STORAGE AND TRANSMISSION OF AMBROSIA FUNGUS IN THE ADULT *XYLEBORUS FORNICATUS* EICHH. (COLEOPTERA: SCOLYTIDAE).

E. F. W. Fernando

A study of the literature on the storage and transmission of the ambrosia fungi in the numerous species of *Xyleborus* has produced many divergent statements so that it seemed that there was room for investigating this problem in *Xyleborus fornicatus* Eichh., the shot-hole borer of tea in Ceylon.

Material and Methods

The adults of *Xyleborus fornicatus* Eichh. bore into the living stems of the tea bush. They bore deep into the branches with a single entrance tunnel, the galleries varying from ¾ inches to 2½ inches in length, and sometimes branched. The walls are stained a "pink" colour due to the associated ambrosia fungus. It does not cause an apparent decay of the tissues of the stem although the cells adjacent to the tunnels take a dark stain.

The specimens of the adult beetles were preserved in Bouin's fluid and Carnoy's fluid. They were doubly embedded, first in celloidin and then in paraffin. Sections were cut at a thickness of 8–15*mu*. The sections were stained in Delafield's Haemtoxylin counterstained with eosin, Ehrlich's Haematoxylin counterstained with eosin, and a modified Gram-Weigert stain as given by Leach (1940). Of these Ehrlich's Haematoxylin gave the best result for detailed histology, while the Gram-Weigert stain clearly demonstrated the fungus tissue. Preliminary serial transverse and sagittal sections were made of the entire adult males and females in order to examine their morphological structures. As no specialized external or internal structures were observed in the thoracic and abdominal regions, the final sections and exminations were confined only to the head region. A careful study of the head region revealed certain specialized structures closely associated with the buccal cavity in the adult female. Such structures were not found in the adult males.

Description

The alimentary canal is divisible into the three usual regions, the fore gut, mid gut and hind gut. The fore gut consists of the buccal cavity, pharynx, oesophagus and crop. Of these, only the buccopharyngeal region and a part of the oesophagus are confined to the head and I am only concerned with this region in the present paper.

Reprinted by permission of the publishers from the Annals and Magazine of Natural History
Ser. 13, vol. ii, p. 475, *August*, 1959,
Published 11/8/1960.

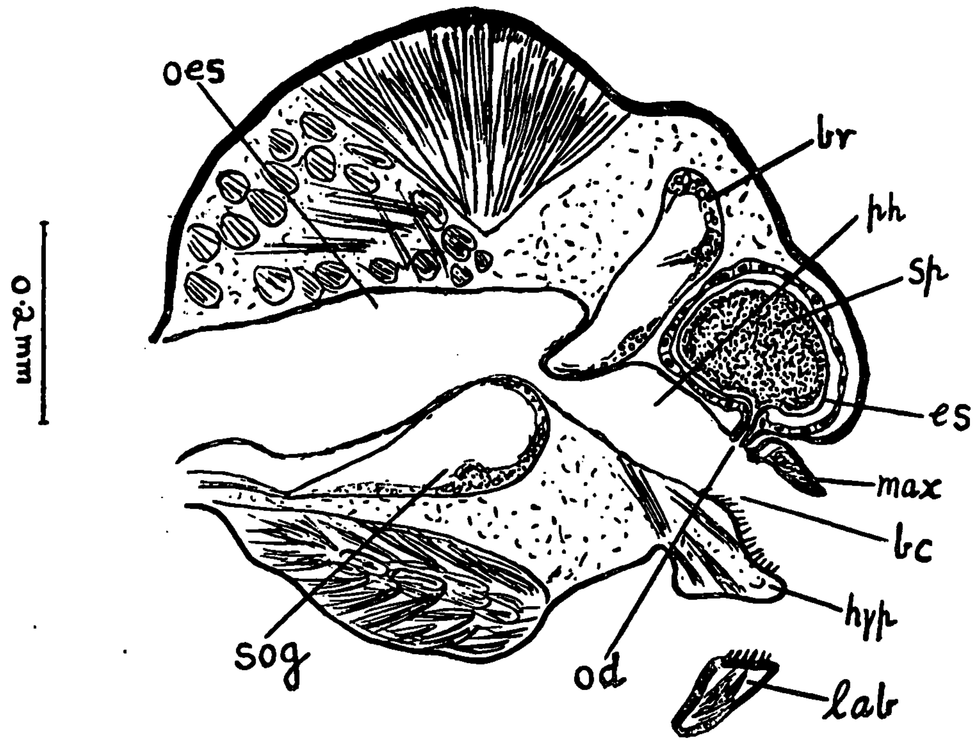

Figure 1. Sagittal section of the head of the female *Xyleborus fornicatus* Eichh. showing fungal storage sac opening into the buccopharyngeal region. br. brain; bc. buccal cavity; es. cuticular lining of sac; hyp. hypopharynx; lab. labium; max. maxilla; od. opening of duct; oes. oesophagus; ph. pharynx; sog. suboesophageal ganglion; sp. spores of ambrosia fungus inside sac.

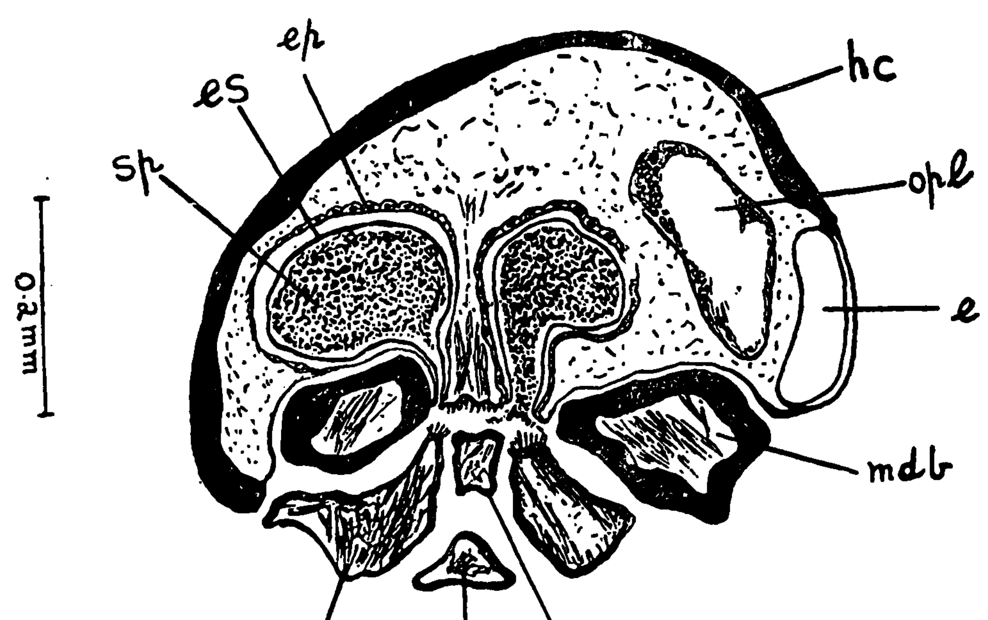

Figure 2. Transverse section of the anterior region of the head of the female *X. fornicatus* showing fungal storage sacs and their ducts opening into the buccopharyngeal region. e. eye; ep. epithelial lining of sac; es. cuticular lining of sac; hc. head. capsule; hyp. hypopharynx; lab. labium; max. maxilla; mdb. mandible; opl. optic lobe; sp. spores of ambrosia fungus inside sac.

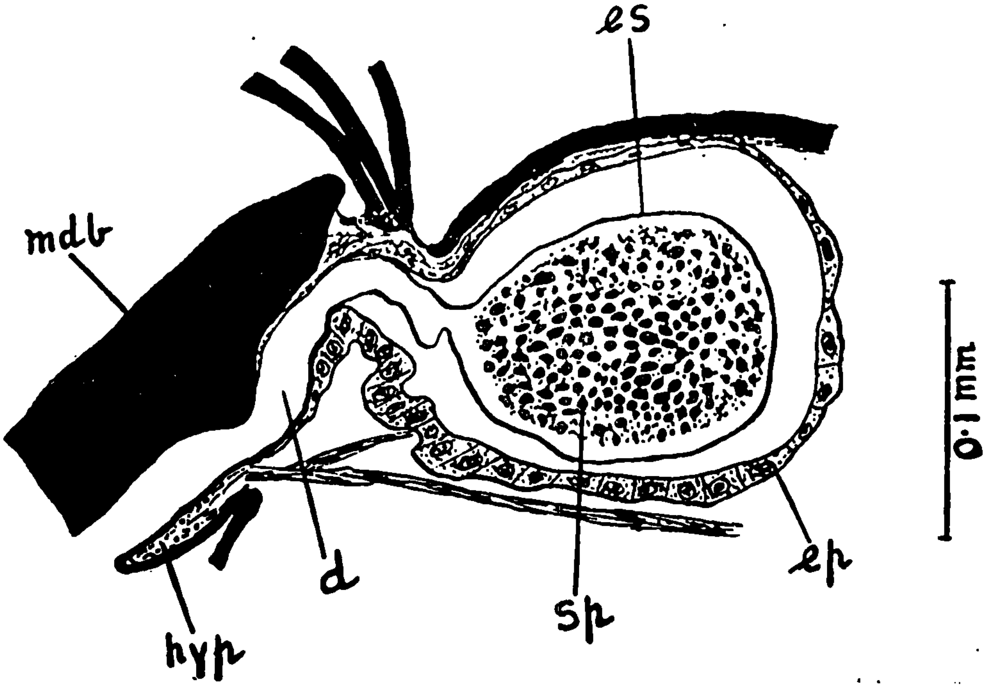

Figure 3. Median sagittal section of the fungal storage sac and duct. d. duct of sac; ep, epithelial lining of sac; es. cuticular lining of sac; hyp. hypopharynx, mdb. mandible; sp. spores of ambrosia fungus inside sac.

The buccal cavity is not structurally differentiated from the pharynx, which is a somewhat elongated sac situated slightly vertically in the head capsule behind its anterior wall, and lying anterior to the nerve connectives between the brain and the sub-oesophageal ganglion. The posterior region of the pharynx is constricted in the region of the oesophageal connectives just before it joins the oesophagus which has a dorsal sac-like projection immediately behind the brain. The oesophagus continues backwards into the crop.

Situated antero-dorsally in the posterior region of the buccal cavity and immediately in front of the brain are a pair of well defined sacs, each sac opening to the exterior by a short duct at the base of the hypopharynx and between the mandibles and the maxillae (figs. 1 and 2). Both transverse and sagittal sections show that the sacs are looped laterally and deflected ventrally. The epithelium of the sac (ep) consists of a well defined single layer of cubical cells with large, slightly oval, deeply staining nuclei. Cell boundaries are evident. The epithelium of the anteroventral region of the sac and of the duct consists of columnar cells with large and elongate nuclei. A distinct duct leads from each sac. The junction of the duct with its sac is slightly constricted (fig. 3, m). These sacs are evidently epidermal pouches and are lined internally by a thin, flexible cuticle (es), which is a continuation of the outer cuticle of the buccal cavity. A certain amount of unequal shrinkage has occurred as a result of fixation, but in many parts of the sacs the endocuticle lines the epithelium. Within the cuticle of the sacs are a mass of spores of the ambrosia fungus and these were clearly revealed by the Gram-Weigert stain (sp). As shown in fig. 2, the fungal spores in one sac are passing into the buccal cavity, but it would appear that the sphincter mentioned above at the junction of the duct and the sac helps to regulate the passage of the spores into the buccal cavity.

Examination of serial sections of the head of the male did not reveal any sacs storing fungal spores in the buccal region.

Discussion

Schmidberger (1836) was the first to observe the association of scolytid beetles with so-called "ambrosia". But the actual fungal nature of the ambrosia was first recognized by Thomas Hartig (1844) when he described the fungus associated with *Xyleborus dispar* Fabr. in *Alnus cordata* and named it *Monilia candida.* Speyer (1923–24) in his studies on the habits of the Ceylonese ambrosia beetles has observed the association of an ambrosia fungus with *Xyleborus fornicatus* Eichh. attacking tea and a number of other plants in Ceylon. The detailed studies of this fungus were described by Gadd and Loos (1947) and it was named *Monacrosporium ambrosium.*

With regard to the transmission and storage of the fungal spores, according to Schneider-Orelli (1911, 1913), the ambrosia fungus of *Xyleborus dispar* Fabr. is transmitted to successive generations in the form of spores stored in the crop of the female beetle which regurgitates them into new galleries. Neger (1908–11), however, thinks that the spores are passed through the body of the beetle and survive in a viable condition in her faecal pellets. According to Strohmeyer (1911) the females of *Xyleborus* from the tropics store the spores within chitinous bristles in front of the head and these are directly transferred into new brood chambers. The most recent worker on the subject, Francke-Grosmann (1956), who worked on several species of ambrosia beetles, found that in *Xyleborus pfeili* Ratzeb., the ambrosia spores are carried in pockets contained in the posterior abdominal tergites. A similar view had already been held by Doane and Gilliard (1929) who worked on three species of ambrosia beetles in California, where the conidia are present in the dorsal pores and on ventral thoracic hairs.

The present investigation shows that in *Xyleborus fornicatus* Eichh. from Ceylon the spores of the ambrosia fungus *Monacrosporium ambrosium* Gadd & Loos are stored within specialized internal sacs situated in the anterior region of the head of the adult female beetle. These spores are transported from the sacs through their ducts into the buccal cavity (fig. 2) and are then probably transmitted into the galleries with the aid of the mandibles and the maxillae to start a new culture for the successive generations. As the larvae, which develop within the galleries, have fairly well-developed mouth-parts, they probably consume a certain amount of cellulose material from the stem tissues, and these probably serve as a supplement to the fungus and provide the growing larvae with a more substantial diet.

In a way not clearly understood, the beetles are able to suppress the development of all extraneous fungi so that the galleries contain only the ambrosia fungus growing in pure culture. When the galleries are deserted by the beetles, their walls assume a grey or black coloration often emitting an unpleasant odour most probably due to the decomposition of the ambrosia fungus and its associated host tissues, together with the growth of secondary saprophytic fungi.

Acknowledgments

I wish to express my most grateful thanks to Mr. G. D. Austin, Entomologist, Tea Research Institute of Ceylon, who gave me all the facilities and encouragement for carrying out this investigation in the Tea Research Institute, Talawakelle. I would also like to express my gratitude to Professor Wilfred Fernando of the University of Ceylon, Colombo, both for his advice and for his kindness in allowing me to complete the investigation in Colombo.

Summary

The present investigation shows that in *Xyleborus fornicatus* Eichh. the spores of the ambrosia fungus *Monacrosporium ambrosium* are contained within two sacs situated anterior to the brain in the head of the female beetle.

These two sacs open by two ducts in the dorsal buccopharyngeal region at the bases of the mandibles and the maxillae.

No fungal spores were detected in any other part of the adult female beetle.

Such sacs or any other structure containing fungal material are not found in the adult males.

This method of storage of the ambrosia fungus by *X. fornicatus* Eichh. differs from the methods of storage described by earlier workers for various species of ambrosia beetles.

References

DOANE, R. W. and GILLIAND, O. J. 1929. Three Californian ambrosia beetles. *J. econ. Entomol.* **22,** 915–921.

FRANCKE-GROSMANN, H. 1956. Hautdrüsen als Trager der Pilzsymbiose bei Ambrosiakafern. *Z. Morph. u. Okol. Tiere.* **45** (3), 275–307.

GADD, C. H. 1947. Observations on the life-cycle of *Xyleborus fornicatus* Eichh. in artificial cultures. *Ann. appl. Biol.* **34,** 197–206.

GADD, C. H. & LOOS, C. A. 1947. The ambrosia fungus of *Xyleborus fornicatus* Eichh. *Trans. Brit. mycol. Soc.* **30.** 13–18.

HARTIG. 1844. Ambrosia des *Bostrychus dispar. Allg. Forst-u Jagdztg. N. F.* **13,** 73–75.

LEACH, J. G. 1940. Insect Transmission of Plant Diseases. New York. 59–67.

NEGER, F. W. 1908. Die Pilzkulturen der Nutzholzborkenkafer. *Zbl. Bakter.* **20,** 279–282.

1909. Ambrosiapilze II. Die Ambrosia der Holzbohrkäfer. *Ber. dtsch. bot. Ges.* **27,** 372–389.

1911. Zur Ubertragung des Ambrosiapilzes *von Xyleborus dispar. Naturwiss. Z. Land-u. Forstw.* **9,** 223–225.

SCHMIDBERGER, J. 1836. Naturgeschichte des Apfelborkenkäfers *Apate dispar.* Beitr. zur Obstaumz. *Naturgesch. Obstbaumen. Inskten (Linz)* **4,** 213–230.

SCHNEIDER-ORELLI, O. 1911. Die Übertragung und Keimung des Ambrosiapizes von *Xyleborus (Anisandrus) dispair* F. Naturwiss. *Z. Land-u. Forstw.* **9,** 223–225.

1913. Üntersuchungen über den pilzzuchtenden Obstbaumborkerkafer *Xyleborus (Anisandrus) dispar* und seinen Nährpilz. *Zbl. Bakter.* **2,** 25–110.

SNODGRASS, R. E. 1935. Principles of Insect Morphology. New York.

SPEYER, E. R. 1923–24. The Habits of the Ceylonese Ambrosia beetles. *Bull. ent. Res.* **14,** 11–23.

STROHMEYER, H. 1911. Die biologische Bedeutung sekundaren Geschlechtscharaktere am Kopfe weibliche Platypodiden. *Entomologische Blatter.* **7,** 103–108.

TROTTER, A. 1933. Il fungo Ambrosia delle galerie di un Xyleborino di Ceylon. *Ann. R. Inst. Sup. Agric. Portici. Ser. III* **5,** 267–275. Abstract in *Rev. app. Mycol.* **14,** 167.