

**A STUDY OF EFFECTS OF SEED EXTRACTS OF SOME PLANTS ON  
DEVELOPMENT OF LARVAE, ADULT OVIPOSITION AND HATCHING  
OF EGGS OF *Aedes aegypti* L.**

**Oluwasola Adebimpe ADEPOJU**

**B. Sc. (Hons) Ife.**

**SCP12/13/R/0035**

**A THESIS SUBMITTED TO THE INSTITUTE OF ECOLOGY AND  
ENVIRONMENTAL STUDIES, OBAFEMI AWOLOWO UNIVERSITY, ILE-IFE,  
NIGERIA IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD  
OF THE DEGREE OF MASTER OF SCIENCE (M. Sc.) IN ENVIRONMENTAL  
CONTROL AND MANAGEMENT**

2016

**AUTHORIZATION TO COPY****OBAFEMI AWOLOWO UNIVERSITY****HEZEKIAL OLUWASANMI LIBRARY****POSTGRADUATE THESIS**

**Author:** Oluwasola Adebimpe ADEPOJU

**Title:** A Study of Effects of Seed Extracts of Some Plants on the Development of Larvae, Adult Oviposition and Hatching of Eggs of *Aedes aegypti* L.

**Degree:** M. Sc. (Environmental Control and Management)

**Year:** 2016

I, Oluwasola Adebimpe ADEPOJU, hereby authorize the Hezekial Oluwasanmi Library to copy my thesis in part or whole in response to request from individuals or organizations for the purpose of private study or research.

---

**ADEPOJU, O. A.**

---

**Date**

**CERTIFICATION**

We certify that this work was carried out by ADEPOJU Oluwasola Adebimpe, (SCP12/13/R/0035) in partial fulfillment of the requirements for the award of the degree of Master of Science (M. Sc.) in Environmental Control and Management, Institute of Ecology and Environmental Studies, Obafemi Awolowo University, Ile – Ife, Nigeria.

---

**Supervisor**  
**Prof. W. A. Muse**  
**Department of Zoology**

---

**Date**

---

**Prof. O. O. Awotoye**  
**Director,**  
**Institute of Ecology and Environmental Studies**

---

**Date**

## DEDICATION

This project is dedicated to Almighty God, the Alpha and Omega and to my parents Dn. and Mrs S. A. Adepoju. Your love throughout the years has greatly enriched my life.

OBAFEMI AWOLOWO UNIVERSITY

## **ACKNOWLEDGEMENT**

I give thanks to God Almighty through Jesus Christ my Lord for being my source of strength and sustenance during this work and in my entire life endeavour. Unto him be all the glory, majesty, honour and praise for ever (Amen).

My sincere gratitude goes to my project supervisor Prof. W. A. Muse for his advice support; guidance and patience that made this work a reality.

I am also very grateful to the Director of the institute; Prof. O. O. Awotoye and I will also like to appreciate all the member of staff of the department of Institute of Ecology and Environmental studies (Academics and non – academics) especially Dr. M. B. Adewole and Dr. O. J. Matthew and also to Mrs Famuyiwa (Department of Pharmacognosy, Ile-Ife).

To my parents, I really appreciate your moral, financial support and your prayers day and night for the success of this work. I cannot but knowledge my siblings Oluwasegun, Adekunbi, Olumuyiwa Adepoju and my beloved niece Ojo Bolaji for their support and contributions.

My special thanks also go to my colleagues who started this M. Sc. programme with me: Akintoye Akinbiyi and Jumoke Oguntoye for their encouragement during the course of study and my appreciation also go to Mr. Andrew Ajayi and my friend Adetutu Omotosho.

**ADEPOJU OLUWASOLA ADEBIMPE**

2016

**TABLE OF CONTENTS**

<b>Title</b>	<b>Page</b>
Title Page	i
Authorization	ii
Certification	iii
Dedication	iv
Acknowledgements	v
Table of Contents	vi
List of Plates	x
List of Table	xi
List of Figures	xiii
Abstracts	xiv
<b>CHAPTER ONE: INTRODUCTION</b>	
1.1 <i>Aedes aegypti</i>	1
1.2 Phytochemicals	2
1.3 <i>Moringa oleifera</i>	3
1.4 <i>Citrus aurantifolia</i>	4
1.5 <i>Carica papaya</i>	4



1.6	Statement of Research	5
1.7	Justification of the Study	6
1.8	Objectives of Research	6

## CHAPTER TWO: LITERATURE REVIEW

2.1	<i>Aedes aegypti</i>	7
2.1.1	Oviposition Behaviour of <i>A. aegypti</i>	8
2.1.2	Life Cycle of <i>A. aegypti</i>	9
2.1.3	Distribution of <i>A. aegypti</i>	12
2.1.4	Resistance of Mosquitoes to Insecticides	12
2.2	<i>Moringa oleifera</i>	15
2.2.1	Importance of <i>M. Oleifera</i>	15
2.3	<i>Citrus aurantifolia</i>	19
2.3.1	Benefits of Lime	19
2.4	<i>Carica papaya</i>	23
2.4.1	The Importance of <i>Carica papaya</i>	23
2.5	Phytochemical Properties of Plant Extracts	27

## CHAPTER THREE: MATERIALS AND METHODS



3.1	Plant Seed Collection and Preparation	32
3.2	Preparation of Plant Extracts	32
3.3	Rearing of <i>Aedes aegypti</i>	33
3.4	Larval Development	34
3.5	Oviposition	34
3.6	Hatching of Eggs	35
3.7	Statistical Analysis	35

#### **CHAPTER FOUR: RESULTS**

4.1	Larval Development	36
4.2	Oviposition	53
4.3	Egg Hatching	59
4.4	Comparative Effects of Boiled and Dissolved Plant Extracts	64

#### **CHAPTER FIVE: DISCUSSION**

Discussion	72
------------	----

#### **CHAPTER SIX: SUMMARY, CONCLUSION AND RECOMMENDATION**

Summary	76
Conclusion	77



Recommendation	78
References	79
Appendices	95

OBAFEMI AWOLowo UNIVERSITY

## LISTS OF PLATES

Plate	Description	Page
1.	Life cycle of <i>A. aegypti</i>	11
2.	Adult male of <i>A. aegypti</i>	14
3.	Dried peeled seeds of <i>M. oleifera</i>	18
4.	Dried seeds of <i>C. aurantifolia</i>	22
5.	Dried seeds of <i>C. papaya</i>	26

## LIST OF TABLES

Table	Description	Page
1.	Effect of dissolved plant extract of <i>Moringa oleifera</i> on the mean survival rates of 2 <sup>nd</sup> instar larvae of <i>A. aegypti</i> maintained in different concentrations of extracts.	38
2.	Effect of boiled plant extract of <i>Moringa oleifera</i> on the mean survival rates of 2 <sup>nd</sup> instar larvae of <i>A. aegypti</i> maintained in different concentrations of extracts.	40
3.	Effect of dissolved plant extract of <i>Citrus aurantifolia</i> on the mean survival rates of 2 <sup>nd</sup> instar larvae of <i>A. aegypti</i> maintained in different concentrations of extracts.	44
4.	Effect of boiled plant extract of <i>Citrus aurantifolia</i> on the mean survival rates of 2 <sup>nd</sup> instar larvae of <i>A. aegypti</i> maintained in different concentrations of extracts.	46
5.	Effect of dissolved plant extract of <i>Carica papaya</i> on the mean survival rates of 2 <sup>nd</sup> instar larvae of <i>A. aegypti</i> maintained in different concentrations of extracts.	49
6.	Effect of boiled plant extract of <i>Carica papaya</i> on the mean survival rates of 2 <sup>nd</sup> instar larvae of <i>A. aegypti</i> maintained in different concentrations of extracts.	51

7. Effects of different concentrations of dissolved plant extracts on oviposition of female *A. aegypti* 55
8. Effects of different concentrations of boiled plant extracts on oviposition of female *A. aegypti* 57
9. Effect of different concentrations of dissolved plant extracts on the hatching of *A. aegypti* eggs 60
10. Effect of different concentrations of dissolved plant extracts on the hatching of *A. aegypti* eggs 62
11. Comparative effects of 12 gL<sup>-1</sup> dissolved and boiled extracts of *M. oleifera*, *C. aurantifolia* and *C. papaya* on adult emergence survival, oviposition and egg hatching of *A. aegypti*. 71

## LIST OF FIGURES

Table	Description	Page
1.	Comparative effects of 8 gL <sup>-1</sup> dissolved extracts of <i>M. oleifera</i> , <i>C. aurantifolia</i> and <i>C. papaya</i> on the survival rate of <i>A. aegypti</i>	65
2.	Comparative effects of 8 gL <sup>-1</sup> boiled extracts of <i>M. oleifera</i> , <i>C. aurantifolia</i> and <i>C. papaya</i> on the survival rate of <i>A. aegypti</i>	66
3.	Comparative effect of 8 gL <sup>-1</sup> dissolved extracts of <i>M. oleifera</i> , <i>C. aurantifolia</i> and <i>C. papaya</i> on oviposition of <i>A. aegypti</i>	68
4.	Comparative effect of 8 gL <sup>-1</sup> dissolved extracts of <i>M. oleifera</i> , <i>C. aurantifolia</i> and <i>C. papaya</i> on oviposition of <i>A. aegypti</i>	69

## ABSTRACT

The study determined larval development of *Aedes aegypti* in the boiled and dissolved extracts of *Moringa oleifera*, *Citrus aurantifolia* and *Carica papaya* and the effects of these plant extracts on oviposition of *A. aegypti*. It also assessed eggs hatchability of *A. aegypti* in the extracts and compared the effectiveness of boiled and dissolved extracts of the three plant materials with a view to providing information on the effectiveness of plant extracts in biological control of mosquitoes.

Eggs of *A. aegypti* were obtained from Department of Pharmacognosy, Obafemi Awolowo University, Ile-Ife. Larvae were fed with edible yeast until adult emergence and adults were fed with glucose and blood meal. The eggs laid by the females were collected, reared and used for main experiment. The fruits of *M. oleifera*, *C. aurantifolia* and *C. papaya* were purchased from a local market in Ile-Ife and the fruits were identified and authenticated at IFE Herbarium. Each of the three seeds was ground into powder with a blender. The powdered seeds were measured out in 4, 8 and 12 g and were dissolved in 2 L of distilled cold water for 24, 48 and 72 hours respectively. The same weights of powdered seeds were also put in 2 L of distilled cold water and boiled for 5, 10 and 15 minutes respectively. Each of the mixture was filtered using muslin cloth to obtain clear solutions that were stored in labelled bottles used for subsequent experiments. Data obtained were analyzed using One-way ANOVA and Students' T-test.

Dissolved extracts of *M. oleifera*, *C. aurantifolia* and *C. papaya* at 24, 48 and 72 hours and in 4, 8 and 12 gL<sup>-1</sup> concentrations of extracts affected the survival of *A. aegypti* at various developmental stages. *M. oleifera* was the most effective causing significant reduction in adult emergence at 4 and 8 gL<sup>-1</sup> concentrations with no emergence of adults at 12 gL<sup>-1</sup> in extracts dissolved for 24, 48 and 72

hours respectively compared with *C. aurantifolia* at the same concentrations with adult emergence of  $13.00 \pm 3.28$ ,  $5.00 \pm 2.00$  and  $0.00 \pm 0.00$  followed by *C. papaya* with  $13.33 \pm 1.52$ ,  $11.67 \pm 4.16$  and  $3.33 \pm 3.05$  respectively. The effects of boiled extracts of *M. oleifera*, *C. aurantifolia* and *C. papaya* were concentration dependent and also increased with increase in period of boiling from 5 - 15 minutes. Number of eggs laid decreased with increase in periods of boiling and duration of dissolved extracts and was also concentration dependent in the three plant seed extracts from 4 - 12 gL<sup>-1</sup> respectively. The eggs deposited in the dissolved extracts were higher than those deposited in the boiled extracts. *M. oleifera* had the lowest number of eggs in 12 gL<sup>-1</sup> at 48 hours dissolved extract with  $9.00 \pm 1.68$  eggs and  $6.33 \pm 3.33$  eggs in 12gL<sup>-1</sup> of 15 minutes boiled extract compared with *C. aurantifolia* ( $55.75 \pm 4.00$  and  $19.67 \pm 2.61$  eggs) and *C. Papaya* ( $78.33 \pm 4.67$  and  $44.67 \pm 1.33$  eggs). There was hatching in the boiled and dissolved extracts of *M. oleifera*, *C. aurantifolia* and *C. papaya* at 4 -12 gL<sup>-1</sup> at various concentrations.

The study concluded that boiled extracts of *M. oleifera*, *C. papaya* and *C. aurantifolia* at different concentrations were more effective than the dissolved extracts in inhibiting the development of larvae and pupae of *A. aegypti*. There was also reduction in oviposition of gravid females of *A. aegypti* in the boiled and dissolved plant extracts. *M. oleifera* was found be the most effective among the three plant materials in controlling the mosquito.

## CHAPTER ONE

### INTRODUCTION

#### 1.1 *Aedes aegypti*

Insects are of importance from public health and agricultural points of view in transmitting diseases while others are pests of stored and field crops (Collins and Paskewitz, 1995). Among species of insects, mosquitoes are very important because they transmit a number of diseases such as malaria, filariasis, dengue, Japanese encephalitis. (Jaswanth *et al.*, 2002). *A. aegypti*, is the primary carrier for viruses that cause dengue fever, dengue hemorrhagic fever, yellow fever, Zika virus and chikungunya fever (Yang *et al.*, 2009).

Mosquito-borne diseases have an economic impact, including loss in commercial and labor outputs, particularly in countries with tropical and subtropical climates (Fradin and Day, 2002). Dengue fever has also increased four-fold since 1970 affecting nearly half the world's population (Gubler, 1998). In 1990, about 1.5 billion people lived in regions where the estimated risk of dengue transmission was greater than fifty percent (Hales *et al.*, 2002). Despite its debilitating effects, lymphatic filariasis is given a very low control priority (Ramaiah *et al.*, 2000). In the Western Pacific and Southeast Asia, *Aedes albopictus* has played a secondary role in transmitting dengue viruses during outbreaks (Gubler, 1998; Guzman and Kouri, 2002; Effler *et al.*, 2003).

Majority of *A. aegypti* breeding places is primarily in man-made containers, including earthenware jars, concrete cisterns, ant traps containers, and other items that collect rainwater nearby houses ( Swaddiwudhipong *et al.*, 1992; Chansaeng *et al.*, 1993; Kittayapong and



Strickman, 1993; Chareonviriyaphap *et al.*, 2003a). *A. aegypti* prefers to rest inside houses, typically in sheltered places such as dark corners, undersides of furniture, hanging objects such as clothes and curtains, and on dark walls (Reiter *et al.*, 1992; WHO, 1999; Scott *et al.*, 2000a).

## 1.2 Phytochemicals

Phytochemicals are botanicals which are naturally occurring insecticides obtained from floral resources. Applications of phytochemicals in mosquito control were in use since the 1920s (Shahi *et al.*, 2010) but the discovery of synthetic insecticides such as DDT in 1939 side-tracked the application of phytochemicals in mosquito control programme. After facing several problems due to over application of synthetic insecticides in nature, there was renewed interest on phytochemicals that are easily biodegradable and have no ill-effects on non-target organisms. Since then, the search for new bioactive compounds from the plant kingdom and an effort to determine its structure and commercial production has been initiated. At present phytochemicals make up to one 1 per cent of world's pesticide market (Isman, 1997).

Several groups of phytochemicals such as alkaloids, steroids, terpenoids, essential oils and phenolics from different plants have been reported previously for their insecticidal activities (Shahi *et al.*, 2010). Insecticidal effects of plant extracts vary not only according to plant species, mosquito species, geographical varieties and parts used, but also due to extraction method adopted and the polarity of the solvents used during extraction.

The efficacy of phytochemicals against mosquito larvae vary significantly depending on plant species, plant parts used, age of plant parts, solvent used during extraction as well as upon the available vector species. Sukumar *et al.* (1991) have described the existence of variations in the

level of effectiveness of phytochemical compounds on target mosquito species *vis-à-vis* plant parts from which these were extracted.

### 1.3 *Moringa oleifera*

*Moringa oleifera* is the most widely cultivated plant species worldwide (Santos *et al.*, 2009) originating from western and sub-Himalayan region, parts of Asia and Africa (Mughal *et al.*, 1999). Its occurrence has thereafter spread to Cambodia, Philippines, Caribbean Islands and Central North and South America (Morton, 1991). The seeds of the plant exhibit anti-inflammatory, anti-tumor effects (Cáceres *et al.*, 1992) and antimicrobial, antispasmodic and diuretic properties (Babu and Murugan, 1998).

*M. oleifera* is the most nutrient-rich plant yet discovered. *M. oleifera* provides a rich and rare combination of nutrients, amino acids, antioxidants, antiaging and anti-inflammatory properties used for nutrition and healing (Fuglie, 1999). *M. oleifera* is a miracle tree with a great indigenous source of highly digestible proteins, Ca, Fe and Vitamin C. It contains all the essential nutritional elements that are essential for livestock and human beings as well (Fahey, 2005).