IOSR Journal of Agriculture and Veterinary Science (IOSR-JAVS)
e-ISSN: 2319-2380, p-ISSN: 2319-2372. Volume 7, Issue 5 Ver. I (May. 2014), PP 50-53
www.iosrjournals.org

Morphogenetic Effects and IGR activity of a botanical Forskolin against last Instar Larvae of Callosobruchus chinensis (Linn.)

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***Abstract*:** *Synthetic insecticides are expensive for subsistence of farmers and they may pose potential risks owing to lack of adequate technical knowledge related to their safer use. There have been lots of search on locally available plant materials that may have shown grain protectant ability.The botanical Forskolin was derived as active alkaloid from the roots of the plantPlectranthus barbatus. Forskolin inhibited the growth and development of the stored grain pest Callosobruchus chinensis.*
Key words: *Forskolin, Callosobruchus chinensis, botanical,IGR activity*

I. Introduction

The stored product pests attacking commodities are many and varied and food commodities with high nutritive values are more susceptible to pest damage. (Odeyemi& Daramola 2000). Botanical insecticides have long been used as alternatives of synthetic chemical insecticides for pest management because chemical insecticides repeatedly posed threat to the environment and human health. Most botanical pesticides of plant origin are non-toxic (Rao *et al.,* 1993) and easily bio-degradable. These plant extracts do not cause problems like bio-accumulation and bio-magnification. Toxicity of certain plant products against insect pestswere reported by Diwedi and Pareek , 2006, Akinkurolere *et al.,* 2006. Various plant extracts were reported to protect bagged grain (Diwedi and Mathur, 2000.)Hence we report here-in the effect of a compound Forskolin isolated from theroots of the plant *Plectranthus barbatus*aginst the stored grain pest*Callosobruchus chinensis.*

On C*allasobruchus chinensis* Forskolin influences general morphological characters, growth and development of insects by associating with the various physiological process.These plant extracts reduce oviposition rate and suppress the adult emergence of bruchids, and also reduced seed damage rate. The morphologically abnormal forms like larval- pupal intermediates and pupal-adult intermediates, which showed immature ovaries and testes in treated insects, thus reducing the oviposition rate, observed in Callasobruchus *chinensis* when compared with the untreated control bruchids.

II. Material And Methods:

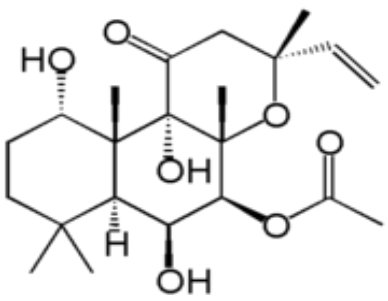
Callosobruchus chinensis

Callosobruchus chinensis larvae are reared in a culture room at 26±1°C , 14 h : 10h light:dark period and 70% RH. Different concentrations of Forskolin 0.1, 0.25, 0.5, 0.75, 1, 2 and 4 µg/ml were prepared in acetone. 30 freshly moulted last instar larvae were seggregated and 1 ml of the concentrations were applied topically on the abdominal region with the Hamilton micro syringe. The experiments were replicated 5 times. Parallel controls treated with 1ml of acetone were maintained. The treated larvae were transferred to the diet and observed daily to note changes.

Forskolin:

Forskolin is a labdane diterpene that is produced by roots of the plant *Plectranthus barbatus*. Chemical Formula of Forskolin : C15 H26O7

Chemical Structure Of Forskolin



III. Results

Severe Morphogenetic abnormolities were observed in Forskolin treated resultant insects at various concentrations (Table – 1). At lower concentrations(0.1 and 0.25%) , the larvae developed into apparantely normal adults. But these forms died within a few hours after moulting. Larvae treated with 0.5 to 2 % concentrations pupate normally but exhibited serious disturbances during adult eclosion, adults developed within the cuticle and were unable to shed exuviae. These abnormal non-viable forms were much smaller and undifferentiated when compared to the controls. At 4% concentration the larvae formed into mosaics. A mixture of larval-pupal cuticle, Adistinct shrinking of the body and slippage of the head are the most important characteristics observed inthese forms.these larval-pupal intermediates and pupal-adult intermediates did not undergo subsequent developmental changes and ultimately died.

Different concentrations of the botanical Forskolin applied on 5 th instar larvae .The phytochemical exhibits the ecdysis inhibition .The inhibition rate is 23.5% at the concentration of 0.10 µg/ml and increasing as per concentration increased . The inhibition rate is 90.0% at the concentration of 4.00 µg/ml (Table 1) The treated resultants developed abnormalities in larvae, pupae and the adults. The adult survival rate is decreased as per the increase of concentration .In treated resultant adult insectss the survival rate is 20.5 % at 0.10 µg/ml concentration and it is 2.7% at 4.00 µg/ml concentration (Table 2).

The following morphological deformities were observed :

- 5th instar larvae were unable to pupate(Plate 1. b,c.).
- Larval-pupal intermediates(Plate 2.b) and Pupal-adult intermediates(Plate 2.c) were formed and failed to develop into successful Adults.
- The abnormal adults developed with deformed wings and appendages(Plate 3.b,c)

The following deformities were observed in ovarian development :

- The adult treated resultants developed the ovarian deformities which influence the reduced fecundity.
- Ovarioles with abnormal oocytes.(Plate 4.b)
- Large chorionated oocytes blocking the oviduct(Plate 4.c)
- Chorionated oocytes large ,irregular unable to ovulateand bloking the passage of other oocytes (Plate 4.d) .

IV. Discussion:

The effect of administering Forskolin topically on the IV and V instar larvae and pupae of *Callasobruchus chinensis* influenced the larval development and molting and pupal development and metamorphosis. There was prolongation of larval period and formation of the larval-pupal intermediates due to treatment of larvae and some of the treated resultants pupated but failed to metamorphose into adult forms. These effects are similar to that of the interference of juvenoid with the production, release and action of molting hormone.(Raja *etal*.,1987).

Our results suggest that the application of Forskolin prevented the normal development of treated larvae and pupae of Callasobruchus chinensis and their resultants. The application of Forskolin resulted in formation of abnormal adults. Most of these forms were unable to mate that decreased the fecundity of the *C. chinensis.* Similar observations were noticed with other insect growth regulators. (Gundersone*t al.,* 1985 ; Koul *et al*., 1987 ; Vardhini *et al*, 2001, Adedire *et al.,* 2004) .

Thus the present study clearly indicates that Forskolin acts as insect growth regulator and it influences the morphogenetic developments and cause to form the morphological deformities like larval-pupal intermediates, pupal-adult intermediates and mal-formed adults. Ovaries also exhibited the deformities like large tropical oocytes, chorionated oocytes , malformed oocytes and ovarioles. The protein levels also were variable with that of control insects. The decreased protein level indicates the influence of Forskolin on the synthesis and uptake of protein during larval pupal transformation and vitellogenesis.

This plant extract inhibited the growth and development of the stored grain pest *C. chinensis* and suggesting its use as a economic, safe and eco-friendly botanical pesticide.

Table 1: Ecdysis inhibition rate in treated insects:

S.No.	Concentration µg/ml	Dosage ml/insect	No of insects	LPI (%)	PAI (%)	Ecdysis Inhibition(%)
1	0.10	1	30	11.5	12.0	23.5
2	0.25	1	30	16.0	14.5	30.5
3	0.50	1	30	20.8	20.0	40.8
4	0.75	1	30	27.3	26.3	53.6
5	1.00	1	30	35.5	33.2	68.7
6	2.00	1	30	42.5	36.0	78.0
7	4.00	1	30	50.5	39.5	90.0
8	Control acetate	1	30	0	0	0

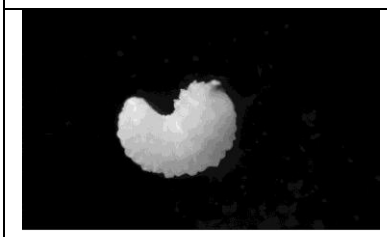
LPI= Larval-pupal intermediates ; PAI= Pupal-adult intermediates.

Table 2: Adult survival rate in treated insects:

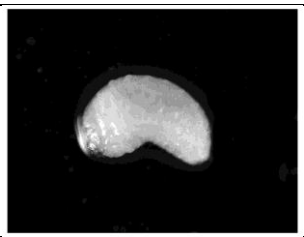
S.No.	Concentration µg/ml	Dosage ml/insect	No of insects	Adult failed to survive (%)	Adult survival (%)
1	0.10	1	30	79.5	20.5
2	0.25	1	30	83.3	17.7
3	0.50	1	30	85.5	14.5
4	0.75	1	30	90.0	10.0
5	1.00	1	30	93.5	6.5
6	2.00	1	30	95.0	5.0
7	4.00	1	30	98.3	2.7
8	Control acetate	1	30	0	100

PLATE - 1

a.Control - larva



b. Treated larva unable to moult



c.V instar larva with malformed Mouth parts and unable to pupate

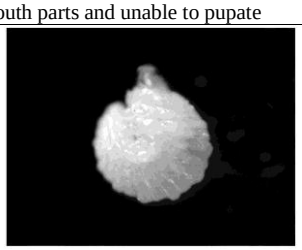
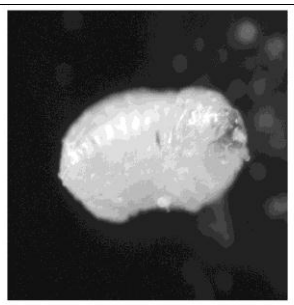
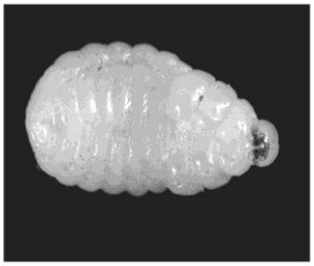


PLATE - 2

a. Control- pupa



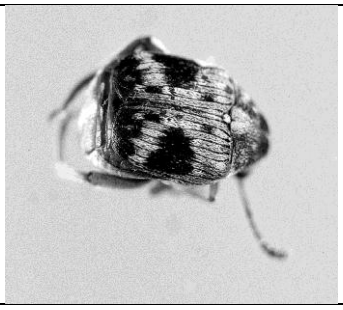
b. Treated-Larval-pupal intermediate



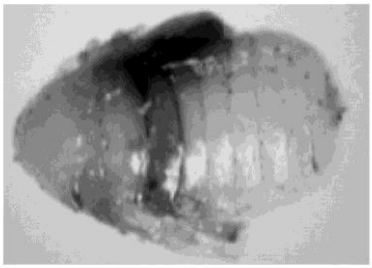
c. Treated-Pupal-adult intermediate

PLATE-3

a. Control-Adult female



b. Treated-Malformed adult



c. Treated- Malformed adult

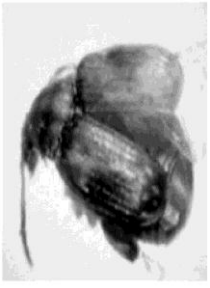
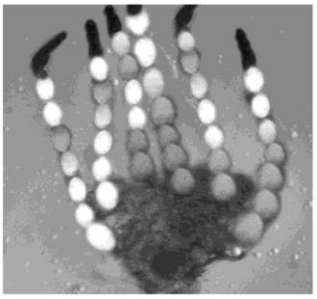
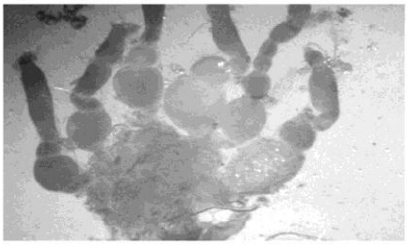
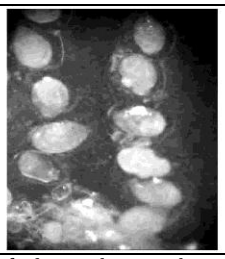


PLATE - 4	
	
a. Control -Ovary with the view of vitellarium ,germarium and ovaries.	**b.** Treated-.Ovarioles with abnormal Oocytes
	
c. Treated-Terminal oocytes abnormally large and unovulated in pupal-adult intermediates	**d.** Treated-Magnified view showing chorionated fully developed unovulated oocytes

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