IDENTIFICATION OF FEEDING STIMULANTS
FOR BOLL WEEVILS FROM COTTON BUDS
AND ANTHERS

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Abstract—Column chromatography of the pentane extract of freeze-dried cotton buds or anthers yielded a wax-sterol ester fraction that exhibited potent feeding stimulant activity for the cotton boll weevil. The waxes of the wax-sterol ester mixture were responsible for the feeding activity. Saponification of the wax-sterol ester fraction yielded about 15% alcohols and 85% sterols. A C_{18} alcohol, dihydrophytol, phytol, and geranylgeraniol constituted 15, 36, 26, and 23%, respectively, of the total alcohols, implicating certain of their long-chain esters as feeding stimulants. Several esters of dihydrophytol, phytol, and geranylgeraniol were identified among the waxes by GC-MS. Certain phytol, geranylgeraniol, and oley alcohol esters containing C_{12} to C_{26} acid moieties were synthesized and were found to induce high feeding stimulant activity in the cotton boll weevil.

Key Words—Boll weevil, Anthonomus grandis, Coleoptera, Curculionidae, feeding stimulants, cotton buds, anthers, phytol, geranylgeraniol esters, phytol oleate, phytol dodecanoate.

INTRODUCTION

Since Keller et al. (1962) reported the presence of feeding stimulant(s) for boll weevils, Anthonomus grandis Boheman (Coleoptera: Curculionidae), in water extracts of cotton squares, considerable work has been done toward the identi-
fication of compounds that stimulate feeding activity. Hedin et al. (1974) fur-
nished an excellent review of the work on insect feeding stimulants in general, 
including compounds that stimulated feeding activity of the boll weevil. Several 
workers reported on a number of compounds from the cotton plant that elicited 
some degree of feeding in the boll weevil (Hedin et al., 1966; Stuck et al., 
1968a, b; Temple et al., 1968). None of the studies, however, have identified 
compounds of sufficient activity to be considered as the feeding stimulant for 
the boll weevil. This paper reports on the identification and synthesis of com-
ponds from cotton buds and anthers that are highly active feeding stimulants 
for the boll weevil.

METHODS AND MATERIALS

Extraction of Cotton Buds and Anthers. Whole cotton buds with bracts 
were chopped in a blender and then freeze-dried. Anthers were dissected out of 
buds and then freeze-dried whole. The freeze-dried materials were extracted in 
a Soxhlet apparatus for 3 hr successively with pentane, ethyl acetate, chloro-
form, and methanol. The solvents were removed under reduced pressure at 50°C, 
and the resulting residues were bioassayed.

Feeding-Stimulant Bioassay. Laboratory reared boll weevils, 1 to 4 weeks 
old, were used in the bioassays. A specific quantity of residue, equivalent to one 
bud or androecium (aggregate of anthers), of an extract or chromatographic 
fraction was dissolved in 1 ml of hexane or methanol. For bioassay of the syn-
thetic esters, a 0.04 M solution in hexane was prepared. Approximately 20 μl 
of each test solution was placed within two of four 1-cm-diameter circles drawn 
around the periphery of 7-cm-diameter qualitative grade filter paper. Similarly, 
 solvent blank was applied to the two remaining 1-cm circles. After the solvent 
evaporated, the filter paper was placed inside an 11-cm-diameter Petri dish on 
a 0.5-cm layer of 2.5% agar. Ten insects were placed into each dish, and the 
dishes were placed in a dark environment overnight at 29°C. Feeding response 
was determined by totaling the number of feeding punctures within the treated 
circles then subtracting the total number of punctures in the blank. Twenty rep-
licas were obtained.

Chromatography. The residue from the pentane extractions was chromato-
graphed over 25 g of hexane-washed Merck silica gel (2.5 × 13 cm), and the 
column was eluted with 100-ml volumes of 0, 3, 10, 25, and 50% ether in hex-
ane. Of the 3% ether in hexane eluent, fractions of 75-ml and 25-ml volume 
were collected. TLC analyses were conducted on silica gel plates. The solvent 
systems were toluene–hexane (1:1) and toluene–ethyl acetate (9:1).

Feeding Stimulant Analyses by GLC and GC-MS. Natural and synthesized 
feeding stimulants were analyzed by GLC and GC-MS. GLC was performed 
with a Varian model 3700 gas chromatograph equipped with a 15-m × 0.32-