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Effect of Plant Resistance in Castor on Endolarval Parasitoid *Snellenius maculipennis* (Szepligate) of Castor Semilooper *Achaea janata* L.

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ABSTRACT

A study on correlation between biological parameters of S. maculipennis with biophysical (epicuticular wax content) and biochemical (total free amino acids, proteins, carbohydrates and phenols) constituents of castor genotypes revealed that wax content was significantly negatively correlated with per cent parasitisation (-0.445); total free amino acids were significantly negatively correlated with per cent parasitisation (-0.702**) and egg and larval period (-0.546**); the proteins were significantly positively correlated with per cent parasitisation (0.938**); carbohydrates were significantly positively correlated with per cent parasitisation (0.957**) and significantly negatively correlated with adult longevity (-0.622**) and phenols were significantly negatively correlated with per cent parasitisation (-0.759**) and egg and larval period (-0.486*).*

Key words: *Snellenius maculipennis, Achaea janata, Genotypes, Correlation, Parasitisation.*

INTRODUCTION

Castor, *Ricinus communis* L. is one of the most important commercial, non-edible oilseed crop in Euphorbiaceae family. The genus, *"Ricinus"* is derived from Latin word meaning "dog tick" because of it's seed resemblance to the common pest of dog. It is reported to have originated in the tropical belt of both India and Africa. Castor is cultivated around the world because of the commercial importance of its oil. Historically, Brazil, China and India have been the key producing countries meeting global requirements. It is cultivated in 30 different countries on commercial scale, of which India, China, Brazil, USSR, Thailand, Ethiopia and Philippines are major castor growing accounts for majority of the world's production (http://www.commoditiescontrol.com).

India is the world's single largest producer of castor seed accounting for more than 85% followed by China with about 7% and Brazil with about 5% of world castor seed output. There has been a consistent increase in output primarily on account of rise in levels in India. In recent years, however, Brazil and China have experienced stagnation in Castor crop (http://www.commoditiescontrol.com).

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India is the world's single largest producer of castor seed accounting for more than 85% followed by China with about 7% and Brazil with about 5% of world castor seed output. There has been a consistent increase in output primarily on account of rise in levels in India. In recent years, however, Brazil and China have experienced stagnation in Castor crop (http://www.commoditiescontrol.com).

Castor oil and its derivatives, besides being used in medicine, are also used in a wide range of sectors including agriculture, textile industry, paper industry, plastic engineering, rubber and pharmaceuticals[4].

Castor oil is also used in many veterinary medicines, as an emollient, soothing medium when dropped into the eyes of animals after removal of foreign bodies. In eri silk-producing areas, castor leaves are fed to eri worms. After extraction of oil, castor cake is valued as manure. It contains 6.4% N, 2.5% phosphoric acid and 1% K and some micronutrients. Castor is also valued for its anti-termite properties. Consequently, there has been a steady increase in the demand for castor oil and its products in the world market owing to their renewable nature, biodegradability and eco-friendliness[4].

Castor (*Ricinuscommunis* L.) is cultivated around the world because of the commercial importance of its oil. India is the world's largest producer of castor seed and meets most of the global demand for castor oil. It contributes about eight lakh tonnes of castor seed and three lakh tonnes of castor oil and sharing 65% and 51% respectively in the world total production. Gujarat, Rajasthan and Andhra Pradesh are the major castor growing states in India. In Andhra Pradesh, castor occupied about 0.33 lakh ha area in 2016-17 and production was estimated about 0.16 lakh tonnes against 0.51 lakh ha area and 0.29 lakh tonnes production in 2015-16(https://www.indiastat.com).

Both biotic and abiotic stresses are the major constraints for decline in castor production. Insect pests are the major biotic factors in lowering of castor yield. The most important insect pests are castor semilooper, *Achaea janata* (Linnaeus); castor shoot and capsule borer, *Dichocrocis punctiferalis* (Guenee); hairy caterpillar, *Euproctisfraterna* (Moore); tobacco caterpillar, *Spodoptera litura* (Fabricius) and sucking pests, such as leafhopper, *Amrasca biguttula biguttula* (Ishida); white fly, *Trialeurodes ricini* (Misra); thrips, *Retithrips syriacus* (Mayet) and mites, *Tetranychus telarius* (Linnaeus)[19].

Castor semilooper *A. janata* is a major defoliator and under severe infestations, completely devour the green foliage, leaving only the veins and enforce the farmers to re-sow the crop. It causes yield reduction to the extent of 20 to 23%[9].

Castor semilooper, *A. janata* is regulated by hymenopteran endolarval parasitoid *Snellenius* (=*Microplitis*) *maculipennis* (Szepligate) (Hymenoptera: Braconidae) which is cosmopolitan in distribution[8]. Under field conditions, it is capable of parasitizing up to 77.31% of semilooper population[29]. The early instars of *A. janata* were attacked by the parasitoid and parasitized larvae did not feed and died later on[40].

Indiscriminate use of pesticides leads to development of resistance in insects to insecticides, toxic effect on natural enemies, pest resurgence and pesticide residues on food and adverse effect on human beings and causes environmental pollution[23]. These negative externalities, though cannot be eliminated altogether, their intensity can be minimized through development, dissemination and promotion of alternative technologies such as integrated pest management as well as good agronomic practices rather relaying solely on chemical pesticides. Integrated Pest Management (IPM) means a careful consideration of all available pest control techniques and subsequent integration of appropriate measures that discourage the development of pest populations by keeping pesticides and other interventions to levels that are economically justified and reduce or minimize risks to human health and the environment.

One of the important components of integrated pest management is the use of natural enemies as biocontrol agents. This has many advantages over the traditional method of chemical control[34]. With regard to parasitoids as biocontrol agents, they assume immense importance as they account for 87% suppression of insect pest in nature in comparison to 12% by predators and 1% by pathogens[20].

Host plant resistance along with natural enemies and cultural practices is a central component of any pest management strategy. Resistance in the form of plant defence is a physiochemical characteristic of plant, which influence the behaviour, biology, survival and reproduction of insects[38].

The characteristics of plant structure and texture can affect herbivores, natural enemies of herbivores, and their interaction. These effects may be genetically variable among plants and induced in individual plants by herbivore attack, and are mediated by primary plant attributes (*i.e*., nutritional quality and physical structure) and defense-related products (*i.e*., secondary chemicals and plant volatiles). The study of tri-trophic interactions is importantin order to understand natural enemies interactions and to manipulate these interactions in pest control[1]. Herbivore-induced plant volatiles have been suggested to function as indirect defence signals that attract natural enemies of herbivores. Several insect parasitoids are known to exploit such plant-provided cues to locate their hosts[13].

Generally, parasitoids are host specific and parasitize the host in a density dependent manner[2], and when exploited successfully this can be an effective way to reduce the frequency of pesticide applications and also reduced environmental pollution[41].

MATERIAL AND METHODS

Maintenance of stock-culture of semilooper *A. Janata*

The stock culture of *A. janata* was maintained in the Insectary, Department of Entomology, S.V. Agricultural College, ANGRAU, Tirupati, Andhra Pradesh at 25 ± 2°C, 75 ± 5% RH during 2017-18. Adults of *A. janata* were collected from college farm and Regional Agricultural Research Station, Tirupati and released into oviposition cages (32 cm × 30 cm × 30 cm) for mating and were provided with 20% honey solution mixed with vitamin - E tablets in cotton swabs. The petioles of castor leaves were kept in conical flasks by placing the cut end of petioles dipped in water containing 10% sugar solution, these leaves acted as substrate for oviposition and old leaves were replaced with fresh leaves as and when required. After one or two days of exposure, leaves having eggs of *A. janata* were shifted to another separate plastic troughs of 25 cm diameter and 10 cm height where the eggs were allowed to hatch. Fresh leaves were provided *adlib* to the newly hatched larvae and the rearing troughs were cleaned regularly to keep a healthy stock culture.

Pupae formed from the stock culture were collected and kept in separate oviposition cages for adult emergence for further rearing as described above. The first instars were used for all the experiments in the present investigations.

Maintenance of nucleus culture of parasitoid, *Snelleniusmaculipennis*

The stock culture of *S. maculipennis* was maintained at Insectary, Department of Entomology, S.V. Agricultural College, Tirupati at 25 ± 2°C, 75 ± 5% RH. Initially, parasitized larvae of *A. janata* with cocoons were collected from college farm and Regional Agricultural Research Station, Tirupati and were kept in Petri plates of 15 cm diameter. Adults emerged from these cocoons were released into oviposition cages for mating and were provided with 5% honey solution mixed with proteinex powder (Nutricia International Pvt Ltd. India) has ingredients namely Vitamin A, D, E, K, B complex group, Folic Acid, Biotin etc. and several minerals like iron, calcium, phosphorus and choline as well, dipped in cotton swab as food material.

Second instar larvae of *A. janata* were released on castor leaves and were offered to adults of *S. maculipennis* for oviposition in oviposition cages. The cut end of petioles of

castor leaves were dipped in water in a conical flask containing 10% sugar solution. After exposing the larvae for 24 to 48 hrs to the adult parasitoids, the parasitised larvae were collected and reared separately in plastic troughs of 20 cm diameter and provided with castor leaves till cocoon formation. After formation of the cocoon from parasitized larvae, parasitized larvae with cocoons still attached to its body were kept in Petri plates of 15 cm diameter for further rearing as described above. The parasitoids were reared and multiplied for two generations to obtain sufficient number of parasitoids before using them for the experiments.

Pot culturing of castor genotypes with different blooms.

Three genotypes of castor *viz*., DPC-9 (Green, zero bloom, susceptible), 48-1 (Red, double bloom, moderately resistant) and GCH-4 (Red, triple bloom, resistant) showing different degrees of resistance to *A. janata*[33] were grown in plastic pots of 23 cm diameter and 20 cm height during *kharif* 2017 under net house conditions at 25-39^{o}C and 48-80% RH . The seeds of these germplasm were procured from Regional Agricultural Research Station, Palem, Telangana, India and The Indian Institute of Oilseeds Research (IIOR), Hyderabad, Telangana, India. The substrate used for growing of plants was red soil : compost in 3 : 1 ratio.

The seeds of test plants were treated with fungicides (Mancozeb 2g Kg^{1}) to prevent seed-borne fungal diseases. All the normal agronomic practices were followed for pot culturing of plants. Leaves from these castor genotypes at peak vegetative stage were used in the experiments. Staggered planting of castor genotypes was done for continuous supply of leaves during the period of experimentation.

Table 1: Bloom characters of castor genotypes used in the present investigation

Genotype	Bloom nature	Character
DPC-9	Green zero	No bloom or waxy material on any part of the plant.
48-1	Red double	Bloom on stems, petioles and lower surface of leaves but not on the upper surface of the leaves.
GCH-4	Red triple	Bloom on every part of plant such as stems, petioles, upper and lower surface of leaves.

Studies on biology of *S. maculipennis* on *A. janata* larvae, reared on castor genotypes with different blooms.

The biology of *S. maculipennis* was studied under laboratory conditions of 25 ± 2˚C temperature and 75 ± 5% RH on excised leaves of three castor genotypes with different blooms *viz*., DPC-9, 48-1 and GCH-4. The petioles of each castor genotype leaves were dipped in water in separate conical flasks containing 10% sugar solution. These conical flasks containing excised leaves of castor genotypes were kept in three separate oviposition cages.

Ten first instar larvae of semilooper *A. Janata* were released on the excised leaves of each genotype and were left for feeding on the leaves for two days. There were total of eight replications. After two days, five pairs (five males and five females) of parasitoids were released in each cage and were allowed to oviposit on *A. janata* larvae feeding on excised leaves of castor with different wax blooms. After exposing the host larvae for 24 to 48 hrs to the adult parasitoids, the parasitized larvae were collected and reared in plastic troughs separately, till cocoon formation and parasitized larvae with cocoons still attached to its body were kept in separate plastic boxes of size 8.5 cm × 4.5 cm × 2.5 cm for adult emergence. After adult emergence they were provided with 5% honey mixed with proteinex powder dipped in cotton swab as food material. Data was recorded on egg+larval duration, number of cocoons formed, cocoon duration and adult longevity of *S.*

maculipennis on *A. janata* reared on castor genotypes having different wax blooms.

Studies on role of castor wax blooms on ovipositional behavior of *S. maculipennis*

A fully developed leaves from each green zero bloom, red double bloom and red triple bloom were excised upto petiole and cut ends of petiole was placed in conical flask with 10% sugar solution and this setup was used for oviposition preference of *S. maculipennis*.

Ten first instar *A. janata* larvae were released on leaves each of zero, double and triple bloom. The larvae were allowed to feed on the leaves of respective genotypes for two days. Care was taken to prevent movement of larvae from leaves of one genotype to another by keeping conical flasks (with leaves) on a 20 cm diameter trough underneath and were arranged in such a way that the leaves do not touch each other.

After two days of larval feeding, five pairs of adult parasitoids were released inside the cage and allowed to oviposit (free choice condition) on the larvae of *A. janata* feeding on different castor genotypes with different blooms. There were total of eight replications. After exposing the larvae for 24 to 48 hrs to the adult parasitoids, the parasitized larvae were collected and reared separately in plastic troughs and provided with leaves of respective castor genotypes till cocoon formation. Emerged adults from cocoons, were provided with 5% honey solution mixed with proteinex powder dipped in cotton swab as food material.

Oviposition preference of *S. maculipennis* towards *A. janata* larvae fed on castor genotypes with different blooms *viz*.,DPC-9, 48-1 and GCH-4 under free choice condition were recorded and expressed as per cent parasitisation.

$$\text{Per cent parasitisation} = \frac{\text{No. of larvae parasitized}}{\text{Total no. of larvae released}} \times 100$$

Studies on biochemical constituents of castor genotypes with different blooms.

The biochemical constituents such as protein, phenol, total free amino acids, and total carbohydrates were estimated at Institute of Frontier Technology (IFT), Regional Agricultural Research Station, Tirupati. The procedures and protocols are enlisted as below. A total of eight replications were used for estimating the biochemical constituents of castor genotypes with different blooms *viz*.,DPC-9, 48-1 and GCH-4.

Total free amino acids (µg/g)

Total free amino acids was estimated by adopting the method suggested by Sadasivam and Manickam[32], Moore and Stein[24]. 500 mg of leaf sample was taken and ground in five ml of 80% ethanol. The homogenate was centrifuged at 6000 rpm for 30 min at 20^{o}C. The supernatant obtained was used for estimation of free amino acids. 0.1 ml of the extract was taken and to it one ml of Ninhydrin solution was added, followed by making up the volume to two ml with distilled water. The test tubes were then heated in boiling water bath for 20 min. Then five ml of diluents was added and the contents were mixed. After 15 minutes intensity of the purple colour was read against a reagent blank in a colorimeter at 570 nm. A standard curve was prepared using leucine and it was linear within the range (R^2 = 0.984). The total free amino acids of the leaf sample was then calculated by substituting the sample Optical Density values in the formula derived from the graph (Y = 0.012X – 0.202), where Y = Optical Density value of the sample and X = concentration (µg ml^{-1}) of free amino acids. Then the concentration of free amino free amino acids in (µg ml^{-1}) was converted to concentration of free amino acids in µg/g for different entries.

Estimation of proteins (mg/g)

The protein content in fresh leaves of all the three castor genotypes such as DPC-9, 48-1 and GCH-4 were estimated as per the method by Wildman and Bonner[43]. 250 mg of leaf sample was macerated with ten ml of phosphate solution. The contents were

centrifuged at 3000 rpm for about ten min and then the supernatant was collected followed by volume make up to 25 ml with distilled water. One ml of supernatant was pipette out and five ml of alkali copper tartarate was added to it. The solution was kept as such for 30 min for colour development. 0.5 ml of phenol reagent was then added and the Optical Density value of the sample was measured at 660 nm in Spectrophotometer. A standard curve was prepared by using bovine serum albumin and it was linear within the range (R^2 = 0.969). The protein content of the sample was calculated by substituting the sample Optical Density values in the formula derived from the graph (Y = 0.0065X – 0.014), where Y = Optical Density value of the sample and X = concentration (µg ml^{-1}) of the protein. Then the concentration of proteins in (µg ml^{-1}) of leaf sample was converted to concentration of proteins in mg/g for different entries.

Total carbohydrates (g/g

Total carbohydrates was estimated by adopting the method suggested by Sadasivam and Manickam (1961), Hedge and Hofreiter[12]. 50 mg of leaf sample was taken in a 50 ml test tube and 2.5 ml of 2.5 N HCl was added to it. The test tubes were kept in hot water bath (100^oC) for three hours and cooled to room temperature. Solid sodium carbonate was then added to the test tubes followed by volume make upto 25 ml using distilled water. The test tubes were then kept at room temperature for 20 min without any disturbance. For analysis one ml of the aliquot was taken, cooled on ice, and four ml of ice cold anthrone reagent was added to it. The test tubes were then kept in hot water bath for eight min and cooled rapidly. The absorbance of the resultant green was measured against a reagent blank at 630 nm. A standard curve was prepared using glucose and it was linear within the range (R^2 = 0.987). The total carbohydrates of the leaf sample was then calculated by substituting the sample Optical Density values in the formula derived from the graph (Y = 0.006X – 0.023), where Y = Optical Density value of the sample and X = concentration (µg ml^{-1}) of carbohydrate. Then the concentration of carbohydrates in (µg ml^{-1}) was converted to concentration of carbohydrates in g/g for different entries.

Phenol content (%)

Phenol content in the castor leaves was estimated by adopting the method suggested by Sadasivam and Manickam[32], Malik and Singh[22], Muthukumaran[25]. One g of the leaf sample was ground in ten time volume of 80% ethanol with a pestle and mortar. Then the homogenate was centrifuged at 10000 rpm for 20 min, supernatant was saved. Then the residue was extracted with five times volume of 80% ethanol and centrifuged. Then the supernatants were pooled and evaporated to dryness. The residue was dissolved in ten ml of distilled water. 0.4 ml of the sample extract was pipetted into test tubes and the volume was made upto three ml with distilled water followed by addition of 0.5 ml of folin-ciacalteau reagent. After three min, two ml of 20% sodium carbonate was added and the test tubes were kept in boiling water bath for one min, cooled and the absorbance was measured against a reagent blank at 650 nm. A standard curve was prepared by using catechol and it was linear within the range (R^2 = 0.963). The phenol content of the sample was calculated by substituting the sample Optical Density values in the formula derived from the graph (Y = 0.397X + 0.013), where Y = Optical Density value of the sample and X = concentration (µg ml^{-1}) of the phenol. Then the concentration of phenols in µg ml^{-1} was converted to concentration of phenols in % for different entries.

Studies on biophysical constituent (wax) of castor genotypes with different blooms.

The biophysical constituent *i.e.,* wax was estimated at Department of Crop Physiology, S.V. Agricultural College, Tirupati.

The wax content in fresh leaves of all the three castor genotypes *viz*., DPC-9, 48-1 and GCH-4 with different blooms were estimated as per the method suggested by Eberco*net al*.[6], with suitable modifications.

Principle

Colour change produced due to the reaction of wax with acidic $K_2Cr_2O_7$ reagent was measured at 590 nm by calorimetry.

Reagent

Acidic $K_2Cr_2O_7$ reagent: 20 g $K_2Cr_2O_7$ was dissolved in 40 ml of distilled water and then mixed with one litre H_2SO_4 and heated to clear solution.

Procedure

The leaf discs (10 no's each of 9 cm diameter) were immersed in 15 ml chloroform for 15 sec. The extract was filtered and evaporated over boiling water bath till there was no smell of chloroform. Five ml of the reagent $K_2Cr_2O_7$ was added to this extract and placed on boiling water bath for 30 min. After cooling, 12 ml of distilled water was added to the resultant extract. The Optical Density was measured at 590 nm after 30 min. Sorghum wax collected from leaf sheaths was dissolved in chloroform and used as standard wax.

Standard curve

Standard solution: 100 mg of sorghum wax was collected from leaf sheaths of 60 days old sorghum plants. The same was dissolved in 100 ml of chloroform.

Working standard: One ml of standard solution was dissolved in 100 ml of chloroform to get minimum concentration of 10 µg in 1 ml.

Different concentration of wax, *i.e.*, 20, 40, 60, 80 and 100 µg was prepared by taking different volume of 2, 4, 6, 8 and 10 ml of working standard solution in different test tubes and evaporated over boiling water bath till there was no smell of chloroform. Five ml of the reagent was added to this working standard and placed on boiling water bath for 30 min. After cooling, 12 ml water was added to the resultant working standard. After 30 min, the Optical Density was measured at 590 nm. To get the standard curve Optical Density values were plotted against concentration.

By comparing the Optical Density value of the sample in the standard curve, the leaf epicuticular wax content in the sample (seven replications) was computed and expressed in µg per unit of cm^2 area.

RESULTS AND DISCUSSION

Biology of endolarval parasitoid (*Snellenius maculipennis*) of castor semilooper *Achaea janata*.

The present study on the biology of *S. maculipennis* Szep. revealed that the newly emerged adult parasitoid mated on the day of emergence and the oviposition commenced from the succeeding day. Twenty four hours after parasitisation, the larvae became sluggish, however it continued feeding at a slower rate till 5-6 days after parasitisation when the parasitoid grub emerged from lateral, posterior end of the host insect body. The grub immediately started forming the cocoon and attached itself to the posterior ventral portion of the host insect. After the emergence of the parasitoid grub the host survived for 4 to 5 days but stopped feeding and died later.

Duration of egg+larva of *S maculipennis*

While studying the biology of *S. maculipennis* on *A. janata* larvae, it was observed that the female parasitoid thursts eggs into the host body (in hemolymph), from the dorsal side of the abdominal segments. The egg after hatching remains in the hemolymph of host body and feeds there in. When the grub completes its larval period, final instar cuts through the body wall of the host larvae on the lateral side, comes out and spins a cocoon around its body within five hours. The grub was apodous, spindle shaped and transparent.

Longest durations of egg+larval stages of *S. maculipennis* (8.02 days) was recorded in *A. janata* larvae reared on GCH-4. Shorter duration of 5.09 days was recorded on 48-1 followed by DPC-9 (6.72 days) which were on par with each other (Table 4.1).

Number of cocoons formed

More number of cocoons of *S. maculipennis* (9.63) was formed on *A. janata* larvae reared on DPC-9. Less number of cocoons of *S. maculipennis* (7.75) was formed on *A. janata* larvae reared on 48-1 followed by GCH-4 (8.75) which were significantly different from each other (Table 4.1).

Duration of cocoon of *S. maculipennis*

The cocoon was initially grey in colour and later turned brown. The cocoons on one side

was firmly attached to rear end of the host larvae and other side to the leaf surface rendering the host immobile.

Longest cocoon duration of *S. maculipennis* (5.33 days) was recorded on *A. janata* larvae when reared on DPC-9. Shortest duration of 4.11 days was recorded on GCH-4 followed by 48-1 (4.81 days) which were on par with each other (Table 4.1).

Longevity of adult parasitoid, *S. maculipennis*

Longevity of adult *S. maculipennis*was calculated as the day when they were emerged from cocoons till death. The adult parasitoid was short, black in colour. The first half of the abdomen was glossy yellowish in colour.

Longest adult longevity of *S. maculipennis* (6.38 days) was recorded when reared on larvae of *A. janata* fed on GCH-4 and shortest adult longevity of 3.64 days was recorded on DPC-9 followed by 48-1 (4.36 days) which were on par with each other (Table 4.1).

Duration of total life cycle of the parasitoid, *S. maculipennis*

Longest duration of total life cycle of parasitoid *S. maculipennis* (18.50 days) was recorded when reared from larvae of *A. janata* fed on GCH-4 and shortest duration of total life cycle of 14.26 days was recorded when reared on 48-1 followed by DPC-9 (15.69 days) which were on par with each other (Table 4.1).

Our results were in conformity with the Prabhakar and Prasad[28] who reported that egg and larval, cocoon, adult longevity, and total life cycle of *S. maculipennis* on *A. janata* larvae as 6.25 ± 0.96 days,5.08± 0.90 days, 3.5 ± 1.02 days and 14.83 ± 2.83 days respectively.

Table 2: Biological parameters of endolarval parasitoid *S. maculipennis* on castor semilooper *A. janata* larvae reared on castor genotypes with different wax blooms

Genotype	Egg and larval period (days)	Number of cocoons formed	Pupal period (days)	Adult longevity (days)	Total life cycle (days)
DPC-9 (Zero bloom, Susceptible)	6.72^{ab}	9.63^{a}	5.33^{a}	3.64^{b}	15.69^{ab}
48-1 (Double bloom, Moderately resistant)	5.09^{b}	7.75^{ab}	4.81^{ab}	4.36^{b}	14.26^{b}
GCH-4 (Triple bloom, Resistant)	8.02^{a}	8.75^{a}	4.11^{b}	6.38^{a}	18.50^{a}
LSD at 0.01	1.90	1.11	1.18	1.83	3.14
CV %	20.31	8.99	17.60	26.92	13.75

Values followed by same letter are not significantly different at 0.01 level.

Role of castor wax bloom on ovipositional behavior of*S. maculipennis*.

Adults of *S. maculipennis* were allowed to oviposit on *A. janata* larvae reared on DPC-9 (Green, zero bloom, susceptible), 48-1 (Red, double bloom, moderately resistant) and GCH-4 (Red, triple bloom, resistant) under free choice condition. Highest parasitisation of 65% was recorded on *A. janata* larvae when reared on DPC-9 and was offered for parasitisation. Lowest parasitisation of 13.75% was recorded on *A. janata* larvae when reared on 48-1 and was offered for parasitisation followed by GCH-4 (18.75 %) which were on par with each other (Table 4.2.1).

Table 3: Parasitisation of *A. janata* larvae by *S. maculipennis* reared on castor genotypes with different wax blooms under free choice condition

Genotype	Per cent parasitisation
DPC-9 (Zero bloom, Susceptible)	65^{a} (53.82)
48-1 (Double bloom, Moderately resistant)	13.75^{c} (21.48)
GCH-4 (Triple bloom, Resistant)	18.75^{bc} (25.54)
LSD at 0.01	9.14
CV %	19.86

Values followed by same letter are not significantly different at 0.01 level and the values in parenthesis are transformed values.

Biochemical and biophysical constituents of castor genotypes with different blooms

Biochemical constituents *viz*., total free amino acids, protein content, carbohydrates, phenol content and biophysical constituent *i.e*., wax from leaves of castor genotypes with different blooms namely DPC-9 (Green, zero bloom, susceptible), 48-1 (Red, double bloom, moderately resistant) and GCH-4 (Red, triple bloom, resistant) were estimated as per protocols in section of material and methods, and are presented in (Table 4.3).

Biochemical constituents

Total free amino acids (TFA)

Highest amount of total free amino acids (307.45 µg/g) was recorded from the leaves of 48-1 followed by GCH-4 (203.43 µg/g). Lowest amount of total free amino acids (175.76 µg/g) was found in DPC-9. All were significantly different from each other (Table 4.3).

Protein content

Highest amount of protein (0.57 mg/g) was recorded from the leaves of DPC-9. Lowest amount of protein (0.30 mg/g) was found in 48-1 followed by GCH-4 (0.37 mg/g). All were significantly different from each other (Table 4.3). The results were in accordance with Kumar *et al*.[18], who reported that amount of proteins (5.19 to 6.78 mg/g) in all susceptible genotypes of rice was found higher compared to resistant genotypes (2.97 to 4.2 mg/g).

Carbohydrate content

Highest amount of carbohydrate (0.24 g/g) was recorded from the leaves of DPC-9. Lowest amount of carbohydrate (0.15 g/g) was found in GCH-4 followed by 48-1 (0.17 g/g). All were significantly different from each other (Table 4.3).

Phenol content

Highest amount of phenol (11.39%) was recorded from the leaves of 48-1 followed by GCH-4 (7.17%). The lowest amount of phenol (5.34%) was found in DPC-9. All were significantly different from each other (Table 4.3). Our results were in conformity with Sharma and Bharadwaj[39], who found that amounts of polyphenols was highest in genotypes which are resistant to *Helicoverpa armigera*.

Biophysical constituent

Wax content

Highest amount of wax (1.43 µg/cm^2) was recorded from the leaves of GCH-4 followed by48-1 (1.20 µg/cm^2) which were significantly different from each other. The lowest amount of wax (0.98 µg/cm^2) was found in DPC-9 (Table 4.3).

Table 4: Biochemical and biophysical constituents of castor genotypes with different wax blooms

Genotype	Total free Amino acids (µg/g)	Proteins (mg/g)	Carbohydrates (g/g)	Phenols (%)	Wax(µg/ cm^2)
DPC-9 (Zero bloom, Susceptible)	175.76^c	0.57^a	0.24^a	5.34^c (13.36)	0.98^b
48-1 (Double bloom, Moderately resistant)	307.45^a	0.30^c	0.17^b	11.39^a (19.72)	1.20^{ab}
GCH-4 (Triple bloom, Resistant)	203.43^b	0.37^b	0.15^c	7.17^b (15.53)	1.43^a
LSD at 0.01	0.58	0.04	0.01	0.39	0.44
CV %	0.18	7.68	5.33	3.45	23.56

Values followed by same letter are not significantly different at 0.01 level and the values in parenthesis are transformed values.

Correlation study of different developmental periods of *S. maculipennis* on *A. janata* larvae reared on castor genotypes with different wax blooms and per cent parasitisation in free choice condition with biochemical and biophysical constituents of castor genotypes with different wax blooms

Biochemical constituents

Total free amino acids were significantly negatively correlated with per cent parasitisation (-0.702**) and egg and larval period (-0.546**) and for the rest of other biological parameters the correlations were non significant.

The protein content were significantly positively correlated with per cent parasitisation (0.938**) and for the rest of other biological parameters the correlations were non significant.

Carbohydrates were significantly positively correlated with per cent parasitisation (0.957**) and significantly negatively correlated with adult longevity (-0.622**) and for the rest of other biological parameters the correlations were non significant.

Phenols were significantly negatively correlated with per cent parasitisation (-0.759**) and egg and larval period (-0.486*) and for the rest of other biological parameters the correlations were non significant. The results were on par with the findings of Ghumare and Mukherjee[10], who reported that lower the amount of total phenolics, better the performance of *Spodoptera litura*and have inverse relationship with preference and larval growth. Muthukumaran[25] reported that phenol content was higher in resistant varieties and it exerted a significant negative correlation with larval feeding and negatively correlated with fruit worm *H. armigera* infestation and leaf caterpillar *S. litura* Fab. incidence in tomato. Biochemical contents of the tomato such as non-reducing sugar and phenols present in higher quantities were found negatively correlated with fruit worm *H. armigera* infestation[35,5,36]. Further, phenol content was higher in resistant accession and it exerted a significant negative correlation with larval feeding[11].

In the present investigations, it was observed that total free amino acids and phenols had a significant negative correlation with and per cent parasitisation. At the same time proteins and carbohydrates had a significant positive correlation with per cent parasitisation.

Phenols which is a plant secondary metabolite is known to exert a negative effect on growth and metabolism of insect herbivores. Several reports have shown negative effect of phenols on insect herbivore, at the same time proteins and carbohydrates tend to act as phagostimulants to most of insect herbivores which increases the susceptibility of host plants to insect herbivores. The results were in close association with Tilakchand[42]. who reported that protein (0.8292 mg/g) content manifested positive and significant correlations while phenol (-0.9459 mg/g) content recorded significantly negative correlation with larval population of *H. armigera*. According to Sharma and Singh[37] plant biochemical characters in okra *i.e*., total sugar and crude protein were positively correlated with fruit borer infestation, whereas total phenols had a negative correlation. Rath and Nayak[31], revealed that the lower level of protein in fruits of tomato varieties were responsible for the low susceptibility of tomato varieties to fruit borer and high amount of phenol were responsible for resistance. Rashmi and Vemadevaiah[30] reported significantly higher amount of total phenol in bollworm tolerant genotype Sahana (6.25 mg/g) in all the stages than Laxmi (5.60 mg/g) indicated its positive role in pest tolerance. Presence of lesser number of survived larvae in Sahana than Laxmi indicated its biochemical basis of tolerance, the genotype Sahana has higher total phenols content than Laxmi. Muthukumaran[25] found that phenol content was high in resistant varieties of tomato against fruit worm, *H. armigera* (Hubner) and exerted a significant negative correlation with larval feeding and negatively correlated with fruit worm *H.*

armigera infestation and leaf caterpillar *S. litura* F. incidence in tomato.

Biophysical constituent.

Wax content were significantly negatively correlated with per cent parasitisation (-0.445*) and for the rest of other biological parameters the correlations were non significant.

In the present investigation, wax content had significant negative correlation with per cent parasitisation. It was also observed from Table 4.2.1 that the per cent parasitisation of *A. janata* larvae by *S. maculipennis* was highest when larvae were allowed to feed on DPC-9 (Green, zero bloom, susceptible) with no wax bloom as compared to that of 48-1 (Red, double bloom, moderately resistant) and GCH-4 (Red, triple bloom, resistant).

In the present investigation, proteins and carbohydrates had a significant positive correlation with per cent parasitisation.

Low phenol (5.34%) and wax (0.98 µg/cm^2**)** contents in susceptible genotype DPC-9 had positive effect on the host insect *A. janata*, which might have resulted in highest per cent parasitisation when larvae reared on DPC-9 were offered for parasitisation to adult parasitoid *S. maculipennis* in free choice, dual choice and no choice condition.

When *A. janata* larvae were reared on a moderately resistant genotype 48-1 having more phenol content (11.39%), the host larvae either might have detoxified the plant secondary metabolites or might have resulted in storage excretion where the phenols might have been stored in fat bodies in the insect hemolymph. Nishio[27] reported that larvae of the monarch butterfly *Danaus plexippus*L. utilize a large volume of gut fluid as a critical factor in the processing of cardenolides ingested from the milkweed *Asclepias humistrata* host plants. Larvae can regurgitate gut fluid readily, giving them cardenolide-rich shield that can be thrust at predators. These bitter-tasting steroids reappear in the teneral adult and are concentrated in the wing scales and hemolymph, providing the imago with a readily available defence against avian predators such as blue jay *Cyano cittacristata.*

The parasitoid used in the present investigation *i.e., S. maculipennis* deposits eggs within hemolymph and it has been postulated by many workers that adult parasitoids generally tries to choose the best host for its offspring during oviposition. Karowe and Schoonhoven[17], reported that vigorous hosts feed more and therefore produce more tissue and/or hemolymph on which the parasitoids chooses to oviposit.

In our present investigation, larvae of *A. janata* feeding on castor genotypes with more phenols probably were least preferred for parasitisation by the adults of *S. maculipennis* as because of phenols in the bodies (hemolymph) of host larvae rendered them unpalatable or unsuitable for egg laying.

Table 5: Correlation of different developmental periods of *S. maculipennis* on *A. janata* larvae reared on castor genotypes with different wax blooms and per cent parasitisation in free choice condition with biochemical and biophysical constituents of castor genotypes with different wax blooms

Correlation of developmental periods of *S. maculipennis*		Biochemical constituents of castor genotypes with different wax blooms				
		Total free amino acids (µg/g)	Proteins (mg/g)	Carbohydrates (g/g)	Phenols (%)	Wax content (µg/cm^2)
Per cent parasitisation	Pearson correlation	-0.702**	0.938**	0.957**	-0.759**	-0.445*
Egg and larval period	Pearson correlation	-0.546**	0.167	-0.124	-0.486*	0.105
Cocoon period	Pearson correlation	-0.062	0.274	0.39	-0.124	-0.294
Adult longevity	Pearson correlation	-0.045	-0.388	-0.622**	0.025	0.313
Total life cycle	Pearson correlation	-0.397	-0.038	-0.328	-0.336	0.15

** Correlation is significant at the 0.01 level (2- tailed).

* Correlation is significant at the 0.05 level (2- tailed).

Summary and Conclusion

Effect of wax bloom of castor genotypes on the biology of endolarval parasitoid *Snellenius maculipennis*

More number of cocoons (9.63) of *S. maculipennis* were formed on *A. janata* larvae reared on DPC-9 and less number of cocoons (7.75) were formed on *A. janata* larvae reared on 48-1 followed by GCH-4 (8.75). Durations of egg and larval stages of *S. maculipennis* in *A. janata* larvae was longest on GCH-4 (8.02 days) and shortest on 48-1 (5.09 days). Cocoon duration of *S. maculipennis* on *A. janata* larvae reared on DPC-9 was longest (5.33 days) and shortest on GCH-4 (4.11 days).Adult longevity of *S. maculipennis* on *A. janata* reared on GCH-4 was longest (6.38 days) and shortest on DPC-9 (3.64 days). Duration of total life cycle of *S. maculipennis* on *A. janata* reared on GCH-4was longest (18.50 days) and shortest on 48-1 (14.26 days).

Role of castor wax bloom on ovipositional behavior of *S. maculipennis*

Under free choice condition highest parasitisation of 65% was recorded on *A. janata* larvae, reared on susceptible genotype DPC-9. Lowest parasitisation of 13.75% was recorded on on*A. janata* larvae, reared on moderately resistant genotype 48-1, followed by resistant genotype GCH-4 (18.75%).

Correlation between biological parameters, per cent parasitisation of *S. maculipennis*on *A. janata* with biophysical and biochemical constituents of castor genotypes

Correlation study of different developmental periods of *S. maculipennis* on *A. janata* larvae reared on castor genotypes with different wax blooms and per cent parasitisation in free choice condition with biochemical and biophysical constituents of castor genotypes with different wax blooms revealed that wax content was significantly negatively correlated with per cent parasitisation (-0.445*), total free amino acids were significantly negatively correlated with per cent parasitisation (-0.702**) and egg and larval period (-0.546**), the proteins were significantly positively correlated with per cent parasitisation (0.938**), carbohydrates were significantly positively correlated with per cent parasitisation (0.957**) and significantly negatively correlated with adult longevity (-0.622**), phenols were significantly negatively correlated with per cent parasitisation(-0.759**) and egg and larval period (-0.486*) and for the rest of other biological parameters the correlations were non significant.

REFERENCES

1. Agrawal, A.A., Mechanisms, ecological consequences and agricultural implications of tri-trophic interactions.*Current Opinion in Plant Biology*. **3:** 329–335 (2000).
2. Annecke, D.P. and Moran, V.C., *Insects and Mites of Cultivated Plants in South Africa*. Butterworths, Durban, South Africa. 338 (1982).
3. Annual Report - DOR. Directorate of Oil Seeds Research, Hyderabad.1-2 (1997).
4. Annual Report – DOR. Directorate of Oil Seeds Research, Hyderabad.1-2 (2003).
5. Dhakshinamoorthy, G., Studies on resistance of tomato against fruit borer, *Helicoverpaarmigera* (Hubner). M.Sc. (Ag.) Thesis, Annamalai University. India (2002).
6. Ebercon, A., Blum, A and Jordan, W.R., A rapid colorimetric method for epicuticular wax content of sorghum leaves. *Crop Science*. **17:** 179-80 (1977).
7. Food and Agriculture Organisation of the United Nations.*Integrated Pest Management.*http://www.fao.org/agriculture /crops/thematicsitemap/ theme/pests/ipm/en/
8. Gaikwad, B.B. and Bilapate, G.G., Field life-tables and key mortality factors of *Achaea janata* Linn. on castor. *Animal Sciences*. **98(5):** 331-339 (1989).
9. Gaikwad, B.B. and Bilapate, G.G., Parasitization of *Achaea janata* and estimation of losses on castor.*Journal of Maharashtra Agricultural Universities*. **17:** 195-196 (1992).
10. Ghumare, S.S. and Mukharjee, S.N., Performance of *Spodopteralitura*Fabricius on different host plants: influence of nitrogen and total phenolics of plants and

mid-gut esterase activity of the insect. *Indian Journal of Experimental biology*. **41:** 895-899 (2003).

11. Gopalakrishnan, R., Induced resistance in tomato against key insect pests. M.Sc. (Ag.) Thesis.*Annamalai University. India*. pp. 146 (2006).
12. Hedge, M. E. and Hofreiter, B. T., *In*: Carbohydrate Chemistry17 (Eds Whistler R L and Be MordaJ N) Acdemic Press, New York (1962).
13. Hoballah, M.E.F. and Turlings, T.C.J., Experimental evidence that plants under caterpillar attack may benefit from attracting parasitoids. *Evolutionary Ecology Research*. **3:** 553-565 (2001).
14. http://www.commoditiescontrol.com
15. http://www.eco-business.com/research/ economic-feasibility-of-sustainable-non-food-biodiesel-castor/
16. https://www.indiastat.com.
17. Karowe, D.N. and Schoonhoven, L.M., Interactions among three trophic levels: the influence of host plant on performance of *Pierisbrassicae* and its parasitoid, *Cotesiaglomerata.Entomologia ExperimentalisetApplicata*. **62(3):** 241-251 (1992).
18. Kumar, V.L., Patil, S.U. and Chakravarthy, A.K., Biochemical basis of resistance in rice against Asian rice gall mid-gut, *Orseoliaoryzae* (Wood-Mason) (Diptera: Cecidomyiidae). *Current Biotica*. **6(2):** 163-170 (2012).
19. Lakshminarayana, M. and Raoof, M.A., Insect pests and diseases of castor and their management. Directorate of Oilseeds Research, Hyderabad, pp: 78 (2005).
20. Lenteren, V.J.C., The development of host discrimination on the preventing of superparasitism in the parasite *Pseudocoliabochei* Weld. *Agriculture Ecosystems and Environment.***10:** 143 (1983).
21. Malick, C. P. and Singh, S. M., Plant Entomology and Histo Entomology. Kalyani Publishers, *New Delhi*. Pp 281(1980).
22. Malick, C. P. and Singh, S. M., Plant Entomology and Histo Entomology. Kalyani Publishers, *New Delhi*. Pp 281 (1980).
23. Metcalf, R.L. and Luckmann, W.H., *The pest management concept, Introduction to Insect Pest Management*. *John Wiley & Sons, New York*. 1 – 31 (1982).
24. Moore, S. and Stein, W. H., Methods in Enzymol (Eds. Colowick, S.P. and Kalpan, N.D) Academic Press New York. Pp 468 (1948).
25. Muthukumaran, N., Biophysical and biochemical factors of resistance in tomato accessions as influenced by selected bioinoculants against fruit worm *Helicoverpa armigera* (Hubner). *International Journal of Current Microbiology and Applied Sciences*. **5(1):** 252-262 (2016).
26. Naik, E., Tri-trophic interactions of castor semilooper, *Achaea janata* L. and it'sendolarvalparasitoid*Snelleniusmaculipennis* (Szepligate). *M. Sc. (Ag.) Thesis.*Acharya N.G. Ranga Agricultural University, Guntur, Andhra Pradesh, India (2017).
27. Nishio, S., The fates and adaptive significance of cardenolides sequestered by larvae of *Danaus plexippus* and *Cycnia inopinatus* (Hy. Edwards). *Ph. D. Thesis*. University of Georgia, Athens, G.A. (1980).
28. Prabhakar, M. and Prasad, Y.G., Biology, and seasonal dynamics of *Snellenius maculipennis (*Szepligate) (Hymenoptera: Braconidae) a larval parasitoid of castor semilooper *Achaea janata*(Linnaeus) (Lepidoptera: Noctuidae). *Journal of Biological Control*. **19(1):** 29-34 (2005).
29. Rai, and Jayaramaiah. The castor semilooper, *Achaea janata* Linnaeus (Lepidoptera: Noctuidae) and its control. *Journal of the Maharashtra Agricultural Universities*. **3 (1):** 73-74 (1978).
30. Rashmi, R. and Vamadevaiah, H.M., Influence of phenolic compounds imparting resistance to cotton (*Gossypium*

hirsutum) against *Helicoverpa armigera*. *International Journal of Tropical Agriculture*. **33(2):** 689-695 (2015).

31. Rath, L.K. and Nayak, V.S., Biochemical basis of resistance in some selected tomato varieties to tomato fruit borer. *Journal of Plant Protection and Environment*. **4(1):** 75-77 (2007).
32. Sadasivam, S. and Manickam, A., *Biometrical methods for Agricultural Sciences. Wiley Eastern Limited, New Delhi*.187-188 (1961).
33. Sarma, A.K., Singh, M.P. and Singh, K.I., Resistance of local castor genotypes to *Achaea janata* Linn. And *Spodoptera litura* Fab. *Journal of Applied Zoological Researches.* **17(2):** 179-181 (2006).
34. Scholler, M. and Flinn, P.W., Parasitoids and Predators. *Alternatives to Pesticides in Stored-Product IPM*. 229-271 (2000).
35. Selvanarayanan, V., Host Plant resistance in tomato against fruit borer, *Helicoverpa armigera* (Hubner). Ph.D. Thesis, Annamalai University, India. pp. 251 (2000).
36. Selvanarayanan, V., Narayanasamy, P., Factors of resistance in tomato accessions against the fruit worm *Helicoverpa armigera* (Hubner). *Crop Protection*. **256(25):** 1075–1079 (2006).
37. Sharma, B.N. and Singh, S., Biophysical and biochemical factors of resistance in okra against shoot and fruit borer. *Indian Jurnal of Entomology*. **72(3):** 212-216 (2010).
38. Sharma, H.C. and Ortiz, R., Host plant resistance to insects: an eco-friendly approach for pest management and environment conservation. *Journal of Environmental Biology*. **23(2):** 111-135 (2002).
39. Sharma, K.C. and Bharadwaj, S.C., Influence of phenological and physical characters of tomato on fruit borer, *Helicoverpa armigera* (Hubner) infestation. *Resistant Pest Management Newsletter*. **19(1):** 4648 (2009).
40. Somasekhar, J., Shekharappa, I., Patil, B.V. and Patil, S.A., Occurrence of castor semilooper, *Achaea janata* Linnaeus and its parasitoid *Microplitis maculipennis. Journal of Agrilculture Science*. **6(2):** 200-202 (1993).
41. Talekar, N.S., Yang, T.C. and Lee, S.T., Introduction of *Diadegma semiclausum* to control Diamond back moth in Taiwan. *Diamond back moth and other cruciferous pests*: *proceeding of the second international work shop*. Eds Talekar, N.S and Griggs, T.D., Asian Vegetable Research and Development Center, Taiwan. 263-270 (1990).
42. Tilakchand, A.A., Morphological and biochemical basis of resistance against fruit borer, *Helicoverpa armigera* (hubner) infesting tomato under South Gujarat condition. *Ph. D. Thesis*. N.M. College of Agriculture, Navsari Agricultural University, Navsari, Gujarat (2014).
43. Wildman, S.G. and Bonner, J., The proteins of green leaves; isolation, enzymatic properties and auxin content of spinach cytoplasmic proteins. *Archives of Biochemistry*. **14(3):** 381-413 (1947).