

STUDIES ON SANDAL SPIKE

Part IV. A Histochemical Study of the Sandal (*Santalum album* Linn.) Root Haustoria in Relation to Mineral Nutrition

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SANDAL (*Santalum album* Linn.) is a root parasite. The formation and morphology of its root haustoria was studied in detail by Barber^{1, 2} and later by Rao³; the former author showed that root-hairs are almost absent on sandal roots. While the plant possibly derives much of its organic nutrition from the host plant, the absence of root-hairs suggests the possibility of its dependence for the inorganic nutrition also on the host plant. In continuation of our investigations on the physiology of sandal in health and disease, a histochemical study of the root haustoria of the plant in relation to its inorganic nutrition has now been undertaken and the results are reported in this paper.

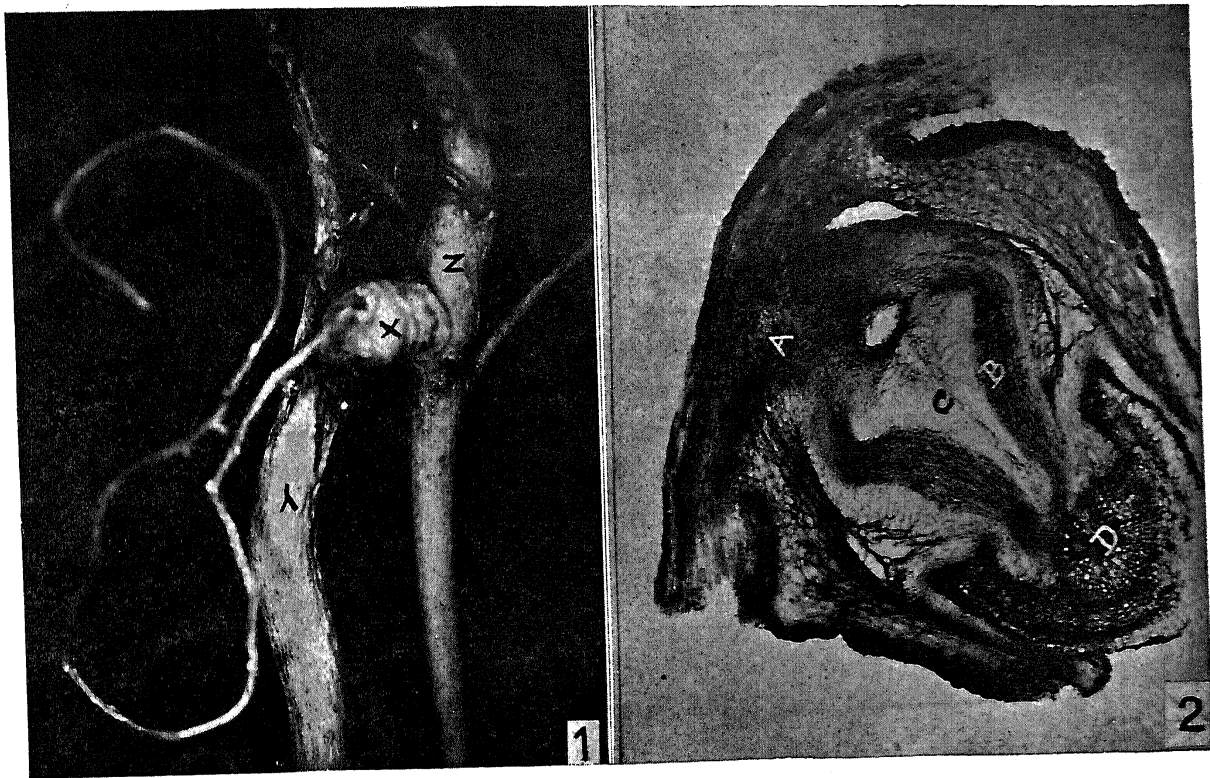
Sandal haustoria in association with *Bauhinia purpurea*, *Tecoma stans*, *Leucaena glauca* and *Broussonetia papyrifera* were taken for sectioning in the present study. Free-hand sections of haustoria fixed in formalin-acetic acid-alcohol were used, after staining with safranin, to see the vascular system, while those of fresh material or material fixed in alcohol wherever necessary, were used for the histochemical study. The elements tested for were K, Ca, Fe, Mg, Zn and Cu, and the tests were done following recognised methods.^{4, 5} For the detection of iron *ortho*-phenanthroline was used for staining (red) after the sections were first treated with acetate buffer (pH 3.5) and hydroquinone solution. This technique was developed basing upon the method of estimation of iron by *ortho*-phenanthroline.⁶

RESULTS AND DISCUSSION

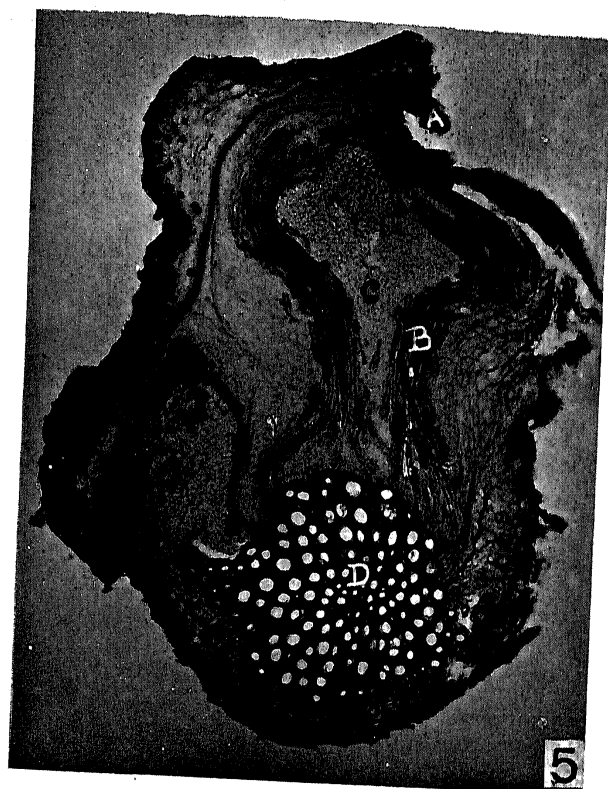
The haustorium in sandal (Fig. 1) is almost conical in shape, the apex rising from the sandal root. There is a direct vascular connection between the sandal and the host roots through the haustorium (Figs. 2-6). The mouth of the vascular cylinder, which bears the shape of an inverted flask, is in the vascular bundles of the host, the body being near the apex of the haustorium. The vascular cylinder spreads into individual strands at the

TABLE I

Sl. No.	Element tested	Reagent used	Response observed	Location in which the response was observed
1	Potassium ..	(a) Sodium cobaltnitrite (b) Platinum chloride	Light yellowish crystals with both the reagents showing the formation of potassium cobaltnitrite and potassium chloroplatinate respectively	Present in the cells in between and around the vascular strands
2	Calcium ..	Oxalic acid	White turbidity indicating the formation of calcium oxalate	In the vascular system of the haustoria and the cells just around the vascular strands
3	Magnesium ..	Quinalizarin	Blue-coloured spots showing the presence of magnesium	Numerous spots were clearly seen near the vascular connections of the haustoria and the host and also in and around the vascular strands of haustoria
4	Iron ..	Potassium ferrocyanide and potassium ferricyanide <i>Ortho</i> -phenanthroline	Bluish-green colouration indicating the presence of iron Reddish colouration showing the presence of iron	Vascular system was clearly coloured in respect of both the reagents
5	Copper ..	Sodium diethyl dithiocarbamate	Yellowish colouration showing the presence of copper	Seen in the vascular system of haustoria and in and around the cells of vascular strands
6	Zinc ..	Sodium nitroprusside and potassium sulphide	Intense reddish-purple colouration indicating the presence of zinc	Clearly seen on the vascular strands and also in the cells present in between the vascular system
7	Sulphydryl groups: (a) Free sulphydryl groups (b) Bound sulphydryl groups	Sodium nitroprusside, ammonium hydroxide, etc. Trichloroacetic acid, sodium nitroprusside, ammonium hydroxide, etc.	Nothing distinct from the controls was seen Reddish-purple-coloured spots were seen	Spots were clearly and uniformly distributed throughout the vascular system and more prominently in the cells between the vascular channels and near the vascular connections of the host and haustorial tissues



FIGS. 1-3



FIGS. 4-5

point of contact with the host plant, the vascular elements thus intimately maintaining contact with those of the host root. It is thus clear that the nutrients, which the sandal derives from the host plant, pass through the vascular system while occasional diffusion of some of these nutrients into the surrounding cells may take place, as the results of the histochemical studies (Table I) show.

It is evident from Table I that all the elements tested for (K, Ca, Mg, Fe, Zn and Cu) are found in the haustorium, specifically located in the vascular strands, thus suggesting that these elements are being taken up by the sandal from the host plant. However, the possibility of the direct absorption also of these nutrients cannot be ruled out. The degree of dependence of the plant for these elements on the host on the one hand and the soil on the other can be studied conveniently by tracer technique which is engaging our attention.

The presence of bound sulfhydryl groups and the absence of free sulfhydryl groups appear to show that cysteine and other free sulfhydryl group-containing compounds are not present in the vascular system of haustoria.

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EXPLANATION OF PLATES

PLATE V & VI

- Fig. 1. Sandal haustorium in association with *Acacia farnesiana*.
(X—Haustorium, Y—Sandal root, Z—Host root).
- Figs. 2-5. The vascular system of the sandal haustoria with different host plants.
- Fig. 2 With *Bauhinia purpurea* (×50).
- Fig. 3. With *Tecoma stans* (×50).
- Fig. 4. With *Leucæna glauca* (×30).
- Fig. 5. With *Broussonetia papyrifera* (×50).
- A—Vascular connections with mother root;
B—Vascular strands leading to host root;
C—Lacunar tissue;
D—host root.
- (Photographs taken by Shri V. N. Ranganathan).