

[CONTRIBUTION FROM THE ARGONNE CANCER RESEARCH HOSPITAL—USAEC, THE UNIVERSITY OF CHICAGO]

The Infrared Spectra of Nitrated Estrones

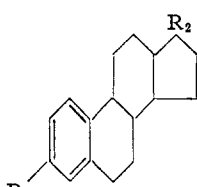
By ROGER A. PICKERING^{1a} AND HAROLD WERBIN^{1b}

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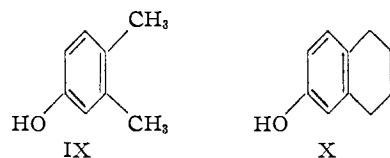
The infrared spectra in KBr of a series of nitrated derivatives of estrone ($\Delta^{1,3,5}$ -estratriene-3-ol-17-one) are discussed and correlations are made based on intermolecular and intramolecular hydrogen bonding effects. New data are given in support of several assignments reported in the literature. On the basis of this study, the structural assignments of 2-nitroestrone and 4-nitroestrone are shown to be the reverse of those previously reported in the literature.

Introduction

This paper discusses the infrared spectra of a series of nitrated derivatives of estrone which were prepared in connection with an investigation of the degradation of the aromatic ring of the steroid.^{2,3} The compounds studied, besides the parent compound estrone ($\Delta^{1,3,5}$ -estratriene-3-ol-17-one) (I), include 2-nitroestrone (II), 4-nitroestrone (III), 2,4-dinitroestrone (IV), several of the corresponding reduced compounds (V and VI), and the acetate derivatives (VII and VIII), together with some

				
	R ₁	R ₂	2	4
I	-OH	=O		
II	-OH	=O	-NO ₂	
III	-OH	=O		-NO ₂
IV	-OH	=O	-NO ₂	-NO ₂
V	-OH	-OH (β)	(-NO ₂)	(-NO ₂)
VI	-OH		(-NO ₂)	(-NO ₂)
VII	-OCOCH ₃	=O	(-NO ₂)	(-NO ₂)
VIII	-OH (or -OCOCH ₃)	-OCOCH ₃	-NO ₂	-NO ₂

model compounds from the 3,4-xylenol (3,4-dimethylphenol) (IX) and the *ar*-2-tetralol (5,6,7,8-tetrahydro-2-naphthol) (X) series.



The central problem involved is the identification and assignment of the proper configuration to the two isomeric mononitrated derivatives. The two compounds have been reported previously in the literature and were assigned their structures on the basis of analogies in the method of preparation.⁴ Subsequently, in another paper, these assignments apparently were confirmed by an interpretation of the infrared spectra.⁵ However, the preparative technique used results in mixtures (the melting points reported are very much lower than those obtained by Werbin and Haloway²), and the inter-

pretation in the second paper is based on the assignment of a single band, a C-H deformation mode, which is quite unreliable in this instance. On the basis of the work reported here it is concluded that the positional assignments of the substituents in 2-nitroestrone (m.p. 183–183.5°) and 4-nitroestrone (dec. 270–280°) are actually the reverse of those previously given. Our assignments, based on the detailed study of the infrared spectra of the series of compounds mentioned, appear conclusive.⁶

Because of the insolubility of key compounds in the usual solvents, the spectra were all taken as solid dispersions in pressed potassium bromide disks. Caution must always be taken in interpreting such solid state spectra, particularly in the empirical assignment of given absorption bands to specific group vibrations.⁷ However, the extensive internal correlations within the series permit conclusions to be drawn and some assignments to be made. Furthermore, the spectra of these solid dispersions present material for a study of infrared spectral effects which are characteristic of intermolecular forces in the crystalline state.

Experimental

The spectra were recorded with a Perkin-Elmer model 21 infrared spectrophotometer using a sodium chloride prism.

TABLE I
O-H STRETCHING BANDS

Compounds	Assignments, cm. ⁻¹	
	3-OH	17-OH
Estrone	3335s
4-Nitroestrone	3230s
2-Nitroestrone	3315m
2,4-Dinitroestrone	3170m
17-Deoxoestrone	3288s
4-Nitro-17-deoxoestrone	3435s
2-Nitro-17-deoxoestrone	3305m
2,4-Dinitro-17-deoxoestrone	3300w
3,4-Xylenol	3180s
2-Nitro-3,4-xylenol	3365s
6-Nitro-3,4-xylenol	3100w
<i>ar</i> -3-Nitro-2-tetralol	3200w
2,6-Dinitro-3,4-xylenol	3220m
<i>ar</i> -1,3-Dinitro-2-tetralol	3290m
17 β -Estradiol	3230s	3415s
2,4-Dinitro-17 β -estradiol	3200m	3570m
2,4-Dinitro-17 β -acetoxyestradiol	3220w ^b

^a s, m, w = strong, medium, weak intensities. ^b Uncorrected.

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(2) H. Werbin and C. Haloway, *J. Biol. Chem.*, **223**, 651 (1956).

(3) H. Werbin, *J. Org. Chem.*, **21**, 1532 (1956).

(4) J. B. Niederl and H. J. Vogel, *THIS JOURNAL*, **71**, 2566 (1949).

(5) E. A. Hillmann-Elies, G. Hillmann and U. Schiedt, *Z. Naturforsch.*, **8B**, 436 (1953).

(6) The same conclusion has been reported independently by Kraychy and Gallagher (*THIS JOURNAL*, **79**, 754 (1957)).

(7) Cf. R. N. Jones and C. Sandorfy, Ch. IV, "Chemical Applications of Spectroscopy," W. West, Ed., Vol. IX, "Technique of Organic Chemistry," A. Weissberger, Ed., Interscience Publishers, Inc., N. Y., 1956, pp. 294–298.

A weighed amount of sample was carefully ground with KBr carrier which had been preweighed (in 150 ± 5 mg. lots) and dried *in vacuo* (at $105\text{--}110^\circ$) to eliminate spurious absorption due to atmospheric moisture. The optimum sample weights in this series of compounds were in the range of 1.3–1.7 mg., weighed to ± 0.2 mg. for good qualitative and, where useful, semi-quantitative comparisons.

The frequencies of the band maxima are calibrated values unless otherwise indicated. The accuracy is generally specified as ± 2 cm.^{-1} below 2000 cm.^{-1} and ± 20 cm.^{-1} above 2000 cm.^{-1} . However, insufficient calibration points, lower resolution of the prism and greater thermal sensitivity in that region (together with the physical state of the samples) combine to make the calibration less reliable for the O–H stretching bands of Table I.

The samples studied were all recrystallized to constant m.p. The preparation of most of them is described by Werbin and Haloway.^{2,3}

Results and Discussion⁸

Phenolic Group.—Table I and Figs. 1–5 present the data on O–H stretching absorptions which contribute to the configurational assignments of

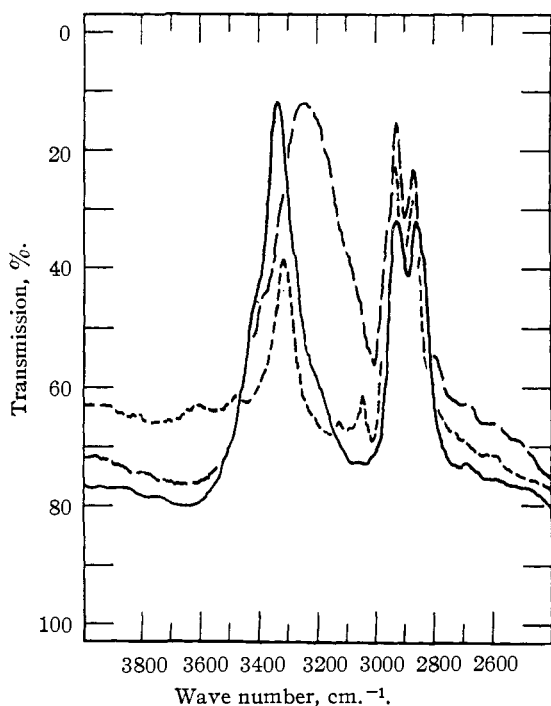


Fig. 1.—Infrared OH stretching absorption in KBr: —, estrone; — —, 4-nitroestrone; - - - -, 2-nitroestrone.

2-nitroestrone and 4-nitroestrone. If coplanarity with the ring is sterically possible, then strong intramolecular hydrogen bonding will occur between the adjacent phenolic and nitro groups. This is achieved in the case of 2-nitroestrone (II) and related compounds but, because of the steric hindrance of the B ring, not in 4-nitroestrone (III). One result of this chelation which might be exhibited in the infrared spectra would be a reduction

(8) To conserve space the spectra of the compounds discussed in the $1650\text{--}650$ cm.^{-1} range, from which the major bands given in Tables III and IV have been taken, have been deposited as Document number 5367 with the ADI Auxiliary Publications Project, Photoduplication Service, Library of Congress, Washington 25, D. C. Copies may be secured by citing the Document number and remitting in advance \$2.50 for photoprints or \$1.75 for 35 mm. microfilm, payable by check or money order to: Chief, Photoduplication Service, Library of Congress.

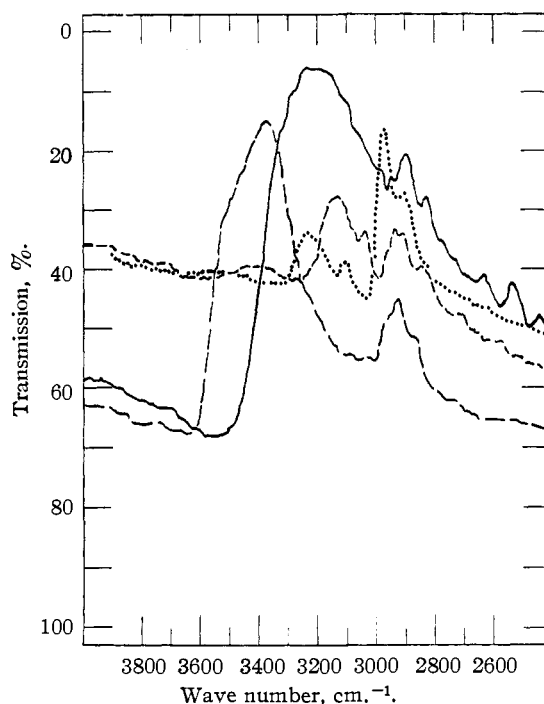


Fig. 2.—Infrared OH stretching absorption in KBr: —, xyleneol; — —, 2-nitroxyleneol; - - - -, 6-nitroxyleneol;, *ar*-3-nitro-2-tetralol.

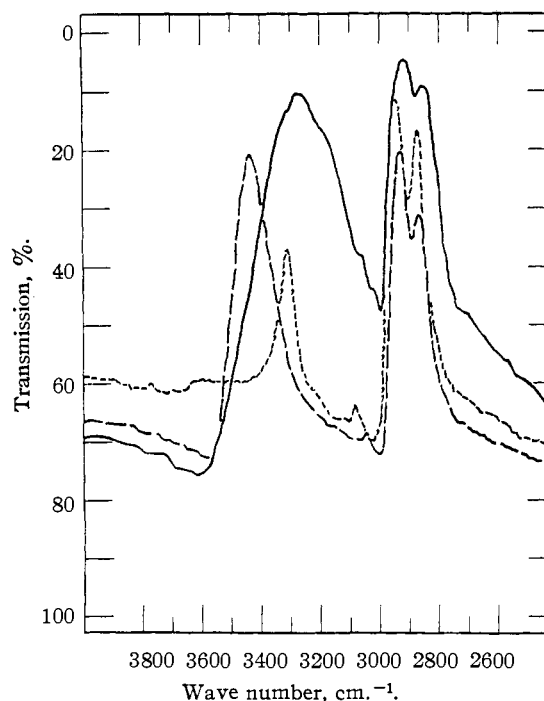


Fig. 3.—Infrared OH stretching absorption in KBr: —, deoxoestrone; — —, 4-nitrodeoxoestrone; - - - -, 2-nitrodeoxoestrone.

in intensity of the O–H stretching absorption.^{9,10} Figure 1 shows this effect markedly, and the argument is reinforced by comparison with the model

(9) Cf. L. J. Bellamy, "The Infrared Spectra of Complex Molecules," Methuen and Co., Ltd., London, 1954, pp. 92–94.

(10) Evidence supporting steric effects and configurational assignments is also provided by ultraviolet spectral data in ref. 2.

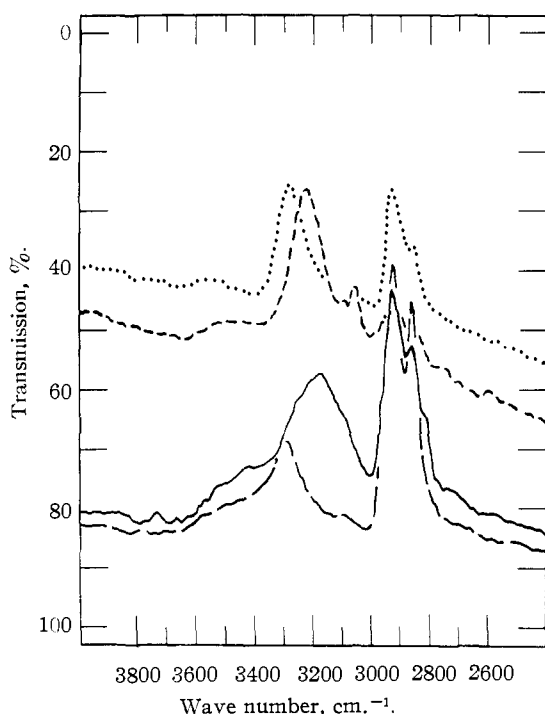


Fig. 4.—Infrared OH stretching absorption in KBr: —, 2,4-dinitroestrone; - - -, 2,4-dinitrodeoxoestrone; - · - ·, 2,6-dinitroxyleneol;, *ar*-1,3-dinitro-2-tetralol.

compounds in Fig. 2. In the case of the dinitro compounds, since one of the nitro groups is in the unhindered position, the O-H band also should be of reduced intensity. This is shown in Fig. 4.

TABLE II
C=O STRETCHING BANDS

Compounds	Assignments, cm. ^{-1a}		
	17-Keto	3-Acetoxy	17-
Estrone	1718
4-Nitroestrone	1718
2-Nitroestrone	1736
2,4-Dinitroestrone	1731
Estrone acetate	1732	1762	..
4-Nitroestrone acetate	1732	1777	..
2-Nitroestrone acetate	1734	1776	..
2,4-Dinitroestrone acetate	1731	1791	..
2,4-Dinitroestradiol 3,17 β -diacetate	..	1786	1728
2,4-Dinitroestradiol 17 β -acetate	1738

^a All bands of strong intensity.

It is not possible to attach much significance to the individual frequencies of the O-H absorption peaks in these solid dispersions. However, the pattern of the relative shifts of the maxima is significant. The difference in these shifts between the estrones (Fig. 1) and the models (Fig. 2) was tested by a comparison with the deoxoestrone (Fig. 3). The results provide evidence for the intermolecular bonding to be expected in the crystalline state between the OH group and the 17-keto group. This interpretation is supported by the predicted shifts in the C=O stretching frequencies.

For comparison with phenolic O-H absorption, the spectra of estradiol ($\Delta^{1,3,5}$ -estratriene-3,17 β -

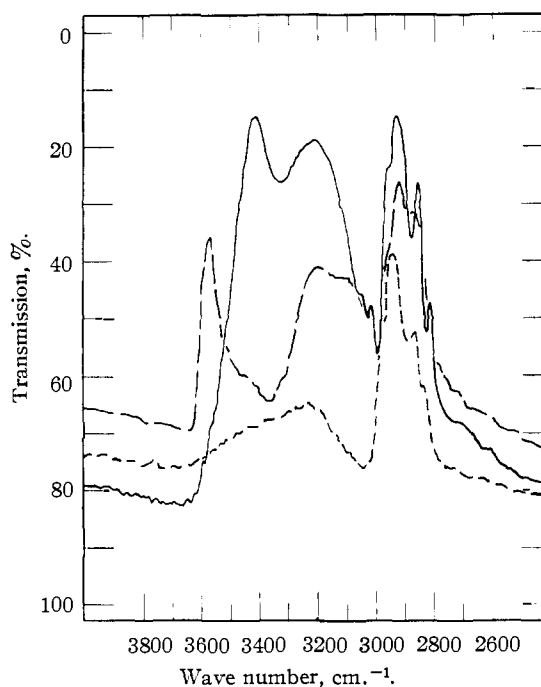


Fig. 5.—Infrared OH stretching absorption in KBr: —, estradiol; - - -, 2,4-dinitroestradiol; - · - ·, 2,4-dinitro-17 β -acetoxyestradiol.

diol), 2,4-dinitroestradiol and a monoesterification product are given in Fig. 5. The latter is assigned the structure of 2,4-dinitro-17 β -acetoxyestradiol by spectral and chemical arguments. The first two compounds have two peaks while the monoacetate exhibits a weak peak in the range of the intramolecularly bonded dinitro compounds of Fig. 4.

In addition to the stretching absorption, two strong characteristic absorption bands in the 1350 cm.⁻¹ and the 1200–1000 cm.⁻¹ regions are generally assigned to the OH group and ascribed to O-H deformation and to C-OH stretching vibrations. Since these bands are in a complex region marked by overlapping and should also be affected very greatly by inter- and intramolecular bonding, assignments are difficult to make. However, in the estrones and deoxoestrone a group of prominent absorption peaks in the 1500–1400 cm.⁻¹ and the 1300–1250 cm.⁻¹ regions are reduced considerably in intensity in the corresponding acetates. This effect is also observed in dinitroestradiol and its diacetate derivative. The acetate C-O stretching near 1200 cm.⁻¹ prevents an additional comparison of the strong bands occurring at 1200–1100 cm.⁻¹ in 2-nitroestrone and its deoxo derivative.

Carbonyl Group.—The C=O stretching bands given in Table II follow a predicted pattern. In estrone and 4-nitroestrone, the 3-OH group is free to form an intermolecular bond with the keto group. This is reflected in the lowering of the frequency to 1718 cm.⁻¹. In the 2-nitro and dinitro estrones, where the OH is involved in strong intramolecular bonding, the frequencies are in the range 1736–1731 cm.⁻¹ in common with the 3-acetoxy derivatives. This range, incidentally, is in excellent agreement with the 1737–1733 cm.⁻¹

TABLE III
 CHARACTERISTIC ABSORPTION BANDS 1650-1000 CM.⁻¹^a

Compounds	A	a	B	a'	b	C	c
Estrone	1621m	1584m	...	1498s
4-Nitroestrone	1624w	1583m	1522s	*	1374s
2-Nitroestrone	1634s	1567ms	1525s	*	1311s
2,4-Dinitroestrone	1627m	1570m	1531s	*	1309m
17-Deoxoestrone	1612s	1591ms	...	1502s
4-Nitro-17-deoxoestrone	1620mw	1580m	1526s	*	1358s
2-Nitro-17-deoxoestrone	1631s	1568ms	1520s	*	1310s
2,4-Dinitro-17-deoxoestrone	1634m	1577m	1553s	*	1310s
3,4-Xylenol	1609m	1587s	...	1496s
2-Nitro-3,4-xylenol	1628wm	1589m	1510s	*	1368s
6-Nitro-3,4-xylenol	1634m	1585ms	1524s	*	1274s
<i>ar</i> -3-Nitro-2-tetralol	1630m	1580ms	1530s	*	1277s
2,6-Dinitro-3,4-xylenol	1630m	1580s	1545s	*	1306s
<i>ar</i> -1,3-Dinitro-2-tetralol	1628m	1575s	1549s	*	1302s
Estrone acetate	1609w	1581vw	...	1494m	...	1203s	1014m
4-Nitroestrone acetate	1611vw	1573vw	1535s	*	1371ms	1193s	1011m
2-Nitroestrone acetate	1615w	1579w	1519s	*	1346s	1197	1003mw
						1180 } ^{sd}	
2,4-Dinitroestrone acetate	1609w	1575m	1525s	*	1346s	1160s	1000mw
17 β -Estradiol	1611m	1587m	...	1501s
2,4-Dinitro-17 β -estradiol	1631m	1570ms	1543s	*	1311s
2,4-Dinitroestradiol 3,17 β -diacetate	1611vw	1578vw	1541s	*	1370m	1165s	1017m
2,4-Dinitroestradiol 17 β -acetate	1634m	1583m	1545s	*	1323m

^a A, a, a' = bands characteristic of aromatic C-C; B, b = characteristic of the nitro group; C, c = characteristic of the acetoxy group; * = obscured by overlapping absorption; d = doublet; s, ms, m, mw, w, vw = strong to medium to weak intensities.

range found in polar CHCl₃ solution compared to the 1745-1741 cm.⁻¹ range in the non-polar solvents CCl₄, CS₂.^{11,12}

The wider range for the 3-acetoxy group C=O vibration is interpreted as paralleling the increasing steric hindrance to resonance interaction with the benzene ring on substituting adjacent nitro groups. In general, the frequencies are also noticeably higher than the assigned range for esters in solution.¹³ Even the vibration assigned to the 17-acetoxy group in the 2,4-dinitro-17 β -estradiol diacetate derivative occurs at 1728 cm.⁻¹ in comparison with 1722-1720 cm.⁻¹ reported by Jones and Herling for one 17 β -acetate ester in CHCl₃.¹¹ 2,4-Dinitro-17 β -acetoxy-estradiol has the band at 1738 cm.⁻¹.

Acetoxy Group.—The 3-acetoxy derivatives of these estrones are characterized readily by the strong C-O stretching band in the range of 1203-1160 cm.⁻¹ (Table III, C). The 1203 cm.⁻¹ maximum for estrone acetate is in excellent agreement with the 1206 cm.⁻¹ peak reported by Jones and co-workers for the compound run in CS₂ solution,^{14,15} while nitro substitution shifts the band to lower frequencies.

The 17-acetate of dinitroestradiol would be expected to absorb near 1240 cm.⁻¹, characteristic

(11) R. N. Jones and F. Herling, *J. Org. Chem.*, **19**, 1252 (1954).

(12) A somewhat similar example of a suggested intermolecular effect observed with cortisone and 11-dihydrocortisone is given by Jones and Sandorfy, ref. 7, pp. 294-295.

(13) A reviewer has kindly pointed out that the higher value for estrone acetate in the solid state is confirmed by "Methods of Biochemical Analyses," H. Rosenkrantz in "Methods of Biochemical Analysis," Vol. 2, D. Gluck, Ed., Interscience Publishers, Inc., New York, N. Y., 1955, p. 34.

(14) R. N. Jones, P. Humphries, F. Herling and K. Dobriner, *THIS JOURNAL*, **73**, 3215 (1951).

(15) R. N. Jones and F. Herling, *ibid.*, **78**, 1152 (1956).

of normal acetates. There is strong absorption in the dinitroestradiol diacetate and the 17-monoacetate at 1244 and 1241 cm.⁻¹, respectively, which is assignable to this C-O vibration.

Jones and Herling have studied extensively the spectra of acetoxy steroids below 1350 cm.⁻¹ for absorptions characteristic of the acetate group in addition to the C-O stretching absorption.¹⁵ They have noted a band at 1013 cm.⁻¹ for phenolic 3-acetates which appears to be confirmed in this series by the absorption bands tabulated as c in Table III. In the dinitroestradiol diacetate and the 17-monoacetate there is absorption at 1038 and 1040 cm.⁻¹, respectively, which is reasonably in line with Jones and Herling's assigned range for the 17-acetoxy group near 1030 cm.⁻¹.¹⁵

Nitro Group.—The asymmetric NO₂ stretching vibration can be assigned without difficulty (Table III, B). Where shoulders or doublets occur, the convention is taken that the strongest absorption peak is due to the NO₂ group. The range of 1553-1519 cm.⁻¹ found for these compounds is in the normal region for the aromatic nitro group.¹⁶

The symmetrical NO₂ stretching vibration, between 1360-1300 cm.⁻¹, is in a more complex region in this series marked by overlapping of the strong OH deformation frequencies. Accordingly, except for the acetates,¹⁷ assignment of a particular peak to the *sym*-NO₂ stretching is less reliable than that of the asymmetrical stretching. Tentative assignments are nevertheless made and are given in Table III, b, because this vibration is more indicative of intramolecular hydrogen bonding and ring

(16) Cf. J. F. Brown, Jr., *ibid.*, **77**, 6341 (1955).

(17) It must be noted, however, that estrone acetate presents an anomaly here in also showing a prominent band of medium intensity at 1368 cm.⁻¹ although it does not have a nitro group.

conjugation. Except for the models 6-nitro-3,4-xylenol and 3-nitro-*ar*-2-tetralol, the results are consistent with the interpretation that strong intramolecular bonding with the adjacent OH group and the resulting conjugation with the ring lowers the frequency to 1311–1302 cm^{-1} from the range of 1374–1358 cm^{-1} characteristic of the unconjugated nitro group. In the acetates, the splitting of the symmetrical stretching absorption into a triplet in the 2-nitro and 2,4-dinitro acetates and the shift of the maxima from 1371 to 1346 cm^{-1} , in comparison with 4-nitroestrone acetate, suggests partial interaction with the aromatic ring by the less hindered nitro group. Dinitroestradiol diacetate does not fit the pattern.

Aromatic Ring.—Despite the prominence of the aromatic C=C stretching absorptions, comparatively little has actually been established on the correlation between their intensities and substituents on the ring. More correlations have been noted for the frequencies of the maxima, but not for highly substituted compounds. In this series the 1500 cm^{-1} absorption is generally obscured by overlapping with the asymmetrical NO₂ stretching vibration, although in many cases a doublet or shoulder is discernible. A good study can be made, however, of the 1600 cm^{-1} absorption and of the 1600–1560 cm^{-1} band. These bands are given in Table III as A and a. For non-nitrated compounds where the vibration can be assigned unequivocally, the 1500 cm^{-1} band is tabulated as III, a'.

An unexpected effect observed is that esterification of the phenolic group consistently reduces the intensities of all three of the C=C stretching bands very markedly. Such an effect has not been previously reported, and no explanation has been deduced for it.

Substitution of a nitro group in the hindered (out-of-ring-plane) position also seems to reduce the III, A band, but its effect is less on the intensity of the III, a, band. A nitro group in resonance with the ring is less consistent in its effect on the intensities although it appears to enhance them somewhat in 2-nitroestrone.

A nitro group conjugated with the ring is more noticeable in affecting the frequencies of the bands. Ignoring the weak bands in the acetates, it consistently displaces the III, A, band to 1634–1630 cm^{-1} . Deoxoestrone and estradiol, like the model 3,4-xylenol (and estrone acetate) have the band at 1612–1609 cm^{-1} . Estrone is exceptional in sharing the intermediate range 1620–1628 cm^{-1} with the compounds having an unconjugated nitro substituent. The range of the III, a, band is 1591–1567 cm^{-1} . The most consistent pattern observed is that conjugation lowers the frequency. This III, a, band has not been found especially indicative of conjugation with the ring as has been variously suggested.¹⁸ It occurs, at least in this series, as an independent and characteristic aromatic band.

In addition to their C=C stretching vibrations aromatic compounds are characterized by the C-H deformation bands in the lower frequencies. Although characteristic ranges for the various

mono-, di- and trisubstitutions have been fairly well established, the effect of four and five substituents is not so well delineated because of the comparatively few such compounds studied. Further, groups which are capable of interaction with the ring can shift the frequencies appreciably. The nitro group is notable in this latter respect.¹⁹

The absorption near 750 cm^{-1} which has been observed in many nitro aromatics²⁰ was found in the course of this work to be highly characteristic of a nitro group intramolecularly bonded with an adjacent OH group (or, in a few instances included for comparison, a NH₂ group). It was found to be quite specific, occurring within $\pm 15 \text{ cm}^{-1}$, with very few exceptions, in various disubstituted, trisubstituted, quadrisubstituted and pentasubstituted aromatics. Subsequently, Kross and co-workers have published a study of the interaction of electrophilic substituents with the aromatic ring and have assigned this absorption in *o*-nitrophenol and *o*-nitroaniline to C-H deformation shifted from the normal range by the nitro group in plane with the ring.²¹ This is a plausible inter-

TABLE IV

Compounds	CHARACTERISTIC LOW FREQUENCY BANDS (CM^{-1}) ^{a,b}				
	A	B	C	D	E
Estrone	877m	818s	788m
Estradiol	874m	821m	787w
Deoxoestrone	867s	822w	797w } 782w } ^d
Xylenol	852m	808s	X
Estrone acetate	(864vw)*	810w	794w
4-Nitroestrone	..	821m	795m
4-Nitrodeoxoestrone	..	828m	796w
2-Nitroxlylenol	..	816s	790s
4-Nitrodeoxoestrone acetate	..	*	*
2-Nitroestrone	903m	761s	660s
2-Nitrodeoxoestrone	904m	763s	659s
6-Nitroxlylenol	X	764s	676m
<i>ar</i> -3-Nitro-2-tetralol	922m	762s } 752s } ^d	670m
2-Nitroestrone acetate	912m	(763w)*	(678w)*
2,4-Dinitroestrone	902ms	744ms	..
2,4-Dinitroestradiol	905ms	747ms	..
2,4-Dinitrodeoxoestrone	901m	744m	..
2,6-Dinitroxlylenol	915s	748s	..
<i>ar</i> -1,3-Dinitro-2-tetralol	909m	746m	..

^a A, B, C, D, E, see text. ^b * = weakened bands. X = no assignable band; d = doublet; s, ms, m, w, vw = strong to medium to weak intensity.

pretation, but before definite assignment of the band to C-H deformation is justified further study seems called for. Alternatively, it may arise from a NO₂ mode of vibration reinforced by the hydrogen bonding. Factors which favor this latter interpretation are its restricted range, its relative intensity and the fact that low frequency absorptions have been reported for alkyl nitrites and nitramines. The use of deuterated compounds would seem to resolve the matter. For the present, it is convenient to

(19) It is for these reasons that the assignments made in ref. 5 on the basis of a single C-H deformation frequency are in error.

(20) Bellamy, ref. 9, p. 253.

(21) R. D. Kross, V. A. Fassel and M. Margoshes, *THIS JOURNAL*, **78**, 1332 (1956).

(18) Cf. Bellamy, ref. 9, pp. 60–61.

group the absorption with the C-H deformations in Table IV (as D).

The IV, A, bands can be assigned to C-H absorption characteristic of an isolated ring hydrogen grouping. The normal range is 900-860 cm^{-1} .²² In the 2-nitro and 2,4-dinitro series the band is shifted slightly above 900 cm^{-1} by the nitro substituent. The grouping of 2 adjacent ring hydrogens, which is given the range 860-800 cm^{-1} , occurs in the estrone series and in the 4-nitroestrones. The generally strong band (IV, B) observed in that range can thus also be assigned to C-H deformation. (The nitro group in 4-nitroestrone, not being

coplanar with the ring, does not shift the frequency appreciably.) In the spectra of these compounds in KBr, there is an additional characteristic band (IV, C) near 790 cm^{-1} .

The 2-nitroestrones are distinguished from the 2,4-dinitroestrones in having strong absorption at 660 cm^{-1} (IV, E).

It is interesting to note, finally, that the C-H absorptions, like the C=C stretching bands, are markedly weakened in the acetates.

Acknowledgment.—Grateful appreciation is expressed to R. Hart for his assistance with the preparation of this paper.

CHICAGO 37, ILL.

(22) Bellamy, ref. 9, pp. 67-69.

[CONTRIBUTION FROM THE LABORATORY OF CHEMICAL PHARMACOLOGY, NATIONAL CANCER INSTITUTE, NATIONAL INSTITUTES OF HEALTH]

Synthetic Polysaccharides. I. Polycondensation of Glucose¹

BY PETER T. MORA AND JOHN W. WOOD

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The polycondensation of α -D-glucose to high molecular weight polymers was studied in the temperature range of 140-170° under vacuum and in the presence of 0.164% phosphorous acid. Under the carefully controlled conditions used, no decomposition was observed and glucose was the only product recovered upon subsequent acid hydrolysis. A melt polymerization with infrared heat gave some insoluble gel indicating condensation at other than the glucosidic hydroxyl, but a melt polymerization plasticized with tetramethylene sulfone, and a process where the solidified polymer was powdered and heated in a second stage gave only water-soluble polyglucoses. The mechanism apparently follows that of the A-R-B₁ polycondensation in which the glucosidic hydroxyl (A) can react only with one of the non-glucosidic hydroxyls (B). The influence of temperature and method of polymerization on residual reducing power (unreacted A end group, indicating 5,000-30,000 number average molecular weight), on intrinsic viscosity (0.02-0.07), optical rotation (+58° to +80°), precipitation behavior by alcohol and percentage of non-dialyzable polymer (70-80%) was studied.

The hydrolysis of polysaccharides is a reversible process and there have been many reports on low molecular weight reversion products, mainly disaccharides, formed in aqueous acid solution.² It was also reported that low molecular weight "polyglucosans" can be formed when glucose is dissolved in water-free acids.³ In a preliminary note Pacsu and Mora have shown that polycondensation of glucose and other simple sugars to high molecular weight polysaccharides can be achieved using an acid catalyst, and applying high vacuum to eliminate water produced during condensation.⁴ Subsequently there were reports from other investigators on similar results.⁵⁻⁸ The scope of the general method for polycondensation of carbohydrates and the early preferred methods for the polymerization of glucose appeared in the patent literature,⁹ but there is no detailed description of methods of preparation of polyglucoses with high conversion and without decomposition. Only the macromo-

lecular investigations were published on the solution behavior of high molecular weight branched polyglucoses.¹⁰

The present studies were undertaken to find new methods of producing polyglucose in optimum yield, without decomposition and free of insoluble gel, and also to study the influence of temperature on the polycondensation and on the resulting polymer. A high degree of polymerization of glucose will be favored by (1) an acid catalyst at a concentration which does not cause side reactions (*i.e.*, decomposition) at the reaction temperature, (2) high temperature, (3) effective elimination of the water produced during the condensation, (4) high concentration of glucose, (5) sufficient mobility of the monomer and of the low molecular weight polymers, (6) absence of oxygen and other conditions which may cause degradation, and finally (7) uniform reaction conditions throughout the reaction medium. This paper describes in detail polymerization experiments in which these conditions were fulfilled. It is necessary to adhere scrupulously to the given set of conditions, since relatively minor deviations can cause great differences in the product and may result in excessive decomposition.

We used three methods designed to achieve both good control of reaction variables and uniform reaction conditions: (1) a melt polymerization where infrared heat was applied; (2) a melt polymerization plasticized with an inert solvent (tetramethylene sulfone); and (3) a two-stage process in which

(1) Presented before the Polymer Division of the American Chemical Society at the 131st National Meeting in Miami, Fla., April 5, 1957.

(2) See, for example, C. N. Tarton, A. Bebbington, S. Dixon and E. Pacsu, *THIS JOURNAL*, **77**, 2565 (1955); A. Thompson, K. Anno, M. L. Wolfrom and M. Inatome, *ibid.*, **76**, 1309 (1954); K. Myrbäck, M. Hammerstand and H. Gelinder, *Arkiv. Kemi*, **1**, 235 (1949).

(3) Cf. B. Helferich and S. Böttger, *Ann.*, **476**, 150 (1929); H. M. Schlubach and E. Luhrs, *ibid.*, **547**, 73 (1941).

(4) E. Pacsu and P. T. Mora, *THIS JOURNAL*, **72**, 1045 (1950).

(5) P. W. Kent, *Biochem. J.*, **55**, 361 (1953).

(6) C. R. Ricketts, *J. Chem. Soc.*, 4031 (1954).

(7) C. R. Ricketts and C. E. Rowe, *ibid.*, 3809 (1955).

(8) C. T. Bishop, *Can. J. Chem.*, **34**, 1255 (1956).

(9) P. T. Mora and E. Pacsu, U. S. Patent 2,719,179 (1955); E. Pacsu and P. T. Mora, Canadian Patent, 530,079 (1956).

(10) P. T. Mora, *J. Polymer Sci.*, **23**, 345 (1957).