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Study of Jha on Lepidopteron Pest

Dr.R.D.Ahirwar

Govt.College Kundum Dep.-Zoology jabalpur MP,India

A B S T R A C T

A new approach to insect pest control is the use substances that adversely affect insect growth and development. Growth and development of insects are under the control of hormones, including pro- thoraco-tropic-hormones (PTTH) (brain hormone), ecdysteroids and juvenile hormones (JH). The peptide hormone PTTH secreted from the brain controls the secretion of the moulting hormone (ecdysone) from the prothoracic gland. Ecdysone is responsible for cellular programming and together with JH, initiating for he moulting process. When JH levels secreted from the corpora allata are high, the epidermis is programmed for a larval, moulting otherwise the epidermis is programmed for metamorphosis. JH is virtually absent in the pupae, but is present in adults to serve some functions in reproduction. Thus, JH suppresses pupation and includes vitellogenesis during the reproductive stage of the insect. There are several known insect JHS synthesized and secreted from the corpora allata viz. JH I- III, JH and Iso-JH.

Keywords: Antifeedant, Casperadaceae, Cleomacea, Cleome viscosa, Diacrisia oblique, Earias fabia, Feeding deterrent, Gycine max, Hibiscus esculentus, Juvabione, Juvenile hormone analogous

1. INTRODUCTION

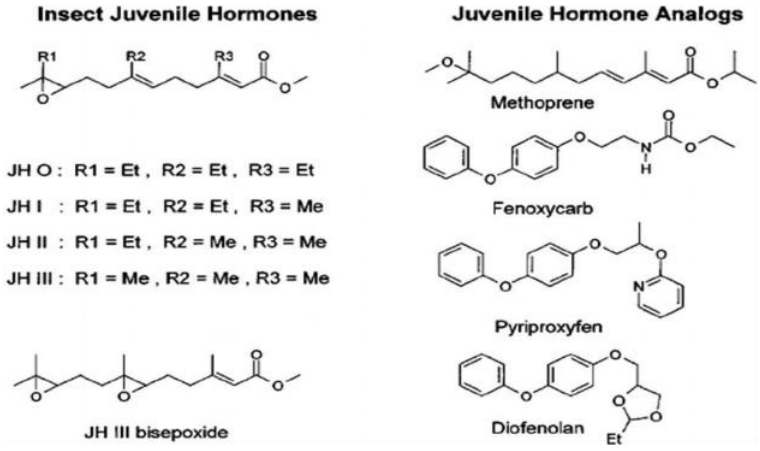
A new approach to insect pest control in the use of substances that adversely affect insect growth and development. These substances are classified as "insect hormone mimics" or "insect growth regulators" (IGRS) Due to their effect on certain logical regulatory process essential to the normal development of insects or their Pro Jhansi coma the are quite selection in their mode of actions in portal really only one target species, IGRS generally control insect either through regulations of metamorphosis or interference with reproduction (Riddiford and Truman), 1978 compounds developed to disturb metamorphosis ensure that no reproductive adult are formed. Those that specifically interfere which reproduction may include the development of adults with certain morphogenetic abnormalities that reduce their reproductive potential adult may be sterile or process abnormally developed genitalia, which hinder the matings process or the capacity to produce fertile offspring. Pesticide regulation emphasized the discoveries or synthesis of compounds (IGRs) that are specific to the target species and do not adversely (or at least minimally) affect beneficial and non-target species. As result, direct approach for discovering selective insecticides are being used, specifically for synthesis of active of biologically active compound guided by the result of quantitative structure-activity relationship (QSAR) analysis for discovery of insecticides from natural products. as well as synthesis of their highly active analogs and for applications of a bio-rational approach to design and synthesize insecticides((Morrrod 1981. Magee et al.,1985).

There are several known insect JHs (i.e. JH I-III, JHand Iso-JH) synthesized and secreted from the corpora allata (Miyamoto et al., 1993). Williams (1967) suggested the compound that mimic the actions of juvenile hormones (JHs) could be used as safe insecticide. Numerous JHanalogs (JHAs) were discovered and certain like methoprene have been used as commercial insecticides (Retnakaran et al., 1985; Staal 1975). Methoprene is approved by the WHO for use in drinking water cisterns to control mosquito larvae. The agriculture use of these JHAs has been limited, because of this left of outdoor stability, the limited insect control spectrum and their so toxin action.

Since the early 1970s, numerous analogs of JH have been tested for insecticidalactivity (Retnakaran et al., 1985; Staal 1975). Most of the early analogs resemble JH in their basis terpenoid structure. The most active ones, Such as methoprene and hydroprene (Staal 1982), however lack the epoxidefunction present in JH(Retnakaran et al., 1985). More recently several highly active compounds that have less apparent similarity to Juvenile

* *Corresponding author.*
E-mail address: rdindian75@gmail.com

hormones have been synthesized: fenoxycarb registered as Insegar, logic, torus, pictyl and varikill (Dorn et at; 1981) pyriproxyfen registered as knack, sumilarv and admiral (Hatakoshi,1986) and diofenolan, registerd as aware; ciba geigy (Sechser,1994). Chemical structures of the Juvenile hormones, terpenoidal (methoprene) and nonterpenodial (fenoxycarb, pyriproxyfen, and diofenloan) Juvenile hormone analogous are given below.



2. REVIEW

India is rich in wide flora that not only consists of plants of medicinal importance but also have several poisonous substances presents in the variety of plants. In the present study, Cleome viscosa of family Casperadaceae have selected to isolate a compound that is analogous to the Juvenile hormones which many interfere with growth and metamorphosis of lepidopteron pest Spodoptera litura .

Some of the workers in the field of crop pest management include Prakash and Tiwari (1989) , Dixit and Saxena, (1991) Saxena et al. (1992, 1994, 1995). Dixit and Saraj (1988) have described insect repellent and juvenile hormone mimicking activity of essential oil of Tridex procumbens Linn. Similarly, Jagganath and Nair (1993) have tested the effect of JHA and Azadirachtin on Spodoptera Litura. Koul (1982) have reported much earlier that natural product have drawn attention of the researcher in the plant protection. Similarly, Dubey (1997) have also reported the management of certain agricultural pest where he has isolated quercetin to compound control the pest of agriculture importance.

Disruption of growth, moulting and metamorphosis as well as the manipulation of insect behavior are being considered as a new innovative approach in the pest management. Nakanishi (1981) have isolated such compounds from Aalmia latifolia against Gypsy moths. Kubo et al. (1981) isolated an active compound from Ajuga remota which is active against various insect pests. Nawrot and Harmatha (1994), Baby (1994), Banerjee (1995), Bowers (1983), Supavarn et al (1974) and Hifnowy et al. (1990) are some prominent workers in the field of pest management through natural products and their active ingredients.

3. MATERIAL AND METHODS

3.1Plantmaterials, Taxonomic positionandits general description:

In the present study, plant Cleome viscosa L. (Fig.1) was selected as plant material which was authenticated. In the present study, fresh leaves of this plant was collected from the surrounding areas of Vidisha and fresh material weight was measured 360 gm, then it was shade dried in the Laboratory of the department of Zoology and after shade drying loss in weight was measured 45 gm and present loss in weight (87.5%) of material was recorded. The material was used for extraction and isolation of plant extract (Table 1). The systemic position of this plant is given below:

Kingdom – Plantae, Division – Angiosperms, Order – Brassicales, Family - Cleomaceae/Casperadaceae Genius – Cleome, Species – viscosa L.

Plant Cleome viscosa of family Cleomaceae (Brassicaceae) widely grown in Bundelkhand region. It is a bushy annual herb commonly known as “Wild Mustard” and is locally famous as “Hurhur”.

3.2 Extraction, Isolation and Purification of plantmaterials

Before Soxhelation the shade dried plant material was powdered of 40-60 mesh size then defatted with n-Hexane. Soxhlet extraction method of Harborne (1984) was followed for the extraction and isolation. Powdered material of the whole plant loaded in the Soxhlet appartaus and extracted in the petroleum ether, benzene and chloroform solvents. 6-8 cycle where run with different solvent to get a good percentage yield (Table 2) and crude extract was filtered in the in breaker. The obtained semi solid crude extract where evaporated in the water bath to get semi solidextract.

Observations and Result

In the present study, fresh leaves of the plant Cleome viscosa L. of family Cleomaceae or Casperadaceae was selected select which is commonly now as "Wild mustard" . About 360 gram of collected in the most of July to September 2011 where kept in the library laboratory for shade drying. After drying the plant material got reduced and loss in weight loss weight but noticed maximum 87.5% bars in . Dried plant material was used for the isolation of crude extract by listen using socks apparatus and tried different solvent viz material methanol from and water according to increasing order of order of polarity. After soxhletion extract filtered and evaporated in water bath for getting semisolid crude and percentage yield of the extract was observed which was maximum in methanol (6.60%), followed by 5.44 % in water, 5.22% in chloroform.

Preliminary phytochemical screening of the plant extracts were also done using various test viz. Meyers, Wagners, Dragendorffs and Hager's test for alkaloids, Molisch Benedict and Fehling tests for carbohydrates, Modified Borntrager and legal test for glycoside, froth formation and foam test for saponins, salkowski and Lieberman- Burchard's test for terpenoids and sterols, Gelatin test for tannins, Alkaline Reagent and lead acetate test for flavonoid and Xanthoproteic and Ninhydrin test for the detection of proteins and amino acids. On the basics of these tests, it was observed that methanol and water extract of this plant was found to be more positive for it but found to be positive for carbohydrates and all the three extract was found to be positive for glycoside saponins, terpenoid, sterols, flavonoid, protein and amino acid which was followed by benzene and petroleum ether also. Chloroform and water extract found to be negative for tannins.

In the present study, solvent viz. toluene; ethyl acetate; glacial acetic acid (5:5:0:3)was subjected to for developing appropriate solvent system thin layer chromatography which was used for isolation of constituted from methanol extract of Cleome viscosa. Four fraction (CV- 1CV-2 CV-3 and CV-4) obtained during column chromatography. Out of them fraction CV-3 was used for juvenile hormone activity (JHA) to see its effect on growth, moulting and metamorphosis lepidopteran pest viz. Diacrisia oblique and Earias fabia. Biologically active fraction CV-3 (Fig.5) was send to SAIF, CDRI Lucknow for getting spectral analytical graph viz. IR,UV, 1HMNMR, 13CNMR and Mass .

Thin layer chromatography of cleome viscosa mathanolic extract was also done by using same solvent system Toluene. ethyl acetate: glacial acetic acid (5:5:0.3) which gave maximum revolution and four sport which the RF value 0.40 0.48 0.75 and 0.82 where obtained. respectively TLC of one gratis sample was also done along with some solvent system it was observed that RF value(0.48)of sports- 3was scene Quite similar to the RF value(0.72) of gratis sample with the same solvent system(Fig.6). Excellent resolution was found of the spot 3 with RF value (0.92). During TLC color characterization of the different fractions were are also observed in visual,UV light the Iodine chamber .

During the observation mentioned in Table (7) and Graph (3), it was noticed that the number ot freshly eggs laid by the female of Diacrisia obliqua and Earias fabia was found to be decreased from the control (T275t2.5 and 740+2.0) to the treated (700+2.I and 467t2.10) as well as standard group (590+2.8 and 368+2.75), respectively, when applied 10-30 µl Extract of Cleome viscosa and 5ul concentration of juvabione. However, all numbers of freshly laid eggs were compared with the average number of eggs laid down by the female of D. oblique (295 eggs) and

E. fabia, (/50 eggs), respectively and eggs hatching percentage were also calculated which were noticed in decreasing order from control (98.45%% and 98.66%), treated (54.05% and 62.26%) and standard group (45.55% and 49.06%) respectively in D. obliqua and E.Fabia (Table 7 and Graph 4).

Results mentioned in the effect of Cleome viscosa extract upon the developmental stages of Diacrisia obliqua and Earias fabia, when 3 different concentrations in, 20 and 30 ul were applied on the pest. The developmental stages were od to be inversely proportional to the concentration caused delayed metamorphosis with several deformities. When, highest 30 µl concentrations of Cleome viscosa extract were applied on developmental 53 stages of D. obliqua and E. fabia, it was found that 60 & 65 % permanent larvae, 30& 30% Larva-pupa intermediate and 10 &5% Headless pupae found in D. obliqua and E. fabia, respectively.

Results in depicted the effect of Cleome viscosa extract on feeding deterrent activity of D. obliqua and E. fabia, and found that as the dose inereases of the extract, the feeding deterrent activity percentage of larvae to go away from the food increases at 30 µl concentration whereas with standard compound Juvabione 5 ul, the feeding deterrent distance percentage was noticed highest (72.20+1.95and 73.20+ 1.25%). respectively in both pest .

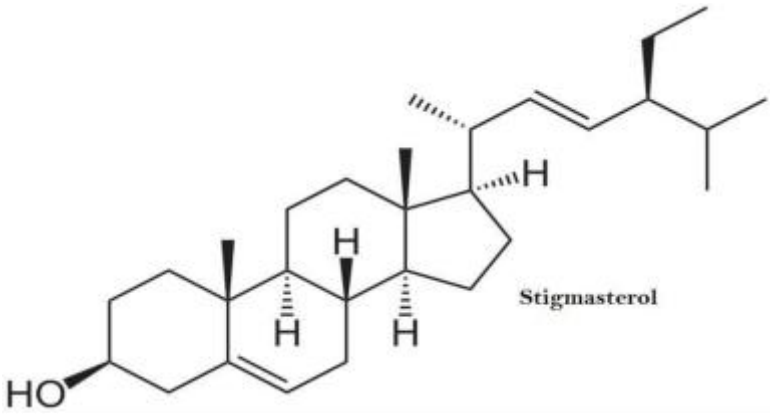
4. PHYTOCHEMISTRY OF PLANT EXTRACTS

In present day school Sylvester fast tested for their juvenile hormone and long activity against *Diacrisia obliqua* and *Earias Fabia* Pest. Then, methanolic extract of this plant were used for preliminary phytochemical screening of the secondary metabolites and Saponins, flavonoids Phenolic compound, glycosides, Lipids, protein Sterols and Alkaloids were found to be confirmed in each plan extract. Then, extracts were purified by three layer and column chromatography and purified fraction were sent to SAIF, CDRI Lucknow for IR, UV, ^{1}HNMR, ^{13}CNMR and Mass spectral analysis (SAIF No.7716). On the basis of a spectral graph interpretation of active principle of this plant extract done and their explanation with structure are as under.

Mass spectral analysis of the plant extract

The proposed structure has been confirmed along with the complete chemical and spectroscopic data in its Support. 1he data of the compound was found in full agreement with the literature (Cong, 1987; Silverstein, 1974) and the different species formed . The significant fragmentation pattern observed in the ESI-MS of *Cleome viscosa* (CV-3) revealed m/z = 279.08 M^{+}, 251.09 $(M\text{-}COH)^{+}$, 207.11 (M-CO-COH), 421.00 (M^{+}), 391.0 ($M\text{-}H_2O^{+}$), 375.0 (M-Me- H_2O^{+}), 359.02 (M-C3H7-H2O), 299.12 (M- C8H16), 279.08 $(M\text{-}H,O\text{-}C9H17)^{+}$, 269.13 (M-C10H21) +. On the

basis of the interpretation of these graphs and available literature (Ranjitha et al., 2009) a compound Stigmasterol with molecular formula of C29H48O and Molecular weight of 412 gm/mol was elucidated from fraction (CV-3) of *Cleome viscosa* that is given below:



Stigmasterol

5. DISCUSSION

India is rich in wide flora which not only consists of medicinally important plants but also consists of several toxic plants. *Cleome viscosa* of family Casperdlaccae was used In the present study to Isolate a compound that was 1ound to be analogous to the juvenile hormone which interferes with growth and metamorphosis of lepidopteron pest. Hence, 360gm fresh leaves of the plant Cleome viscosa L. of family Cleomaceae or Casperadaceae that is commonly known as "Wild mustard" was shade dried and loss in Weight was measured 87.5% then it was used for the isolation of crude extract by distillation using Soxhlet apparatus and used methanol, chloroform and water for soxhletion and percentage yield of the extract was calculated which was to be maximum in methanol (6.60a), 1ollowed by 5.44o in water, 5.22 % in chloroform, respectively. Niraimalhi, Karunanithi and Brindha (2012) have reported percentage yield of Cleome viscosa various extracts and maximum percentage yield was found to be l1.41% in ethanol, followed by 9.57% in chloroform and 3.89% n-Hexane.

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