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ENHANCED REPRODUCTIVE POTENTIAL OF *NEOCHETINA BRUCHI* HOSTACHE FED ON WATER HYACINTH PLANTS FROM POLLUTED WATER BODIES

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CLOSE relations are known to exist between the phytophagous insects and their food plants. Only among female insects special nutritional requirements for reproduction are observed in addition to energy. The effects of various chemicals on plants may affect the insect species feeding on them. Sex ratio, reproduction and survival of the females varied in several species of insects with the food plant of their host¹⁻⁵. We report here the enhanced reproductive activity of *Neochetina bruchi* fed on water hyacinth plants grown in polluted water, rich in several inorganic molecules.

Eichornia crassipes (Mart) solms, commonly known as waterhyacinth has become a serious concern as a weed pest in recent years. Several pathogens, mites, insects and other arthropods are found attacking this weed, of which two species of weevils, *Neochetina eichorniae* and *N. bruchi* are found as promising bio-control agents⁶. Hence, the above two species of *Neochetina* have been introduced in India and host specificity tests were carried out under quarantine conditions at the Commonwealth Institute of Biological Control and the National Centre for Biological Control, Bangalore. The above weevils collected from Bangalore are being successfully reared in the laboratory glass

house. Adults of *N. bruchi* were selected for the present study. Water hyacinth plants were collected from two different polluted areas around Hyderabad. The above water bodies are heavily contaminated with industrial effluents from nearby industries like starch and brewery. The plants along with the polluted water were placed in 5 l beakers. Five pairs of freshly emerged adults of *N. bruchi* were introduced into each of these beakers. The insects released on plants collected from the laboratory pond were kept as control. The experiments were done in triplicates. The insects were examined for egg laying and hatching. The plants were replaced and the eggs and surviving females were counted daily. Their numbers and observations were recorded and the average mean fecundity rates determined.

For anatomical study, ovaries of the insects were collected from the polluted water bodies and the laboratory pond. Dissections were performed in the insect ringer solution. The material was stained with borax carmine, progressively dehydrated, cleared in cedarwood oil and fixed in canada balsam. Total amino acid and protein content of the leaves from the polluted water body were estimated and compared with that of the laboratory pond^{7,8}.

The effect of industrial pollutants on the fecundity of *N. bruchi* is shown in table 1. Females of *N. bruchi* laid tiny oval-round creamish eggs in the petioles of water hyacinth plants. The eggs hatched after 3-4 days into minute first instar larvae which tunneled into the petiole for further development. The larval period lasted for a month inside the petiole. The 3rd instar larvae pierced out through the base of the petioles and pupated among the roots of water hyacinth. The life span of the adults was 3-4 months. The pre-oviposition period was 3-4 days.

Table 1 Fecundity of *N. Bruchi* fed on water hyacinth plants from different bodies*

Place	Fecundity during			
	1st week	2nd week	3rd week	4th week
Laboratory pond (control)	285 ± 5.19	236 ± 8.83	128 ± 5.49	102 ± 2.86
Area 1 (Banjara hill)	286 ± 0.86	254 ± 4.10	243 ± 8.28	180 ± 6.73
Area 2	289 ± 5.92	286 ± 4.70	266 ± 4.67	212 ± 4.90

* Average of triplicate experiments: The number of pairs of insets studied was 5.

The number of eggs laid on the plants from control pond during the first two weeks of oviposition ranged from 7 to 10 eggs/female/day (table 1). Thereafter the number declined to 3–5 eggs/female/day. In the case of females from polluted water bodies the fecundity rate almost remained constant at 7–10 eggs/female even after three weeks of oviposition. Hence the total number of eggs was almost doubled in the plants collected from polluted water bodies.

Females of *N. bruchi* have a telotrophic type of ovary with trophocytes in the germarium region. The reproductive system comprised of a pair of ovaries, each of which is made up of two ovarioles. The two lateral oviducts of the two ovaries joined to form a common median oviduct that broadened posteriorly into the vagina (figure 1). The females from polluted water bodies showed an active phase of oocyte growth even after two weeks of the oviposition cycle in comparison to control females which showed declined activity (figures 2 and 3).

Our previous studies on the biochemical estimations of proteins and various inorganic minerals showed an increase in protein content as well as certain minerals⁹. Usually, fecundity of insects increases with increase of dietary proteins. The aminoacid and total sterol content of the leaves from polluted water showed an increase in comparison to control (table 2). Evidence regarding the role of sterols in adult nutrition is meagre. Sang King¹⁰

Table 2 Estimation of total aminoacid and total sterol content of the leaves from polluted and laboratory (control) water bodies

	Aminoacid/g dry wt. of the leaves (mg/g)	Sterol/g fresh wt. of the leaves (mg/g)
Laboratory pond (control)	1.8 ± 0.09	2.0 ± 0.01
Polluted water body	2.4 ± 0.09	5.4 ± 0.06

showed that fecundity of *D. melanogaster* decreased on a sterol free diet, but the female produced its normal number of eggs when supplemented with $4 \times 10^6\%$ cholesterol. Monroe¹¹ reported that cholesterol deficiency in *M. domestica* had no apparent effect on adult survival, ovarian growth or fecundity but caused nearly 80% reduction in egg hatch. The fecundity of *Leptinotarsa decemlineata* increased with increase in the lecithin content of host plant².

The investigation of mineral requirements in insect nutrition is a neglected area of research. However, there are reports of manganese, zinc and potassium that accelerated fecundity in various insects¹². The mineral analysis of water hyacinth leaves from polluted water bodies predominantly showed the presence of magnesium, aluminium and calcium. The increase in protein and sterol content



Figures 1–3. Reproductive system of: 1. One-day-old female showing telotrophic type of ovary; 2. Female from laboratory pond (during the third week of oviposition); 3. Female from polluted water body (during the third week of oviposition) [OV, ovariole; G, germarium; O, oocyte; LOD, lateral oviduct; CMD, common median oviduct].

of the leaves from polluted water bodies coupled with various inorganic minerals appeared to have resulted in an increased fecundity of the weevils.

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MULTIPLE SHOOT FORMATION IN EMBRYO CULTURE OF *SOLANUM MELONGENA*

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THE effectiveness of *in vitro* methods in improving crop plants makes them an attractive practical alternative to conventional techniques. For rapid *in vitro* multiplication of plants along with seedling explants, embryos too have been successfully used^{1,2}. In *Solanum melongena*, a vegetable crop plant regeneration from callus³⁻⁶, anthers^{7,8} and seedling explant^{9,10} was reported. This investigation outlines the formation of multiple shoots in embryo cultures of *S. melongena*, var. Pusa Purple Long (PPL) and further development into complete plantlets.

Wet seeds of *S. melongena* were surface-sterilized by 0.1% mercuric chloride for 5 min and washed repeatedly in sterilized distilled water. Excised embryos were cultured on Murashige and Skoog's (MS)¹¹ medium consisting of auxins and cytokinins. The pH of the medium was adjusted to 5.8 by 0.1% NaOH. All the cultures were kept under 16/8 photoperiod at 25 ± 2°C.

The growth and morphogenetic response of excised embryos (figure 1) to MS media supplemented with various growth regulators are represented in table 1. 2,4-D 0.5 mg/l in combination with Kn 1 mg/l or BAP 1 mg/l produced extensive, vigorous, friable, white-coloured callus all over the explant (figure 2), while it alone formed moderate callus. Root formation was predominant in MS media

Table 1 Morphogenetic response of embryos from seeds of *Solanum melongena* to MS medium with various growth regulators

Media (mg/l)	Morphogenetic response
MS + 2,4-D (1)	Callus
MS + NAA (1)	Callus + roots
MS + IAA (1)	Roots + single shoot
MS + Kn (1)	Callus + multiple shoots
MS + BAP (1)	Roots + multiple shoots*
MS + 2,4-D (0.5) + BAP (1)	Callus
MS + NAA (0.5) + BAP (1)	Callus + roots + single shoot
MS + IAA (0.5) + BAP (1)	Callus + roots + multiple shoots**
MS + 2,4-D (0.5) + Kn (1)	Callus
MS + NAA (0.5) + Kn (1)	Callus + single shoots

The data scored at the end of four weeks in culture of seven replicas, 2,4-D: Dichlorophenoxy acetic acid; NAA: Naphthalene acetic acid; IAA: Indole acetic acid; Kn: Kinetin; BAP: Benzylamine purine; * about 50%; ** more than 50%.