

CRYSTALLINE CHEMICAL COMPONENTS OF THE
SEEDS OF *CASSIA TORA* LINN. IDENTITY OF
TORA SUBSTANCE C WITH RUBROFUSARIN
AND TORA SUBSTANCE B WITH
NOR-RUBROFUSARIN

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THE isolation of three crystalline substances from the seeds of *Cassia tora* Linn. designated as Tora Substances A, B and C was reported in a brief note from these laboratories some time back.¹ From the properties of these substances and some typical derivatives it appeared probable that tora substance C might be identical with rubrofusarin, a metabolic product of the fungus *Fusarium culmorum*² and tora substance B with nor-rubrofusarin, the demethylation product of rubrofusarin. This probable identity was also indicated in the earlier note. Detailed work on the tora substances has not been possible so far, since the yield of the substances is very poor and their separation from one another and from other companion substances is very difficult.

Raistrick and Coworkers² who reported the results of their investigations on rubrofusarin in 1937 considered it to be a monomethylmonomethoxy-dihydroxyxanthone, the exact locations of the substituent groups being left undefined. Though this assignment of the compound to the xanthone group remained uncontradicted there was no positive support for it from the chemical or physical angle for some years. On the other hand there has been a growing feeling that it may not be a xanthone compound at all. Mull and Nord³ suggested two possible structures on spectral grounds, which were subsequently considered to be insufficient by Lund *et al.*⁴ Recently interest in the structure of rubrofusarin has been actively revived and it has now been definitely established from chemical evidence^{5, 6} that it is a 2-methylnaphtho- γ -pyrone with the alternative structures (I) and (II), of which the former has been proved to be the right one by X-ray crystallography.^{5, 7} In view of this current interest in the subject, an attempt has been made to establish the identity or otherwise of tora substance C with rubrofusarin by a direct comparison of the two substances employing their infra-red spectra. The sample of rubrofusarin kindly provided by Prof. Birkinshaw from Prof. Raistrick's collection and tora substance C have been found to possess

identical I.R. spectra, thus confirming their identity suspected earlier on the basis of chemical data.

As has already been reported¹ the trimethyl ether of tora substance B and the dimethyl ether of tora substance C have identical composition and properties and their mixed m.p. is undepressed. Also the dimethyl ether of tora substance B and the monomethyl ether of tora substance C have identical composition and properties and their mixed m.p. is undepressed. It has now been found that these two ethers have also identical infra-red spectra. Thus these two substances are identical in all respects. From the observed relationship between tora substances B and C it can be safely concluded that tora substance B is indeed nor-rubrofusarin.

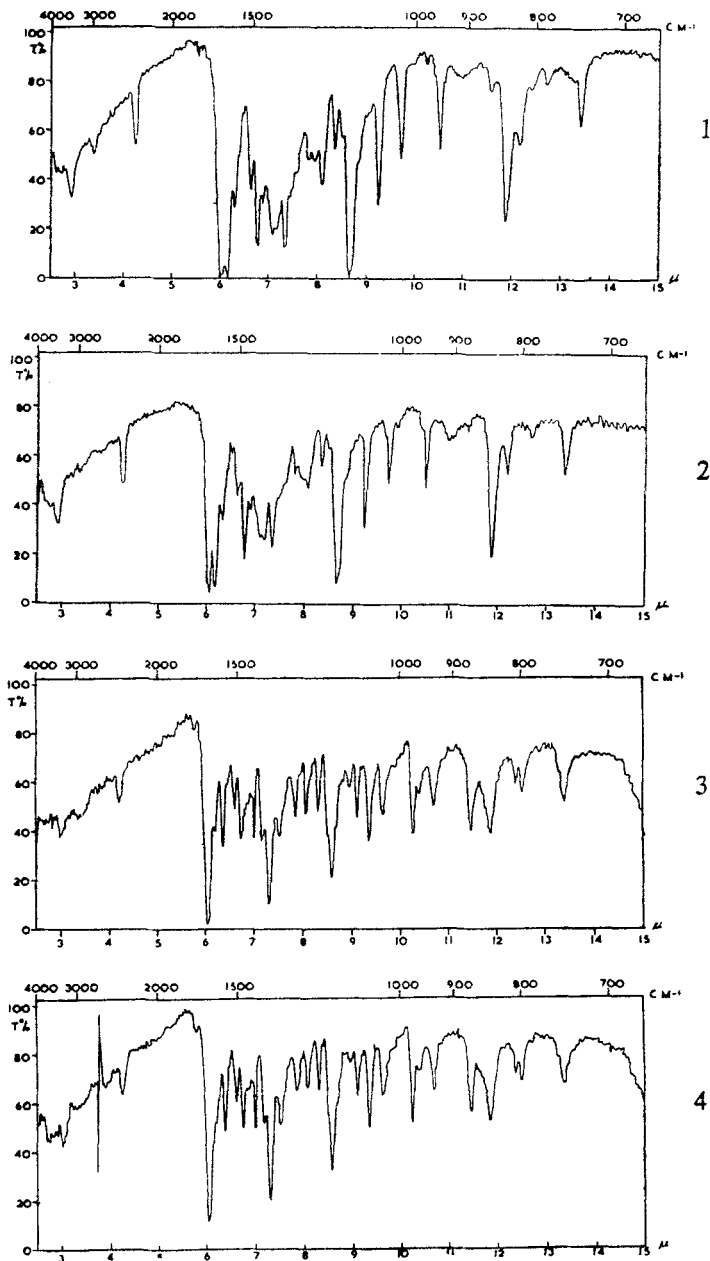
Rubrofusarin affords one of the rare instances where the same chemical substance has been found both as a mould product and as a product of higher plants. In view of the occurrence of the nor compound in the higher plant it is quite possible that it occurs in the mould also and has been missed.

Comparative data on the physical constants of the substances and derivatives in the two (fungal and higher plant) series are presented below in a compact form.

		Tora substance C	Rubrofusarin
Formula	..	$C_{16}H_{12}O_5$	$C_{15}H_{12}O_5$
m.p.	..	208-09°	210-11°
m.p. of diacetate	..	267-69°	260°
m.p. of monomethyl ether	..	207-09°	203-04°
m.p. of dimethyl ether	..	183-85°	187-88°

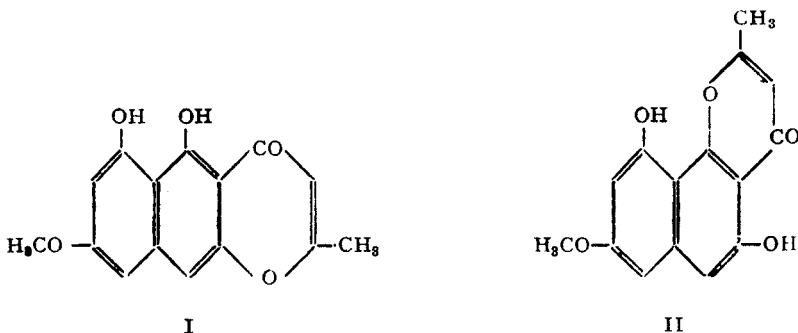
		Tora substance B	Nor-rubrofusarin
Formula	..	$C_{14}H_{10}O_5$	$C_{14}H_{10}O_5$
m.p.	..	286-288° (<i>d</i>)	280° (<i>d</i>)
m.p. of diacetate	..	206-09°	204°
m.p. of dimethyl ether	..	207-09°	203°

The infra-red spectra referred to earlier are reproduced in Figs. 1-4.



FIGS. 1-4. Infra-red spectra of tora substance C (Fig. 1); Rubrofusarin (Fig. 2); Dimethyl ether of tora substance B (Fig. 3); Monomethyl ether of tora substance C (Fig. 4). All spectra were taken with a Perkin-Elmer Infra-cord spectrophotometer using KBr disc. *N.B.*—The big wave between $3.7\ \mu$ and $3.8\ \mu$ in Fig. 4 was caused by current fluctuation.

Details of the preparation and properties of the derivatives of the tora substances have not been published so far and are therefore presented here.



EXPERIMENTAL

Tora Substance C (= Rubrofusarin)

The isolation of this substance from the seeds of *C. tora* has already been described.⁸ It was obtained as shining red needles from chloroform-acetone, m.p. 208–09°. It was sparingly soluble in aqueous sodium hydroxide but dissolved in concentrated ammonium hydroxide giving a deep yellow solution which slowly turned orange. With concentrated sulphuric acid it gave a greenish-yellow solution in which the rim slowly turned orange; addition of a crystal of potassium dichromate yielded a reddish-violet colour which rapidly turned brown. Alcoholic ferric chloride gave a greenish-brown colour. Found: C, 66.7; H, 4.8; —OCH₃, 11.0%. C₁₅H₁₂O₅ requires C, 66.1; H, 4.4; —OCH₃ (1), 11.4%. For I.R. spectrum see Fig. 1.

Diacetate.—It was prepared by heating the substance (0.2 g.) with acetic anhydride (3 ml.) and pyridine (1.6 ml.) at 100° for 4 hours. The product crystallised from benzene-petroleum ether as almost colourless rods, m.p. 267–69°. Found: C, 64.6; H, 4.8; —OCH₃, 9.0; —COCH₃, 23.9%. C₁₈H₁₆O₇ requires C, 64.0; H, 4.5; —OCH₃ (1), 8.7; —COCH₃ (2), 24.2%.

Monomethyl ether.—This was prepared by treating a benzene solution of the substance with excess of ethereal diazomethane and leaving overnight. After crystallisation from benzene-petroleum ether and then from absolute alcohol it was obtained as clusters of pale yellow needles, m.p. 207–09°. For I.R. spectrum see Fig. 4. Found: C, 66.9; H, 4.7; —OCH₃, 22.1%. C₁₆H₁₄O₅ requires C, 67.1; H, 4.9; —OCH₃ (2), 21.7%.

Dimethyl ether.—This was prepared by heating tora substance C (300 mg.) in acetone solution (60 ml.) with excess of dimethyl sulphate

(1.2 ml.) and anhydrous potassium carbonate (4.5 g.) under reflux for 12 hours. The dimethyl ether obtained by evaporation of the acetone filtrate crystallised from benzene-petroleum ether as glistening colourless needles, m.p. 183–85°. Found: C, 68.6; H, 5.8; $-\text{OCH}_3$, 29.8%. $\text{C}_{17}\text{H}_{16}\text{O}_5$ requires C, 68.0; H, 5.4; $-\text{OCH}_3$ (3), 31.0%.

Tora Substance B (= Nor-rubofusarin)

The isolation of the substance from the seeds of *C. tora* has already been described.⁸ It was obtained as yellow needles from alcohol or acetone, m.p. 286–88° (decomp.). It gave a yellow solution with aqueous sodium hydroxide or sodium carbonate and a light pink solution with concentrated sulphuric acid. Alcoholic ferric chloride gave a greenish-brown colour. It contained no methoxyl. Found: C, 65.0; H, 4.4%. $\text{C}_{14}\text{H}_{10}\text{O}_5$ requires C, 65.1; H, 3.9%.

Diacetate.—This was prepared by heating the substance (50 mg.) with acetic anhydride (1 ml.), fused sodium acetate (200 mg.) and glacial acetic acid (0.5 ml.) under reflux for 20 minutes and pouring into water. It crystallised as golden yellow needles from rectified spirit, m.p. 206–09°. Found: C, 63.3; H, 4.4; $-\text{COCH}_3$, 23.6%. $\text{C}_{18}\text{H}_{14}\text{O}_7$ requires C, 63.2; H, 4.1; $-\text{COCH}_3$ (2), 25.2%.

Triacetate.—This was prepared by heating tora substance B (0.1 g.) with acetic anhydride (2 ml.) and pyridine (1 ml.) at 100° for 5 hours. The product obtained by pouring into a large excess of water and filtering was taken up in benzene and the ice-cold solution washed with ice-cold 2% aqueous sodium hydroxide. The alkali-insoluble portion was recovered and crystallised from benzene-petroleum ether, when light yellow needles, m.p. 216–18°, were obtained. Found: C, 63.2; H, 4.7; $-\text{COCH}_3$, 32.3%. $\text{C}_{20}\text{H}_{16}\text{O}_8$ requires C, 62.5; H, 4.2; $-\text{COCH}_3$ (3), 33.6%.

Dimethyl ether.—This was prepared by treating a benzene solution of the substance with excess of ethereal diazomethane and leaving overnight. It was crystallised from benzene-petroleum ether and then from absolute alcohol as pale yellow needles, m.p. 207–09°. Found: C, 67.0; H, 4.8; $-\text{OCH}_3$, 20.7%. $\text{C}_{16}\text{H}_{14}\text{O}_5$ requires C, 67.1; H, 4.9; $-\text{OCH}_3$ (2), 21.7%. For I.R. spectrum see Fig. 3. This ether did not depress the m.p. of the monomethyl ether of tora substance C.

Trimethyl ether.—This was prepared by heating tora substance B (200 mg.) in acetone solution (60 ml.) with excess of dimethyl sulphate (1 ml.) and anhydrous potassium carbonate (4 g.) under reflux for 12 hours. The ether obtained by evaporation of the acetone filtrate

crystallised from benzene-petroleum ether as colourless needles, m.p. 186–89°. Found: C, 67·8; H, 5·6; —OCH₃, 30·0%. C₁₇H₁₈O₅ requires C, 68·0; H, 5·4; —OCH₃ (3), 31·0%. This substance did not depress the m.p. of the dimethyl ether of tora substance C.

SUMMARY

The identity of tora substance C with rubrofusarin and of tora substance B with nor-rubrofusarin has been established.

ACKNOWLEDGEMENT

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