate with 50% $H_2SO_4$ revealed the presence of at least seven highly mobile compounds. The most mobile component and the least mobile component were isolated in yields of 48 mg and 24 mg respectively. U.v. (hexane) $\lambda_{max}$ 238, 305, 317, 333 mμ ( ε 6,000 ); and 240, 299, 308, 323 mμ (ε 10,000 respectively). Both components melted in the range 75-95°.

(b) Mild Acid*

(i) 20 mg portions of 3β-acetoxy-5-hydroxy-5α-cholest-6-ene 55, 3β-acetoxy-7α-hydroxy-5-cholestene 59 and 3β-acetoxy-7β-hydroxy-5-cholestene 60 were dissolved in acetic acid (10 ml, 80%) and ether (0.5 ml) and the three solutions were stirred at room temperature. The reactions were followed by t.l.c. and the results are given in TABLE 5 (p. 62).

(ii) A solution of 3β-acetoxy-5-hydroxy-5α-cholest-6-ene 55 (100 mg) in acetic acid (10 ml, 80%) and ether (0.5 ml) was stirred at room temperature for 2 hours. Water (20 ml) was added and the solution extracted with ether. The ethereal solution was washed with $NaHCO_3$ solution and water, then dried and evaporated. The residue (90 mg) was

*The products from the reactions in this section were identified by comparison of their n.m.r. spectra, t.l.c.'s and m.p.'s with authentic samples obtained from the reactions in Experimental Section 3.

chromatographed on SilicaR (5 g) with benzene as the eluent. The first compound eluted (15 mg) was identified as 3β, 7α-diacetoxy-5-cholestene 61. The second compound eluted (72 mg) was identified as 3β-acetoxy-7α-hydroxy-5-cholestene 59.

(iii) A solution of 3β-acetoxy-7α-hydroxy-5-cholestene 59 (100 mg) in acetic acid (10 ml, 80%) and ether (0.5 ml) and a similar solution of the 7β-isomer 60 (90 mg) were stirred at room temperature for 6 hours. Workup and chromatography as usual afforded in both cases an unidentified compound A ( 3 - 5 mg), an epimeric mixture of 3β, 7ξ-diacetoxy-5-cholestene 61 and 62 (52 - 60 mg) and the respective starting materials 59 and 60 (20 - 30 mg).

(iv) Four solutions [80% acetic acid (20 ml) and ether (1.0 ml)] containing 50 mg portions of 3β-acetoxy-7α-hydroxy-5-cholestene 59, 3β, 7α-diacetoxy-5-cholestene 61, 3β-acetoxy-5, 7-cholestadiene 93 and 3β-acetoxy-4, 6-cholestadiene 94 were heated on steam baths for 2 hours. The 5, 7-diene 93 proved to be unreactive (t.l.c.). The usual workup and chromatography of the remaining three reactions afforded polyene mixtures. M.p.'s 56-65°; u.v.'s (hexane) $\lambda_{max}$ 296-298, 309-310, 323-324 mμ (ε 1000).

(v) A solution of 3β, 17β-diacetoxy-5-hydroxy-5α-androst-6-ene 56 (100 mg) in acetic acid (10 ml, 80%) was stirred at room temperature for 2 hours. The usual workup and chromatography afforded 3β, 7α, 17β-triacetoxy-5-androstene 58 (17 mg) and 3β, 17β-diacetoxy-5-androstene 65 (75 mg). When a similar reaction mixture was heated on a steam bath for 2 hours a polyene was obtained after workup and chromatography. M.p. 130-131°; n.m.r. (60 Mc, $CDCl_3$) δ 0.83 (18-$CH_3$, s, 3H),

0.95 (19-$CH_3$, s, 3H), 2.01 (17β-OAc, s, 3H), 4.55 (17α-H, m, 1H), 5.30-6.02 (olefinic protons, m, ~5H); u.v. (hexane) $\lambda_{max}$ 297, 309, 324 mμ (ε 10,000).

Anal. Calcd. for $C_{21}H_{28}O_2$ : C, 80.73; H 9.03.

Found : C, 80.72; H, 9.07.

(vi) A solution of 3β-acetoxy-5-hydroxy-5α-androst-6-en-17-one 57 (100 mg) in acetic acid (10 ml, 80%) was stirred at room temperature for 2 hours. The usual workup and chromatography afforded 3β, 7α-diacetoxy-5-androsten-17-one 64 (15 mg) and 3β-acetoxy-7α-hydroxy-5-androsten-17-one 63 (82 mg).

8. Allylic Rearrangement of 3β,19-Diacetoxy-5-hydroxy-5α-androst-6-en-17-one 77

A solution of 3β,19-diacetoxy-5-hydroxy-5α-androst-6-en-17-one 77 (200 mg) in acetic acid (20 ml, 80%) was stirred at room temperature for 1.5 hours. The usual workup and chromatography on SilicaR (10 g) with ether-benzene (1:9) as eluent afforded 3β,7α,19-triacetoxy-5-androsten-17-one 95 (25 mg) as an oil, $[\alpha]_D$ -103° (C 0.66, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 0.87 (18-$CH_3$, s, 3H), 1.99 (3β,19,7α-OAc, s, 9H), 3.80, 4.00, 4.55, 4.75 (19-$CH_2$-, q, 2H), 4.80 (3α-H, m, 1H), 5.10 (7β-H, t, 1H), 5.82 (6-H, d, 1H, J = 5.0 Hz), starting material (33 mg) and 3β,19-diacetoxy-7α-hydroxy-5-androsten-17-one 94 (123 mg). Crystallization of the latter compound from ether-pet.ether (30-60) afforded an analytical sample. M.p. 139-140°; $[\alpha]_D$ -94.7° (C 0.3, $CHCl_3$); n.m.r. (60 Mc, $CDCl_3$) δ 0.89 (18-$CH_3$, s, 3H), 2.00 (3β,19-OAc, s, 6H), 3.80, 4.00, 4.50, 4.70 (19-$CH_2$-, q, 2H), 3.90 (7β-H, m, 1H), 4.60 (3α-H, m, 1H), 5.86 (6-H, d, 1H, J = 5.5 Hz).

Anal. Calcd. for $C_{23}H_{32}O_6$ : C, 68.29; H, 7.97.

Found : C, 67.81; H, 8.27.

9. Attempted Synthesis of 1α-Hydroxy-$\Delta^4$-3-keto-androgens

(a) $\Delta^{1,4}$-3-Keto Androgens

(i) A mixture of 17β-acetoxy-4-androsten-3-one 97 (500 mg) and DDQ (350 mg) in benzene (25 ml) was refluxed for 24 hours. The mixture was cooled and filtered, and the filtrate was washed with dilute NaOH and water, then dried and evaporated. The residue was crystallized from ether-pet. ether (30-60), yielding 17β-acetoxy-1,4-androstadien-3-one 98 (320 mg); m.p. 152-153° (lit.[113], m.p. 151-152°).

A mixture of the dienone 98 (160 mg) in methanol (12 ml) and methylene chloride (12 ml) was cooled in an ice bath to 5°. Hydrogen peroxide (3 ml, 30%) and 1N sodium hydroxide (6 ml) were added and the solution maintained below 10°. The reaction was followed by t.l.c. After 24 hours (reaction kept in fridge at 5°) water (20 ml) was added and the mixture was extracted with methylene chloride. The extract was washed with water, dried and evaporated and the residue (150 mg) was chromatographed on SilicaR (20 g) with ether-benzene (1:19) as the eluent. The first compound eluted (75 mg) was 17β-acetoxy-1β,2β;4β,5-dioxido-5β-androstan-3-one 100. M.p. 220-221°; $[\alpha]_D$ +56° (C 0.27, $CHCl_3$); (lit.[92], m.p. 213-215°; $[\alpha]_D$ +65°). The second compound eluted (74 mg) was 17β-hydroxy-1β,2β;4β,5-dioxido-5β-androstan-3-one 99; m.p. 236-238°. The diepoxide 99 (10 mg) was acetylated with acetic anhydride in pyridine and after the usual workup and crystallization from acetone-hexane yielded the diepoxide 100; m.p. 220-221°.

Identical results were obtained at room temperature with methanol as the solvent. There was no reaction at temperatures below -5° other than slow hydrolysis of the 17β-acetoxy group.

(ii) Benzene (50 ml) was refluxed for 1 hour through a Soxhlet extractor equipped with a calcium carbide thimble. 17β-Acetoxy-1,4-androstadien-3-one 98 (180 mg), ethylene glycol (0.4 ml) and p-toluenesulphonic acid (10 mg) were added and the mixture refluxed for 3 hours. The benzene solution was cooled and washed with $NaHCO_3$ solution and water, then dried and evaporated. The residue was identified by n.m.r. as starting material 98. The same results were obtained when less catalyst was used and also when toluene was substituted for benzene. Prolongation of the refluxing was also ineffective.

(b) $\Delta^{1,4,6}$-3-Keto-Androgens

(i) A mixture consisting of 17β-acetoxy-4-androstene-3-one 97 (5.65 g), triethylorthoformate (5.6 ml) and p-toluenesulphonic acid (100 mg) in dioxane (56 ml) was stirred at room temperature for 2.5 hours. Water (65 ml) and pyridine (10 ml) were added and the mixture cooled in an ice bath and then filtered. The crude precipitate was crystallized from methanol containing a few drops of pyridine yielding pure 3-ethoxy-17β-acetoxy-3,5-androstadiene 101 (4.3 g). M.p. 130-132° (lit.[114], m.p. 129-132°).

The enol ether 101 (2.6 g) in dry acetone (80 ml) was treated rapidly ( 5 min) with a solution of DDQ (4.34 g) in dry acetone (70 ml). After stirring at room temperature for 10 minutes the mixture was poured onto an alumina column (200 g, activity Grade I) and the column was eluted with acetone until the yellow coloured band had been completely removed off the column. The yellow coloured acetone fraction was decolorized with charcoal and was evaporated. Crystallization of the crude material from acetone-hexane afforded pure 3β-acetoxy-1,4,6-androstatrien-3-one 102 (1.05 g). M.p. 149-150° (lit.[115], m.p. 151-153°).

A solution of the trienone 102 (1.3 g) in methylene chloride (10 ml) and methanol (20 ml) was cooled to -10° in an acetone-dry ice bath. 7N NaOH (0.6 ml) and hydrogen peroxide (2.25 ml, 30%) were added and the mixture placed in a fridge at 4° for 20 hours. The mixture was neutralized with dilute acetic acid and water (20 ml) was added. After filtering, the crude product was crystallized from acetone-hexane affording pure 17β-acetoxy-1α,2α-oxido-4,6-androstadien-3-one 103 (1.1 g). M.p. 204-205° (lit.[95], m.p. 205-206°).

The monoepoxide 103 (200 mg) was dissolved in benzene (10 ml). To this solution was added freshly prepared* catalyst (5% $PdCl_2/SrCO_3$) and the mixture was connected to a hydrogenation apparatus. After the expulsion of air, hydrogen was introduced into the system (atmospheric pressure) and the hydrogenation was carried out at room temperature for 1.5 hours.

The mixture was filtered and the benzene evaporated. Crystallization of the residue from acetone-hexane yielded 17β-acetoxy-1α,2α-oxido-4-androsten-3-one 104 (200 mg). M.p. 167-16 ° (lit.[92], m.p. 167-168°).

The epoxide 104 (150 mg) in dioxane (2 ml) was treated with triethylorthoformate (0.2 ml) and p-toluenesulphonic acid (5 mg) at room temperature for 30 minutes. The usual workup afforded only starting material. The addition of excess triethylorthoformate was ineffective. When the reaction was heated the epoxide appeared (t.l.c.) to decompose into several compounds.

---

* The catalyst was prepared by dissolving palladium in conc. $HNO_3$ and evaporating the acid solution to dryness. The residue was picked up in HCl and the resulting mixture was evaporated to dryness. This step was repeated 5 times. The $PdCl_2$ thus obtained was mixed with the required amount of $SrCO_3$ and a sludge of these compounds was made by the addition of water. The intimately mixed sludge was placed in an oven at 100° for 2 hours, thus affording the required catalyst $PdCl_2/SrCO_3$.

To the epoxide 104 (50 mg) in hot spectral grade methanol (0.5 ml) under nitrogen was added pyrrolidine (0.5 ml). After 5 minutes the mixture was cooled and filtered. The crude material appeared as six spots on the t.l.c. plates, indicating that decomposition had occurred.

(ii) A mixture of 3β-hydroxy-5-androsten-17-one 8 (3.5 g) and DDQ (12 g) in dioxane (100 ml) was refluxed for 16 hours. The mixture was cooled and filtered and the filtrate was poured onto an alumina column (200 g, activity Grade I). Elution with benzene afforded starting material 8 (2.5 g). Further elution with benzene-acetone (1:1) afforded 1,4,6-androstatriene-3,17-dione 105 (500 mg). M.p. 164-165° (lit.[116], m.p. 164.5-166°) and 4,6-androstadiene-3,17-dione 106 (200 mg). M.p. 170-171° (lit.[61], m.p. 169.5-171°).

The trienone 105 (500 mg) was dissolved in methylene chloride (3 ml) and methanol (7 ml) and the solution was cooled in a dry-ice bath to -10°. 7N NaOH (0.2 ml) and hydrogen peroxide (0.75 ml, 30%) were added and the reaction mixture was placed in a fridge at 4° for 20 hours. The mixture was neutralized with dilute acetic acid and the methylene chloride was evaporated. Water (5 ml) was added and the mixture filtered. Crystallization of the crude from acetone yielded 1α,2α-oxido-4,6-androstadiene-3,17-dione 107 (500 mg). M.p. 220-223° (lit.[94], m.p. 225-227°).

The epoxide 107 (100 mg) in chloroform (6 ml) was treated with HBr (1.6 ml, 50%) for 15 minutes at room temperature. The chloroform solution was washed with dilute sodium sulphite solution and water, then dried and evaporated. Crystallization of the crude afforded 2-bromo-1,4,6-androstatriene-3,17-dione 110 (105 mg). M.p. 235-238° (lit.[94], m.p. 234-239°); n.m.r. (60 Mc, $CDCl_3$) δ 1.00 (18-$CH_3$, s, 3H), 1.30 (19-$CH_3$, s, 3H), 6.0-6.55 (4-H, 6-H, 7-H, m, 3H), 7.50 (1-H, s, 1H).

A solution of the epoxide 107 (300 mg) in chloroform (9 ml) was cooled in an ice bath to 5°. Cold HBr (6 ml, 50%) was added and the mixture stirred rapidly, to ensure intimate mixing, for 15 minutes. Aqueous sodium carbonate was added slowly and the chloroform solution was separated and washed with a $NaHCO_3$ solution, water, then dried and evaporated. Crystallization of the residue from acetone-hexane afforded 1α-hydroxy-2β-bromo-4,6-androstadiene-3,17-dione 109 (200 mg). M.p. 187-190° decomp. N.m.r. (60 Mc, $CDCl_3$) δ 1.00 (18-$CH_3$, s, 3H) 1.50 (19-$CH_3$, s, 3H), 4.40 (1β-H, d, 1H), 4.50 (2α-H, b, 1H), 6.02 (4-H, s, 1H), 6.45 (6-H, 7-H, s, 2H). The bromohydrin is unstable (t.l.c. evidence) even when standing in the fridge at 4°.

The bromohydrin 109 was hydrogenated as earlier described, in benzene under atmospheric hydrogen. The ratio of steroid to catalyst (wt.:wt.) was varied as was the length of time of the reaction and the results have been depicted in FIGURE 26 (p. 77).

(iii) 1α-Hydroxy-2β-bromo-4,6-androstadiene-3,17-dione 109 (20 mg) was dissolved in N,N-dimethylformamide and another 20 mg portion was dissolved in ethanol. To each solution was added zinc powder (300 mg);both reaction mixtures were heated on a steam bath. No reaction appeared to be occurring (t.l.c. evidence) until the temperature was above 65°. Above this temperature both reactions proceeded at approximately the same rate. After 2 hours both mixtures were filtered and the solvents evaporated. The n.m.r. spectra of the crude products showed that both reactions had resulted in the formation of 1,4,6-androstatriene-3,17-dione 105.

(iv) 1α, 2α-Oxido-4, 6-androstadiene-3, 17-dione 107 (250 mg) in benzene (25 ml) was hydrogenated under [redacted] hydrogen* with 5% $PdCl_2/SrCO_3$ (300 mg) as the catalyst, for 2.5 hours. The mixture was filtered and the benzene evaporated, and the residue was crystallized from acetone-hexane affording 1α, 2α-oxido-4-androstene-3, 17-dione 108 (250 mg). M.p. 201-203°; n.m.r. (60 Mc, $CDCl_3$), δ 0.95 (18-$CH_3$, s, 3H), 1.28 (19-$CH_3$, s, 3H), 3.33 - 3.62 (1β-H, 2β-H, m, 2H), 5.71 (4-H, m, 1H).

A mixture of the epoxide 108 (120 mg), ethylene glycol (1.0 ml) and p-toluenesulphonic acid (10 mg) in benzene (200 ml) was refluxed for 3 hours. The usual workup afforded crude material (80 mg). The n.m.r. spectrum showed no signal in the range for the epoxidic protons. Refluxing of the epoxide 108 in benzene with a few grains of p-toluenesulphonic acid also resulted in destruction of the 1α, 2α-epoxidic group.

* atmospheric pressure

## CLAIMS TO ORIGINAL RESEARCH

1. The mild acid catalyzed allylic rearrangement of $\Delta^6$-5α-hydroxy steroids was studied and applied as a route to 7α,19-disubstituted androgens.

2. The efficiencies of the rearrangements of several $\Delta^6$-5α-hydroperoxy steroids was investigated.

3. The non-stereospecificity of the reaction of lead tetraacetate with a $\Delta^5$-7-hydrazone was demonstrated.

4. The epoxidation of a $\Delta^6$-5α-hydroxy steroid with peracid was shown to result in a 6α, 7α-epoxide.

5. Molecular rotations and n.m.r. data were correlated for some 7α- and 7β-substituted steroids.

6. The following new compounds were prepared and characterized:
   a) 3β,19-Diacetoxy-5-androstene-7,17-dione;
   b) 7α-Acetoxy-3β,17β,19-trihydroxy-5-androstene;
   c) 7β-Acetoxy-3β,17β,19-trihydroxy-5-androstene;
   d) 3β,17β,19-Triacetoxy-5-androsten-7-one;
   e) 3β-Acetoxy-5-hydroxy-5α-androst-6-en-17-one;
   f) 3β-Acetoxy-7α-hydroxy-5-androsten-17-one;
   g) 3β,17β-Diacetoxy-5-hydroxy-5α-androst-6-ene;
   h) 3β,17β-Diacetoxy-7α-hydroxy-5-androstene;
   i) 3β-Acetoxy-5-cholestene-7α,19-diol;
   j) 3β,19-Diacetoxy-5-cholestene*;

---

* This compound has been reported[68] but no elemental analysis was given.

k) 3β,19-Diacetoxy-5-cholesten-7-one;

l) 5-Cholestene-3β, 7β,19-triol;

m) 3β-Acetoxy-5,19-dihydroxy-5α-androst-6-en-17-one;

n) 3β,19-Diacetoxy-5-hydroxy-5α-androst-6-en-17-one;

o) 3β,17β,19-Triacetoxy-5-hydroxy-5α-androst-6-ene;

p) 3β, 7α-Diacetoxy-5, 6α-oxido-5α-cholestane*;

q) 3β, 7α,17β-Triacetoxy-5-androstene;

r) 3β, 7α,17β-Triacetoxy-5, 6α-oxido-5α-androstane;

s) 3, 5-Cholestadiene-7-N,N-Dimethylhydrazone;

t) 3β,17β-Diacetoxy-5-hydroxy-6α, 7α-oxido-5α-androstane;

u) 3β,17β-Diacetoxy-5, 7α-dihydroxy-6β-bromo-5α-androstane;

v) 3β-Acetoxy-5-hydroxy-7α-bromo-6β,19-oxido-5α-androstan-17-one;

w) 3β-Acetoxy-5-hydroxy-6β,19-oxido-5α-androst-7-en-17-one;

x) 3β,19-Diacetoxy-7α-hydroxy-5-androsten-17-one.

7. The following compounds were prepared and identified by spectral methods but no elemental analyses were obtained:

a) 3β, 7α,17β,19-Tetraacetoxy-5-androstene;

b) 3β, 7β,17β,19-Tetraacetoxy-5-androstene;

c) 3β, 7α,19-Triacetoxy-5-cholestene;

d) 3β, 7β,19-Triacetoxy-5-cholestene;

e) 3β, 7α-Diacetoxy-5, 6β-oxido-5β-cholestane;

f) 3β, 7α,17β-Triacetoxy-5, 6β-dihydroxy-5α-androstane;

g) 3β, 7α,19-Triacetoxy-5-androsten-17-one;

h) 1α-Hydroxy-2β-bromo-4, 6-androstadiene-3,17-dione.

i) 1α, 2α-oxido-4-androstene-3,17-dione

---

* This compound was reported[60] in 1936 but the configuration of the epoxide was not known.

## REFERENCES


1. E.A. Doisy, C.D. Veler and S.A. Thayer, Am.J.Physiol., 90, 329 (1929).
2. A. Butenandt, Naturwiss., 17, 879 (1929).
3. L.F. Fieser and M. Fieser, "Steroids", Reinhold Publishing Corp., New York, N. Y., 1959, p. 44.
4. G.F. Marrian, J.Endocrinol., 35, vi (1966).
5. A. Girard, G. Sandulesco, A. Fridenson and J.J. Rutgers, Compt.Rend., 194, 909, 195, 981 (1932).

6.a. E.T. Engel and P. Carter, "Physical Properties of the Steroid Hormones", L.L. Engle, Ed., The MacMillan Co., New York, N. Y., 1963, p. 1.

b. E. Heftmann and E. Mosettig, "Biochemistry of Steroids", Reinhold Publishing Corp., New York, N. Y., 1960, p. 155.

7.a. C. Huggins, Cancer Res., 25, 1163 (1965).

b. A. Segaloff, "Methods in Hormone Research," Vol. V., R. Dorfman, Ed., Academic Press Inc., New York, N. Y., 1962, p. 205.

8.a. J. Mamortsen, F.J. Moore, C.E. Hopkins, O.T. Kuzna and J. Werner, J.Proc.Soc.Exptl.Biol.Med., 110, 400 (1962).

b. J. Stamler, L.N. Katz, R. Pick, L.A. Lewis, I.H. Page, A. Pick, B.M. Kaplan, D.M. Berkson and D. Century, "Drugs affecting Lipid Metabolism", Elsevier, Amsterdam, 1961, p. 432.

9.a. Chem. Eng. News, 45, 44 (March 27, 1967).

b. G. Pincus, Science, 153, 493 (1966).

10. P. Morand and J. Lyall, Chemical Reviews, 68, 85 (1968).

11. J.A. Hogg and J. Korman, "Medicinal Chemistry", 2, F.F. Blicke and C.M. Suter, Ed., Wiley, 1956, p. 34-217.

12. IUPAC-IUB-Commissions, Biochim. Biophys. Acta, 164, 453-486 (1968).

13. K. Bloch and D. Rittenberg, J. Biol. Chem. 145, 625 (1942).

14.a. K. Bloch, Angew. Chem., 77, 944 (1965).

b. K. Bloch, Science, 150, 19 (1965).

c. R.B. Clayton, Quart. Rev., (London), 19, 168 (1965).

d. J.H. Richards and J.B. Hendrickson, "The Biosynthesis of Steroids, Terpenes and Acetogenins", W.A. Benjamin Inc., New York, N.Y., 1964, p. 305.

e. E. Staple, "Biogenesis of Natural Compounds", P. Bernfield, Ed., The MacMillan Co., New York, N.Y. 1963, p. 155.

15.a. I.R. Dorfman and F. Unger, "Metabolism of Steroid Hormones", Academic Press Inc., New York, N.Y., 1965, p. 123.

b. See ref. 14d. p. 341.

c. See ref. 14e. p. 168.

16. A.S. Meyer, Experientia, 11, 99 (1955).

17. A.S. Meyer, Biochim. Biophys. Acta, 17, 441 (1955).

18. J.E. Longchampt, C. Gual, M. Ehrenstein and R.I. Dorfman, Endocrinol., 66, 416 (1960).

19. R.B. Wilcox and L.L. Engel, Steroids, Suppl. I, 49 (1965).

20. R.B. Wilcox and L.L. Engel, Steroids, Suppl. II, 249 (1965).

21. T. Morato, M. Hayano, R.I. Dorfman, L.R. Axelrod, Biochem. Biophys. Res. Comm., 6, 334 (1961).

22. L.R. Axelrod, C. Matthijssen, P.N. Rao and J.W. Goldzieher, Acta Endocrinol., 48, 383 (1965).

23. S.J.M. Skinner and M. Akhtar, Biochem. J., 114, 75 (1969).

24. L. Stárka and H. Breuer, Hoppe-Seylers Z. Physiol. Chem. 344, 124 (1966).

25.a. K.J. Ryan, Biochim. Biophys. Acta, 27, 658 (1958).

b. K.J. Ryan, J. Biol. Chem., 234, 268 (1959).

26. T. Morato, K. Raab, H.J. Brodie, M. Hayano and R.I. Dorfman, J. Am. Chem. Soc., 84, 3764 (1962).

27. H.J. Brodie, K.J. Kripalani and G. Possanza, J. Am. Chem. Soc., 91, 1241 (1969).

28.a. J. Fishman and H. Guzik, J. Am. Chem. Soc., 91, 2805 (1969).

b. J. Fishman, H. Guzik and D. Dixon, Biochem., 8, 4304 (1969).

29. Y. Osawa and D.G. Spaeth, Biochem., 10, 66 (1971).

30.a. H.J. Brodie, K. Raab, G. Possanza, N. Seto and M. Gut, J. Org. Chem., 34, 2697 (1969).

b. E.J. Corey and R.A. Sneen, J. Am. Chem. Soc., 78, 6271 (1956).

31. H.J. Brodie, G. Possanza and J.D. Townsley, Biochim. Biophys. Acta, 152, 770 (1968).

32. J.D. Townsley and H.J. Brodie, Biochem. 7, 33 (1968).

33. C. Gual, T. Morato, M. Hayano, M. Gut and R.I. Dorfman, Endocrinol., 71, 920 (1962).

34. R.A. Meigs and K.J. Ryan, J. Biol. Chem., 246, 83 (1971).

35. J.F. Bagli, P. Morand, K. Wiesner and R. Gaudry, Tet. Let., 387 (1964).

36.a. R. D. H. Heard and M. M. Hoffman, J. Biol. Chem., 135, 801 (1940).

b. R. D. H. Heard and M. M. Hoffman, J. Biol. Chem., 138, 651 (1941).

37.a. R. D. H. Heard, E.G. Bligh, M. C. Cann, P. H. Jellinck, V. J. O'Donnell, B. G. Rao and J. L. Webb, Recent Progr. Hormone Research, 12, 45 (1956).

b. K. Savard, K. Andrec, B. W. L. Brooksbank, C. Reyneri, R. I. Dorfman, R. D. H. Heard, R. Jacobs and S.S. Solomon, J. Biol. Chem., 231, 765 (1958).

38. R. D. H. Heard, R. Jacobs, V.J. O'Donnell, F.G. Peron, J. C. Saffran S.S. Solomon, L.M. Thompson, H. Willoughby and C.H. Yates, Recent Progr. Hormone Research, 9, 383 (1954).

39.a. J. J. Schneider and M.L. Lewbart, Recent Progr. Hormone Research, 15, 201 (1959).

b. See ref. 15a., p. 66.

40. L. Stárka and O. Haupt, Naturwiss., 51, 164 (1964).

41.a. J. Sulcova and L. Stárka, Experientia, 19, 609 (1963).

b. L. Stárka, Naturwiss., 52, 499 (1965).

42. R. Knuppen, O. Haupt and H. Breuer, Steroids, 3, 123 (1964).

43. H. Inano and B. Tamaoki, Biochem., 10, 1503 (1971).

44.a. W. C. Heinrichs, R. C. Mushen and A. Colas, Steroids, 9, 23 (1967).

b. J. Sulcova and L. Stárka, Steroids, 12, 113 (1968).

45.a. S.H. Eppstein, P.D. Meister, H. C. Murray and D.H. Peterson, "Vitamins and Hormones", 14, 359 (1956).

b. R. M. Dodson, R. T. Nicholson and R. D. Muir, J. Am. Chem. Soc., 81, 6295 (1959).

c. M. Opadu and M. Hasunuma, J. Pharm. Soc. (Japan), 86, 67 (1966).

d. R. Abraham, E. Balke, K. Krisch, S. Leonhäuser, K. Leybold, K.A. Sack and H. Staudinger, "Handbuch Physiol.-u. Pathol. Chem. Anal.," 6A, 917 (1964) and references cited therein.

46. L. Cédard, B. Fillman, R. Knuppen, B. Lisboa and H. Breuer, Hoppe-Seylers Z. Physiol. Chem., 338, 89 (1964).

47. L. Cédard, S. Yannotti and J. Varagnot, Compt. Rend. Soc. Biol., 155, 458 (1961).

48. L. Stárka and H. Breuer, Biochim. Biophys. Acta, 115, 306 (1966).

49. V. Grenville, D.K. Patel, V. Petrow, I.A. Stuart-Webb and D.M. Williamson, J. Chem. Soc., 4105 (1957).

50. J. Kalvoda, K. Heusler, H. Ueberwasser, G. Anner and A. Wettstein, Helv. Chim. Acta, 46, 1361 (1963).

51. J. Tadanier, J. Org. Chem., 28, 1744 (1963).

52. A. L. J. Beckwith and G. W. Evans, Proc. Chem. Soc. 63, (1962).

53. L. Stárka, Coll. Czech. Chem. Comm., 26, 2452 (1961).

54. M.S. Kharasch, G. Sosnovsky and N. C. Yang, J. Am. Chem. Soc., 81, 5819 (1959).

55. L. F. Fieser, M. Fieser and R.N. Chakravarty, J. Am. Chem. Soc., 71, 2226 (1949).

56.a. H. J. E. Lowenthal, Tetrahedron, 26, 269 (1959).

b. C. W. Shoppee and B. C. Newman, J. Chem. Soc. (C), 981 (1968).

57.a. G.O. Schenck, Angew. Chem., 69, 579 (1957).

b. G.O. Schenck, O.A. Neumüller and W. Eisfeld, Ann., 618, 202 (1958).

c. A. Nickon and J.F. Bagli, J.Am. Chem.Soc., 83, 1498 (1961).

58. H. B. Henbest and E. R. H. Jones, J.Chem.Soc., 1792 (1948).

59. H. B. Henbest and E.R.H. Jones, J.Chem.Soc., 1798 (1948).

60. T. Barr, I. M. Heilbron, E.G. Parry and F.S. Spring, J.Chem.Soc., 1437 (1936).

61. See ref. 3., p. 161.

62. M. Karplus, J.Chem.Phys., 30, 11 (1959).

63. See ref. 3., p. 90 - 168 and references cited therein.

64. See. ref. 3., p. 153 - 161.

65. M. Stefanović, A. Jobić, Z. Maksimović, Lj. Lorenc and M.Lj. Mihailovic, Helv. Chim. Acta, 53, 1895 (1970).

66. W. Fenical, D.R. Kearns and P. Radlick, J.Am. Chem.Soc., 91, 3396, 7771 (1969).

67.a. A. Nickon and W.L. Mendelson, Can.J.Chem., 43, 1419 (1965).

b. A. Nickon, N. Schwartz, J. B. DiGiorgio and D.A. Widdowson, J.Org.Chem. 30, 1711 (1965).

c. A. Nickon and W.L. Mendelson, J.Org.Chem., 30, 2087 (1965).

68. M. Kaufman, Ph.D. Thesis, University of Ottawa, Canada, August, 1968.

69. B. Lythgoe and S. Trippett, J.Chem.Soc., 471 (1959).

70. L.F. Fieser and M. Fieser, "Reagents for Organic Synthesis", John Wiley and Sons, Inc., New York, N.Y., 1967, p. 136.

71. H. B. Henbest and R.A.L. Wilson, J.Chem.Soc., 1958 (1957).

72.a. M. Mousseron-Canet, B. Labeeuw and J.C. Lanet, Compt.Rend., 262, 1438 (1966) and references cited therein.

b. M. Mousseron-Canet and J.C. Guilleux, Bull.Soc.Chim. France, 12, 3853, 3858 (1966).

73. P. Morand and M. Kaufman, Can. J. Chem., 49, 3185 (1971).

74. A.D. Cross, E. Denot, R. Acevedo, R. Urquiza and A. Bowers, J. Org. Chem., 29, 2195 (1964).

75. A.D. Cross, J. Am. Chem. Soc., 84, 3206 (1962).

76.a. A. Petit and J. P. Mathieu, "Tables de Constantes et Données Numériques-6. Constantes Selectionnées. Pouvoir Rotatoire Naturel. I. Steroïds", Masson et Cie., Paris, 1956.

b. A. Bowers, L.C. Ibáñez and H.J. Ringold, Tetrahedron, 7, 138 (1959).

c. T. Ruzicka, E. Hardegger and L. Kauter, Helv. Chim. Acta, 27, 1164 (1944).

77. C.W. Shoppee, R.J.W. Cremlyn, D.E. Evans and G.H.R. Summers, J. Chem. Soc., 4364 (1957).

78. D.H.R. Barton, J.F. McGhie and P.L. Batten, J. Chem. Soc., (C), 1033 (1970).

79. M. Debono and R.M. Molloy, J. Org. Chem., 34, 1454 (1969).

80. G.R. Newkome and D.L. Fishel, J. Org. Chem., 31, 677 (1966).

81. J. Mauthner and W. Suida, Monatsh., 17, 579 (1896).

82. A. Fürst and P.A. Plattner, Helv. Chim. Acta, 32, 275 (1949).

83.a. K. Tori, T. Komens and T. Nakagawa, J. Org. Chem., 29, 1136 (1964).

b. J.F. Bagli, P. Morand and R. Gaudry, J. Org. Chem., 28, 1207 (1963).

84. J. Bredt, Ann., 437, 1 (1924).

85. G.O. Schenck and O.A. Neumüller, Ann., 618, 194 (1958).

86. T. Westphalen, Ber., 48, 1064 (1915).

87. P. B. D. de la Mare in "Molecular Rearrangements", Vol. 1, Ed. P. de Mayo, Interscience Publishers, New York, N. Y., 1963, p. 30 - 86.

88. L. Ruzicka, V. Prelog and E. Tagmann, Helv. Chim. Acta, 27, 1149 (1944).

89. See ref. 3., p. 17.

90. D.R. Idler and B. Truscott, J. Fish. Res. Bd. Canada, 23, 615 (1966).

91. H. J. Ringold and A. Turner, Chem. Ind., 211 (1962).

92. G. Eggart, P. Keller, C. Lehmann and H. Wehrli, Helv. Chim. Acta, 51, 940 (1968).

93. H. J. Ringold and S.K. Malhotra, Tet. Let., 15, 669 (1962).

94. B. Pelc, J. Hodkova and J. Holubek, Coll. Czech. Chem. Comm., 31, 1363 (1966).

95. J. Pfister, C. Lehmann and H. Wehrli, Helv. Chim. Acta, 51, 1505 (1968).

96. H. Wehrli, C. Lehmann, P. Keller, J. Bonet, K. Schaffner and O. Jeger, Helv. Chim. Acta, 49,2218 (1968).

97. S.K. Pradhan and H.J. Ringold, J. Org. Chem., 29, 601 (1964).

98. R.M. Dodson, A.H. Goldkamp and R.D. Muir, J. Am. Chem. Soc., 82, 4026 (1960).

99. L. L. Smith, T.J. Foell and D.M. Teller, J. Org. Chem., 30, 3781 (1965).

100. B. Pelc and E. Kodicek, J. Chem. Soc. (C), 1568 (1971).

101. B. Pelc and J. Hodkova, Coll. Czech. Chem. Comm., 32, 410 (1967).

102. J. Romo and A. Romo de Vivar, J. Am. Chem. Soc. 81, 3446 (1959).

103. N.A. Milas and D.M. Surgenor, J. Am. Chem. Soc., 68, 642 (1946).

104. J.K. Norymberski and G.F. Woods, J. Chem. Soc., 3426 (1955).

105. A. Bowers (to Syntex Corp). CA., 61, 4436a (1964); U.S. Patent, 3,079,381.

106. J.F. Bagli (to American Home Products Corp)., U.S. Patent, 3,162, 655 CA., 62, 16330d (1965).

107. H.J. Ringold, J. Am. Chem. Soc., 82, 961 (1960).

108. D.H.R. Barton, J. Chem. Soc., 1116 (1946).

109. M. Akhtar and D.H.R. Barton, J. Am. Chem. Soc., 86, 1528 (1964).

110.a. R.M. Moriarty and T.D.J. D'Silva, Tetrahedron, 21, 547 (1965).

b. B. Berkoz, E. Denot and A. Bowers, Steroids, 1, 251 (1963).

111. J. Joska and F. Sorm, Coll. Czech. Chem. Comm., 23, 1377 (1958).

112. L. Ruzicka and V. Prelog, Helv. Chim. Acta, 26, 975 (1943).

113. H.H. Inhoffen, G. Zühlsdorff and Huang-Minlon, Ber., 73, 451 (1940).

114. A. Bowers, E. Denot and R. Becerra, J. Am. Chem. Soc., 82, 4007 (1960).

115. G. Rosenkranz, C. Djerassi, S. Kaufman, J. Pataki and J. Romo, Nature, London, 165, 814 (1950).

116. S. Kaufman, J. Pataki, G. Rosenkranz, J. Romo and C. Djerassi, J. Am. Chem. Soc., 72, 4531 (1950).