

PART IV

CONSTITUTION OF DESOXYMORELLIN AND TWO
NEW PIGMENTS FROM GARCINIA MORELLA

Desoxymorellin

The alcoholic mother liquor after the crystallization of morellin from the total colouring matters of the pericarp yields an orange-yellow resin on evaporation. BRINGI¹ chromatographed the hexane-soluble fraction of the resin on a column of Florex XXX (an extruded fuller's earth) and developed the column with hexane. The first deep orange band on elution and evaporation gave a crystalline pigment, provisionally named as desoxymorellin on the basis of elementary analysis and mol. weight. BRINGI also recorded the following data. Desoxymorellin, for which the formula $C_{33}H_{40}O_6$ was suggested on the assumption of $C_{33}H_{38}O_7$ for morellin, formed a monomethyl ether, a mono-p-toluene-sulphonyl ester, and a stable monoacetate. It did not condense with o-phenylenediamine. Hydrogenation of desoxymorellin in the presence of palladium on calcium carbonate yielded a crystalline tetrahydro derivative. By alkali fusion of desoxymorellin phloroglucinol, isovaleric acid and homophthalic acid were isolated. BRINGI concluded that a carbonyl group in morellin was replaced by a methylene group in desoxymorellin, because the main feature of the IR spectrum of desoxymorellin was the disappearance of the band in the 1670 cm^{-1} region.

The UV spectrum of morellin showed three maxima at 236, 289 and 360 $m\mu$; in desoxymorellin the latter two bands were very similar in wavelength and intensity, but the short wave band was much lower in intensity.

In the present work the investigation of the structure of desoxymorellin was continued.

Ethylenic bonds. Desoxymorellin absorbed 4 moles of hydrogen in presence of palladium on charcoal (12 per cent). The first mole was absorbed in 5 min, the second in 10 min, the third in 30 min, and the last in 90 min.

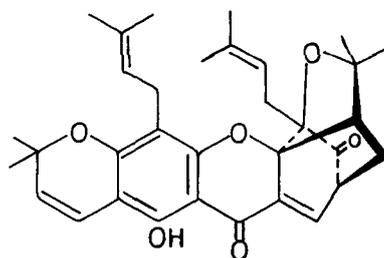
Copper complex. A dioxane solution of desoxymorellin on treatment with excess of aqueous copper acetate gave a red copper complex, which crystallized from chloroform-hexane. Tetrahydro- and octahydro-desoxymorellin, the acetate, and the methyl ether did not form the copper complex. It was therefore obvious that a phenolic hydroxyl group and a readily reducible ethylenic bond in conjugation with a carbonyl group are involved in the copper complex formation, exactly as in morellin.

Mozingo reduction. Desoxymorellin on refluxing with Raney nickel in cyclohexanol for 24 hours gave a crystalline compound, m.p. 112° , identical with the product (MRP-I) obtained by the action of Raney nickel in boiling ethanol

on morellin. The IR spectrum of a 2 per cent solution of either in CCl_4 is recorded in Fig. 19.

Alkali fusion of octahydrodesoxymorellin. Octahydrodesoxymorellin was fused with 5 times the weight of potassium hydroxide pellets and zinc dust at 280° . The phenolic fraction yielded the C_{16} -phenol and 8-isoamyl-5,7-dihydroxy-2,2-dimethylchroman characterized as its dimethyl ether (cf. XXIV - Part III).

Constitution of desoxymorellin. The isolation of 8-isoamyl-5,7-dihydroxy-2,2-dimethylchroman as a major product of alkali fusion, the formation of the copper complex, the behaviour on catalytic hydrogenation, and the UV spectrum demonstrate the identity of desoxymorellin and morellin in the chromenochromanone parts of the molecules. The only difference in fact appears to be in the replacement of the aldehyde group of morellin by a methyl group in desoxymorellin, confirmed by the identity of two Mozingo reduction products. Desoxymorellin may therefore be assigned the structure I, and it probably represents an intermediate stage in the biosynthesis of morellin discussed earlier.



DESOXYMORELLIN

Desoxymorellin - NMR Data

The NMR spectrum of desoxymorellin is given in Fig. 18. It shows that there is no aldehyde function in the molecule. In comparison with morellin, desoxymorellin has one additional C-methyl group giving a singlet absorption. However, this absorption does not occur in the region for methyl groups on double bonds. The actual positions of the methyl signals are 8.23 (1), 8.32 (2), 8.57 (2), 8.64 (1), 8.73 (1) and 8.97 (1). The vinyl triplet occurring at 3.90 and 3.68 in the spectra of morellin and isomorellin is replaced here by one at 5.7 showing that the double bond on which it is situated is no longer conjugated to a carbonyl function. The 2,2-dimethylchromene doublets appear nearly in the same place as in morellin and isomorellin. The vinyl doublet representing the proton on the double bond in the bicyclo-octenone part of the molecule shows up at 2.6. In the region from 6.3 to 8 the spectrum of desoxymorellin is similar to that of isomorellin as may be expected from the absence of the aldehyde function. The chelated hydroxyl appears at -2.84.

All the features of the spectrum, except the absorption position of the additional methyl group, are consistent with the notion that desoxymorellin and morellin differ only in that the aldehyde group of the latter is

replaced by a methyl group in the former. The signal at 8.64 is probably the one due to this additional methyl group. The abnormal position of the signal is probably due to diamagnetic shielding effects.

The change from morellin to isomorellin may involve something more than cis-trans isomerism. The methyl group affected by the isomerization does not show any accompanying large chemical shift, while the aldehyde group does. A simultaneous conformational change is also probably involved which may be responsible for the large diamagnetic shift of the aldehyde absorption as well as the abnormal position of the methyl absorption discussed above. If this turns out to be correct, desoxymorellin may more properly be called desoxyisomorellin. Further clarification of this point is required.

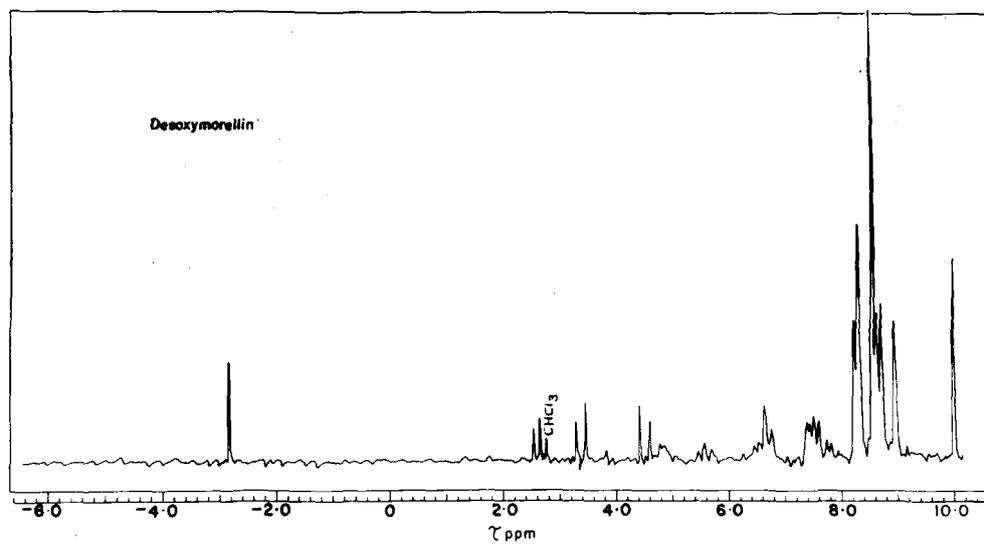
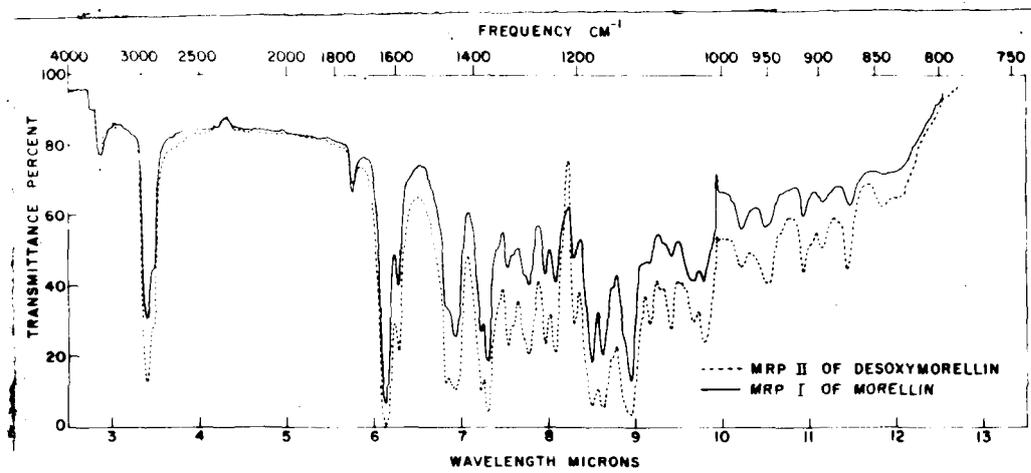


FIG. 18.

FIG. 19. Spectra in CCl₄.

Isolation of two new pigments from *Garcinia morella*

After the isolation of desoxymorellin by the Florex chromatography of the hexane extract of the resin, the second band gave a lemon-yellow crystalline pigment, provisionally called Morellin X. The subsequent band also gave a lemon-yellow pigment provisionally designated as Morellin Y.

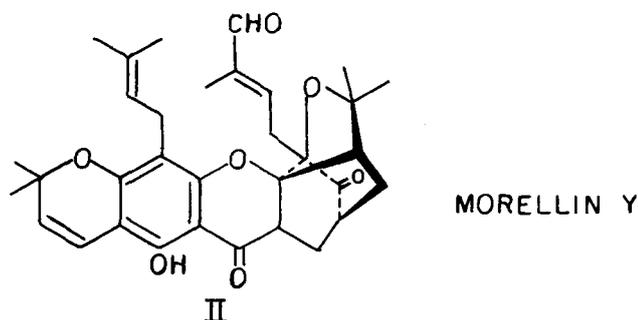
Morellin Y

Morellin Y crystallized in large lemon-yellow needles, m.p. 167° . The elementary analysis and mol. wt. (X-ray method), kindly determined by Professor G. N. Ramachandran, agreed with the molecular formula $C_{33}H_{38}O_7$. It resembled dihydro and other hydrogenated derivatives of morellin in its green ferric colour and absence of copper complex formation. It was evident that the double bond conjugated with the chromanone carbonyl was reduced. This was also confirmed by the reduction in the intensity of the band at $360\text{ m}\mu$ in the UV spectrum. The positive Tollens test and the band at 1685 cm^{-1} ($\alpha\beta$ -unsaturated aldehyde) in the IR spectrum (Fig. 23) suggested the presence of an aldehyde group as in morellin, which was also confirmed by the NMR spectrum.

The NMR spectrum of Morellin Y has many common features with those of morellin and isomorellin. The chelated hydroxyl and aldehyde groups absorb at -1.87 and 0.57. The chromene doublets appear at 3.4 and 4.5. The vinyl proton of the prenyl side-chain of the aromatic nucleus shows up at 5.0. The doublet absorption, representing the vinyl proton on the bicyclo-octenone system of morellin and isomorellin is missing in the spectrum of Morellin Y, suggesting that this double bond is saturated. The triplet absorption at 3.17 represents the vinyl proton of the C₅ side-chain containing aldehyde function. The position of this signal suggests that the side-chain of this compound is the same as in isomorellin. As in morellin and isomorellin seven methyl groups are indicated, three of which are on double bonds.

The absence of copper complex formation, similarities in ferric colour, UV and NMR data, suggested that the compound is probably a dihydro derivative of morellin or isomorellin. Dihydroisomorellin, m.p. 161^o, was prepared by absorbing 1 mole of hydrogen in methanol in presence of palladized carbon, and its properties compared with Morellin Y. The mixed m.p. was undepressed. However, the NMR spectrum of the dihydro derivative of isomorellin

showed that the sample was a mixture of isomers. The chelated hydroxyl signal was split into two. Presumably isomorellin has hydrogenated from either side of the double bond conjugated with chromanone carbonyl. In the methyl region also there were small differences between the two spectra. In other respects the two spectra were identical. It is provisionally suggested that Morellin Y (II) is one of the two dihydro derivatives of isomorellin expected from catalytic hydrogenation of the double bond conjugated with the chromanone carbonyl.



Morellin X

Morellin X crystallized in large lemon-yellow rods, m.p. 143° . Repeated elementary analysis and the mol. wt. (585 ± 1), kindly determined by Professor G. N. RAMACHANDRAN, corresponded to the molecular formula $C_{35}H_{40}O_8$. It resembled dihydro and other hydrogenated derivatives of morellin in its green ferric colour. In presence of palladium on charcoal it absorbed 3 moles of hydrogen, the first in 12 min, the second in 1.25 hr, and the third in about 4 hr. Alkali fusion of hexahydromorellin X yielded the C_{16} -phenol (5%) and 8-isoamyl-5,7-dihydroxy-2,2-dimethylchroman (30%), the latter being isolated as the dimethyl ether. Unlike morellin and desoxymorellin,

and like dihydromorellin, it did not form a copper complex, indicating the absence of the double bond in conjugation with the chromanone carbonyl which is present in morellin. This was confirmed by the marked reduction in the intensity of the band in the 360 $m\mu$ region of the UV spectrum in comparison with morellin. The positive Tollens test, formation of a diacetate, the 1690 cm^{-1} band in the IR spectrum (Fig. 21) ($\alpha\beta$ -unsaturated aldehyde), and the signal at 0.57 in the NMR spectrum indicated the presence of an aldehyde group as in morellin. These data show that Morellin X is very similar in structure to morellin, and further work is in progress to determine the character of the C_2H_2O residue.

Morellin X - NMR data

The NMR spectrum of morellin resembles that of Morellin Y very closely. Four out of the five vinyl absorptions and seven out of the eight \underline{C} -methyl absorptions are almost exactly in the same place. The chelated hydroxyl and aldehyde groups appear with very minor shifts at -1.95 and 0.57. The additional vinyl proton gives a doublet at 5.55 ($J=5$ cps) and the additional methyl signal is a triplet ($J=6.5$ cps) superposed on tertiary methyl signal at 8.86. The triplet methyl absorption shows that Morellin X has an ethyl group. In the region from 6 to 8

the spectrum is quite complex. The total proton count for this molecule has not been made very accurately. Further data have to be obtained before a structure for the compound can be suggested. However, it is clear that the left half of the structure of morellin is retained in this pigment. The C_5 side-chain containing the aldehyde group is also present in it.

In a note in 1953 MURTHY and RAO² mentioned the isolation from the seed coat of Garcinia morella of a second pigment named moreollin in lemon-yellow hexagonal prisms, m.p. 168° , which isomerized to isomoreollin, m.p. 142° , by treatment with 5 per cent alcoholic sodium hydroxide at 40° . They have also referred to another pigment, guttiferin ("a highly active antibiotic present throughout the plant"). The impossible formula $C_{24}H_{27}O_6$ (with an odd number of hydrogen atoms) was assigned to moreollin, and some colour reactions were described. No further details concerning moreollin and guttiferin appear to have been published.

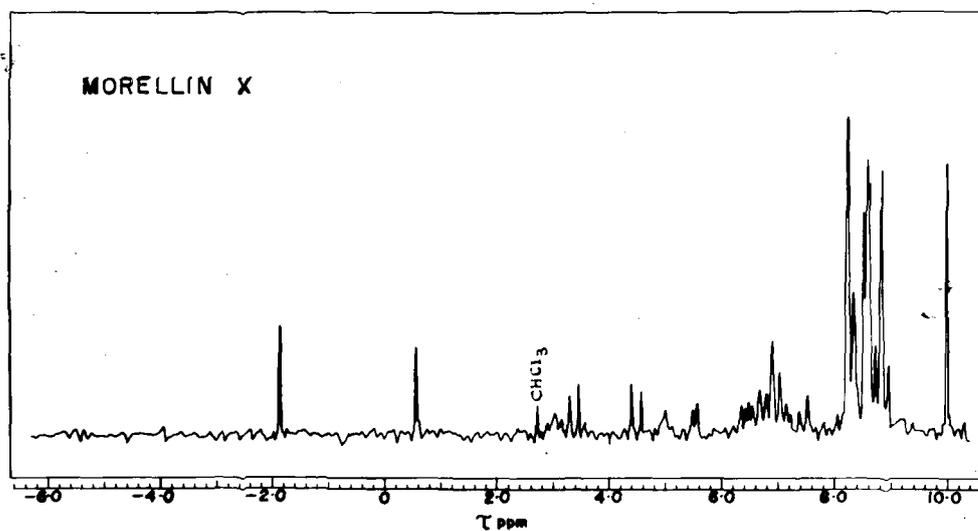
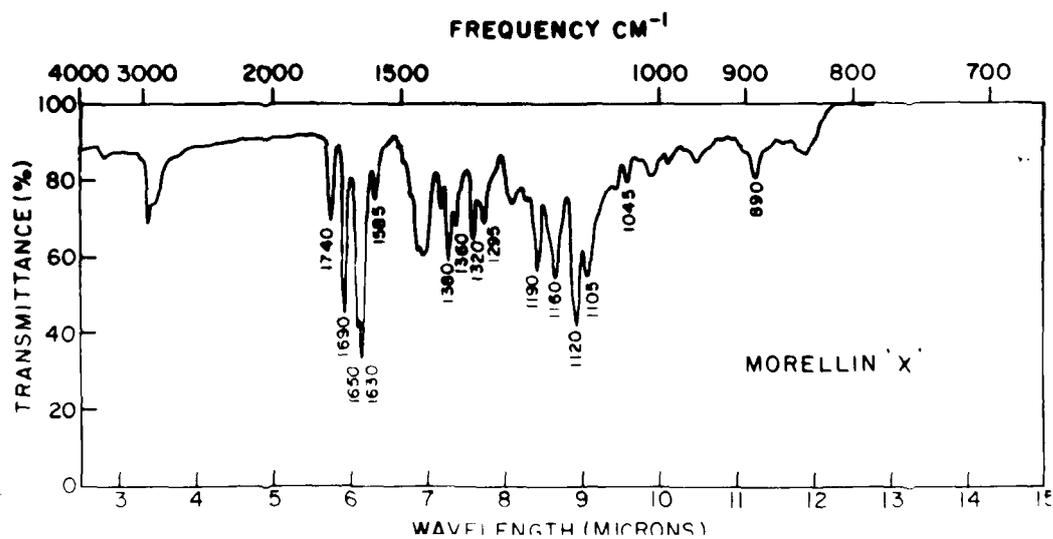


FIG. 20.

FIG. 21. Spectrum in CCl₄.

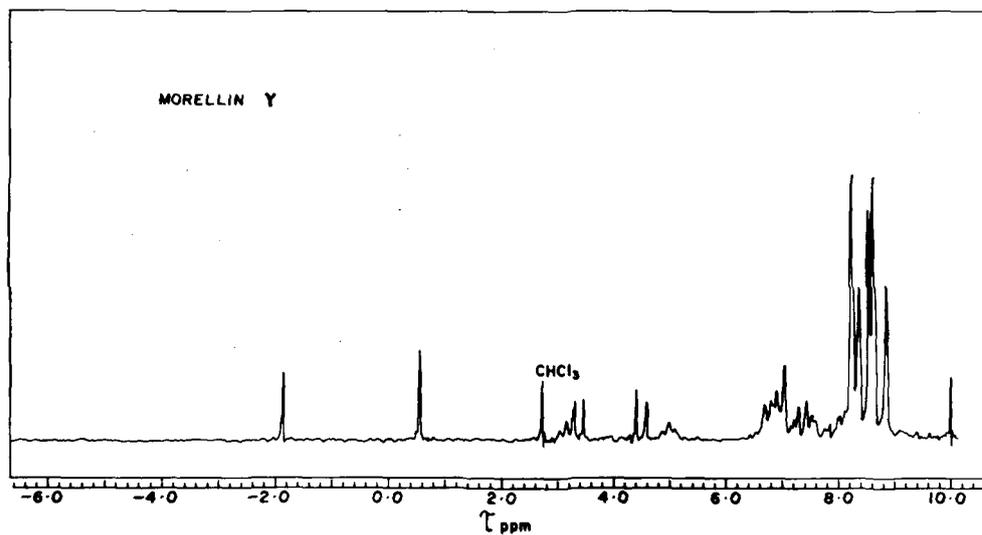
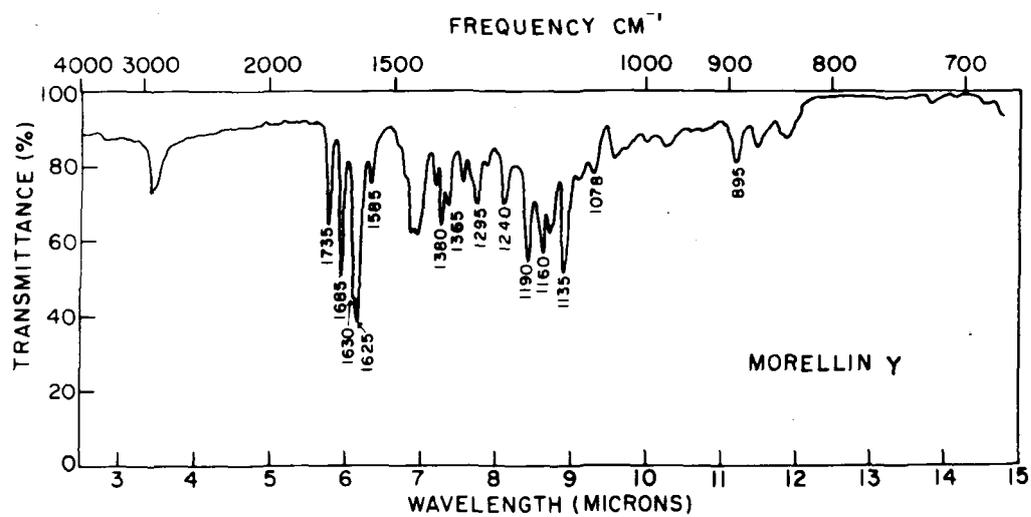


FIG. 22.

FIG. 23. Spectrum in CCl_4 .

EXPERIMENTAL

Isolation of Morellin X and Morellin Y

The orange-yellow resin isolated after the evaporation of the alcoholic mother liquor of crude morellin obtained by the extraction of the pericarp of Garcinia morella, was extracted with hot hexane (60-80°). The extract was cooled to room temp and was chromatographed through a column of Florex XXX (900 g packed in a column of 6 x 1050 cm). The first deep orange band on elution and distillation gave an orange gum which readily crystallized from methanol as orange needles, m.p. 126°, undepressed when mixed with an authentic sample of desoxymorellin (3.0 g).

The next pale-yellow band gave a lemon-yellow gum, which on crystallization from methanol yielded large rods of Morellin X, lemon yellow in colour (2.0 g), m.p. 143°. (Found: C, 71.4, 71.6; H, 7.0, 7.4. \underline{M} by X-ray crystallography, 585 ± 1. $C_{35}H_{40}O_8$ requires: C, 71.4; H, 6.8%. \underline{M} , 588. $C_{35}H_{38}O_8$ requires: C, 71.7; H, 6.5%. \underline{M} , 586). It gave a green ferric colour and did not answer Mg-HCl test. It did not form a copper complex.

The third band gave pale-yellow crystalline needles of Morellin Y, which were recrystallized from methanol in rods (0.5 g), m.p. 167°. (Found: C, 72.4, 72.5; H, 6.6, 6.6. \underline{M} by X-ray crystallography method, 545 ± 1. $C_{33}H_{38}O_7$ requires: C, 72.5; H, 7.0%. \underline{M} , 546). It gave a green

ferric colour and no colour with magnesium and hydrochloric acid. It also did not form a copper complex.

Copper complex of desoxymorellin

The pigment (0.2 g) in dioxane (2 ml) was treated with excess of copper acetate solution (25 ml) prepared by dissolving 5.0 g of copper acetate monohydrate and 2 g of crystalline sodium acetate in 200 ml of water. The red-brownish precipitate was collected and crystallized from chloroform-hexane mixture in blood-red needles. (Found: C, 70.2; H, 6.3. $C_{33}H_{37}O_6 \cdot \frac{Cu}{2}$ requires: C, 70.6; H, 6.6%).

Raney nickel-cyclohexanol reduction of desoxymorellin

In a 150 ml 3-neck flask, equipped with a mechanical stirrer and a distillation condenser, Raney nickel (12 g) over alcohol was placed and toluene (80 ml) was added and distilled with continuous stirring. When all the water and alcohol were distilled by azeotropic distillation, most of the toluene was distilled. Anhydrous cyclohexanol (30 ml) and desoxymorellin (1.0 g) were added and the mixture refluxed for 24 hr under stirring.

The mixture was filtered hot over a bed of Hyflo Super-Cel and washed with warm toluene (50 ml). The combined filtrate was steam-distilled. The colourless residue on treatment with methanol gave colourless needles (0.6 g), m.p. 110° . Mixed m.p. with MRP-I of morellin was undepressed

and the IR spectra of the two in CCl_4 were superposable (Fig. 19).

Alkali fusion of octahydrodesoxymorellin

Desoxymorellin (3.0 g) was hydrogenated in methanol (30 ml) in presence of palladium on carbon (12%; 0.8 g). When the absorption ceased, after 4 moles it was filtered into a 10 ml 3-neck flask and evaporated to dryness. Potassium hydroxide (17.0 g) and zinc dust (1.5 g) were added and the mixture slowly heated in a metal-bath under stirring with nitrogen atm. A rubber-like mass floating in milk-white fluid separated at 190° . A clear chocolate-yellow melt was obtained at 255° , when peppermint odour was noticed. After keeping it at 255° for 2 min, it was cooled and diluted with water (150 ml). It was filtered and acidified with 1:1 sulphuric acid when a pale-yellow sticky mass separated with a strong odour of fatty acids. The mixture was extracted with ether and washed with 10% aqueous sodium bicarbonate, sodium carbonate and sodium hydroxide. The bicarbonate extract was acidified and the mixture of acids kept aside.

Sodium carbonate soluble fraction

The carbonate solution was acidified and saturated with salt and extracted with ether. Distillation of the solvent gave a pale-yellow semi-crystalline mass which was

recrystallized from acetone-hexane mixture (0.1 g), m.p. 162° ; mixed m.p. with $C_{16}H_{24}O_4$ phenol was undepressed. The mother liquor was mixed with the sodium hydroxide soluble fraction.

Sodium hydroxide soluble fraction

The solution was acidified and extracted with ether. Distillation of ether extract gave a pale-yellow gum (0.6 g). Thin layer chromatography of the gum on silica gel with 5% ethyl acetate-benzene as solvent showed a small stationary spot corresponding to C_{16} -phenol and a major spot corresponding to the major spot in the similar fraction from the alkali fusion of octahydromorellin. The mother liquor of C_{16} -phenol was mixed with the gum and refluxed with acetone (50 ml), potassium carbonate (10 g) and dimethyl sulphate (3 ml) for 8 hr. After removal of acetone, the residue was diluted with water. The gum was extracted with ether and washed. The product obtained after removal of ether was chromatographed on alumina in hexane. The bluish fluorescent band was eluted and evaporated. A colourless viscous oil was obtained which crystallized from dil methanol in colourless fluffy needles (0.3 g), m.p. 43° . Mixed m.p. with the synthetic sample of 8-isoamyl-5,7-dimethoxy-2,2-dimethylchroman was undepressed and the IR spectra of the two were superposable.

Neutral fraction

Distillation of ether extract containing the neutral fraction, yielded a gum (0.2 g) with a green ferric colour. It had a strong peppermint odour.

Acetylation of Morellin X

Morellin X (0.2 g) was gently refluxed with acetic anhydride (3 ml) and pyridine (3.0 ml) for 5 hr and the pale-red solution poured into crushed ice (20 g). The pale-yellow solid that separated was readily crystallized from methanol in pale-yellow needles (0.15 g), m.p. 178°. (Found: OAc, 12.5. $C_{39}H_{44}O_{10}$ requires: OAc, 12.0%).

Alkali fusion of Hexahydromorellin X

The pigment (2.0 g) was hydrogenated in methanol (20 ml) in presence of palladized carbon (0.6 g; 10%). When the absorption stopped after 3 moles, it was filtered and evaporated. The pale-yellow product was fused with potassium hydroxide at 270°. On working up the phenolic fraction as in the case of octahydromorellin and octahydro-desoxymorellin, 8-isoamyl-5,7-dimethoxy-2,2-dimethyl-chroman (0.2 g) and C_{16} -phenol (50 mg) were obtained.

Dihydroisomorellin

Isomorellin (0.5 g) was hydrogenated in methanol (15 ml) in presence of palladized carbon (0.2 g; 10%). When it

absorbed (22.5 ml; 1 mole), it was filtered and concentrated, when pale-yellow needles readily separated (0.4 g), m.p. 160-161°. Mixed m.p. with Morellin Y was 166-167°.

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- ² D. V. KRISHNAMURTHY and P. L. N. RAO, J. Sci. Ind. Res. (India) 12B, 565 (1953).