

THE ABUNDANCE OF THE ENTOMOPATHOGENIC NEMATODES STEINERNEMA CARPOCAPSAE ON VARIOUS CULTURE MEDIA

by Dyah Indriyanti

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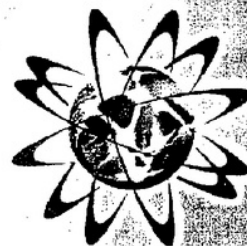
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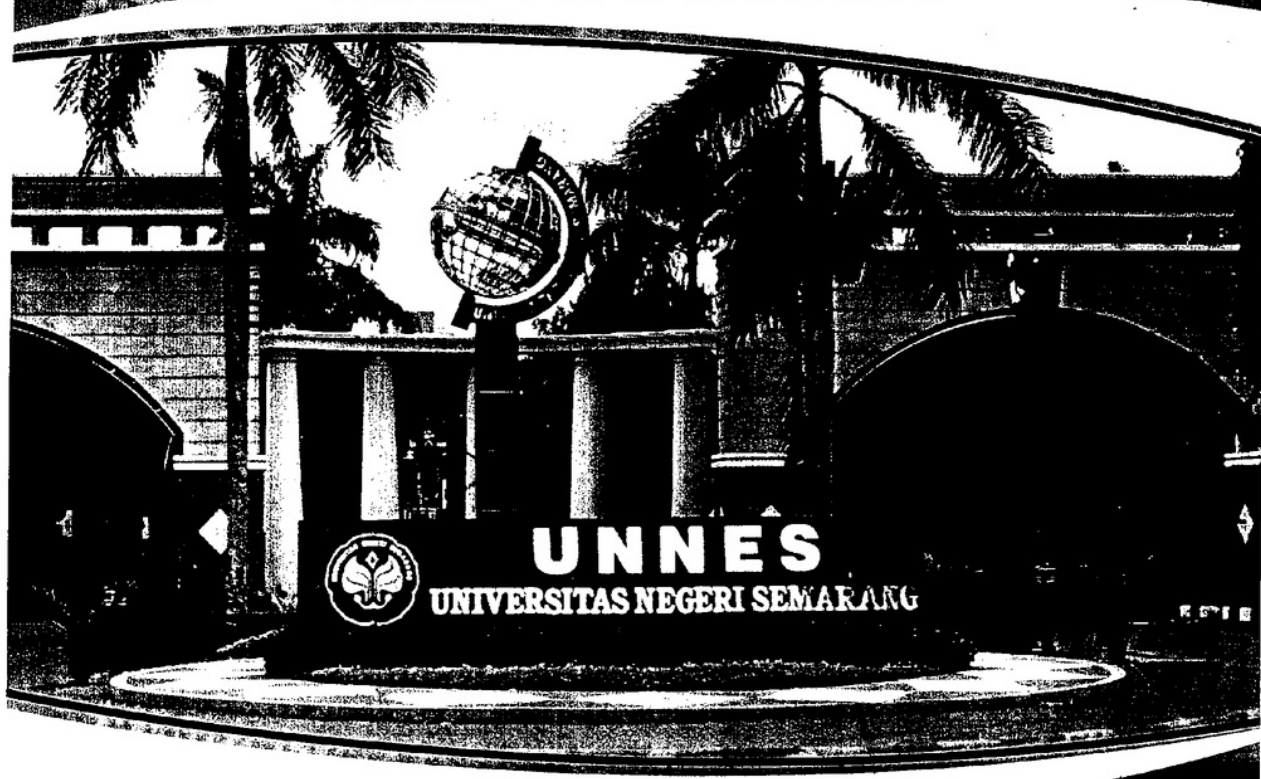
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PROCEEDING
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AND EDUCATION



Theme :

Optimizing The Role of Science and Science Education
in Global Cooperation

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PREFACE

Thanks to God Almighty this International Conference Proceeding could be completed. All articles in this proceeding are presented in International Conference On Mathematics, Science, and Education – Optimizing The Role of Science and Science Education in Global Cooperation on September 19-21st, 2014 at Patra Jasa Semarang Convention Hotel. This Conference is organized by Faculty of Mathematics and Natural Science. This proceeding has been reviewed of Mathematics and Science experts before it is published.

This conference is designed to improve the discussion and research scope in mathematics, science, and education area in the international level. Sub topics in this proceeding cover mathematics, applied mathematics, and mathematics education in accelerating character building. Enhancing biology and biology education research for a better life. Green chemistry in research and education. Physics and physics education for trending research.

Hopefully this publication of proceeding will be profitable for all of us.

Semarang, 3 December 2014

Regards
Committee of ICMSE 2014

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MESSAGE FROM THE DEAN OF FMIPA UNNES

Dear Participants of ICMSE 2014,

It is a pleasure to welcome all of you in the first International Conference on Mathematics and Science Educations (ICMSE 2014) held by Faculty of Mathematics and Natural Sciences, Semarang State University.

Faculty of Mathematics and Natural Science Semarang State University or more popularly known as FMIPA Unnes has 6 departments and 11 study programs of Mathematics and Natural Sciences education backgrounds and non education backgrounds. FMIPA Unnes has the mission of being an excellent and meaningful faculty by improving human resources through scientific activity.

One of efforts to result excellent and meaningful human resources through scientific activity is by performing discussion and knowledge sharing. To widen discussion of science and research development in mathematics and science educations scopes in national and international level, ICMSE 2014 was initiated as the medium of that discussion. I believe that ICMSE 2014 as the first international conference held by FMIPA Unnes can facilitate the knowledge sharing in mathematics and science educations area in order to establish a global cooperation among experts and researchers.

With the hope that this conference will be the medium to optimize the role of Mathematics, Science and Education in global cooperation, I am proud to welcome all of you and I wish you a pleasant sharing and discussion in this conference and enjoyable stay in Semarang, Indonesia.

Prof. Dr. Wiyanto, M.Si.
Dean of Faculty of Mathematics and Natural Sciences
Semarang State University

MESSAGE FROM CONFERENCE CHAIRMAN

My pleasure, welcome to you today on the occasion of this International Conference on Mathematics, Science, and Education (ICMSE 2014). I would like to extend my warmest welcome to all of the distinguished participants, especially those who have travelled long distances to be present here. This conference has already established itself as a key event to offer various thoughts and knowledge in enhancing our understanding in fundamental sciences and education.

This conference focus on "Optimizing The Role of Science and Science Education in Global Cooperation", offers all of us the opportunity to explore exciting information. The aim of the conference is to provide an interdisciplinary forum for scientist engaged in the full spectrum of research and development activities. The meeting intends to bring together researchers, scientists, and scholars to exchange and share their experiences, new ideas, and research result in related fields and discuss the practical challenges encountered and the solutions adopted. I invite all of you to approach this year's events to take advantage of the many ways in which you too might explore the unfamiliar - and discover a great deal in the process.

First, the various sessions that have been organized for the next day promise exciting revelation for all who attend them. Each speakers who are experts in their respective fields, will address a major topic or issue related to Fundamental Sciences,. You might learn more about a topic with which you were already familiar; or you might also find yourself discovering a whole new world of ideas and information you didn't know existed. Either way, you'll have many opportunities to explore fascinating new terrain with these reputable speakers.

Second, the key note speakers will provide, for all of us, an important window into the world of the future. We are privileged to have them as our key note speakers Prof. Barke, Munster University Germany, Prof. Martin Stein, Munster University Germany, Prof. Simone Krees, Munster University Germany, Prof. Matthias Ludwig, University Frankfurt Germany, Prof. Van Horsen, Delf Univesity Netherland, Prof. Rahim Sahar, UTM Malaysia and Dr. Margareta Rahayuningsih, M.Si experience has taken them through the whole cycle of Life and General science.

Finally, as you attend these various events, keep in mind that other people can also serve as doorways to new worlds. Hearing of someone else's background and experiences can often make for fascinating discoveries that can educate and profoundly affect us. So take advantage of this rare gathering of hundreds of people working in various fields to meet one another, talk with one another, and learn from one another.

In conclusion, I hope that you will find your time with us exciting. We have a great agenda for you with esteemed speaker and presenters from our profession. I do hope you will enjoy the next couple of days. I would like to once again extend my gratitude to all the participants, generous sponsor and I look forward to a most successful and fruitful conference.

Professor Dr. YL.Sukestiyarno
Chairman of ICMSE 2014

INTERNATIONAL CONFERENCE ON MATHEMATICS, SCIENCE, AND EDUCATION
"Optimizing The Role of Science and Science Education in Global Cooperation"

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THE ABUNDANCE OF THE ENTOMOPATHOGENIC NEMATODES STEINERNEMA CARPOCAPSAE ON VARIOUS CULTURE MEDIA

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ABSTRACT

The aim of the study was analyzing the abundance of entomopathogenic nematodes (ENPs) *Steinernema carpocapsae* on various artificial culture media. Infective juveniles of *S. carpocapsa* (1200 IJs/ml) were reared on seven different culture media for four weeks. Media A contained 2 gram (g) yeast extract, 0.2 g agar and 30 ml water. Media B contained 2 g egg yolk, 0.2 g agar and 30 ml water. Media C contained 2 g chicken intestine, 0.2 g agar and 30 ml water. Media D contained 0.5 g yeast extract, 1.5 g egg yolk, 0.2 g agar and 30 ml water. Media E contained 0.5 g yeast extract, 1.5g chicken intestine, 0.2 g agar and 30 ml water. Media F contained 1.5 g egg yolk, 0.5 chicken intestine, 0.2 g agar and 30 ml water. Media G contained 0.5 g yeast extract, 1 g egg yolk, 0.5 g chicken intestine, 0.2 g agar and 30 ml water. *S. carpocapsa* population from each media were calculated every week. The experiment design used a completely random sampling, seven treatments and five replicates. Result show that the average abundance population infective juveniles nematode (IJs/ml) rearing from medium B, D, F and G increased on the second week after inoculation, yielded 31296, 20400, 24876 and 14568 IJs/ml respectively. The population of ENPs decreased on the third till fourth weeks inoculation on all culture media. The highest population of ENPs happened on the egg yolk culture media for four weeks i.e. 7236 IJs/ml.

Keywords: Entomopathogenic nematodes; rearing; culture medium

INTRODUCTION

Entomopathogenic nematodes (ENPs) are small round worms that can be used as natural biopesticides. Unlike plant parasitic nematodes, which can be serious crop pests, the entomopathogenic nematodes only kill insects. Entomopathogenic nematodes used in biological pest control kill insects with the help of symbiotic bacteria (Mile et al 2012). ENPs has emerged as excellent candidate for biological control of insect pest (Somwong & Petcharat 2012). ENPs is one of the biological control agents for insect pests. They have several advantages that very quickly killed the host, wide host range, actively looking for a host that is effective for controlling insects (Wartono and Priatno 2009). The Problems in the field was mass rearing of ENPs. The application of ENPs in the field need a lot of ENPs

population, so culture ENPs on the artificial media is important.

According to Mulyaningsih (2010), ENPs can be produced in vivo and in vitro culture. Culture of Nematode in vivo performed using insect larvae, while in vitro is done on artificial media. Artificial media sought are easily available and cheap.

The aim of the study was analyzing the abundance of entomopathogenic nematodes *Steinernema carpocapsae* on various artificial culture media.

METHODS

Seven artificial culture media were used in rearing of *S. carpocapsae* (5 replications for each media). *S. carpocapsa* were reared on seven different culture media for four weeks. The media culture composition as follows:

Table 1: Composition of Various culture media of ENPs

Composition Treatment	Yeast extract	Egg yolk	Chicken intestine (powder)	Agar	Water (Aquades)
Yeast extract (A)	2 g	-	-	0.2 g	30 ml
Egg yolk (B)	-	2 g	-	0.2 g	30 ml
Chicken intestine (C)	-	-	2 g	0.2 g	30 ml
Yeast extract + egg yolk (D)	0.5 g	1.5 g	-	0.2 g	30 ml
Yeast extract + chicken intestine (E)	0.5 g	-	1.5 g	0.2 g	30 ml
Egg yolk+ chicken intestine (F)	-	1.5 g	0.5 g	0.2 g	30 ml
Yeast extract + egg yolk + chicken intestine (G)	0.5 g	1 g	0.5 g	0.2 g	30 ml

Remarks

1. The composition of yeast extract media was modified from Lunau et al. in Chaerani et al. (2012).
2. The composition of egg yolk media was modified from Han et al. in Chaerani et al. (2012).
3. The Composition of chicken intestines media was modified from Bedding et al. (2012) in Chaerani (2012).

The in vitro culture media of each treatment was mixed evenly with blender.

Each of culture media puts in bottles sized 6x10 cm, and absorbed into pieces of sponge sized 6x1 cm. The sponge was placed on the base of the bottle. The media were autoclaved at 121°C and 15 lbs for 30 min. After the media was cooled, 1 ml of water containing *S. carpocapsa* suspension (1200 IJs) was inoculated into each media. The nematodes was taken from Biopesticides "Coleonema" production by Nemadic. The ENPs culture media were incubated at Laboratory

27-30 °C, RH 65-87% for four weeks (28 days). The total number of nematodes produced per week was counted and the data recorded was analyzed of variance.

RESULTS AND DISCUSSION

The mean number of ENPs populations on a various culture media for weekly were presented in Figure 1-7.

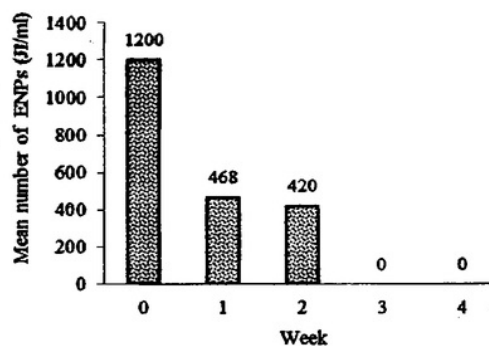


Figure-1. Comparison of *S. carpocapsa* population on extract yeast media culture (A) for four weeks

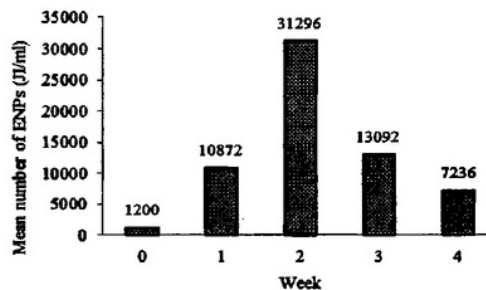


Figure-2. Comparison of *S. carpocapsa* population on egg yolk culture media (B) for four weeks

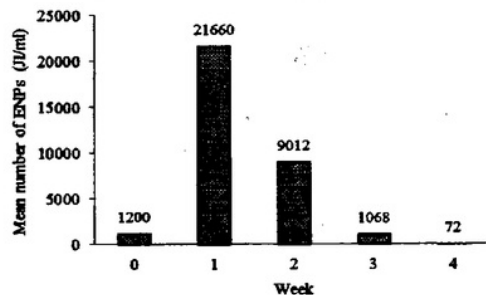


Figure-3. Comparison of *S. carpocapsa* population on chicken intestine culture media (C) for four weeks

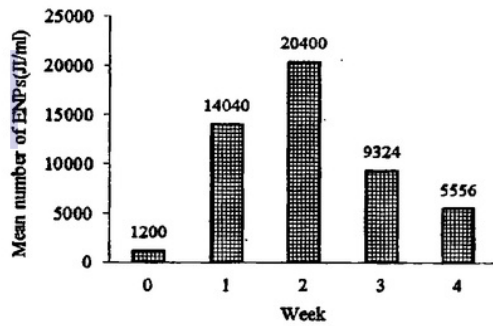


Figure-4. Comparison of *S. carpocapsa* population on mixture yeast extract and egg yolk culture media (D) for four weeks

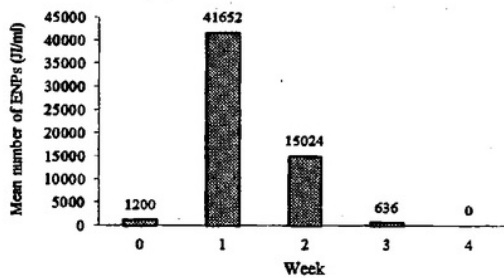


Figure-5. Comparison of *S. carpocapsa* population on mixture yeast extract and chicken intestine culture media (E) for four weeks

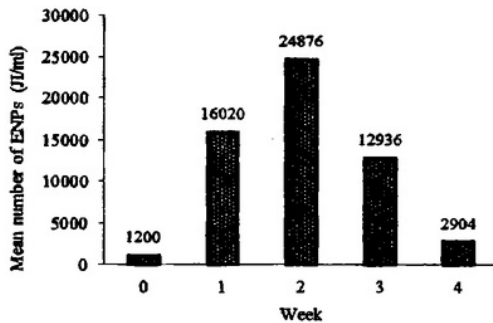


Figure-6. Comparison of *S. carpocapsa* population on mixture egg yolk and chicken intestine culture media (F) for four weeks

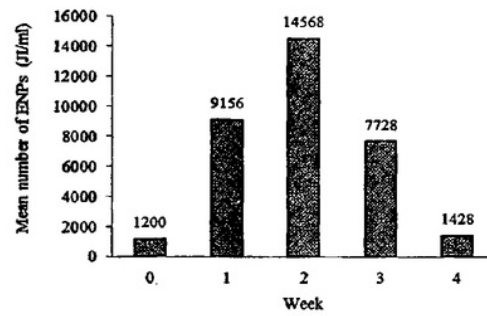


Figure-7. Comparison of *S. carpocapsa* population on mixture yeast extract, egg yolk and chicken intestine culture media (G) for four weeks

Data from figure 1-7 show that population ENPs from six culture medias have increased for the first week. The population of the ENPs increased due to the medium contain of a source of nutrients such as carbohydrates, proteins and lipids, the ENPs in adapting to reproduce (Shapiro & Gaugler 2002).

The population ENPs increased on the second week after inoculation, there were four medias i.e egg yolk culture medium (31296 IJs/ml), mixture yeast extract and egg yolk culture medium (20400 IJs/ml), mixture egg yolk and chicken intestine culture medium (24876 IJs/ml), mixture yeast extract, egg yolk and chicken intestine culture medium (14568 IJs/ml). The increasing of *S. carpocapsa* population due to they could adapted and taken the nutrients from medium to breeding.

ENPs population data among the medias on the fourth week were analyzed one-way ANOVA test. Results of one-way ANOVA test with a level of 5% was obtained value of $F = 10,168$; $df = 34$; $Sig. = 00$. Value $Sig. P < 0.05$, it means that the population of the ENPs on various culture medias on the fourth week significantly different.

The nematode yields on the fourth weeks, show that Nematodes reared in egg yolk medium produced highest population ENPs than other medias. The egg yolk media was the best medium that can be used for ENPs culture media. Source of nutrients such as carbohydrates, proteins and lipids in egg yolk media contained 44.8% water, 17.7% protein, 35.2% lipid , 1.1 % carbohydrat and 1.2% ash (Romanoff & Romanoff 1993). While Extract yeast contained 50-52% protein, 30-37% carbohydrat and 4-5% lipid and 7-8% mineral (Reed 1991 in Ahmad 2005). Chicken intestine contained of 65.90% collagen protein, 22.93% crude protein, crude

lipid 5.60%, 3:44% ash content, 6.68% 2:03% minerals and other materials (Baihaki et al. 2010). Egg yolk proved suitable for ENPs culture media due to it contained high lipid than others. According to Somwong & Petcharat (2012), lipids are necessary for development and reproduction.

CONCLUSION

The population of ENPs generally breeding increased in the first week until the second on the artificial media. The highest population of ENPs happened on the egg yolk culture media for four weeks i.e. 7236 IJs/ml.

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