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Research Article

Role of Adjuvants in the Formulation

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ABSTRACT

The formulation used in the control of the crop pests such as *Helicoverpa armigera, Spodoptera litura* require one or more adjuvants depending upon their efficacy in the bio control. Several adjuvants were tested for the management of these pests mention above in different parts of the world. Many adjuvants were used in past 10 years but all the four adjuvants used in this formulation were found comparatively better over others in suppressing the population of *H. armigera* and *S. litura.* The opportunity to use EPN is promising because more than 90% of insect pests spend part of their life cycle in the soil. EPN have been formulated commercially in various carriers along with various adjuvants.

1. Introduction

Nematodes are found in almost all types of ecosystems and occur in unimaginable numbers in wide variety of shapes and sizes. They are termed based on habitat in which they are found like free living marine and freshwater, soil, saprophytes, parasitizing plant, microphagous or animals. Those nematodes which parasitize insects are regarded as Entomopathogenic nematodes (EPN). Entomopathogenic nematodes possess many attributes of an excellent biological control agent. EPN on the other hand are beneficial nematodes parasitizing crop insect pests, and are used as a bio pesticide agents a wide variety of insect pests. The impressive attributes of EPN have stimulated strong commercial interest in nematodes as biological insecticides and are perceived as viable alternative to chemicals in integrated pest management (IPM) programme. Desiccation significantly reduces the survival of nematodes and is one important factor affecting commercial use of the nematodes at every stage, from their mass production to application in the field. They possess many positive attributes including their broad host range, safety to non-target organisms and the environment, exemption from registration in many countries , ease of mass-production, ease of application, ability to search for pests, rapid host mortality , potential to recycle in the environment, and compatibility with most agricultural chemicals. These positive attributes and the need to find alternative methods of pest control to chemical insecticides have led to the rapid commercialization of the nematodes. Progress has been made that Entomopathogenic nematodes are now available commercially. Formulations have been made to enhance the activity of the EPN. Entomopathogenic nematodes (EPN) from the families Steinernematidae and Heterorhabditidae are lethal parasites of insects that are widely distributed in soils throughout the world (Gaugler 2002). Their ability actively to locate their insect hosts, specific association with highly virulent bacteria, high reproductive potential, the possibility of mass production and harmless impact on vertebrates and plants make these nematodes highly suitable for the development of environmentally friendly alternatives for the control of insect pests (Kaya & Gaugler 1993; Boemare 2002; Gaugler 2002). Various carriers, such as clay (Bedding, 1991), activated charcoal (Yakawa and Pitt, 1985), sponge, vermiculite, andpeat (Georgis, 1990), have been used to formulate infective juvenile nematodes. A formulation based on immobilization of nematodes in calcium alginate gel has also been developed (Georgis, 1990; Kaya and Nelsen, 1985).

2. Material and Methods

2.1 Preparation of samples:

To extract nematodes, first we took soil samples from different localities. We kept them separately in perforated plastics boxes. All the boxes were labeled with locality and date. Insect larvae of same size and age were picked from insect culture. We use *Helicoverpa armigera* (Hubner), *Corcyra cephalonica* or *Galleria mellonela* larvae for this purpose.

The samples were processed by Cobb's (1918)| sieving and decantation technique. About 500 cc soil was placed in a bucket and thoroughly mixed with a small amount of water. The debris and stones were removed and soil lumps, if present, were broken by hand. The bucket was then filled with water to about 3/4th of its volume and then the suspension was stirred to make it homogeneous. The bucket was left undisturbed for about ½ a minute to allow the heavy soil particles to settle at the bottom. The muddy suspension was then poured in to another bucket through a coarse sieve (2mm

pore size) which retained debris, roots and leaves. The suspension in the second bucket was then poured through a 300 mesh sieve (pore size 53 µm). The nematodes and find soil particles were retained on this sieve. The process was repeated thrice for better recovery of nematodes.

2.2 Isolation:-

The residue on the sieve on the sieve was collected into a beaker and poured on a small coarse sieve lined with tissue paper. The sieve was then placed on a Bearmann's funnel containing water sufficient to touch the bottom of the sieve and water level. The stem of the funnel was fitted with rubber tubing provided with a stopper. The nematodes migrated from the sieve into the clear water of the funnel and settled at the bottom. After about 24 hours a small amount of water was drawn from the funnel through the rubber tubing into a cavity block. The nematodes isolated as above were fixed and processed for mounting on slide.

2.2 **Nematode culture:**

The four potential strains of *Steinernema masoodi* were cultured in the fifth instar larvae of *G. mellonella* following the Dutky et al., (1964) technique. The infective juveniles were collected using White trap method (White, 1927) and were stored at 15ºC in BOD incubator for further analysis. The EPN suspension consisting of IJs stored in sterile distilled water was first examined under stereoscopic microscope to check the activity of the juveniles and diluted with a known quantity of sterile distilled water for making the suspension according to the required number of IJs. The EPN suspension consisting of IJs stored in sterile distilled water was first examined under stereoscopic microscope to check the activity of the juveniles and diluted with a known quantity of sterile distilled water for making the suspension according to the required number of IJs. Ten larvae of *Galleria mellonella* were placed on Whatman's filter paper glass petri plates together with IJs of EPN at 250, 500, 750, and 1000 IJs / petri plate. The treatments were replicated three times.

3. Results and Discussion

3.1 Methods for transforming the EPN into various Formulations

The formulation made with the adjuvants require, an EPN and an additive or adjuvant. Adjuvants are the material used to enhance the work of EPN; they can be anti-desiccant, phagostimulant & UV protectant etc. Different adjuvants were mixed in adequate amount in the liquid culture of EPN & formulations were made. These formulations then were tested for their effect on the survival & infectivity.

Table 1: Adjuvants used in the formulation

S. N	Adjuvants	Utility
1	Glycerin	Anti-desiccant
2	Sugar solution	Phagostimulant
3	Robin blue	UV protectant
4	Sodium bicarbonate	pH regulator

Various adjuvants at different concentration were combined with EPN & check for their effect. The main purpose of the additives used in the formulations has been to increase the survival and maintain the virulence of the EPNs.

Glycerin: It is an *anti-desiccant* which means it acts as a protective coating of the leaf or needle of the plant, substantially reducing water loss during high periods of stress. Anti-desiccants, also called anti-transpirants, are sprays that provide a protective coating to evergreen foliage that reduces the amount of water that escapes.

Sugar solution: It is a phagostimulant which is used for even spray of the formulation on foliage. The phagostimulant is preferably selected from a carbohydrate and/or and amino acid. The application also discloses a method of controlling or reducing populations of pest insects. This comprises the application of an effective amount of a preparation comprising an EPN associated with a phagostimulant and all other adjuvants. The experiment was conducted to measure the increase of larval mortality following the addition of sugar solution. The sugar solution is easily available made easily with water and sugar.

Sodium bicarbonate: It is used as pH regulator thus regulates formation of malic acid on the foliage.

Robin blue: It is a fabric whitener and hence used as ultraviolet protectant. Addition of glycerin as anti-desiccants, Robin blue as UV protectant, sugar solution as phagostimulant and sodium bicarbonate as pH regulator they all together prolonged the activity of EPN IJs and their survival on leaves of the leguminous crops, Nickel and Shapiro 1992 has also emphasize that usefulness of UV retardant in their study.

3.2 Formulation applied:

Liquid suspension contained EPN IJs, 1% glycerin, 0.01% robin blue, sugar solution and 0.5% sodium bicarbonate. Three nematode applications were made at 10-days interval during fruiting and podding stage using hand sprayer in crop field. There were three treatments having 3 replications in each.

3.3 Observation recorded:

In the experiments, Formulation enhances the work of EPN so strongly that it decreases the pod damage and the increases the yield. It gave good results. As the treatment with EPN, glycerin, sugar solution, robin blue & sodium bicarbonate with the different concentration the mortality recorded increased to 94%.

4. Conclusion:

Formulation must be used as all together they are nontoxic, eco-friendly and causes no harm to the crop thus control the pests to a large extent. EPN along with adjuvants become much more effective comparative to EPN alone. Adjuvants play the vital role in protecting the foliage, increase the yield & decrease the pod damage. Together with the adjuvants EPN proves to be the best biopesticides among all as they enhance the moisture, prevent desiccation and control the pH and UV radiations. When these formulations were applied in the fields of leguminous crops it showed the great difference in the yield. Thus, these adjuvants can be used in future for large scale control of insect pests.

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Competing Interests

The authors have declared that no competing interests exist.

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