hydrolysis, as well as the low Zeisel number, indicates that the methyl groups are probably attached to nitrogens.

Dimethyldianilinogossypol.—By a similar procedure to the preparation of the oxo compound but refluxing overnight rather than allowing the reaction mixture to stand at room temperature resulted in a methylated but unoxidized product. It was purified from benzene and formed red crystals, m. p. 253–258°. It gave a dark red-brown color in sulfuric acid.

Anal. Calcd. for $C_{44}H_{42}N_2O_6$: C, 75.83; H, 6.36; N, 4.02; OCH_3, 8.89. Found: C, 75.70; H, 6.11; N, 4.09; OCH_3, 1.01.

Concentrated sulfuric acid does not hydrolyze this product. This and the low Zeisel number indicate the probability that the methyls are attached to nitrogen.

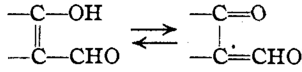
Summary

Gossypol condenses with β-naphthylamine or o-phenylenediamine in the same way as with aniline. Two molecules of amine react and two moles of water are eliminated.

Several other aniline derivatives gave products which did not analyze for simple condensation products. All of these hydrolyzed with sulfuric acid to give gossypol.

Dianilinogossypol can be acetylated to a hexaacetate. Two acetyls are attached to nitrogen. Methylation gives sometimes a dimethyldianilinogossypol or more commonly a dimethyldianilinooxogossypol.

It is deduced that there exist in gossypol two of the tautomeric residues



$$\begin{array}{l}\text{—C—OH}\\ \;\;\|\\ \text{—C—CHO}\end{array} \rightleftarrows \begin{array}{l}\text{—C=O}\\ \;\;|\\ \text{—C=CHOH}\end{array}$$

URBANA, ILLINOIS RECEIVED JUNE 27, 1938

[CONTRIBUTION FROM THE CHEMICAL LABORATORY OF THE UNIVERSITY OF ILLINOIS]

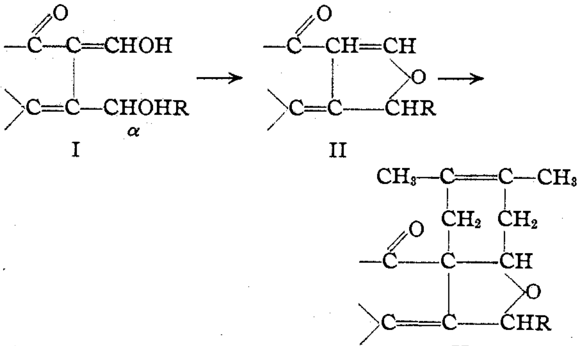
Structure of Gossypol. VI.[1] Addition Products with Butadienes

BY ROGER ADAMS, B. S. FRIEDMAN, C. C. PRICE, R. C. MORRIS AND E. C. KIRKPATRICK[2]

From a study of anhydrogossypol[3] and its reaction products and of the diamino- and dianilinogossypols[1] it was deduced that gossypol contains two similar groupings which are probably vinyl alcohols. Since the most common type of relatively stable vinyl alcohols possesses a carbonyl in place of one of the hydrogens of the methylene group $\left(\text{—}\overset{\text{O}}{\overset{\|}{\text{C}}}\text{—CH=CHOH}\right)$, the possibility of two such residues in gossypol has now been studied. The reaction used for this purpose was the Diels–Alder and the reagent, 2,3-dimethylbutadiene.

Gossypol condenses very readily in alcohol solution with the addition of two molecules of dimethylbutadiene and elimination of two molecules of water to give a beautifully crystalline product. In a similar way, anhydrogossypol or diaminogossypol in benzene condenses with two molecules of dimethylbutadiene to give the same derivative.

In the case of gossypol, the reaction involves the elimination of water; in the case of diaminogossypol the loss of ammonia; in the case of anhydrogossypol, however, the same derivative is formed by simple addition. Since anhydrogossypol adds the diene, it is a safe conclusion that the molecule probably contains α,β-unsaturated carbonyl linkages. The addition product, unlike anhydrogossypol itself, is not hydrolyzed. Consequently, the easily-opened oxygen ring which was postulated previously as being present in anhydrogossypol, must have been stabilized. This is no doubt due to the fact that the carbon–carbon double bond to which the diene adds is part of the oxygen-containing ring of the anhydrogossypol. Suggested nuclei which will account for the changes follow: gossypol (I), anhydrogossypol (II), dimethylbutadiene addition product (III).



I II III

It is probable that gossypol and diaminogossypol

---

[1] For the previous article in this field see Adams, Price and Dial, THIS JOURNAL, **60**, 2158 (1938).

[2] A portion of a thesis submitted in partial fulfilment of the requirements for the degree of Doctor of Philosophy by E. C. Kirkpatrick, Solvay Fellow, 1936–1938.

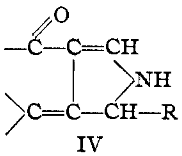
[3] Miller and Adams, THIS JOURNAL, **59**, 1736 (1937).

first lose water and ammonia, respectively, and then the diene adds to the anhydrogossypol formed.

The nucleus II accounts for the ease with which anhydrogossypol is hydrolyzed to gossypol, methylated or acetylated to gossypol hexamethyl ether or gossypol hexaacetate and converted by ammonia or aniline to the corresponding gossypol derivatives. Nucleus III indicates why the addition product is stable to hydrolysis as it no longer contains an easily-opened oxygen ring.

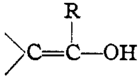
The dimethylbutadiene anhydrogossypol product is soluble in alkali and contains two acidic hydroxyls which are readily acetylated or methylated to form stable crystalline derivatives. It also gives a red color with pyroboroacetate, indicative of a phenol group *ortho* or *peri* to a carbonyl.

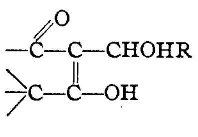
Still further deductions concerning the oxygen-containing ring formation when gossypol is converted to anhydrogossypol may be made. Liquid ammonia[3] converts anhydrogossypol to diaminogossypol and not to a molecule containing an imino nitrogen in place of the oxygen of the oxygen-containing ring in anhydrogossypol. It is difficult to believe that the nitrogen compound corresponding to II shown in IV would not be



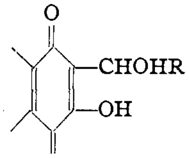
IV

stable if the hydroxyl marked α in formula I were alcoholic in character. On the other hand, if it were phenolic, the non-formation of IV is explicable. For this reason, it is assumed that the oxygen-containing ring in anhydrogossypol is formed by elimination of water between the hydroxyl of an hydroxymethylene group and a phenolic group.

The ease of condensation of anhydrogossypol or gossypol with the diene and the fact that neither gossypol hexamethyl ether nor tetramethyl ether undergoes such a reaction, makes it probable that the hydrogen on the carbon holding the vinyl hydroxyl is unsubstituted. The grouping should therefore be represented preferably by >C=CHOH and not by  $>C{=}\overset{\overset{\displaystyle R}{|}}{C}{-}OH$. The ease of the reaction with the diene also tends to nullify the possibility of a structure isomeric with I shown in V and hence eliminates the likelihood of the presence of residues of a quinone-like structure (VI) in the gossypol molecule.



V



VI

The reaction is not limited to dimethylbutadiene. Anhydrogossypol also condenses with 2,3-butadiene to give an analogous product in which two molecules of diene have added, and two acidic hydroxyls remain unaffected. The product apparently is not as stable as the dimethylbutadiene derivative, as it dissociates upon standing in solution. It gives a diacetate similar to the dimethylbutadiene derivative.

From the study of the aniline and ammonia derivatives[1,3] and of the diene addition products, the presence in gossypol of two tautomeric hydroxyaldehyde residues seems fairly well established.

Experimental

The Addition of 2,3-Dimethylbutadiene-1,3 to Gossypol.—To a solution of 1 g. of gossypol in 25 cc. of absolute alcohol was added 1.5 cc. of freshly prepared dimethylbutadiene. The reaction mixture was refluxed for eight hours on the steam-bath, the condenser was then removed and the alcohol allowed to evaporate. After concentrating for a short time, crystals began to separate, the flask was cooled and the precipitate filtered. On further concentration of the mother liquor more product was obtained; yield 0.8 g. The material crystallized from acetone–methanol in bright yellow cubes, m. p. 244–245° with decomposition.

Anal. Calcd. for $C_{42}H_{46}O_6$: C, 78.00; H, 7.12. Found: C, 77.83; H, 7.09.

The crystals gave a bright yellow-brown color in concentrated sulfuric acid. Alcoholic ferric chloride gave a deep red color. Aqueous sodium hydrosulfite solution had no effect on an alcoholic solution of the addition compound. It gave a deep red with pyroboroacetate.

The addition reaction takes place equally well using benzene as the solvent.

The addition product would not condense with aniline. It was unaffected by heating for one-half hour on the steam-bath with 40% aqueous alkali. It decomposed when dissolved in concentrated sulfuric acid and allowed to stand overnight. It did not reduce catalytically with hydrogen and platinum oxide platinum black.

Methylation of the Dimethylbutadiene Addition Product.—To a solution of 0.4 g. of the addition product in 10 cc. of 30% methanolic potassium hydroxide was added an excess of dimethyl sulfate, in portions. When the reaction was complete the solution was made alkaline, diluted with water, and the precipitate filtered and washed. The product was air dried and crystallized from acetone–methanol; bright yellow needles, m. p. 227–229°, yield 0.32 g. It gave a yellow-brown color in concentrated sulfuric acid and gave no color with phenol reagents.

Anal. Calcd. for $C_{44}H_{50}O_6$: C, 78.3; H, 7.42. Found: C, 78.11; H, 7.35.

Acetylation of the Dimethylbutadiene Addition Product.—By heating for a half-hour a mixture of 0.075 g. of addition product, excess of acetic anhydride and sodium acetate, and then pouring into water, 0.065 g. of the acetylated compound was obtained. It crystallized in light yellow plates from acetone–methanol, m. p. 227–229°, and gave a brown color in concentrated sulfuric acid.

Anal. Calcd. for $C_{46}H_{50}O_8$: C, 75.5; H, 6.84. Found: C, 75.22; H, 6.95.

Saponification of this product yielded the original material as shown by melting point and mixed melting point.

The product was not affected by long refluxing (two and one-half hours) with acetic anhydride and sodium acetate.

Condensation of 2,3-Dimethylbutadiene-1,3 with Anhydrogossypol.—To a solution of 0.5 g. of anhydrogossypol in 25 cc. of dry benzene was added 1.5 cc. of freshly prepared dimethylbutadiene. The solution was refluxed overnight on the steam-bath, then evaporated to dryness. The residue was purified by crystallization from acetone–methanol; m. p. 244–245° with decomposition. A mixed melting point with the product from the addition of dimethylbutadiene to gossypol showed no lowering.

Condensation of 2,3-Dimethylbutadiene-1,3 with Diaminogossypol.—A solution of 0.425 g. of diaminogossypol in 25 cc. of dry benzene was refluxed on the steam-bath overnight with 1.5 cc. of freshly distilled dimethylbutadiene.

The material dissolved to give the red solution characteristic of diaminogossypol in benzene. At the end of eight hours the red color had faded to the yellow color common to a solution of gossypol.

The solution was evaporated and the residue crystallized from acetone–methanol, m. p. 244–245° with decomposition. This product was identical with that obtained in the reaction above using gossypol or anhydrogossypol.

Attempted Condensations with 2,3-Dimethylbutadiene-1,3.—Using the same procedure that was employed in the condensation of gossypol with dimethylbutadiene, attempts were made to condense hexaacetylgossypol, the white hexamethyl ethers of gossypol and apogossypol. In every case the starting material was recovered unchanged.

Condensation of Butadiene-1,3 with Anhydrogossypol.—A mixture of 3 cc. of pure butadiene-1,3,5 cc. of pure dry benzene and 0.5 g. of anhydrogossypol was heated in a sealed tube on a steam-bath for twelve hours. After evaporation of the excess butadiene by warming, the red solution was cooled and filtered from the precipitate. The solid thus obtained was purified by rapid crystallization from acetone–methanol; yellow crystals which darkened at 242° and melted with decomposition at 245–246°.

Anal. Calcd. for $C_{38}H_{38}O_6$: C, 77.2; H, 6.43. Found: C, 76.95; H, 6.46.

The product gave an orange color in concentrated sulfuric acid. If the addition compound was allowed to remain standing in solution, there was a reversion with regeneration of anhydrogossypol.

Acetylation of Butadiene Addition Product.—By a procedure similar to that described for the dimethylbutadiene product, a white diacetate was obtained. It was purified from aqueous methanol, m. p. 250–251° with decomposition. It gave a yellow color in concentrated sulfuric acid.

Anal. Calcd. for $C_{38}H_{36}O_4(OCOCH_3)_2$: C, 74.8; H, 6.24. Found: C, 74.91; H, 6.50.

Summary

Gossypol, anhydrogossypol, and diaminogossypol react with dimethylbutadiene to give a product which analyzes for the addition of two moles of diene to one of anhydrogossypol. During the reaction with gossypol or diaminogossypol, two moles of water or two moles of ammonia are eliminated simultaneously. The product contains two hydroxyls which can be methylated or acetylated.

Butadiene gives a similar product with anhydrogossypol though it is less stable than the dimethylbutadiene derivative.

The deduction from these experiments is that two α,β-unsaturated carbonyl groups are present in the anhydrogossypol molecule. Gossypol in forming anhydrogossypol probably loses water between hydroxymethylene groups and phenolic hydroxyls.

URBANA, ILLINOIS RECEIVED JUNE 27, 1938