of 135°. During this period, vigorous evolution of carbon dioxide occurred. After the decomposition reaction subsided, the temperature was raised gradually during 3 hr. to 160°, when crude cyclopentenone together with lactone, dicarboxylic acids, and the acid anhydride[2] were distilled. From the ethereal solution of this crude cyclopentenone acidic material was removed with 5% potassium carbonate solution. The ether layer was washed with water, dried, and evaporated.

In order to separate the cyclopentenone from the mixture, Girard P reagent[15] was employed. To a boiling mixture of 1.4 g. of Girard P reagent, 16 ml. of ethanol, 2.7 ml. of methanol, and 2 g. of acetic acid, 1.2 g. of the crude cyclopentenone was added. After boiling for 1 hr., the mixture was poured into a solution of 3 g. of sodium carbonate in 100 ml. of ice–water. The neutral solution was extracted twice with ether and 12 ml. of 12 *N* sulfuric acid was added. The ether layer was washed with sodium bicarbonate solution as well as saturated sodium chloride solution, dried, and evaporated. On redistillation of the residue there was obtained 0.6 g. (32% based on VIb) of the isomer of *trans*-jasmone, b.p. 85° (1.5 mm.), $n^{20}D$ 1.5069; ν_{max} (neat) 980 (cross-conjugated *trans* double bond), 970 (*trans* double bond), 1698 (carbonyl), and 1640 and 1660 cm.$^{-1}$ (ethylenic bond).

On heating this material at 200° for 2 hr., double bond migration was noticed on an examination of the infrared spectrum.

Anal. Calcd. for $C_{11}H_{16}O$: C, 80.44; H, 9.83. Found: C, 79.77; H, 9.79.

2,4-Dinitrophenylhydrazone of the cyclopentenone melted at 213–215°.

Anal. Calcd. for $C_{17}H_{20}O_4N_4$: C, 59.29; H, 5.85. Found: C, 59.37; H, 6.15.

Similar treatment of 1.82 g. (0.01 mole) of γ-methyl-γ-(3-hexenyl)butyrolactone (VIIb) with polyphosphoric acid afforded 0.65 g. (41%) of the cyclopentenone, b.p. 90° (2 mm.). The infrared spectra were identical with those of the material prepared from VIb.

(15) A. Girard and G. Sandalesco, *Helv. Chim. Acta*, **19**, 1095 (1936).

Dehydration of γ-Methyl-γ-(3-butenyl)paraconic Acid (VIa).—Reaction of γ-methyl-γ-(3-butenyl)paraconic acid (VIa) with polyphosphoric acid was examined under 10 mm. at 135–145°, but the mixture became too sticky, and no volatile material was obtained.

γ-Methyl-γ-(3,4-dibromohexyl)butyrolactone.—Bromination of 18.2 g. (0.1 mole) of γ-methyl-γ-(3-hexenyl)butyrolactone (VIIb) was carried out in 30 ml. of carbon tetrachloride at −5° with 16 g. of bromine. On removal of the solvent *in vacuo*, there was obtained 34 g. of γ-methyl-γ-(3,4-dibromohexyl)butyrolactone, $n^{20}D$ 1.5332, ν_{max} (neat) 1770 cm.$^{-1}$ (lactone carbonyl). Attempted purification failed because the substance decomposed on heating. This material, however, gave satisfactory analyses without further purification.

Anal. Calcd. for $C_{11}H_{18}Br_2O_2$: C, 38.62; H, 5.30. Found: C, 38.98; H, 4.99.

Dehydration of γ-Methyl-γ-(3,4-dibromohexyl)butyrolactone.—A mixture of 1.5 ml. of polyphosphoric acid and 3.4 g. (0.01 mole) of γ-methyl-γ-(3,4-dibromohexyl)butyrolactone was stirred at 145° for 30 min. After treating the reaction mixture in the same way as described above on the paraconic acid (IIIb), there was obtained 2.1 g. of the reaction products which contained the desired cyclopentenone IVb together with unchanged lactone. In order to separate the cyclopentenone, Girard P reagent was employed. From 2.1 g. of the mixture, 1.3 g. (40%) of IVb was obtained. The infrared spectra of the dibromocyclopentenone were superimposable in every fine detail with those of the compound obtained from the corresponding paraconic acid IIIb.

Allethrone (Va) by Dehydration of γ-Methyl-γ-(3,4-dibromobutyl)butyrolactone.—A mixture of 5 ml. of polyphosphoric acid and 6.8 g. (0.022 mole) of γ-methyl-γ-(3,4-dibromobutyl)butyrolactone (prepared from VIIa by bromination as described above) was heated at 130–145° for 30 min., and to this mixture 30 ml. of water and 10 ml. of benzene were added. On debromination of the reaction mixture in the same way as described above, there was obtained 0.6 g. (21% based on dibromobutyrolactone) of allethrone (Va), b.p. 120° (30 mm.). The infrared spectra were identical with those of the compound described in a preceding paragraph.

The Structure of Carnosol

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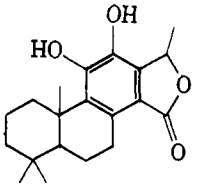
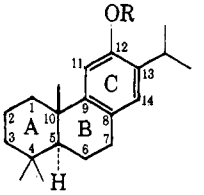
Carnosol is shown to be a phenolic diterpenic lactone of the ferruginol type. Its biosynthesis as well as that of the related tanshinones is discussed. A proton magnetic resonance study of the derivatives of carnosol, ferruginol, and totarol is presented.

Over 20 years ago a bitter principle was isolated from sage, *Salvia carnosa* Dougl.[4] The natural substance was named carnosol and noted to be a diphenolic, ester-containing $C_{19}H_{26}O_4$ hydrophenanthrene. While nothing further has been reported on this compound, another bitter principle, picrosalvin, was isolated recently from two other species of sage, *Salvia officinalis* L. and *Salvia triloba* L.,[5] as well as from rosemary, *Rosmarinus officinalis* L. (see Experimental), and recorded to be a $C_{20}H_{26}O_4$ *o*-diphenolic hydrophenanthrene lactone of structure I. The similarity of the physical and chemical properties of the two principles suggested that they may be the same natural product. As a consequence, a direct comparison of the two substances and their derivatives was made (see Experimental)[6] and their identity established. In view of the priority of the work of



I



IIa, R=H
b, R=Bz
c, R=Ac
d, R=Me

(1) Universität Würzburg.
(2) Indiana University.
(3) National Science Foundation Cooperative Predoctoral Fellow, 1962–.
(4) A. I. White and G. L. Jenkins, *J. Am. Pharm. Assoc., Sci. Ed.*, **31**, 33, 37 (1942).
(5) (a) C. H. Brieskorn and A. Fuchs, *Chem. Ber.*, **95**, 3034 (1962). (b) Undoubtedly this substance is identical with the $C_{20}H_{26}O_4$ compound isolated from *Salvia officinalis* L. by M. M. Janot, H. Pourrat, and J. LeMen [*Ann. pharm. franc.*, **10**, 433 (1952)].

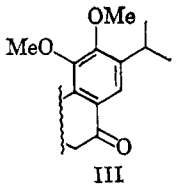
(6) The authors are most grateful to Professor A. I. White for his gift of carnosol and its derivatives.

White and Jenkins,[4] the name carnosol will be used for the bitter principle henceforth.

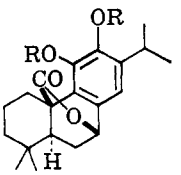
Various results from the last structure analysis militated against formula I for carnosol. The chemical shifts of the hydrogen signals of the methyl groups (0.88, 0.93, 1.15, and 1.26 p.p.m.) in the proton magnetic resonance spectrum of carnosol diacetate could not account for the placement of a methyl group on a phthalide nucleus. The carbonyl absorption, 5.73 μ, in the infrared spectra of carnosol, its diacetate, and its dimethyl ether lay between the regions characteristic of the carbonyl bands of γ- and δ-lactones and, hence, was doubtful support for the phthalide moiety in I. Finally, the possible identity of one of the selenium dehydrogenation products of carnosol with retene[7] was incompatible with formula I and more in line with a carnosol structure based on the ferruginol (IIa) nucleus.

Further strengthening of this view came from the observation of the alkaline hydrogen peroxide oxidation of carnosol yielding isobutyric acid as well as from closer inspection of the p.m.r. spectra of the carnosol derivatives. The spectra of both the diacetate and dimethyl ether revealed signals characteristic of an aromatic hydrogen and isopropyl hydrogens (*vide infra*).[8] These facts in conjunction with the previous data[5] indicated carnosol to be an 11-hydroxyferruginol derivative.

This formulation limited the lactone carbonyl carbon of carnosol to a nuclear position in the ferruginol skeleton, replacing one of the three remaining methyl groups. The oxygen terminus of the lactone group could be located safely at C-7 on the basis of the downfield 5.4 p.p.m. methine signal present in the p.m.r. spectra of the natural product and its two derivatives. Furthermore, the presence of oxygen at C-7, a benzylic position, was confirmed by the hydrogenolysis of carnosol dimethyl ether to a carboxylic acid and by the preparation of a sugiol derivative (III) from carnosol dimethyl ether on alkaline hydrolysis, esterification with diazomethane, and manganese dioxide oxidation. The reported ease of decarboxylation of carnosol and its quinone[5] indicated both endings of the lactone moiety to be benzylic. On the assumption of carnosol being a diterpene biosynthetically related to ferruginol and hence of A–B *trans* configuration, the further structure analysis was predicted on IVa as a reasonable, working formula.

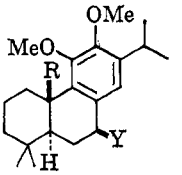


III



IVa, R = H
b, R = Me
c, R = Ac

In order to check the implied carnosol–ferruginol relationship, the following experiments were performed. Carnosol dimethyl ether was converted to a diol (Va) on lithium aluminum hydride reduction and the diacetate (Vb) of the diol was hydrogenolyzed catalytically to a monoacetate (Vc). Hydrolysis of the latter and Sarett oxidation of the resultant alcohol (Vd) gave an aldehyde (Ve) whose Wolff–Kishner reduction led to a degradation product whose structure had to be that of 11-methoxyferruginol methyl ether (Vf) on the basis of the above carnosol formula (IVa). As a consequence, authentic Vf was synthesized.



Va, R = CH_2OH; Y = OH
b, R = CH_2OAc; Y = OAc
c, R = CH_2OAc; Y = H
d, R = CH_2OH; Y = H
e, R = CHO; Y = H
f, R = Me; Y = H
g, R = CO_2H; Y = H
h, R = CO_2Me; Y = H, 7-keto

Treatment of ferruginol benzoate (IIb) with sodium methoxide and thereafter with *p*-nitrobenzenediazonium chloride yielded 11-*p*-nitrophenylazoferruginol. Sodium dithionite reduction of the methyl ether of the azophenol followed by diazotization of the resultant, air-sensitive 11-aminoferruginol methyl ether in methanolic sulfuric acid led to 11-methoxyferruginol methyl ether (Vf), identical in all respects with the carnosol degradation product. Thus, the absolute configuration of carnosol is that shown in IVa.[9]

Biosynthetic Considerations.—As the structure IVa indicates, carnosol represents an interesting high state of oxidation of the ferruginol (II) skeleton. Its oxidized C-7 is reminiscent of similar groupings in the naturally occurring oxidation products of ferruginol: dehydroferruginol (VIa),[10] sugiol (VIb),[11] and cryptojaponol (VIc).[12,13] Since C-7 is benzylic, it would be expected to be a readily oxidizable site in the ferruginol skeleton. An equally easy site of oxidation, in view of its *ortho* relationship to a phenolic hydroxyl group, is C-11. As a consequence, the catechol moiety of cryptojaponol (VIc) and carnosol (IVa) is illustrative of an early intermediate in the oxidative metabolism of the sensitive phenolic ring of ferruginol. Further oxidation of the catechol system would be expected to lead to the hydroxyhydroquinone unit VII and, thereafter, the hydroxyquinone system represented by royleanone (VIIIa),[14] acetoxyroyleanone (VIIIb),[14] dehydroroylea-

(7) The hydrocarbon fraction (B^5) of the dehydrogenation mixture now has been shown by gas phase chromatography to be composed of retene, pimanthrene, and two minor constituents (see Experimental).

(8) An exhaustive study of the p.m.r. spectra of ring-C aromatic, tricarbocyclic, diterpenic substances (R. W. J. Carney, Ph. D. Dissertation, Iowa State University, 1962) has yielded the needed models for the present work.

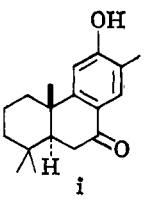
(9) The strain in the six-membered lactone ring accounts for the unusual infrared carbonyl absorption of the natural product.[5]

(10) J. B-son Bredenberg, *Acta Chem. Scand.*, **11**, 932 (1957).

(11) C. W. Brandt and B. R. Thomas, *J. Chem. Soc.*, 2442 (1952).

(12) T. Kondo, M. Suda, and M. Teshima, *J. Pharm. Soc. Japan*, **82**, 1252 (1962).

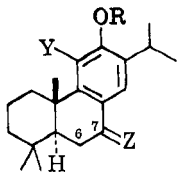
(13) Nimbiol (i), another diterpenic, ring-C phenolic, natural product [E. Wenkert, V. I. Stenberg, and P. Beak, *J. Am. Chem. Soc.*, **83**, 2320 (1961), and references therein] also has its C-7 in a high state of oxidation.



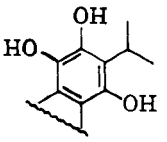
i

(14) O. E. Edwards, G. Feniak, and M. Los, *Can. J. Chem.*; **40**, 1540 (1962).

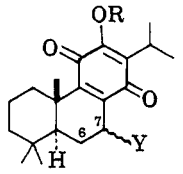
none (VIIIc),[14] and horminone (VIIId).[15] Predictably, *Salvia officinalis* L., the plant whose leaves had yielded carnosol, now was shown to contain royleanone (VIIIa) and the two derivatives VIIIb and VIIIc in its roots.



VIa, R = Y = H; Z = H_2, 6,7-dehydro
b, R = Y = H; Z = O
c, R = Me; Y = OH; Z = O
d, R = Me; Y = NH_2; Z = H_2
e, R = H; Y = *p*-$O_2NC_6H_4N_2$; Z = H_2
f, R = Me; Y = *p*-$O_2NC_6H_4N_2$; Z = H_2
g, R = Ac; Y = H; Z = O
h, R = Me; Y = H; Z = O

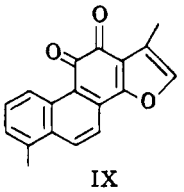


VII

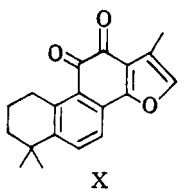


VIIIa, Y = R = H
b, Y = OAc; R = H
c, Y = R = H, 6,7-dehydro
d, Y = OH; R = H
e, Y = H; R = Me

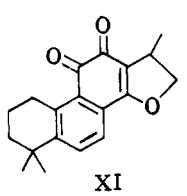
The unusually high state of oxidation of the angular C-10 substituent of carnosol is a clue to the biosynthesis of the natural quinones of *Salvia miltiorrhiza* Bge., tanshinone-I (IX), tanshinone-II (X), and cryptotanshinone (XI).[16] Oxidative decarboxylation of the carnosol system would lead to the aromatic ring B common to the tanshinones, while oxidation of its isopropyl side chain (by benzylic oxidation followed by dehydration or by prototropic rearrangements of the quinone) and subsequent oxidation of the isopropenylcatechol yield an isopropenyl product (XII). Hydration of the latter affords a metastable alcohol (XIII) expected to undergo intramolecular addition to the quinone unit thereby producing the leuco cryptotanshinone functionality (XIV). Further oxidation of the latter would lead to the tanshinones. Thus it appears that carnosol may be an important intermediate in the metabolic paths of oxidative transformations of the ferruginol ring system.



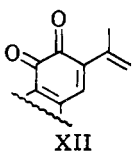
IX



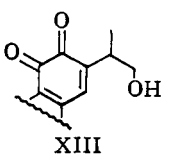
X



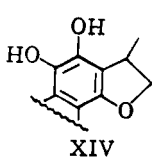
XI



XII

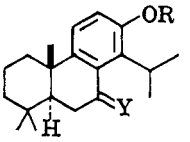


XIII

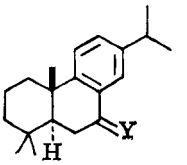


XIV

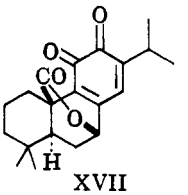
Proton Magnetic Resonance Spectra.—The present study necessitated an inspection of the p.m.r. spectra of carnosol (IVa) and its derivatives. For comparison the spectra of ferruginol (IIa) and totarol (XVa)[17] and their derivatives were studied also. The spectral investigation of 32 compounds—dehydroabietane (XVIa)[18] and its 7-keto derivative (XVIb),[18] ferruginol derivatives IIa–d and VId–f, sugiol derivatives VIb,g,h,[11] totarol derivatives XVa–d,[17,19,20] royleanone derivatives VIIIa,b,d,e,[14,15] and carnosol derivatives IVa–c, Va–h, and XVII—yielded interesting data of diagnostic value in structure determination.



XVa, R = H; Y = H_2
b, R = Ac; Y = H_2
c, R = H; Y = HOH
d, R = Ac; Y = O



XVIa, Y = H_2
b, Y = O



XVII

Ring-A substituents at C-4 and C-10 gave characteristic p.m.r. signals. In the absence of neighboring polar functions the C-4 methyl groups showed a chemical shift of 0.91–0.98 p.p.m. often in form of a six-proton singlet. The methyl signals of the 7-keto compounds appear as two three-proton singlets, one at the normal position (0.92–0.94 p.p.m.) and the other downfield at 0.99–1.01 p.p.m. A similar separation of the methyl signals can be observed in the spectra of the substances containing 10-carbonyl functions. In the cases of the nonrigid 10-carbonyl examples, the equatorial 4-methyl signals appear at normal positions (0.97–0.99 p.p.m.), while the axial 4-methyl groups are shifted diamagnetically to 0.81–0.83 p.p.m. The shielding effect of *ca.* 0.1 p.p.m. by the 10-carbonyl groups on the hydrogens of the axial 4-methyl group is similar to the diamagnetic shielding of the 10-methyl function by the axial C-4 carbonyl-containing groups in podocarpic acid derivatives (XVIII).[8] The 4-methyl signals of the rigid lactones are less well separated (0.86–0.87, 0.90–0.91 p.p.m.). The involvement of the 10-carbonyl

XVIII

(15) This diterpenic quinone was isolated originally by A. Goris from *Horminum pyrenaicum* L. and was shown to be 7-hydroxyroyleanone by J. LeMen and P. Potier. (The authors are most grateful to Professor LeMen for this information.)

(16) (a) R. H. Thomson, "Naturally Occurring Quinones," Academic Press, New York, N. Y., 1957; p. 260; (b) Y. Okumura, H. Kakisawa, M. Kato, and Y. Hirata, *Bull. Chem. Soc. Japan*, **34**, 895 (1961).

(17) The authors are most grateful to Dr. J. C. Dacre (University of Otago, Dunedin, New Zealand) for a sample of totarol acetate.

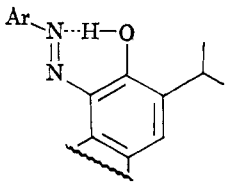
(18) E. Wenkert, P. Beak, R. W. J. Carney, J. W. Chamberlin, D. B. R. Johnston, C. D. Roth, and A. Tahara, *Can. J. Chem.*, **41**, 1924 (1963).

(19) The two 7-oxygenated totarol derivatives XVc and d were kindly furnished to us by Dr. B. R. Thomas.

(20) Y. Chow and H. Erdtman, *Acta Chem. Scand.*, **16**, 1305 (1962), and references therein.

groups in these cases in a new ring distorts them from their original axial conformation and thus alters their nonbonded interaction with the C-4 substituents.

The normal chemical shift of 10-methyl groups is 1.14–1.20 p.p.m., while the 10-methyl signals in 7-keto compounds and in substances containing 11-methoxy and amino groups appear at 1.22–1.24 p.p.m. and 1.31–1.34 p.p.m., respectively. 7-Ketototarol acetate (XVd), a compound whose ketonic function is buttressed seriously by the ring-C substituents and whose ring B, as a consequence, would be expected to be distorted when compared with sugiol acetate (VIg), has its 10-methyl group more shielded, 1.12 p.p.m. While the 10-methyl group in the azo derivative VIf is shielded in a manner similar to that of the 11-amino compound VId, the C-10 substituent in the azophenol VIe is greatly deshielded, 1.63 p.p.m. Presumably, the bulky arylazo group is nonplanar with ring C in the case of the methyl ether VIf (probably protruding perpendicularly on the α-side of the diterpenic framework), while being held in the ring-C plane by hydrogen bonding (*cf.* XIX) in the azophenol (VIe). The 10-methyl signal of royleanone methyl ether (VIIIe), 1.31 p.p.m., is reminiscent of that of the 11-methoxy and amino cases. Hydrogen bonding in the other royleanones appears to be responsible for the diamagnetic shift (1.22–1.25 p.p.m.) of their 10-methyl signals.[21]



XIX

As the above data on methyl groups indicate, the chemical shift of the hydrogens of the C-10 substituents in the presence of 11-methoxy groups is *ca.* 0.3–0.4 p.p.m. downfield of the corresponding hydrogens in a saturated environment. A similar paramagnetic shift was observed for the hydrogens of a 10-aldehydo group, from δ = 9.9 to 10.23 p.p.m. (*cf.* Ve)[8]; of 10-hydroxymethyl groups, from δ = 3.7 to 4.01–4.02 p.p.m. (two-proton, broad singlet)[22]; and of 10-acetoxymethyl groups, from $\delta_A + \delta_B/2$ = 4.1 to 4.55, 4.65 p.p.m. (AB pattern, J = 11.6 c.p.s.).

The two-proton signal characteristic of the 7-methylene group is a broad triplet at 2.7–3.1 p.p.m., while that of the 6-methylene group in 7-keto cases is the AB part of an ABX pattern at 2.6–3.0 p.p.m.[23] The aromatic hydrogen at C-14 of C-7-saturated cases suffers a paramagnetic shift of 1.0–1.3 p.p.m. upon introduction of a 7-keto function. The normal position of the doublet (J = 7 c.p.s.) of the 13-isopropyl methyl groups is 1.16–1.26 p.p.m. The introduction of 7-keto groups causes a paramagnetic shift within this range, while O-methylation or -acetylation of 11- or 12-phenolic groups results in a diamagnetic shift. While the isopropyl methyl signals of the quinones of the royleanone type (VIII) fall within the above range, the doublet of the isopropyl methyl groups of carnosol quinone (XVII) appears at 1.11 p.p.m. Substitution of the 12-hydroxy groups leads to twinning of the isopropyl methyl signals with a separation of the doublet of up to 0.04 p.p.m. Presumably, an increase of the steric requirement of the neighboring function causes a preferred conformation of the isopropyl group wherein its methyl constituents reside in different shielding zones of the aromatic nucleus. This conformation is probably one in which a methyl group and the methine hydrogen scissor the C-12 oxygen-containing function. Even this conformation is destabilized in a fair number of cases of further crowding by C-11 substituents. Thus, in contrast to ferruginol methyl ether (IId) which shows two doublets, its 11-arylazo derivative (VIf) reveals only one.

The isopropyl hydrogen signals of totarol and its derivatives (XV) are anomalous. The downfield shift from within the range of 1.16–1.26 to 1.33 p.p.m. of the methyl doublet in the p.m.r. spectrum of the natural product (XVa) and its similarity with that of 7-hydroxytotarol (XVc), 1.33 and 1.45 p.p.m. (one methyl being further deshielded by the C-7 hydroxyl oxygen), suggest that in these cases of nonbonded interaction with the *peri*-methylene substituents at C-7 the preferred conformation of the isopropyl group is one wherein its methyl groups scissor the phenolic hydroxyl function while the C-7 substituents scissor the isopropyl methine hydrogen. As a consequence, it is not surprising that the septet signal of the central hydrogen of the isopropyl group of totarol (XVa) appears at the extreme downfield end of the 2.9–3.3-p.p.m. range common to all ferruginol-type compounds and that this signal occurs at a more striking downfield position, 3.54 p.p.m., in the spectrum of 7-hydroxytotarol (XVc). As expected, the isopropyl methine hydrogen signal of 7-ketototarol acetate (XVd) is shifted even more paramagnetically, 3.75 p.p.m. The identity of position of the methine hydrogen signal of the isopropyl groups of totarol (XVa) and totarol acetate (XVb) shows the conformation of the alkyl groups of these two substances to be the same. However, the slight twinning of the methyl doublets, 1.24 and 1.26 p.p.m., in the spectrum of the latter indicates a preferred conformation of the acetyl group out of the plane of the aromatic ring. Probably this still applies to the case of 7-ketototarol acetate (XVd, δ_{Me} = 1.23 and 1.33 p.p.m.), although the wider separation of the two doublets suggests that the orientation of the isopropyl group is slightly off its aforementioned conformation.

Experimental[24]

Isolation of Carnosol and the Royleanones.—A survey of the *Labiatae* genera closely related to *Salvia* revealed the presence of carnosol only in *Rosmarinus officinalis* L. Extraction of the leaves[5] and the roots (*vide infra*) gave results parallel with those ob-

(21) Y. Kondo, I. Ikenoue, and T. Takemoto [*Chem. Pharm. Bull.* (Tokyo), **11**, 678 (1963)] reported the chemical shifts of the methyl groups of ferruginol acetate (IIc) and sugiol acetate (VIg), but assigned them incorrectly.

(22) A. Gaudemer, J. Polonsky, and E. Wenkert, *Bull. soc. chim. France*, 407 (1964).

(23) *Cf.* J. B-son Bredenberg and J. N. Shoolery, *Acta Chem. Scand.*, **14**, 556 (1960).

(24) All melting points are uncorrected. Infrared spectra were obtained on Perkin-Elmer spectrophotometers, Model 21 or Model 137 B, with sodium chloride optics. The p.m.r. spectra were taken in dilute deuteriochloroform solution, with tetramethylsilane as internal standard, on a Varian Associates Model A-60 spectrometer. Ultraviolet spectra were obtained with either a Perkin-Elmer spectrophotometer, Model 202, or Zeiss spectrophotometer, PMQ II. Optical rotations are of 95% ethanol solutions recorded on a Rudolph and Sons polarimeter, Model 70. Thin-layer chromatography (t.l.c.) was run on 5 × 20 cm. glass plates coated with silica gel G and developed with 10% either in hexane or 5% ethyl acetate in chloroform. The spots were detected by the use of iodine vapor.

tained for *Salvia officinalis* L.[5]: 1.2% (by dry weight) carnosol in the leaves and the royleanones in the roots. (Unfortunately insufficient root material was available for isolation and characterization, but t.l.c. of the extract gave a pattern similar to that of *Salvia officinalis* L.)

Dried root material 500 g., of *Salvia officinalis* L. was extracted with hexane yielding 10.5 g. of a dark brown resin. This was dissolved in 95% ethanol and allowed to stand. Yellow crystals separated from the solution and were filtered off, yielding 1.5 g. of crystalline material. Its recrystallization from ethanol gave yellow crystals of 7-acetoxyroyleanone, m.p. 211–213°; spectra: infrared (KBr), OH 3.05 (m), C=O and C=C 5.83 (s), 6.05 (s), 6.12 (s), 6.22 (w) μ; ultraviolet, λ_{max}^{MeOH} 272 mμ (log ϵ 4.07) and 406 (2.88); Rast mol. wt. 370. (calcd., 374.5) Direct melting point and spectral comparison with authentic acetoxyroyleanone proved them to be identical.[25]

Evaporation of the filtrate and chromatography of the residue on silica showed the presence of three more hydroxyquinones. The first, eluted by 19:1 hexane–ether, was identified as royleanone by direct comparison with material prepared by hydrogenolysis of acetoxyroyleanone.[14] The second minor hydroxyquinone could not be obtained in pure form by chromatography or crystallization. However, it was identified as 6,7-dehydroroyleanone by comparison of its spectral (infrared and ultraviolet) properties and its t.l.c. behavior with those of authentic 6,7-dehydroroyleanone, prepared by elimination of the acetoxy group of acetoxyroyleanone.[14] The third minor hydroxyquinone has not been obtained in sufficient quantity to allow identification.

Identification of Carnosol.—The p.m.r. and infrared spectra of carnosol and picrosalvin were superimposable and their mixture melting point gave no depression.

Anal. Calcd. for $C_{20}H_{26}O_4$: C, 72.69; H, 7.93. Found: C, 72.91; H, 7.94.

The p.m.r. and infrared spectra of carnosol diacetate and picrosalvin diacetate were superimposable and their mixture melting point gave no depression.

Anal. Calcd. for $C_{24}H_{30}O_6$: C, 69.54; H, 7.29. Found: C, 69.47; H, 7.17.

The p.m.r. and infrared spectra of carnosol dimethyl ether and picrosalvin dimethyl ether were superimposable.

TABLE I

G.P.C. ANALYSIS OF DEHYDROGENATION PRODUCT B[5]

Compound	Reported R_L[a]	Product B R_L[b]
Phenanthrene	1.00	1.00
1-Methylphenanthrene	1.52	1.60
Pimanthrene	2.13	2.14[c]
1-Methyl-7-ethylphenanthrene	2.83	2.74
Retene	3.33	3.30[d]

[a] A. J. Solo and S. W. Pelletier, *Anal. Chem.* **35**, 1584 (1963). [b] Column: 20% silicone gum rubber SE-30 on Chromosorb P 60–80, 6-ft. × 0.25-in. o.d.; temperature of 250° isothermal; He flow rate of 70 ml./min. (on an Aerograph A-90-P). [c] Shown to be identical with pimanthrene by addition of authentic pimanthrene. [d] Shown as identical with retene by addition of authentic retene.

Oxidation of Carnosol.—A solution of 100 mg. of carnosol in 50 ml. of 0.1 *N* aqueous sodium hydroxide was treated with 0.5 ml. of 30% hydrogen peroxide at 75°. The temperature was maintained for 3 hr., during which time four additional 0.5-ml. portions of peroxide were added. The brown-red color of carnosol in alkaline solution disappeared after about 1 hr. The solution was made acid to congo red with phosphoric acid and distilled, the volume being maintained at 20–50 ml. by periodic addition of water. Approximately 100 ml. of distillate was collected. This distillate was made basic with 0.1 *N* sodium hydroxide and concentrated to about 2 ml. Reacidification of the concentrate with phosphoric acid was followed by extraction with five 3-ml. portions of ether. The combined extracts were dried, concentrated, and analyzed by g.p.c. (Perkin-Elmer fractometer 116E, 2 m. × 6.35 mm., silicone oil D.C. 200 at 130°, and 90 ml./min. flow rate of He). The major peak was shown to be identical with isobutyric acid by relative retention time and by admixture of authentic acid.

(25) The authors are indebted to Dr. O. E. Edwards (National Research Council, Ottawa, Canada) who graciously supplied authentic acetoxyroyleanone for comparison.

Reductions of Carnosol Dimethyl Ether.—A mixture of 50 mg. of 10% palladium–charcoal, 80 mg. of carnosol dimethyl ether, and 1 drop of concentrated sulfuric acid in 10 ml. of glacial acetic acid was hydrogenated at 20° for 36 hr. The catalyst was filtered and the solution was diluted with 50 ml. of water and extracted with ether. The extract was washed with water, dried, and evaporated to dryness. Crystallization of the residue from 95% ethanol yielded 45 mg. of colorless crystals of acid Vg, m.p. 210–211°; $[\alpha]^{25}D$ +114° (*c* 0.1); infrared spectrum (Nujol), C=O 5.92 (s) μ.

Anal. Calcd. for $C_{22}H_{32}O_4$: C, 73.30; H, 8.95. Found: C, 73.19; H, 8.87.

To a solution of 1.5 g. of carnosol dimethyl ether in 100 ml. of dry ether was added 0.9 g. of lithium aluminum hydride, and the mixture was refluxed for 3 hr. The excess hydride was destroyed by addition of ethyl acetate followed by 50 ml. of water and 10 ml. of 1 *M* sulfuric acid. The organic layer was separated, washed with water, and dried. Evaporation of the extract and crystallization of the residual gum from methanol–water gave 1.35 g. of colorless crystals of diol Va, m.p. 197–198°, $[\alpha]^{25}D$ +94° (*c* 0.22).

Anal. Calcd. for $C_{22}H_{34}O_4$: C, 72.89; H, 9.45. Found: C, 72.63; H, 9.49.

Acetylation of 1.2 g. of the diol with excess acetic anhydride and pyridine (overnight at room temperature) gave 1.22 g. of a colorless, oily diacetate (Vb) which could not be crystallized; p.m.r. spectrum, 3-proton singlets at 1.90 and 2.14 p.p.m. (acetyl methyls), 2-proton AB pattern $\delta_A + \delta_B/2$ = 4.65 p.p.m. (J = 11.6 c.p.s., acetoxymethyl methylene), 1-proton triplet at 6.08 p.p.m. (J = 8.4 c.p.s., C-7 methine).

A mixture of 0.2 g. of 10% palladium–charcoal and 1.2 g. of the diacetate in 50 ml. of glacial acetic acid was hydrogenated for 12 hr. at room temperature and atmospheric pressure. The catalyst was filtered and the solvent was removed *in vacuo*. The remaining colorless oil of acetate Vc, 1.02 g., showed one spot by t.l.c. but could not be induced to crystallize; spectra: infrared (film), C=O 5.77 (s) μ; p.m.r., 3-proton singlet at 1.86 p.p.m. (acetyl methyl), 2-proton AB pattern $\delta_A + \delta_B/2$ = 4.55 p.p.m. (J = 11.6 c.p.s., acetoxymethyl methylene), 2-proton multiplet at 2.88 p.p.m. (C-7 methylene).

A solution of 0.98 g. of the acetate (Vc) in 10 ml. of methanol and 50 ml. of 5% methanolic potassium hydroxide was refluxed for 0.5 hr. The solution was cooled, diluted with 50 ml. of water, and extracted with ether. The residue, 0.88 g., was crystallized from 95% ethanol yielding colorless needles of alcohol Vd, m.p. 85–86°; $[\alpha]^{25}D$ +98° (*c* 0.18); infrared spectrum (KBr), OH 2.82 (w) C=C 6.26 (w) μ.

Anal. Calcd. for $C_{22}H_{34}O_3$: C, 76.26; H, 9.89. Found: C, 76.35; H, 10.01.

Oxidation of Carnosol Derivatives.—A solution of crude alkaline hydrolysis product of carnosol dimethyl ether,[5] 300 mg., in 25 ml. of ether was treated with excess diazomethane in ether for 2 hr. The solvent was removed *in vacuo* and the residue was chromatographed on silica. Elution with chloroform yielded a colorless oil, 0.270 mg., which could not be crystallized. Treatment of a solution of 250 mg. of the crude hydroxyester in 30 ml. of dry ether with 2.5 g. of activated manganese dioxide for 12 hr. at room temperature, removal of the suspended material by filtration, and evaporation of the solvent gave 245 mg. of colorless, oily keto ester III (showing one spot by t.l.c.), which could not be crystallized; spectra: infrared (film), C=O 5.79 (s), 5.94 (s), C=C 6.29 (s) μ; ultraviolet (methanol), λ_{max} 220 mμ (log ϵ 4.45), 273 (4.12). Its 2,4-dinitrophenylhydrazone melted at 152–155°.

Anal. Calcd. for $C_{29}H_{36}N_4O_8$: N, 9.85. Found: N, 9.66.

A solution of 850 mg. of alcohol Vd was treated with 0.5 g. of chromium trioxide in 20 ml. of pyridine. The mixture was stirred overnight at room temperature, then diluted with 100 ml. of water and extracted with ether. The extract was washed with 1 *N* hydrochloric acid solution and dried; the ether was evaporated. Crystallization of the residue, 750 mg., from methanol gave colorless needles of the aldehyde Ve, m.p. 116–117°; $[\alpha]^{25}D$ +59° (*c* 0.19); spectra: infrared (Nujol), aldehyde CH 3.64 (w), C=O 5.74 (s), C=C 6.35 (w) μ; p.m.r., 1-proton singlet at 10.23 p.p.m. (aldehyde methine).

Anal. Calcd. for $C_{22}H_{32}O_3$: C, 76.70; H, 9.36. Found: C, 76.67; H, 9.32.

11-*p*-Nitrophenylazoferruginol (VIe).—A solution of 1.00 g. of ferruginol benzoate and 5.0 g. of sodium methoxide in 100 ml. of methanol was warmed on a steam bath for 0.5 hr. and then cooled in an ice bath at 0–5°. A solution of *p*-nitrophenyldiazonium

chloride, prepared by the addition of a solution of 210 mg. of sodium nitrite in 10 ml. of water to a cold solution of 400 mg. of *p*-nitroaniline in 20 ml. of 1 *N* hydrochloric acid, was added slowly with stirring. The dye was precipitated by the addition of excess water and filtered. Crystallization from 95% ethanol afforded 640 mg. of 11-*p*-nitrophenylazoferruginol, m.p. 163–165°; spectra: infrared (Nujol), C=C 6.22 (m) μ; ultraviolet (methanol), λ_{max} 283 mμ (log ϵ 2.31), 379 (3.97), 479 (2.54); p.m.r., 1-proton singlet at 6.98 p.p.m. (C-14 H), 4-proton multiplet at 8.05 p.p.m. (aromatic hydrogens).

Anal. Calcd. for $C_{26}H_{33}N_3O_3$: C, 71.69; H, 7.64. Found: C, 71.22; H, 7.72.

11-*p*-Nitrophenylazoferruginol Methyl Ether (VIf).—A solution of 435 mg. of VIe and 1.0 ml. of dimethyl sulfate in 100 ml. of dry acetone was refluxed for 18 hr. over 20 g. of anhydrous potassium carbonate. The carbonate was filtered and the solution was evaporated *in vacuo*. Crystallization of the residue from absolute alcohol gave 385 mg. of the azo compound VIf, m.p. 165–167°; spectra: infrared (Nujol), C=C 6.21 (m) μ; ultraviolet (methanol), λ_{max} 281 mμ (log ϵ 3.87), 335 (2.62), 490 (2.09); p.m.r., 3-proton singlet at 3.43 p.p.m. (O-methyl).

Anal. Calcd. for $C_{27}H_{35}N_3O_3$: C, 72.13; H, 7.85. Found: C, 71.97; H, 7.94.

11-Aminoferruginol Methyl Ether (VId).—A mixture of 350 mg. of VIf and 5.0 g. of sodium hydrosulfite in 100 ml. of 95% ethanol was refluxed on a steam bath. Enough water was added to form a homogeneous solution. After 3 hr. the color of the solution changed from blood red to pale yellow. The cooled solution was then poured into an equal volume of water and extracted with three 50-ml. portions of chloroform. The extract was dried and the solvent was removed *in vacuo*. Chromatography of the residual light brown oil, 319 mg., on neutral alumina (activity I) gave 119 mg. of a yellow oil on elution with 1:1 hexane–benzene, whose p.m.r. spectrum was compatible with 11-aminoferruginol methyl ether: 6-proton singlet at 0.98 p.p.m. (C-4 methyls), 3-proton singlet at 1.34 p.p.m. (C-10 methyl), 6-proton doublet at 1.19 p.p.m. (J = 7.0 c.p.s., i-Pro methyls), 1-proton septet at 3.18 p.p.m. (J = 7.0 c.p.s., i-Pro methine), 2-proton multiplet at 2.78 p.p.m. (C-7 methylene), 3-proton singlet at 3.70 p.p.m. (O-methyl), 2-proton multiplet at 3.85 p.p.m. (amino hydrogens), 1-proton singlet 6.35 at p.p.m. (C-14 H).

11-Methoxyferruginol Methyl Ether (Vf).—A solution of 550 mg. of the aldehyde Ve in 8 ml. of diethylene glycol with 0.5 g. of sodium hydroxide and 1 ml. of 90% hydrazine hydrate was heated to 120°. Methanol, 1 ml., was added and the solution refluxed for 10 hr. Water, methanol, and the excess hydrazine hydrate were removed by distillation, and the residue was refluxed for another 8 hr. at 195–205°. The reaction mixture was cooled, diluted with water, and extracted with ether. The extract was dried and the solvent was evaporated. The dark, viscous residue was chromatographed on 15 g. of neutral alumina (activity I). Elution with 20% ether in hexane gave 60 mg. of a white solid. Crystallization from methanol yielded colorless needles of VIf, m.p. 89–90.5°; $[\alpha]^{25}D$ +104° (c 0.13); spectra: infrared (Nujol), C=C 6.24 (w) μ; p.m.r., 3-proton singlet at 1.31 p.p.m. (C-10-methyl).

Anal. Calcd. for $C_{22}H_{34}O_2$: C, 79.95; H, 10.37. Found: C, 79.78; H, 10.29.

A solution of 119 mg. of VId in 25 ml. of methanol was acidified with 25 drops of concentrated sulfuric acid, cooled to 0–5° in an ice bath and mixed with a solution of 35 mg. of sodium nitrite in methanol. The mixture was allowed to warm slowly to 25° and then was refluxed on the steam bath for 0.5 hr. The cooled solution was neutralized with saturated sodium bicarbonate and extracted with methylene chloride. The extract was dried and the solvent was removed *in vacuo*. A chloroform solution of the residual dark oil was filtered through a short alumina column and the eluted pale yellow oil was distilled. The distillate solidified and was crystallized from methanol yielding 43 mg. of Vf, m.p. 89.5–90.5°; $[\alpha]^{25}D$ +100° (c 0.42); spectra identical with those of Vf above.

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Synthesis of Isoquinolines. I.[1] Copyrine and Isoquinoline Systems Derived from 3-Cyano-4-methylpyridine[2,3]

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3-Cyano-4-methylpyridine was converted to 3-phenylcopyrine and 8-oxo-6-phenyl-5,6,7,8-tetrahydroisoquinoline. In the course of the work, an apparently anomalous intramolecular Ritter reaction was observed.

The synthesis of isoquinoline systems from a preformed pyridine nucleus offers numerous possibilities for placing substituent groups in either ring. As part of a continuing effort along these lines, some further reactions of 3-cyano-4-methylpyridine[5] (**1**) have been explored. The ready availability of this compound[5] as well as its two reactive functions make it an attractive starting material for construction of a second ring. Since some fruitless effort had already been expended upon the preparation of an isoquinoline system,[5] it seemed more reasonable to direct this work toward copyrine (2,7-diazanaphthalene). Thus, a copyrine system was realized and even, finally, an isoquinoline system.

The nitrile function was chosen as the first point of attack (Scheme I). Accordingly, **1** was reduced catalytically to 3-(aminomethyl)-4-methylpyridine (**2**) in 84% yield. The formation of secondary amines[6] was suppressed by saturating the solvent with gaseous ammonia. The amine **2** was characterized by two derivatives, a benzamide and an acetamide (**3**). Attempts to convert **3** to the dicyclic product **4** by boiling in acetic anhydride or by treatment with sodium hydride were not successful.

Treatment of the amine **2** with *m*-nitrobenzaldehyde converted it to the imine **5** in 95% yield. An attempt to convert **5** to the dicyclic product **6** by refluxing it with acetic anhydride and sodium acetate was not successful. However, a product was isolated which

(1) This paper represents the beginning of a new series. For our preceding work on isoquinoline alkaloids, see J. M. Bobbitt, R. Ebermann, and M. Schubert, *Tetrahedron Letters*, 575 (1963).

(2) This paper was presented in part at the 145th National Meeting of the American Chemical Society, New York, N. Y., Sept., 1963.

(3) This investigation was supported in part by Research Grant No. CY-3905 from the National Cancer Institute of the National Institutes of Health, U. S. Public Health Service.

(4) Abstracted in part from the Ph.D. Dissertation of R. E. Doolittle, The University of Connecticut, Storrs, Connecticut, 1963.

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