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Title:

Histological Study of Mosquito Larvae (*Aedes caspius*) Treated with Essential Oils of *Thymus serpyllum* and *Origanum majorana* in the El Oued and El Kala Region.

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الشكر والعرفان

بسم الله الرحمن الرحيم

إلى من ساندوني خلال رحلتي العلمية، وكانوا نورًا لطريقي، أُهدي ثمرة جهدي المتواضع...
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بسم الله الرحمن الرحيم

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Abstract

The mosquito *Aedes caspius* poses a health threat in many areas, including Algeria, where it is widely spread and causes epidemics of vector-borne diseases such as malaria and dengue fever. This study aims to evaluate the efficacy of essential oils extracted from Thyme (*Thymus serpyllum*) and marjoram (*Origanum majorana*) in controlling *Aedes caspius* larvae. Essential oils were extracted using a Clevenger apparatus, with the yield of thyme oil being 1.10% and marjoram oil 0.74%. Toxicity tests were conducted on larvae at different concentrations for 24, 48, and 72 hours. The results showed that both oils had a toxic effect on mosquito larvae, with mortality rates increasing with higher concentrations over the different time periods. The lethal concentrations (LC50 and LC90) of the oils were as follows: marjoram oil (LC50 = 175 µL/L, LC75 = 268.71 µL/L, LC90 = 325 µL/L) and thyme oil (LC50 = 39.62 µL/L, LC75 = 145.02 µL/L, LC90 = 208.26 µL/L). Histological examination revealed faster and stronger destruction of the midgut epithelium and cytoplasmic cells in the larvae treated with thyme oil compared to those treated with marjoram oil. The results suggest that *Thymus serpyllum* oil is more effective in eliminating *Aedes caspius* larvae than *marjoram* oil, supporting the potential use of both oils as natural alternatives for mosquito control.

Key words:

Essential oils - *Aedes caspius* - Toxicity - Histological examination

Résumé

Le moustique *Aedes caspius* représente une menace pour la santé dans de nombreuses régions, y compris l'Algérie, où il est largement répandu et cause des épidémies de maladies transmises par les vecteurs telles que le paludisme et la dengue. Cette étude vise à évaluer l'efficacité des huiles essentielles extraites de thym sauvage (*Thymus serpyllum*) et de marjolaine (*Origanum majorana*) dans le contrôle des larves de *Aedes caspius*. Les huiles essentielles ont été extraites à l'aide d'un appareil Clevenger, le rendement de l'huile de thym étant de 1,10 % et celui de l'huile de marjolaine de 0,74 %. Des tests de toxicité ont été réalisés sur les larves à différentes concentrations pendant 24, 48 et 72 heures. Les résultats ont montré que les deux huiles avaient un effet toxique sur les larves de moustiques, avec des taux de mortalité augmentant avec des concentrations plus élevées au fil des différentes périodes de temps. Les concentrations létales (LC50 et LC90) des huiles étaient les suivantes : huile de marjolaine (LC50 = 175 µL/L, LC75 = 268,71 µL/L, LC90 = 325 µL/L) et huile de thym (LC50 = 39,62 µL/L, LC75 = 145,02 µL/L, LC90 = 208,26 µL/L). Les examens histologiques ont révélé une destruction plus rapide et plus forte de l'épithélium de l'intestin moyen et des cellules cytoplasmiques dans les larves traitées avec de l'huile de thym par rapport à celles traitées avec de l'huile de marjolaine. Les résultats suggèrent que l'huile de thym sauvage est plus efficace pour éliminer les larves de *Aedes caspius* que l'huile de marjolaine, soutenant ainsi l'utilisation potentielle des deux huiles comme alternatives naturelles pour le contrôle des moustiques.

Mots-clés :

Huiles essentielles -Aedes caspius –Toxicité- Examen histologique

ملخص:

تشكل البعوضة Aedes caspius تهديدًا صحيًا في العديد من المناطق، بما في ذلك الجزائر، حيث تُسجل انتشارًا واسعًا وتسبب أوبئة للأمراض المنقولة مثل الملاريا وحمى الضنك. تهدف هذه الدراسة إلى تقييم فعالية الزيوت العطرية المستخلصة من نبات الزعيترة (*Thymus serpyllum*) والمردقوش (*Origanum majorana*) في مكافحة يرقات *Aedes caspius*. تم استخراج الزيوت العطرية باستخدام جهاز كليفيجر، حيث بلغت نسبة الزيت المستخلص من الزعيترة 1.10% ومن المردقوش 0.74 % تم إجراء اختبارات سُمّية على اليرقات في تراكيز مختلفة لمدة 24، 48، و72 ساعة. أظهرت النتائج أن كلا الزيتين لهما تأثير سام على يرقات البعوض، حيث زادت معدلات الوفاة مع زيادة التركيز عبر الفترات الزمنية المختلفة. كانت قيم التركيز القاتل LC50 و LC90 للزيتين كما يلي: زيت المردقوش (LC50 = 175 µL/L)، (LC75 = 268.71 µL/L)، (LC90 = 325 µL/L) وزيت الزعيترة (LC50 = 39.62 µL/L، LC75 = 145.02 µL/L، LC90 = 208.26 µL/L). أظهرت الفحوصات النسيجية تدميرًا أسرع وأقوى لظهارة المعدة الوسطى والخلية السيتوبلازمية في اليرقات المعالجة بزيت الزعيترة مقارنة بزيت المردقوش. النتائج تشير إلى أن زيت الزعيترة أكثر فعالية في القضاء على يرقات *Aedes caspius* من زيت المردقوش، مما يعزز الإمكانية لاستخدامهما كبديل طبيعي لمكافحة البعوض

الكلمات المفتاحية :

الزيوت العطرية – *Aedes caspius* البعوضة - السمية – الفحص النسيجي

General Scientific Abbreviations

1. LC - Lethal Concentration

2. LC_{50} - Lethal Concentration for 50% mortality

3. LC_{90} - Lethal Concentration for 90% mortality

4. µL/L - Microliter per liter (concentration unit)

5. H&E - Hematoxylin and Eosin (histological stain)

6. RH - Relative Humidity

7. R^2 - Coefficient of Determination (statistical fit)

8. Q_2 - Emberger's Pluviothermic Quotient (bioclimatic index)

9. A. (mosquito genus)

10. L4 - Fourth-instar larvae

11. PCOS - Polycystic Ovary Syndrome

12. NaOH - Sodium Hydroxide

13. EO - Essential Oil

14. EOs - Essential Oils

15. GC-MS - Gas Chromatography-Mass Spectrometry

16. AChE - Acetylcholinesterase

17. WHO - World Health Organization

18. UNESCO - United Nations Educational, Scientific and Cultural Organization

19. PNEK - Parc National d'El-Kala (El Kala National Park)

20. ONM- Office National de la Météorologie (Algeria)

21. IUCN - International Union for Conservation of Nature

22. MAB - Man and Biosphere Programme

23. HEOs - Hydrodistilled Essential Oils

24. LD_{50} - Lethal Dose for 50% mortality

25. AMCA- American Mosquito Control Association

26. DDT - DichloroDiphenylTrichloroethane

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TABLE OF FIGURES

LIST OF TABLES

Table of Contents

Practical Part

CHAPTER III

MATERIALS AND METHODS

Introduction

Insects represent the most diverse group in the animal kingdom, with more than two million species described worldwide to date, and thousands of new species discovered every year. They appeared on Earth approximately 400 million years ago—long before the emergence of modern humans, estimated at around 200,000 years ago (Firas Ahmed Al-Zyoud, 2018). Thanks to their incredible adaptability, insects thrive in a variety of ecological niches, particularly in tropical and subtropical climates (Okogun et al., 2003).

Among the most significant orders of the insect class is **Diptera**, due to the richness and diversity of the species it includes. The **Culicidae** family, which belongs to this order, is particularly feared for its role as a vector of numerous infectious diseases (Harwood & James, 1979; Service, 1993; Rueda, 2008; González et al., 2023). In medical and veterinary entomology, **Culicidae**—commonly known as mosquitoes—are considered the primary vectors of pathogenic agents, followed by ticks and fleas, and are also recognized as major pests (Lecointre & Hèrvé, 2001; WHO, 2016; Dris, 2019; Lecollinet et al., 2022; López et al., 2023).

To date, nearly **3,600 species** of Culicidae have been identified, with the most recent described in Thailand by Taai & Harbach (2015). Three main genera are particularly involved in human and veterinary pathology: **Aedes**, **Anopheles**, and **Culex** (Brunhes et al., 1999; Fontenille, 2010; Alves et al., 2010; Delaunay et al., 2012; Boudmegh, 2015; Djeddar et al., 2022). These mosquitoes are competent vectors of a wide range of pathogens responsible for severe and sometimes fatal diseases in humans and animals (Aouinty et al., 2006; Kosone et al., 2008; Zarroug, 2017).

Among the most concerning diseases is **malaria**, mainly transmitted by mosquitoes of the genus *Anopheles*. According to the WHO World Malaria Report (2021), about **241 million malaria cases** were recorded in 2020, resulting in **627,000 deaths**—an increase of 14 million cases and 69,000 deaths compared to 2019. On the other hand, **dengue**, an arboviral disease primarily transmitted by *Aedes* mosquitoes, is also a major public health issue, particularly in tropical and subtropical regions. Recent estimates report around **390 million cases per year**, including 96 million symptomatic cases and 40,000 deaths (Bhatt et al., 2013; WHO, 2020; Martinet, 2021).

Other important mosquito-borne arboviruses include **West Nile fever**, historically endemic in sub-Saharan Africa but later detected in Egypt (1940s), Romania (1950), and the Camargue region in France (1960), before spreading to North Africa, Europe, the United States, Canada, and Mexico (Zeller & Schuffenecker, 2004; Krida et al., 2015). Additional viruses such as **Rift Valley fever**, **Bancroftian filariasis microfilariae**, and **Sindbis virus**are also transmitted by mosquitoes (Moutailler et al., 2008; WHO, 2018; Gaüzère, 2013; Dris, 2019; Candeloro et al., 2020; Amdoni et al., 2022).

In this context, **mosquito control** has become a critical priority (Baba-aissa et al., 2021). For decades, humans have relied on conventional chemical insecticides—including organophosphates, carbamates, and pyrethroids—to control vector and pest insects. However, these chemicals have led to resistance development, negatively affected non-target organisms, and caused adverse impacts on the environment and human health, including neurotoxicity in vertebrates (Mathew & Thanuja, 2008; Cuervo-Parra et al., 2016; Belkhiri, 2023).

In light of these disadvantages, research has turned toward **alternative control methods**, especially the use of **plant-based biopesticides**, which are considered promising ecological and cost-effective alternatives (Thomas et al., 2014; Seghier et al., 2020; Chansang et al., 2018). Plants are a rich source of bioactive compounds with larvicidal, antimalarial, and insecticidal properties—such as terpenes, alkaloids, flavonoids, tannins, and polyacetylenes (Zirihi, 2006; N'guessan et al., 2009; Mansouri, 2015; Dahchar, 2017; Bouguerra & Soltani, 2018; Amira & Boudjelida, 2018).

Algeria, with an estimated flora of about **2,000 plant species**, boasts remarkable floral biodiversity. In an effort to valorize this wealth and explore natural alternatives to chemical insecticides, the present study focused on evaluating the insecticidal effects of **essential oils** extracted from two medicinal plants: *Thymus serpyllum* (wild thyme) and *Origanum majorana* (sweet marjoram), against fourth-instar larvae of the mosquito *Aedes caspius*.

The main objectives of this study are twofold:

1. To valorize the insecticidal activity of these two medicinal plants against *Aedes caspius*, a recognized vector of dengue.
2. To evaluate the developmental and histological effects of these essential oils on larvae treated with lethal concentrations, with the aim of developing a safer, biodegradable biopesticide that respects aquatic ecosystems.

This study is divided into five chapters: The firs chapter, of which provides a general of mosquitoes, and the second is dedicated medicinal herbs test. The third chapter explaining the materials and methods used in the experiments and protocols applied, followed by a chapter presenting the main results and their discussion. Finally, the study concludes with general conclusions.

Theoretical Part

Chapter 1:

Overview of Mosquitoes

Mosquitoes share three key characteristics with other arthropods: jointed appendages, an exoskeleton, and segmented bodies. While many mosquito species do not pose a direct threat to humans, certain types act as true parasites, transmitting various diseases. Others cause direct harm through their bites and related activities. Some species can function both as parasites and vectors of pathogens simultaneously (Molan, 2012).

Insect:

Insects belong to the class Insecta, a diverse group of invertebrate animals within the phylum Arthropoda. They represent one of the most widespread and abundant taxonomic groups under this phylum. With over one million described species, insects account for more than half of all known living organisms on Earth, making them the most biologically diverse group of terrestrial creatures. While they inhabit nearly all terrestrial environments, only a limited number of species are adapted to aquatic ecosystems (Younes, 2013).

Mosquitoes:

Among disease-transmitting arthropods, mosquitoes (Diptera: Culicidae) represent the most significant threat to public health due to their global distribution and vector competence. These insects serve as efficient vectors for numerous pathogens, including *Plasmodium* spp. (malaria), Wuchereria bancrofti (lymphatic filariasis), and various arboviruses responsible for yellow fever, dengue, and viral encephalitides (Belkhiri et al., 2023) (Coosemans & Van Gompel, 1998).

1. Classification of mosquito life cycle and morphology:

1.1Classification of mosquitoes:

The family Culicidae (mosquitoes) belongs to the order Diptera, suborder Nematocera, characterized by elongated, filamentous antennae. This family is subdivided into three sub-families: Anophelinae**,** Culicinae, and Toxorhynchitinae**.**

The subfamily Toxorhynchitinae consists of a single genus, *Toxorhynchites*, which includes large, non-hematophagous mosquitoes whose larvae are predaceous, feeding on other mosquito larvae (Dleng, 1995).

1.2 Taxonomic Classification:

Based on the globally accepted taxonomy by Harbach (1985, 1988) and subsequent revisions:

- Kingdom: Animalia
- Phylum: Arthropoda
- Subphylum: Hexapoda
- Superclass: Insecta
- Class: Insecta
- Subclass: Pterygota
- Division: Endopterygota (Holometabola)

1.3 Order Diptera:

- **SubOrder**: Nematocera (long-horned flies with multi-segmented antennae).
- **Family**: Culicidae (mosquitoes).
- **Subfamily**: Culicinae.
- **Genus**: *Culex* (abbreviated as *Cux* in some taxonomic contexts; Ward, 1989).

1.4 Mosquito Morphology:

Mosquitoes are holometabolous insects, undergoing complete metamorphosis through three distinct life stages: larva, pupa, and adult. The external morphology of each developmental stage serves as a key diagnostic tool for differentiating between genera, as their anatomical features vary according to their ecological adaptations (aquatic vs. aerial lifestyles)(Carnevale et al,2009)

A-Egg Stage:

Mosquito eggs are spindle-shaped (fusiform), measuring approximately 0.5 mm in length. Initially white, they undergo rapid chromatic transition to brown or black due to chemical oxidation (Berchi, 2000). Eggs are typically deposited on the water surface. Species of the genera *Anopheles* and *Aedes* lay eggs individually, while *Culex* and *Culiseta* oviposit in clustered aggregates (egg rafts). The eggs adhere to substrates via a viscous secretion. A single female may lay 250–500 eggs per oviposition cycle. Notably, most female mosquitoes require a blood meal to complete vitellogenesis and attain reproductive capacity. Embryonic development concludes within 2–3 days post-oviposition, after which eclosion occurs (Mustafa, 2017). Mosquito eggs are highly susceptible to desiccation, with complete dehydration resulting in mortality.

Figure 1: Image illustrates mosquito eggs (BALENGHIEN, 2007)

B- Larval Stages (Instars):

The larval phase is aquatic, with mosquito larvae appearing as legless and wingless worms. Their development can be divided into four primary stages: L1, L2, L3, and L4. These larvae become distinctly segmented during the fourth developmental stage (Arnaout, 2020). The larval stages typically last 5–10 days but may extend to several months under certain conditions. Larval size ranges from 1 to 15 mm, depending on species, developmental stage, and physiological factors. No visible sexual dimorphism is observed in larvae (Pagés et al., 2022; Failloux, 2022).

Body Segmentation :

The larval body is divided into three distinct regions, most notably in the fourth instar:

1. **Head**:

- Composed of a pair of antennae, two compound eyes (one non-functional), and precursors of adult ocular structures (Goulu, 2015).
- Encased in a completely sclerotized, spherical chitinous capsule (Berchi, 2000).
- Bears sensory antennae and mouthparts.
- Connects to the thorax via a flexible cervical membrane (cervix) (Abu Qurin & Murad, 1992).

2. **Thorax**:

- Consists of three segments, one of which is larger than the others.

3. **Abdomen**:

- Comprises ten segments.
- Contains the respiratory organ (tracheal system) and rudimentary reproductive structures.

Figure 2: Image illustrates mosquitoes in the larval stage (Ahmed S.Abdel-Aty ;2020)

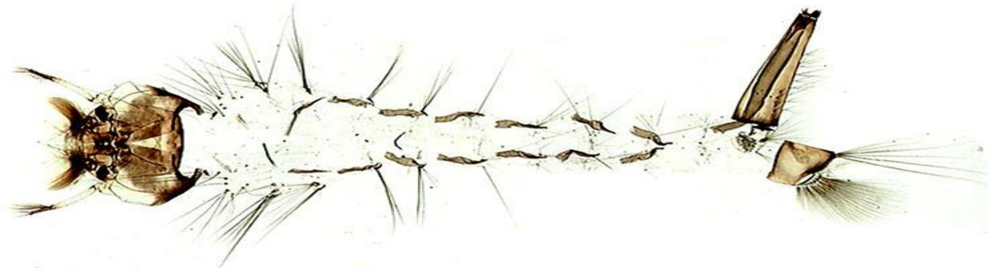


Figure 3 : Image illustrates morphology of a larva of Culex pipiens(Schaffner *et al*,2001)

C- The Pupal Stage:

The pupa (also called *tumbler*) emerges from the fourth larval instar. It is aquatic and non-feeding due to the closure of both oral and anal apertures (Clements, 1999). This stage typically lasts 1–3 days. During this phase, the head and thorax fuse to form a cephalothorax, which bears a dorsal pair of short, trumpet-shaped respiratory siphons (measuring a few millimeters in length, with size varying by species). Pupae undergo profound morphological and physiological restructuring to prepare for adulthood, as they do not feed (Nadji, 2011).

The pupal stage culminates in the final metamorphosis, enabling the transition from aquatic to aerial life. Upon completion, the pupal exuviae are shed, and the fully developed adult mosquito emerges.

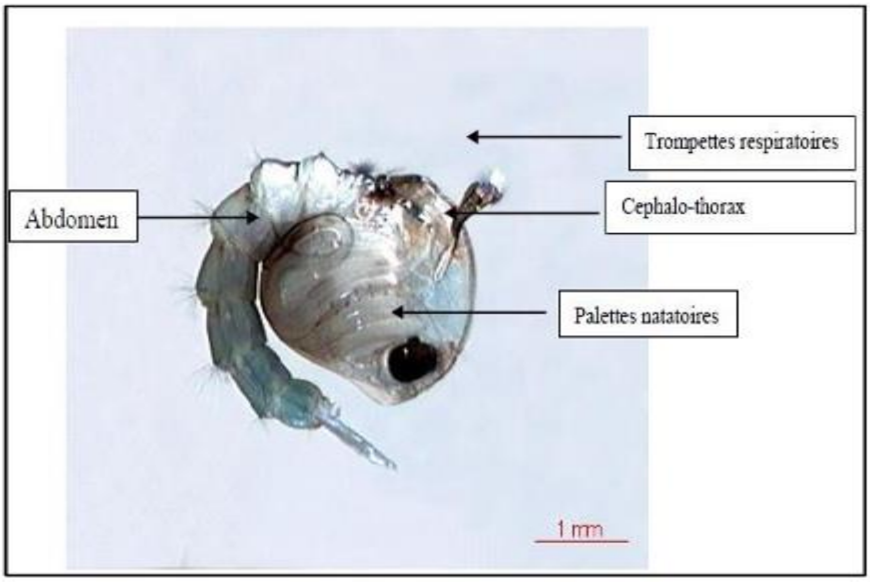


Figure 4: Image illustrates General morphology of the Mosquito Pupa l Houaous(Arnout,2020)

D- Adult Stage (Adult or Imago):

Adult mosquitoes are elongated insects measuring 5–20 mm in length (Rodhain & Perez, 1985). Their body is divided into three distinct regions:

- **Head**:

The head is spherical and bears a pair of kidney-shaped compound eyes separated by a narrow frontal band. The eyes consist of hexagonal, light-sensitive units (ommatidia) that appear blue-green in coloration. Between the compound eyes arises a pair of segmented antennae, each comprising 14–16 segments. The first segment (scape) is small and partially concealed by the second segment (pedicel). Subsequent segments (flagellomeres) are uniform in shape and covered with setae. Males exhibit denser and longer antennal setae compared to females, a key feature for sex differentiation (Ben Nasser & Dardouri, 2021).

- **Thorax**:

The thorax is the central body region, slightly convex and connected to the head via a short membranous neck. It bears three pairs of legs: the fore- and midlegs display ventral scales, while the hindlegs are characterized by dark-scaled regions interspersed with pale longitudinal stripes (TABTI, 2017).

- **Wings**:

As members of the order Diptera, mosquitoes possess one pair of membranous, transparent forewings. The hindwings are reduced to club-shaped halteres (balancing organs), which vibrate synchronously with the wings during flight to maintain stability (Ben Nasser & Dardouri, 2021)

- **Abdomen**:

In both sexes, the abdomen consists of ten segments. The first eight segments are flexible and externally differentiated, each composed of a dorsal tergite and a ventral sternite connected by lateral membranes. The arrangement, density, and coloration of scales and setae on the ventral segments are highly variable among species and serve as diagnostic taxonomic features.

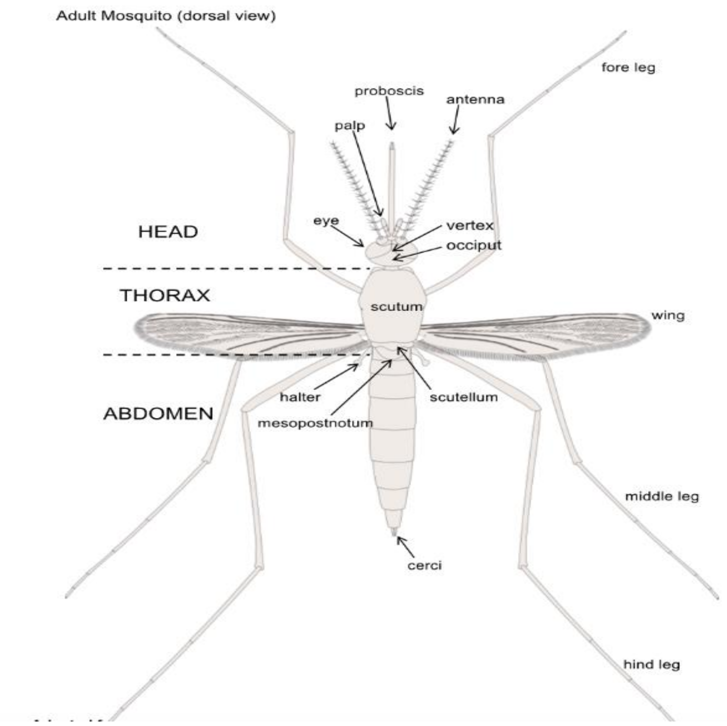


Figure 5: Image Anatomical diagram illustrating the body parts of an adult mosquito (head, thorax, abdomen)(Nathan Burkeett ;2013)

1.5Life Cycle:

The life cycle of mosquitoes typically spans approximately 12–20 days and comprises four stages: eggs, larval stage, pupal stage, and adults. These transformations occur through two distinct phases: aquatic and aerial (Alia et al., 2005). Mating usually occurs within 24–48 hours after emergence. In some species, males form swarms or aggregations, often positioned at variable or clearly defined locations. A single male can mate with multiple females, while females typically mate only once. Post-mating, females store sperm in specialized structures called spermathecae, retaining viable sperm for up to 10 months (Matile, 1955; Seguy, 1993). Females are capable of laying up to 2,000 eggs over a three-week period (Hawley, 1988). Upon maturation, eggs hatch into first-instar larvae (1–2 mm in length), which develop through four instars until reaching the fourth stage (1.5 cm). Larvae feed on organic matter and microorganisms (Maryse, 2008). The pupal stage (comma-shaped) is a non-feeding transitional phase lasting 3–5 days (Perez and Rodhain, 1985). Adult mosquitoes of both sexes primarily feed on nectar and plant sugars. However, females require a blood meal from vertebrates (mammals, amphibians, or birds) to obtain proteins necessary for egg development, a behavior linked to their oviposition phase (Guillaumot, 2006) .

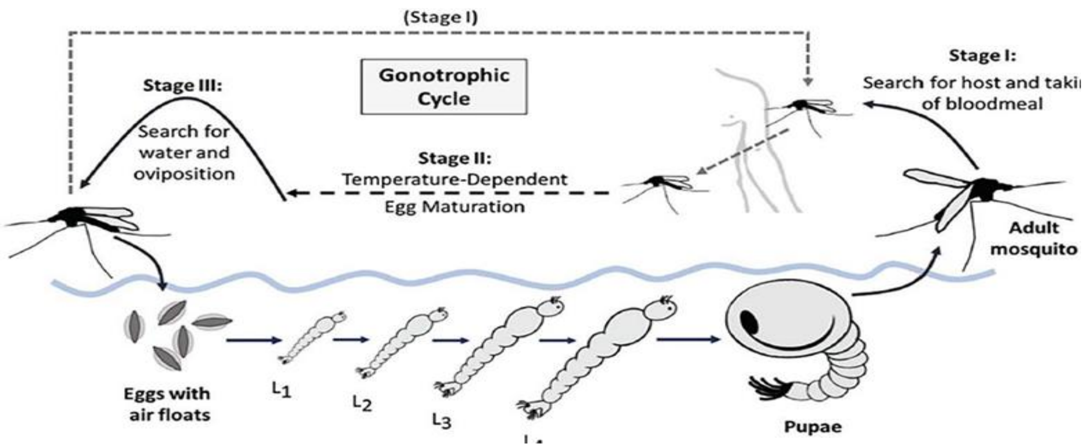


Figure 6: Mosquito lifecycle (Adopted from Okuneye *et al*., 2019)

2- Nuisance Aspect and Vector Role of Mosquitoes:

2-1 Mosquitoes and Health Risks:

Mosquitoes significantly impact both external and internal environments. Externally, their persistent biting creates a nuisance that restricts outdoor activities, leading to decreased enjoyment of recreational spaces. Internally, mosquitoes are vectors for pathogens capable of causing severe diseases, such as malaria, dengue, and Zika virus. These health risks have substantial economic consequences, including loss of tourism revenue due to reduced outdoor engagement and increased costs associated with mosquito control measures and medical treatment for infections. The interplay of these factors underscores the importance of effective mosquito management strategies to mitigate their impact on public health and the economy.

Mosquito-borne diseases pose a significant threat to public health, causing millions of infections and resulting in substantial economic costs. According to recent estimates, the global economic burden of mosquito-borne diseases is approximately $12 billion annually, which includes direct costs such as medical treatment and indirect costs related to lost productivity due to illness (CDC, 2024)

These diseases, which include malaria, dengue, Zika, and West Nile virus, are transmitted through the bites of infected mosquitoes. The prevalence of these diseases varies by region and season, with certain areas experiencing higher rates of infection due to environmental factors that favor mosquito breeding (Benelli & Mehlhorn, 2023).

Preventive measures are crucial in controlling the spread of these diseases. Effective strategies include the use of insect repellents, wearing protective clothing, and eliminating standing water where mosquitoes breed (AMCA, 2024)

In summary, the impact of mosquito-borne diseases on health systems and economies underscores the need for continued public health efforts and research to mitigate their effects (Nadji, 2011).

2-2 Nuisance Problems:

Many mosquito species, even those not involved in pathogen transmission, cause substantial nuisance issues due to their aggressive biting behavior. Their impact depends on:

- Host-seeking strategies (e.g., ambush vs. hunting).
- Human and animal reactions to bites (itching, allergic responses).

Female mosquitoes require blood meals to produce eggs, increasing human exposure. Species linked to wetlands (e.g., daytime ambush feeders like *Aedes*) are particularly problematic, as their activity coincides with human outdoor routines, heightening demands for control measures.

 Aedes: This is a genus of mosquitoes known for being aggressive biters and for transmitting various diseases, including dengue fever and Zika virus. Species within this genus, such as *Aedes aegypti*, are particularly problematic because they are active during the day and often bite humans.

 Host-seeking strategies: This term refers to the methods mosquitoes use to locate their hosts (humans or animals) for feeding. There are generally two strategies:

- **Ambush**: This strategy involves waiting for a host to come close before attacking. Mosquitoes that use this method tend to remain still and then quickly bite when a host is near.
- **Hunting**: This strategy involves actively searching for hosts, which can include flying around and using sensory cues to locate potential blood sources.

Human and animal reactions to bites: This refers to the physiological responses that occur after a mosquito bite, which can include:

- **Itching**: A common reaction due to the body's immune response to the saliva injected by the mosquito.
- **Allergic responses**: Some individuals may experience more severe reactions, including swelling or hives, due to allergies to mosquito saliva.

2-3 Potential Health Problems:

For disease transmission, a female mosquito must:

1. Acquire pathogens from an infected host during feeding.
2. Survive long enough for pathogen replication (e.g., in salivary glands).
3. Transmit pathogens to a new host during subsequent feeding.

- Medical Importance of Mosquitoes :

The nuisance impact of mosquitoes ranks secondary compared to their role as vectors of pathogens that cause severe diseases in humans and animals. Key mosquito-borne diseases include:

A. Plasmodium *Parasites:*

- Cause human malaria (*Plasmodium* spp.), a life-threatening disease transmitted primarily by *Anopheles* mosquitoes.

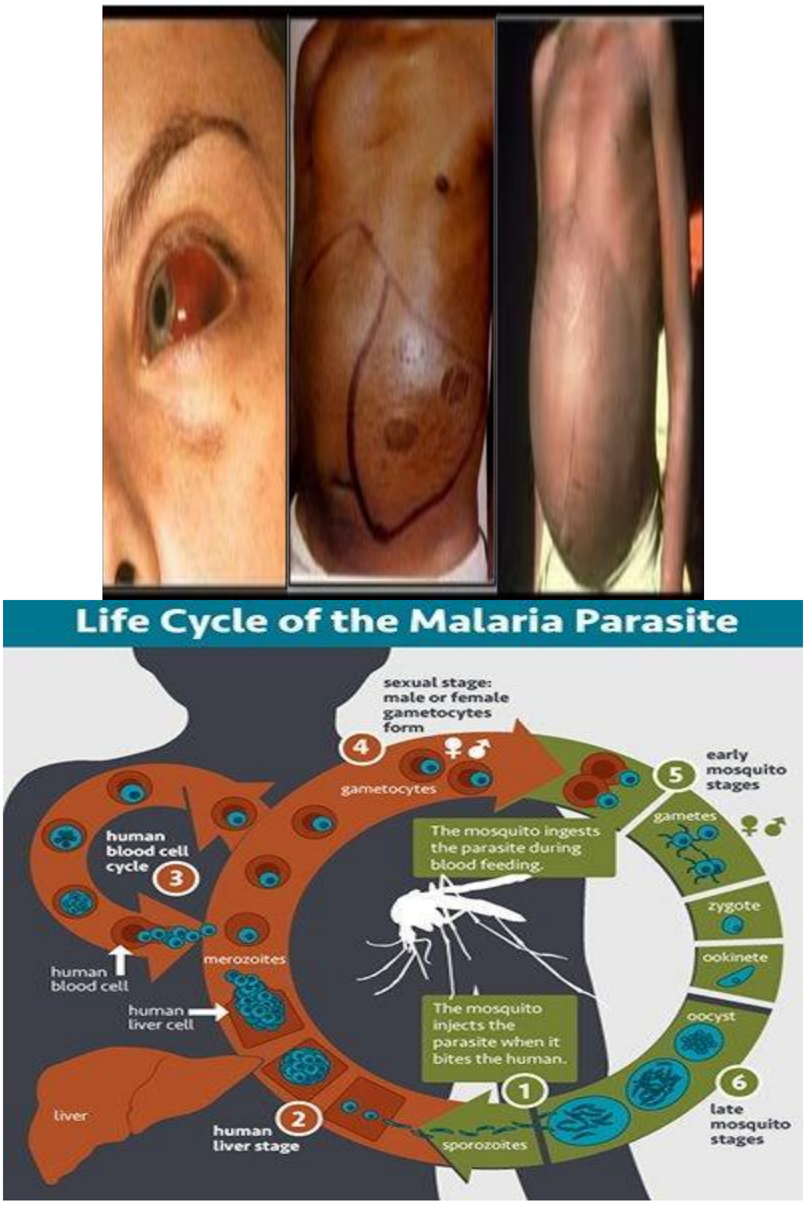


Figure 8:Image showing some of the symptoms of malaria (boubidi ,2008)

B. Filarial Worms (**Wuchereria bancrofti*****)**:*

***Elephantiasis (Lymphatic Filariasis)**:*

- Caused by the nematode *Wuchereria bancrofti*.
- Prevalent in continental Africa and other tropical regions.

Transmission Mechanism:

- Mosquitoes (e.g., *Culex*, *Anopheles*) ingest microfilariae from infected humans.
- Infective larvae migrate to the mosquito's proboscis and enter new hosts during feeding

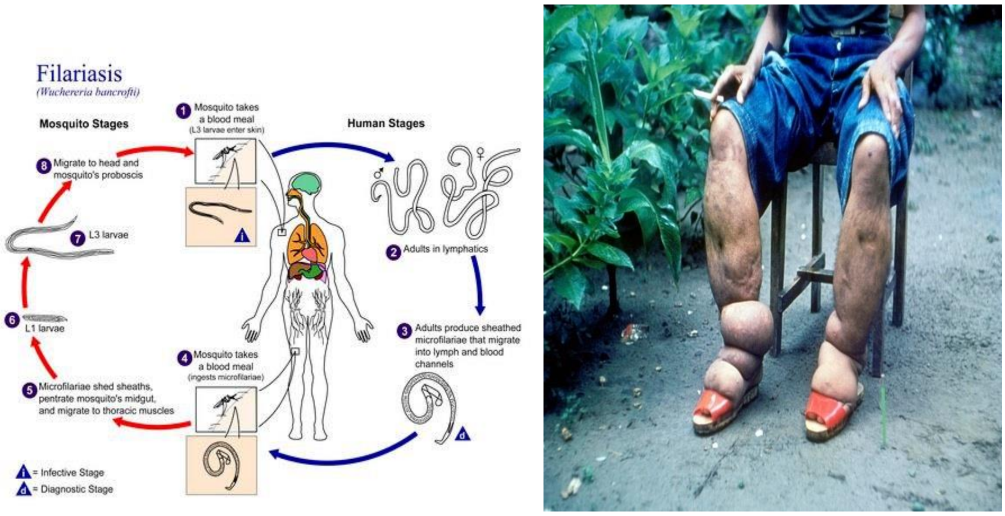


Figure 9: Image showing the symptoms of elephantiasis caused by filarial worms (Daniel Murrell, 2023)

Transmission Cycle:

1. Mosquitoes ingest microfilariae from infected human blood.
2. Microfilariae develop into infective larvae within the mosquito.
3. Larvae are transmitted to new human hosts during subsequent blood-feeding.

C. Arboviruses*:*

- **Yellow fever virus**: Causes hemorrhagic fever.
- **Dengue virus**: Causes dengue fever, a flu-like illness.
- **Encephalitis viruses**: Trigger brain inflammation (e.g., West Nile virus).

Figure 10: The image illustrates the symptoms of yellow fever (centers for disease control ,2024).

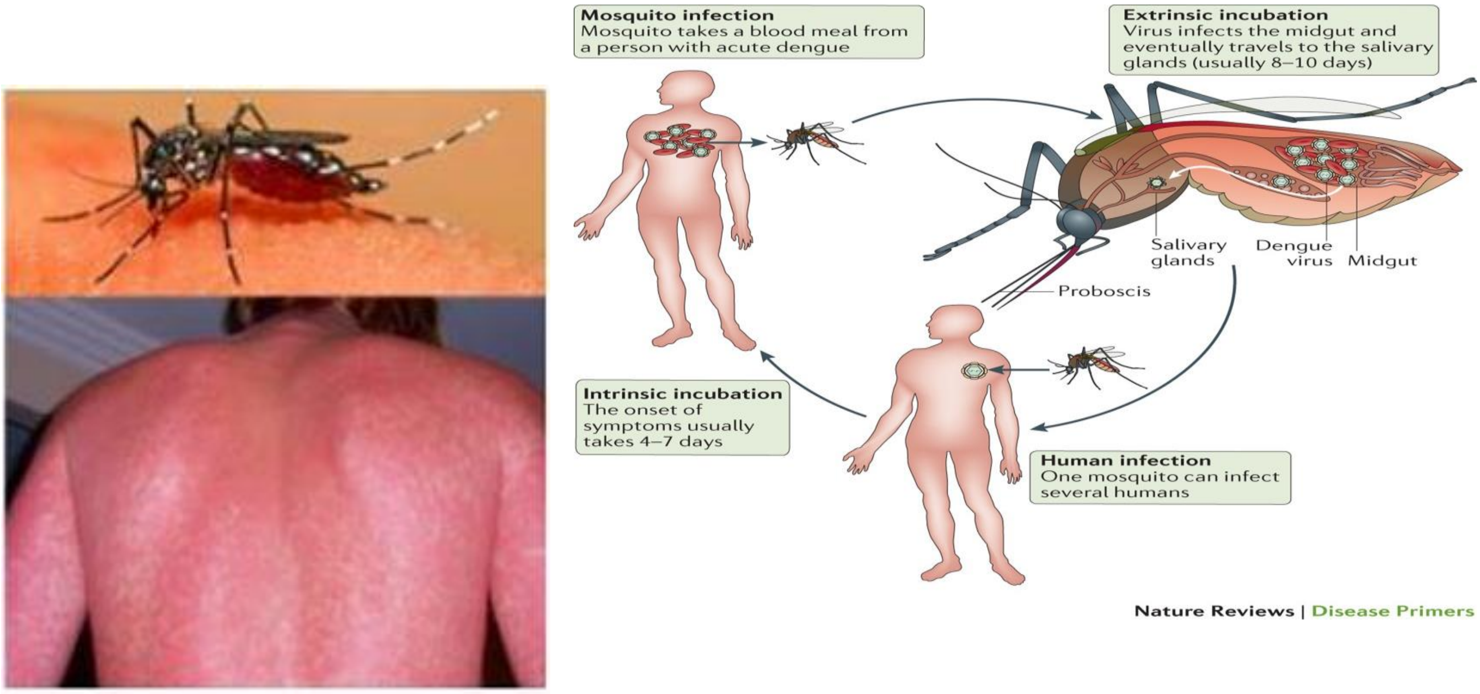


Figure 11 : Image showing the presentation of dengue fever (Maria Guzman,2016)

These diseases highlight mosquitoes' dual role as pests and vectors of global health significance (*Ughasi et al., 2012*).

The Ecological Role of Mosquitoes:

While mosquitoes are vectors of deadly diseases such as malaria and dengue, they hold ecological significance as keystone species in aquatic and terrestrial food chains, serving as prey for fish, birds, and amphibians (*Bernard et al., 2000*). Thus, control strategies must balance public health priorities with ecosystem preservation to avoid unintended ecological disruptions

3- Methods of Mosquito Control:

3.1. Physical Control:

- **Personal Protection**: Use of loose, long-sleeved clothing and light-colored attire, combined with insecticide-treated bed nets (ITNs) and indoor residual spraying (IRS) (Fontenille et al., 2006).
- **Mechanical Elimination**: Removal or modification of larval breeding sites (e.g., draining stagnant water). Though effective historically, urbanization has reduced its applicability (Hamon & Mouchet, 1967; Brown, 1967).

3.2. Chemical Control:

- **Organochlorines (e.g., DDT)**: Neurotoxic agents disrupting K+/Na+ ion channels, causing paralysis and death in mosquitoes. Widely used but linked to environmental persistence (Boyer, 2006).
- **Pyrethroids and Organophosphates**: Broad-spectrum insecticides for vector control. However, widespread resistance has emerged across mosquito populations (Al-Sarar, 2010).
- **Challenges**: High costs in developing nations, environmental contamination, and non-target toxicity (Koua et al., 1998 Aouinty et al., 2006). Overuse of chlorine-based pesticides (e.g., DDT) has disrupted agricultural ecosystems (Mörner et al., 2002).

3.3. Biological Control:

Biological control is an effective and environmentally friendly method for managing mosquito populations, focusing on the use of living organisms such as microorganisms, fungi, fish, and plant extracts. This approach aims to reduce the impact of mosquitoes as disease vectors without relying on harmful chemical pesticides.

3.3.1. Plant Extracts

Plant extracts have demonstrated toxic effects against various dipteran species. Historically, substances like pyrethrum, nicotine, and rotenone have been recognized as insecticides. In certain African regions, mashed tobacco leaves mixed with water have been used to combat mosquitoes, while the scents of plants such as basil (*Ocimum basilicum*) and *Corrigiola telephiifolia* are effective repellents (Aouinty et al., 2004). Recent studies have shown the efficacy of extracts from five different plants against species like *Culex pipiens* and *Culiseta longiareolata* (Aouinty et al., 2006).

3.3.2. Entomopathogenic Bacteria

The use of entomopathogenic microorganisms is a promising alternative for effective pest management due to their natural ubiquity in ecosystems, diversity, and environmental persistence. Notably, *Bacillus thuringiensis* (var. israelensis) and *Bacillus sphaericus* are widely used to target mosquito larvae (Zahiri et al., 2002; Monnerat et al., 2004).

3.3.3. Entomopathogenic Fungi

Several species of entomopathogenic fungi have been isolated and tested against major mosquito vectors, capable of killing both larval and adult forms. Commonly used genera include *Beauveria*, *Metarhizium*, and *Lagenidium* (Scholte, 2004).

3.3.4. Larvivorous Fish

Indigenous fish species have been utilized globally to control mosquito larvae. *Gambusia affinis* remains the most commonly used species in biological control efforts (Schleier et al., 2007).

3.3.5. Other Natural Predators

The use of natural predators is a recurrent strategy in vector control. Certain arthropods, such as copepods, prey on mosquito larvae. For instance, in Vietnam, copepods have been effective in eliminating *Aedes* and *Anopheles* larvae from large water reservoirs (Kay and Vu, 2005).

4. Integrated Vector Management (IVM)

Integrated Vector Management combines chemical, physical, and biological methods to maintain mosquito populations below critical thresholds. The effectiveness of IVM relies on a solid ecological understanding, particularly regarding the spatio-temporal variations in mosquito development and activity (Himmi et al., 1998; Lounaci, 2003)

Chapter II

Medicinal Plants

Medicinal plants are defined as plant species with therapeutic properties that Collected from their natural habitats or cultivated . These plants are used either fresh (tender or dried) or as raw material in the manufacture of various liquid and solid extracts (Boubakhti,2010).

The study of plants is no longer limited to medicinal uses as in the past, but now extends to broader fields such as economics, industry and the pharmaceutical industry. With the increasing biodiversity of plants and the spread of diseases, scientists are looking for them as effective sources of medicines. The interest in medicinal plants has led to the discovery of their therapeutic components and their use in the manufacture of medicines for chronic diseases, as well as the extraction of essential oils that are medically and economically useful. These plants are now considered a promising source for the development of new medicines that are more effective and less harmful to humans (Amekran,2015). A medicinal plant is defined as any plant organism that contains bioactive compounds for medical use, while an aromatic plant is classified as a plant species that contains essential oils (or what is known as volatile oils) in one of its plant parts, so that these oils can be extracted using approved techniques in this field (Mekhedmi,2014).

1 : The marjoram *Origanum majorana*(Linnaeus,1753)

Figure 12: Portrait of an *Origanum majorana L.*

from the Hassi Khalifa region of Oued Souf (original photo,2024)

1.1) Scientific Description of Marjoram :

Origanum majorana L. is an annual aromatic herb belonging to the Lamiaceae family, widely cultivated as a culinary plant for its fragrant leaves. Native to Southwest Asia, this species has become naturalized throughout Mediterranean regions, where it is commonly grown as a seasonal crop in kitchen gardens.

Pharmacognostic studies classify marjoram as both a medicinal and aromatic plant, alternatively referred to as sweet marjoram or knotted marjoram. Therapeutically, it demonstrates significant bioactive properties. While morphologically similar to oregano (*Origanum vulgare*), marjoram is distinguished by its more subtle aromatic profile and lower phenolic content (Clément, 2020).

1.2) Plant names :

Depending on the language, there are several names for Marjoram. These include :

- **German name :** Garten-Majoran
- **French name :** Marjolaine du jardin
- **Italian name :** Maggiorana
- **Arabic name :** Merdeqouch ou Merdaqouch selon les magrébines) (Baba Aissa F., 2011) ; Merdgouch. (Beloued A., 2009)
- **English name :** Marjoram
- **Berber or Teurgui name :** Arzema, M'loul (Beloued A., 2009).

1.3) Scientific Classification:

Table 01 : Scientific classification of *Origanum marjorana* (Carl Linnaeus,1753) (Al Daher T et *al.,* 2024)

Origanum majorana	Scientific name
Flowering Plants	Division
Dicotyledons	Class
Lamiaceae	Family
Origanum	Genus
O.majorana (Linnaeus,1753)	Species

1.4) Description of plant :

This plant is characterized by small white or purple flowers with a calyx shape, and oval, gray-green leaves covered with fine down, growing oppositely along the stem. The plant typically reaches a height of about **30 cm**, with a woody base and a pubescent texture. The root is taprooted, twisted, and somewhat branched. The stem is erect, branched, quadrangular in cross-section, and reddish in color.

The leaves are simple, opposite, petiolate, oval to elongated in shape, with smooth margins. They measure approximately 2 cm in length and 1 cm in width and are covered with a whitish down. The inflorescences are globular spikes, both axillary and terminal, usually arranged in groups of three. The flowers are small, zygomorphic, white or pink, and enclosed at the base by broad, shell-like bracts.

The calyx is gamosepalous and bilabiate, composed of five sepals. The corolla is gamopetalous with five petals forming a notched upper lip and a trilobed lower lip. There are four stamens, two of which are longer, bearing reddish anthers with divergent lobes. The ovary is formed of two biovulate carpels and is topped with a bifid stigma (Burt, 2004).

Figure 13: Marjoram flowers and stems (Anonyme 1)

1.5) Geographical distribution :

Origanum majorana is commonly known as native to Cyprus, Antalya (Turkey), distributed in different parts of Mediterranean countries as Serbia, Italy, Corsica, southern Spain and Portugal, Morocco, and Algeria (Ietswaart, 1980). Including, all over the world where it is cultivated in many countries in Europe such as France, US, Asia, in different parts of India, Hungary, and United States (Prerna and Vasu deva, 2015).

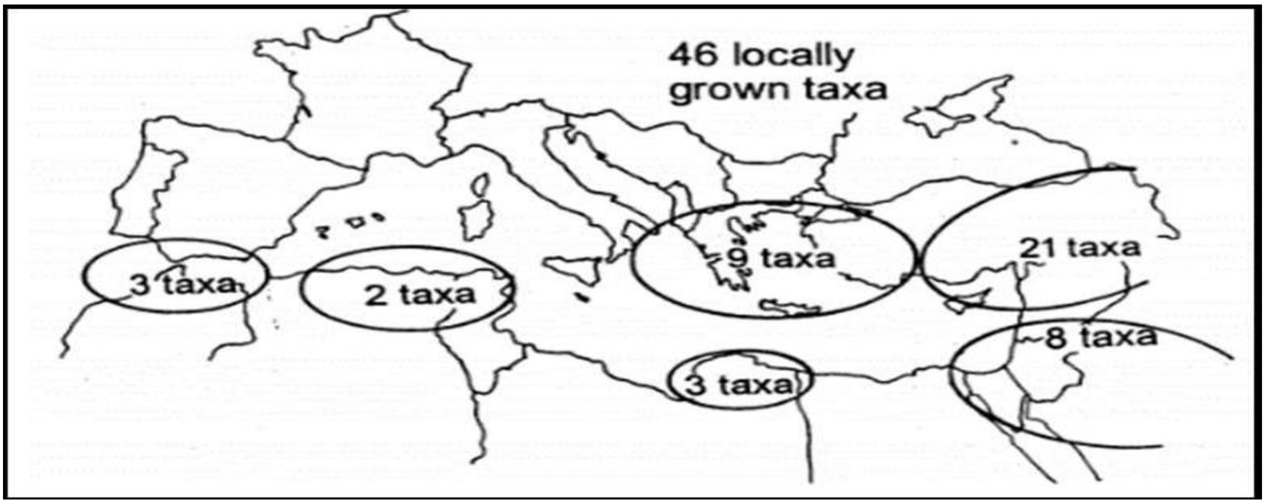


Figure 14: Distribution of the genus Origanum across the continents (Ietswaart, 1980)

1.6) Chemical composition of *Origanum majorana* essential oil :

The essential oil of marjoram (*Origanum majorana*) exhibits a complex chemical composition dominated by oxygenated monoterpenes, particularly terpinen-4-ol (22.85%) and (E)-sabinene hydrate (15.94%) as primary components (Kahouli., 2010).The monoterpene fraction includes γ-terpinene (12.60%), sabinene (7.65%), and α-terpinene (7.73%), while sesquiterpenes are represented by β-caryophyllene (2.49%) and bicyclogermacrene (1.22%). The oil also contains significant oxygenated compounds such as α-terpineol (4.88%), (Z)-sabinene hydrate (4.40%), and linalyl acetate (1.70%) (FATHY *et al*., 2009) . Minor constituents include p-cymene (1.57%), limonene (1.76%), terpinolene (2.92%), and β-phellandrene (1.90%), along with trace amounts of α-pinene (0.77%), α-thujene (0.77%), β-pinene (0.43%), and α-phellandrene (0.56%) (Triantaphyllouk *et al*, 2001) . According to Brada (2013), the major compounds accounted for (1.6%) of the total composition, distributed as follows: λ-terpinene (34.1%), p-cymene (27.6%), and carvacrol (9.6%). However, studies by Daoudi Merbah (2016) and Kerbouche (2015) reported that the dominant compounds were carvacrol (31.8–60.8%), γ-terpinene, and p-cymene. Notably, in some populations, carvacrol was replaced by thymol at a concentration of 33.6%.

This chemical profile demonstrates the oil's rich diversity of bioactive compounds, with terpinen-4-ol emerging as the predominant phytochemical marker.The volatile oil is characterized by the presence of camphor, borneol, terpinane, and sabinane among its principal terpenoid components (Encyclopedia of Plants,2019) .

Figure 15: *Origanum majorana* essential oil (Al Daher T et *al..,* 2024)

1.7) Traditional use of the plant *Origanum majorana* :

Origanum majorana, a member of the Lamiaceae family, is utilized in the food industry as a flavoring agent and natural preservative due to its antimicrobial essential oil (Renta،2012). While in traditional medicine, it is employed for its potential therapeutic effects against oncological, neurological, rheumatologic, cardiovascular, respiratory, and digestive disorders, attributed to bioactive compounds such as terpinene-4-ol, linalool, sabinene, and rosmarinic acid, which exhibit antimicrobial, anti-inflammatory, antioxidant, and antispasmodic properties (Kovaaevia،2001 ; Arafa *et al*،2023).

1.8) Anti-toxic effect :

Marjoram (*Origanum majorana*) is a herb rich in bioactive compounds, including phenolic antioxidants such as rosmarinic acid, flavonoids (apigenin and luteolin), and carnosic acid, which play a significant role in neutralizing free radicals and protecting cells from oxidative damage. Additionally, marjoram provides essential nutrients, including vitamin K (critical for blood clotting and bone health), iron (necessary for hemoglobin synthesis and oxygen transport), calcium (vital for

bone integrity and muscle function), manganese (a cofactor for antioxidant enzymes and metabolic processes), and vitamin E (which protects cell membranes from oxidative stress).
The essential oil of marjoram contains bioactive compounds such as camphor, esters, and terpenes, contributing to its antibacterial, nervine relaxant, vasodilatory, and antispasmodic

properties (Schaal,2010). These components make marjoram a valuable functional herb with potential benefits for cardiovascular, neurological, and immune health.

The plant is also used to alleviate rheumatism, cough, cold, and digestive disorders (Mechergui et *al*., 2015). Indeed, oregano is known for its bactericidal properties, sedative effects, antispasmodic action, and antiseptic benefits for the respiratory tract (Allane, 2009). Additionally, it exhibits appetizing, stomachic, expectorant, parasiticidal, and analgesic properties, as well as serving as a culinary spice (Saimi, 2014).

1.9) Uses of the plant:

- **Antioxidant properties of marjoram extract :**

Marjoram's bioactive compounds, particularly carvacrol, exhibit potent antioxidant effects by neutralizing free radicals and preventing oxidative damage (Bina and Rahimi,2016 ; Ramazan et *al*.,2016), while also demonstrating anti-inflammatory properties through modulation of inflammatory pathways (Ramazan et *al*.,2016 ; Palanisamy et *al*.,2016). These dual actions may help mitigate chronic inflammation-associated diseases like diabetes, cancer, and autoimmune disorders (Hazem and Ippolito,2017 ; Camillo et *al*.,2015).

- **Antimicrobial Properties of Plant Extracts :**

Plant extracts exhibit broad-spectrum antimicrobial activity. Studies demonstrate that Gram-positive bacteria are more susceptible to these extracts than Gram-negative bacteria. This difference is attributed to the outer membrane of Gram-negative bacteria, which contains lipopolysaccharides that act as a barrier against hydrophobic molecules and large compounds (Walsh et *al*., 2003; Starlper et *al*., 2015).

These bioactive-rich extracts primarily target the bacterial plasma membrane (Hyldaard et *al*., 2012), explaining their effectiveness as antimicrobial agents.

- **Potential Hormonal Regulation Effects of Marjoram :**

Studies suggest marjoram may help regulate menstrual cycles and restore hormonal balance in women with irregular periods. Its extract or tea appears to stimulate menstrual flow and could benefit women with polycystic ovary syndrome (PCOS). Research involving 25 PCOS patients demonstrated that marjoram tea consumption improved both hormonal profiles and insulin sensitivity. These findings indicate marjoram's potential as a natural therapeutic agent for hormonal disorders, though further research is needed to confirm these effects (Bina and Rahimi,2016 ; Alkazaleh et *al*..,2016).

2.1) Scientific Description of Thymus :

Thymus serpyllum L., commonly known as wild thyme, is a perennial herbaceous plant belonging to the Lamiaceae family. According to the latest taxonomic updates, this family includes over 7,000 genera, with the genus *Thymus* comprising a taxonomically complex group of perennial aromatic plants. Estimates of the number of *Thymus* species vary significantly across sources: Jaric et al. (2015) report approximately 111 recognized species, while other taxonomic authorities suggest a broader range, citing up to 250 (Mabberley, 1997; Zeljković and Maksimović, 2015) or even **over** 400 species globally (Leal et al., 2017). This variation reflects differing classification systems and ongoing revisions in species delimitation.

These species show considerable morphological diversity and notable variations in their chemical composition. *Thymus serpyllum* is native to Northern Europe (Jaric et al., 2015). The genus is generally divided into eight sections, with a large number of species endemic to the Mediterranean region. The major bioactive constituents found in thyme include thymol, carvacrol, and a range of flavonoids (Aziz et al., 2008).

Thymus , Thymus serpyllum L (Carl Linnaeus, 1753) :

figure 16 :Photo of a Thymus serpyllum the Kala region (personal 2025)

figure 17: Photo of a Thymus in serpyllum leaves from the Kala

2.2) Plant names :

- **Sientific name:***Thymus serpyllum*
- **Common names:** Wild thyme - Creeping thyme - Attractive thyme - Sa'tar - Za'tar
- **In Amazigh:** Azukni (Omayar N,2018)

2.3) Classification of plant :

- **Kingdom:** Plantae
- **Class:** Magnoliopsida
- **Order:** Lamiales
- **Family:** Lamiaceae
- **Genus:** *Thymus.*
- **Species:***T. serpyllum* (Santa; Quezel1963 ; Prasanth Reddy et al،2014)

2.4) Description of plant :

Thymus serpyllum is a perennial aromatic herb reaching 5–15 cm in height and approximately 50 cm in spread, with dense branching and a prostrate growth habit. The root system is superficial and fibrous. Stems are short, erect, and woody, with ridged gray surfaces covered in brown trichomes. Leaves are small, simple, and oppositely arranged, lanceolate in shape with entire margins, numerous, and grayish in color due to dense pubescence. Flowers are borne in spicate inflorescences, purple or pink in color, with a flowering period from May to September. The species thrives in dry habitats, particularly rocky areas (Omayar N,2018).

Figure 18: *Thymus serpyllum* flowers (Anonyme 2)

2.5) Geographical distribution :

This species is native to the Mediterranean region but is now cultivated worldwide (Premrov Bajuk et al., 2022). Its natural distribution includes southern Europe and Mediterranean coastal countries, with populations extending into temperate climate zones globally. Notably, it also occurs in semi-arid regions of southeastern Spain (Alcaraz and Delgado, 1999).

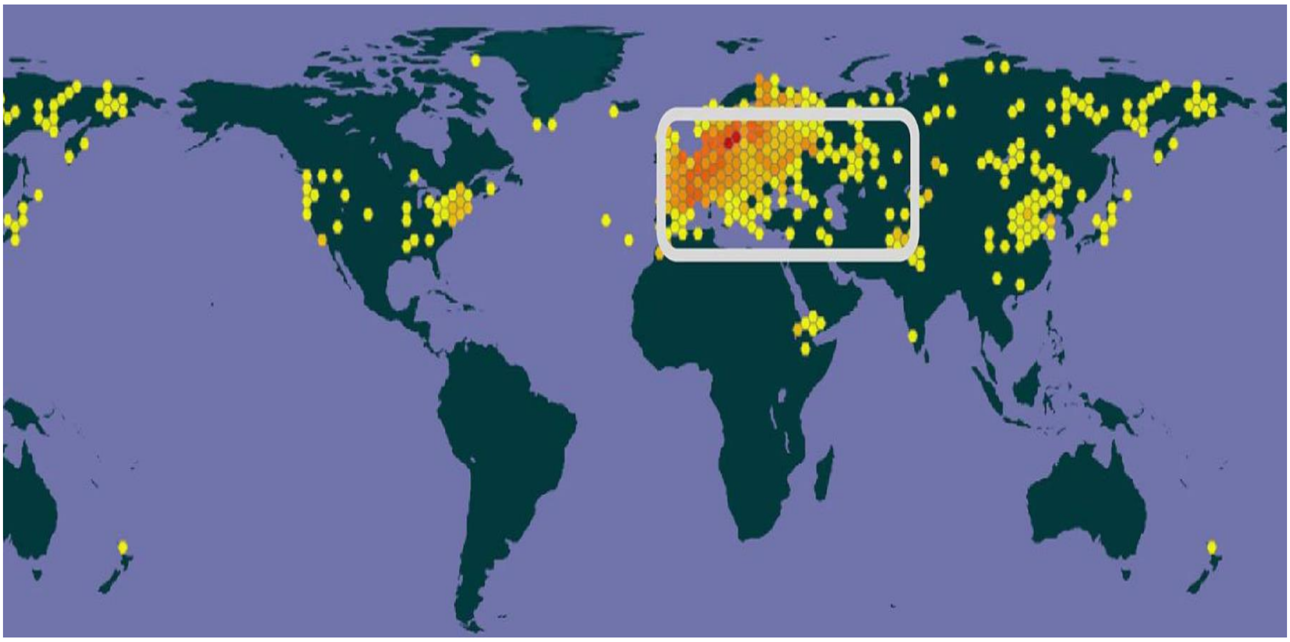


Figure 19: Geographical distribution of plant *Thymus serpyllum* (GBIF Secretariat,2023)

2.6) Chemical composition of Thymus serpyllum L essential oil :

The essential oil of wild thyme (*Thymus serpyllum*) represents its primary bioactive component, dominated by thymol and carvacrol - phenolic monoterpenes exhibiting potent irritant, rubefacient, anti-inflammatory, antihistaminic, antifungal, and antiseptic properties. The leaves and flowering tops contain diverse phytochemicals including volatile compounds (eucalyptol with its expectorant and antiseptic effects, and borneol), phenolic acids (nicotinic acid with purported cerebrovascular effects and acetic acid), B-complex vitamins (nicotina-mide, folate, thiamine, pyridoxine, riboflavin), ascorbic acid, β-carotene, various flavones, and essential minerals (potassium, iron, manganese, calcium, magnesium, and selenium), col-lectively contributing to its significant nutritional and therapeutic value in both traditional and modern medicine. (Omayar ,2018)

Figure 20: Essential oil of *Thymus serpyllum* (Anonyme 3)

2.7) Traditional use of the plant Thymus serpyllum L :

The genus *Thymus*, comprising over 110 to more than 400 species depending on taxonomic interpretation, includes both annual and perennial herbaceous plants widely used for medicinal and culinary purposes. These species are predominantly distributed across the Old World and have been extensively studied due to their historical use in traditional medicine (Leal et al., 2017).

Thymus serpyllum L. (wild thyme) has a long history of traditional use, particularly for its aerial parts, which are rich in essential oils. These parts are known for their strong aroma and slightly pungent, bitter taste, making them popular in both therapeutic and culinary contexts (Saad, 2018; Anab Baladi Newspaper, 2016). In traditional medicine, wild thyme has been used for treating respiratory ailments, digestive disorders, and as an antiseptic.

Contemporary studies have validated many of these traditional applications. The genus *Thymus* demonstrates pharmacological activity against neoplastic conditions, psychiatric disorders, and inflammatory diseases (Zahra et al., 2022). Additional properties identified include analgesic, thyroid-stimulating, cardioprotective, antioxidant, and anti-diabetic (type 2) effects. Beyond its medicinal importance, thyme continues to play a vital role as a culinary herb, highlighting its dual role in both nutrition and phytotherapy (Omayar N., 2018).

2.8) Anti-toxic effect :

Thyme essential oil (*Thymus spp.*) possesses significant pharmacological properties, demonstrating anti-inflammatory, cardioprotective, and anticancer activities while enhancing immune function. Clinically, it alleviates sinusitis, provides analgesic effects, and treats dermatological conditions. Its rich mineral content (calcium, manganese) and vitamin K contribute to bone health, while vitamin A supports vision. Cosmetically, it nourishes hair and skin while stimulating circulation (Jukic and Milos, 2005 Tea et al., 2006 and Meshkatalsadat, 2007).

This oil ranks among the top ten essential oils due to its potent antioxidant, antimicrobial (antibacterial/antifungal), and food preservative properties, with additional anti-aging benefits (Al-Sheibany et al., 2005). Flowering aerial parts yield 1.3-1.7% volatile oil, though concentration varies with climatic, geographic, and genetic factors that influence both oil composition and bioactivity. Thyme oil contains approximately 70% phenolic compounds (thymol 30%, carvacrol 26%, p-cymene 13%), alongside flavonoids, tannins, and other bioactive constituents including linalool, eugenol, terpineol, and tannins (Tepe et al., 2004).

2.9) Uses of the plant :

Thymus serpyllum is a medicinally valuable plant with scientifically validated pharmacological properties, including acetylcholinesterase (AChE) inhibitory activity that enhances cognitive function and may protect against Alzheimer's disease, along with potent antifungal and antibacterial effects effective against dandruff when applied topically with carrier oils. Its therapeutic spectrum encompasses relief of dysmenorrhea, gastric ulcer treatment through proton pump inhibition, and improvement of irritable bowel syndrome symptoms via serotonin receptor modulation, primarily attributed to bioactive compounds thymol and carvacrol which exhibit antineoplastic, antidiabetic (via insulin sensitivity enhancement), and cardioprotective (through nitric oxide-mediated vasodilation) properties. Additionally, it demonstrates anti-inflammatory, antiparasitic (against intestinal helminths), and immunomodulatory activities, while its dermatological applications leverage antioxidant-rich composition for skincare and hair treatments, and culinary uses benefit

from flavor-enhancing, digestive, and natural food preservation qualities, though current evidence primarily derives from in vitro and animal studies, necessitating further clinical trials to establish therapeutic efficacy and dosage parameters in humans (Omayar ,2018) .

3 : Essential oils

Figure 21: Volatil oils (Anonyme 4)

Essential oils are volatile aromatic compounds naturally produced by certain plants, known as complex volatile plant metabolites present in minute concentrations (Adam et al., 2009). Their content varies significantly among plant species and they are extracted through various methods. Including distillation, where the oils are carried by water vapor (hydrodistillation) or steam (steam distillation) from either whole plants or specific parts. (Bakkali et al., 2008, Da Porto and Decorti, 2009) . The most significant constituents of medicinal and aromatic plants include alkaloids, volatile essential oils, tannins, and resins. These aromatic medicinal plants are characterized by their distinctive aroma and flavor, primarily attributed to their volatile oil content. These bioactive compounds serve two fundamental purposes: enhancement of food flavor and fragrance, and pharmaceutical applications as therapeutic additives (Rubin, 2004).

These aromatic compounds are distributed throughout different plant organs (flowers, leaves, stems, roots, barks, fruits, etc.) (Bruneton, 1999 Hijjawi et *al.*, 2004, Bakkali et al., 2008).

Fields of Application of Essential Oils:

Since ancient times, essential oils have been recognized for their significant therapeutic potential and have been utilized across various domains, including medicine, pharmacy (as medicinal preparations or drug excipients), cosmetics, cleaning and disinfection products, and the food industry as preservatives and flavoring agents (Adhia,2009).

- **Pharmaceutical and Medical Field:**

Essential oils have been used in therapeutics since antiquity due to their broad therapeutic applications. Their bioactive compounds exert notable effects on the human body. Despite significant advancements in the synthesis of organic and chemical compounds in pharmaceutical manufacturing, the use of essential oils in medicine remains indispensable, as they represent a valuable source of irreplaceable bioactive compounds.

Many essential oils are incorporated into pharmaceutical formulations and are also administered as infusions (e.g., peppermint (Mentha), thyme (Thymus), among others. Additionally, essential oils exhibit anti-inflammatory, antioxidant, deodorizing (Désodorisante), and insecticidal (Insecticides) properties (Mekhedmi,2014).

- **Food Industry:**

Essential oils are widely employed in food processing as flavorings, colorants, or fragrances. They are added to baked goods, confectioneries, dairy products, alcoholic and non-alcoholic beverages. Moreover, due to their antimicrobial properties, essential oils serve as natural preservatives, inhibiting microbial and fungal growth—an effect attributed to their terpenoid compounds.(Derabla,2016). Among the most widely used essential oils globally is orange essential oil (Zerdoum,2015).

- **Perfumery Industry:**

This sector represents one of the primary applications of essential oils, owing to their aromatic volatile compounds. Perfumes are classified into two types: alcohol-free fragrances and alcohol-based fragrances (Seddik،2010).

Practical Part

CHAPTER III

MATERIALS AND METHODS

Background: This study involved the collection of plant and animal samples from distinct regions in Algeria to analyze their unique characteristics and responsiveness to environmental conditions. Botanically, ***(Thymus serpyllum)*** (L.)was sampled from **El Kala**, a biodiverse coastal area near Algeria's eastern coast, where its humid climate and fertile ecosystems may enhance its bioactive properties. In contrast, Marjoram (*Origanum majorana*) (L.)was collected from **El Oued Province** in southeastern Algeria, characterized by arid, semi-desert conditions that likely shape its chemical composition and adaptive resilience.

Zoologically, mosquito samples were obtained from **El Oued**, a region conducive to insect proliferation due to its oases and seasonal watercourses, offering insights into their ecological behavior and public health implications. This comparative approach underscores the interplay between organisms and their environments, supporting applications in ecological research and sustainable resource utilization.

1.Description of Study Areas:

In this chapter, the selected study areas El Tarf Province (El Kala) and El Oued Province will be presented:

1.1 First Study Area (Oued Souf):

The term "Souf" is a Berber-derived word referring to a river and is synonymous with "valley." It denotes a cluster of palm trees surrounded by sand dunes. Traditionally, local communities relied on agriculture governed by the *Ghout* system—a traditional irrigation method involving excavated basins near the water table. This practice implies that settlements originated from excavation processes, complicating land development plans and necessitating costly interventions (M. N. O., 1999).

1.2 Geographical Location:

Biskra; and to the south, the province of Ouargla. The geographical coordinates of Oued Souf Province are approximately longitude 6.9667°E and latitude [Z]°N (Source: WordPress, D. Abbass).

The province covers an area of 54,573 km² (representing 0.29% of the national territory), with a population of approximately 134,699 inhabitants as of 2023 (National Census, 2023).

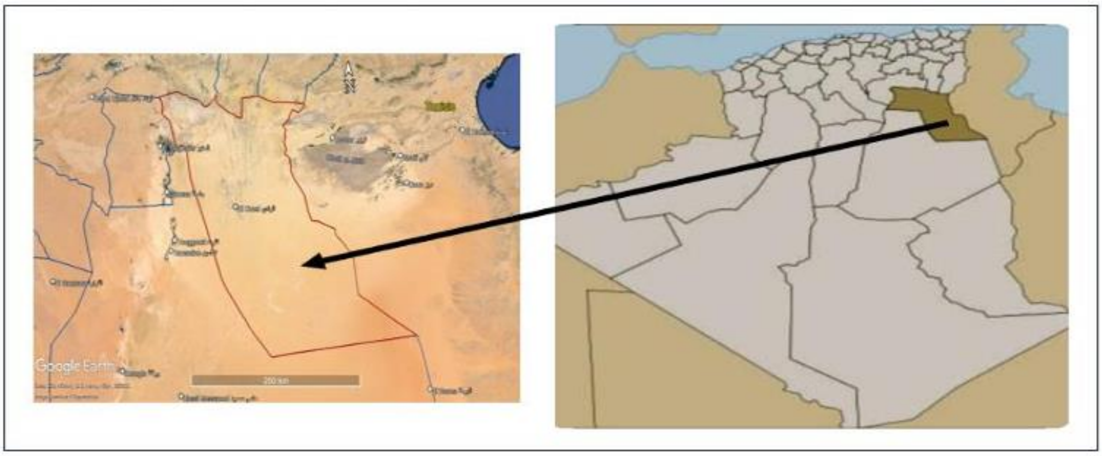


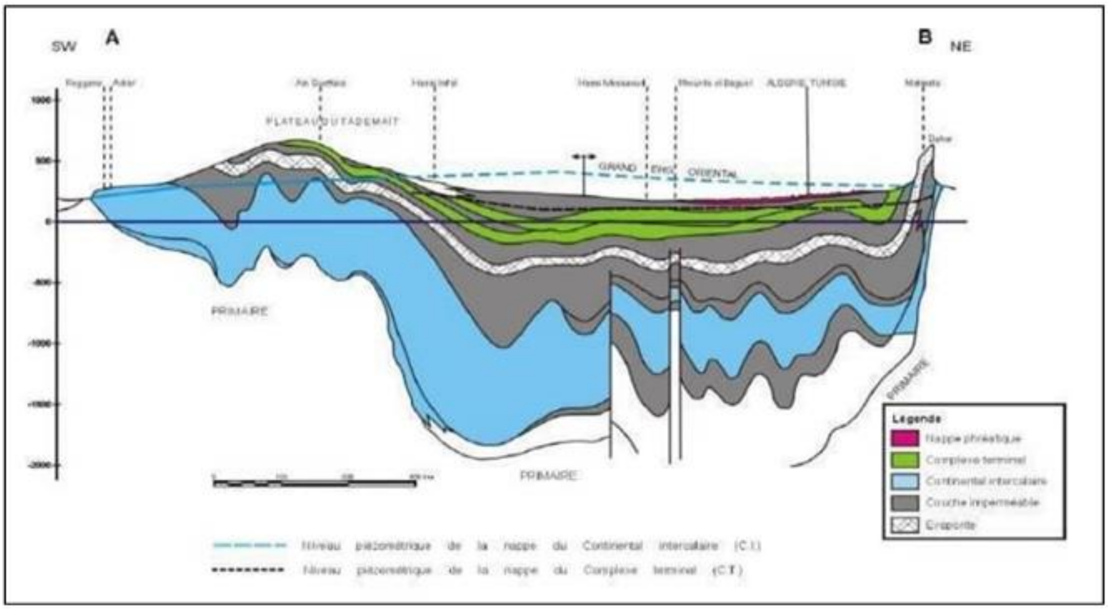
Figure 22: Geographical location of Oued Souf Province (southeastern Algeria) Document .17 (Wikiwand, 2024)

1-3 Cross-Section of Intercalary Continental Aquifers:

The groundwater reservoirs are located in **sandy-clay layers** and **continental sandstone formations**. These waters, known as **"Intercalaire"**, are ancient waters stored during **Quaternary Ice Age** rainfall periods.

Key Characteristics:

- **Geographic Extent:** Situated between the **Tassili Mountains** and the **Saharan Atlas,** covering an area of **600,000 km².**
- **Aquifer Thickness:** Reaches **hundreds of meters** in depth.
- **Hydrogeological Properties:**
  - **Artesian (confined) groundwater** with a flow rate of **200–250 L/s**.
  - **Temperature:** Ranges between **58–70°C**.
  - **Salinity:** Varies from **1.5–2 g/L.***(D.R.E, 2010)*.



.Figure 23: **The Hydrogeological Section Across the Sahara (UNESCO, 1972)**

1-4 Climatic Characteristics:

1-4-1Climate:

The Wilaya of El Oued is characterized by **a desert climate** due to its location within low-latitude zones near the tropics, resulting in **high temperatures** and a wind regime that generates **hot, dry air currents** (Ozenda, 1991). This climate is particularly marked by:

- **Low precipitation,** rarelyexceeding **80–100 mm annually,**
- **High evaporation rates,**
- **Extreme temperature fluctuations,** ranging from **below 0°C in winter to up to 50°C in summer.**

However, a distinct **local microclimate** exists within the palm groves, which helps **moderate climatic conditions** to some extent (Anonymous, 2006; A.N.D.I., 2013).

Key Climatic Factors

In practice**, temperature** and **precipitation** are among the most influential climatic factors, as their interaction shapes the **fundamental climate characteristics** of any region.

To determine the climatic features of the study area, we analyzed a **10-year climatic dataset (2011–2020),** which serves as the basis for understanding the regional climate.

1-4-2 Temperature:

El Oued Province experiences **extreme heat**, ranking among the **hottest provinces in Algeria**. In 2023, the **annual average temperature** reached **29.71 °C**. Climatic data reveal that the **hottest July** recorded an average temperature of **38 °C**, while the **lowest temperature** was observed in January at **10.2 °C**.

Table 02: Temperature data for the Oued Souf region (2023).
(Sources: National Office of Meteorology (ONM), El Oued; www.tutiempo.com

Annual	Dec	Nov	Oct	Sep	Aug	Jul	Jun	May	APR	Mar	Feb	Jan	**Month**
29.71	**12.5**	**18.4**	**25.1**	**30.2**	**33.5**	**38**	**31.6**	**25**	**21.9**	**19.1**	**19.2**	**10.2**	**Temp**

1-4-3 Precipitation:

Precipitation

Precipitation refers to all forms of water—liquid or solid—that fall from the atmosphere. It varies spatially and exerts significant effects on the distribution and composition of local organisms (Benazia, 2021).

Key Observations:

- **Annual precipitation: 29.71 mm**
- **Peak monthly precipitation: 10.16 mm** (May)
- **Months with no recorded precipitation**: January, March, July, September, and October (*Table N: .....*)

Table 03: Precipitation Data for the Oued Souf Region (2024)(Sources: National Office of Meteorology (ONM), El Oued; *www.tutiempo.com*)

Annual	Dec	Nov	Oct	Sep	Aug	Jul	Jun	May	APR	Mar	Feb	Jan	**Month**
29.71	**5.08**	**2.03**	**0**	**0**	0.5	**0**	1.02	10.16	4.06	**0**	6.86	**0**	P(mm)

1-4-4 Wind Patterns:

The prevailing wind directions in the Oued Souf region are **easterly and northeasterly**, with less frequent **westerly** and **southwesterly** flows. These winds are often coupled with **extremely high temperatures** (e.g., *Sirocco* winds), particularly during spring when wind intensity peaks.

Key Characteristics:

- **Seasonal Strength:** Strongest winds occur during the **palm tree pollination period** in spring.

- **Sand Transport:** Winds carry significant sand loads, imparting **a yellowish hue to the sky**.
- **Duration & Speed**: Episodes can persist for **up to 3 consecutive days**, with speeds ranging from **30–40 km/h** (Chikima et al., 2021).

Table 04: Monthly Mean Wind Speed in the Oued Souf Region (2023).

(Sources: National Office of Meteorology and Environment (ONMEI), El Oued; www.tutiempo.com)

Annual	Dec	Nov	Oct	Sep	Aug	Jul	Jun	May	APR	Mar	Feb	Jan	Month
11.4	9.5	10	8.8	11.6	11.3	11.5	14.4	15.1	13	12.2	9.9	9.3	Wind speed (km/h)

1-4-5Relative Humidity:

Relative humidity (RH) refers to the concentration of water vapor in the air, indicating the potential for precipitation, dew, or fog formation.

The Oued Souf region is characterized by **arid conditions**, with an **annual average RH of 36.9%.** Humidity exhibits significant seasonal variation:

- **Highest monthly average: 60%** (December**)**
- **Lowest monthly average: 17.5%** (July)

(See Table N for monthly data)

Table 05 : Monthly Averages of Relative Humidity (2023)

(Sources: National Office of Meteorology (ONM), El Oued; www.tutiempo.com)

Annual	Dec	Nov	Oct	Sep	Aug	Jul	Jun	May	APR	Mar	Feb	Jan	Month
36.9%	60	46	34.6	32.9	27.5	17.5	29.3	35.9	32	32.3	47.6	47.9	(%)RH

1-4-6Climatic Summary:

a. Ombrothermal Diagram Methodology

The climatic growing season and aridity thresholds were analyzed using the **ombrothermic diagram** method, as conceptualized by Bagnouls and Gaussen (Figure 24). This approach integrates monthly temperature and precipitation averages to delineate bioclimatic zones, with the x-axis representing months and the y-axis plotting both thermal and pluviometric parameters.

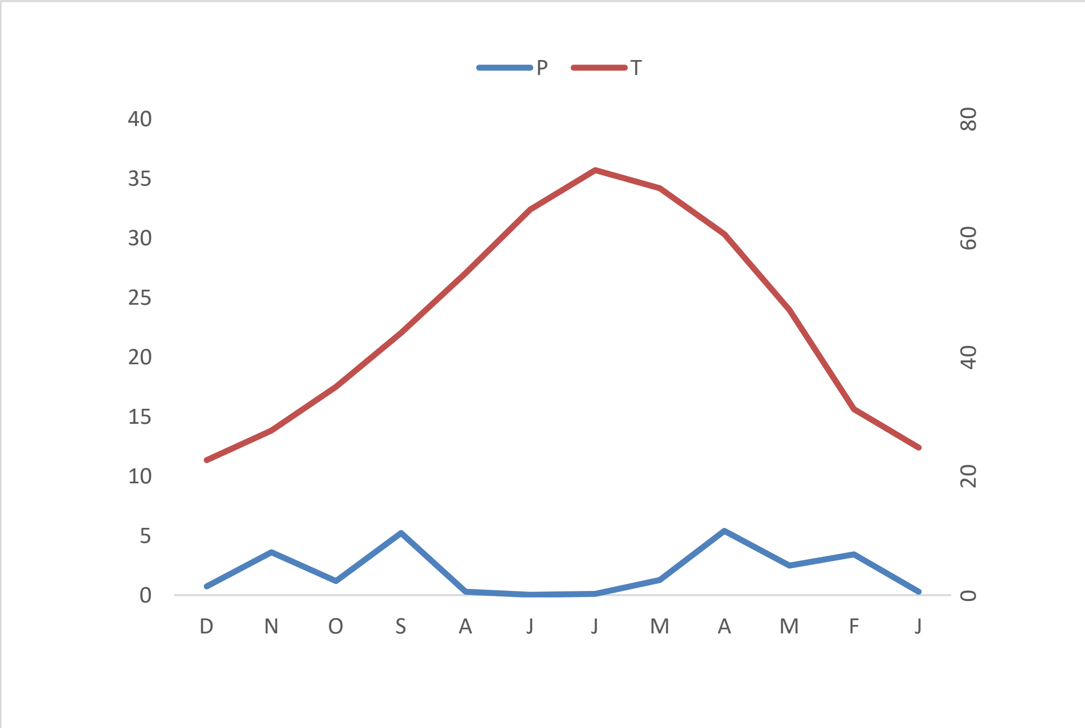


Figure24: Bagnouls and Gaussen umbrothermal diagram for the El Oued region (period 2014-2024)

The climatic data are used to draw up the Bagnouls and Gaussen umbrothermal diagram. These show that in 2024 and over the period 2014-2024, the Oued Souf region experienced drought throughout the year, from January to December **(Figure24).**

1-4-7 Bioclimatic Analysis Methodology:

To evaluate the monthly variability of dry and humid periods in the study area (2012–2023), we constructed a thermopluviometric diagram based on the Bagnouls-Gaussen method. The analysis reveals that arid conditions prevail year-round, with a brief humid phase lasting approximately one month (mid-January to mid-February in 2023).

Emberger's Bioclimatic Climagram:

The bioclimatic zonation of the region was determined using Emberger's pluviothermic quotient (Q2*Q*2), calculated as:

Q2=P×100M+m*Q*2=*M*+*mP*×100

where:

- P*P* = Mean annual precipitation (mm)
- M*M* = Mean maximum temperature of the hottest month (°C)
- m*m* = Mean minimum temperature of the coldest month (°C)

For the Souf region (2012–2023), the parameters are:

- P=29.71 mm*P*=29.71mm
- M=38.0 °C*M*=38.0°C
- m=10.2 °C*m*=10.2°C

The calculated Q2=3.6*Q*2=3.6 classifies the area within the Saharan bioclimaticstage with a semi-arid transitional subtype (*FigureN*).

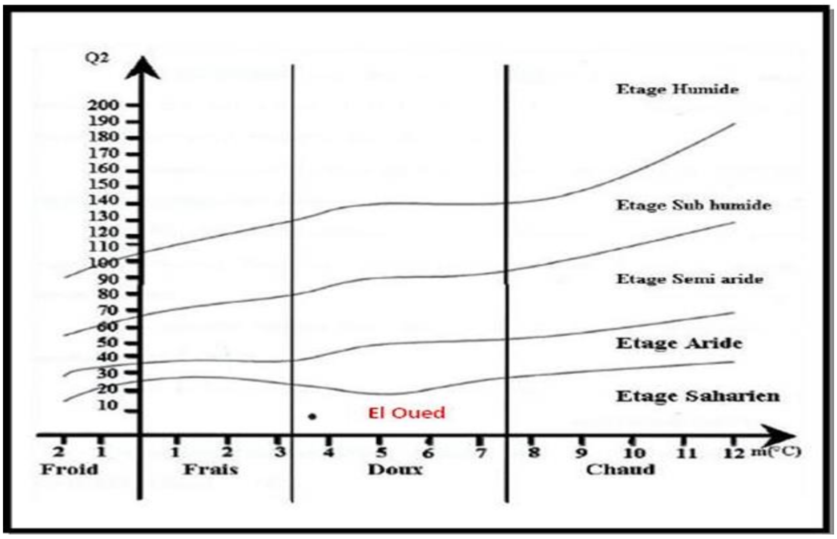


Figure 25: Position of the El Oued Region in Emberger's Pluviothermic Climagram(period 2012-2023)

1-4-8 the Biotic Factors:

The biotic factors analyzed in this study encompass the flora and fauna of El Oued region

:Overview of the Vegetation in El Oued

According to EMBERGER (1955), vegetation serves as a faithful indicator of climatic conditions, with plant cover playing a critical role in species distribution (Ozenda). As highlighted by Hilss (2007), the vegetation in Souf is characterized by sparse, low-density coverage, dominated by small, rapidly growing plants adapted to arid environments. Agricultural practices in the area primarily involve small-scale farms organized in funnel-shaped plots, locally termed "*ghout*" (Nadjah, 1971).

Faunal Diversity in El Oued:

Cattalization (1986) notes that desert ecosystems typically host fewer species per unit area compared to other biomes. Nevertheless, the Algerian Sahara, including El Oued, supports a surprising diversity of invertebrates, fish, amphibians, reptiles, and mammals, though research remains limited (LE Berre, 1989). The region's fauna predominantly comprises Mediterranean and Sudanese-origin arthropods and reptiles (Voisin, 2004). Key taxa include the Agamidae family, represented by *Acanthinurus acanthinurus* (Bell, 1825).

a. Dominant Invertebrates:

Invertebrate surveys (BEGGAS, 1992; Khechkouche et al., 2011; Alia, 2008) identified 113 arthropod species in El Oued , with Tenebrionid beetles such as *Pimelia angulata* and *Ateuchus sacer* (Linnaeus, 1758) being prevalent. Fish diversity is limited to the Poeciliidae family, notably *Gambusia* species (Le Berre, 1989).

b. Avian Species:

Avian inventories (Isenmann & Moali, 2000) documented 28 bird species across 13 families in Wadi Souf. The Sylviidae family, including *Sylvia deserticola* (Tristram, 1859) and *Sylvia nana* (Scopoli, 1769), dominates the region.

c. Reptilian Diversity:

Reptilian surveys (Le Berre, 1990; Frazin, 2004; Khachkhouch & Mostefa, 2008) recorded 20 species spanning 7 families and 6 orders, reflecting a mix of Mediterranean and Saharan adaptations.

2. Second Study Area (El Kala):

Description and Localization of El Kala Region:

2-1 Geographical Location:

El Kala is a coastal city bordering Tunisia, ranking as the second-largest urban center in El Tarf Province. It hosts the El Kala National Park (PNEK), one of Algeria's most significant ecological complexes and a key protected area in the western Mediterranean Basin.

Established by governmental decree in 1983, the park was designated a UNESCO Biosphere Reserve in 1990 under the Man and Biosphere (MAB) program (Messikh, 2016; Alayat, 2024).

Geographical Coordinates:

The study area is located in the extreme northeastern sector of Algeria (36.89°N, 8.43°E) at an elevation of 30 meters above sea level, within the Tell Atlas mountain system. It spans an area of 292 km², representing approximately 26% of the total land area of El Tarf Province, and is situated approximately 70 km east of the city of Annaba.

The region is bounded by the Algerian-Tunisian border to the east, the Mediterranean Sea to the north, Annaba Province to the west, and the provinces of Souk Ahras and Guelma to the south.

This area holds significant biogeographical importance due to its transitional position between Mediterranean coastal ecosystems and inland mountainous terrains, making it a critical zone for ecological and environmental research (Messikh, 2016; Loucif et al., 2020). Its unique geographical and climatic characteristics contribute to its rich biodiversity and ecological sensitivity, warranting further scientific investigation.

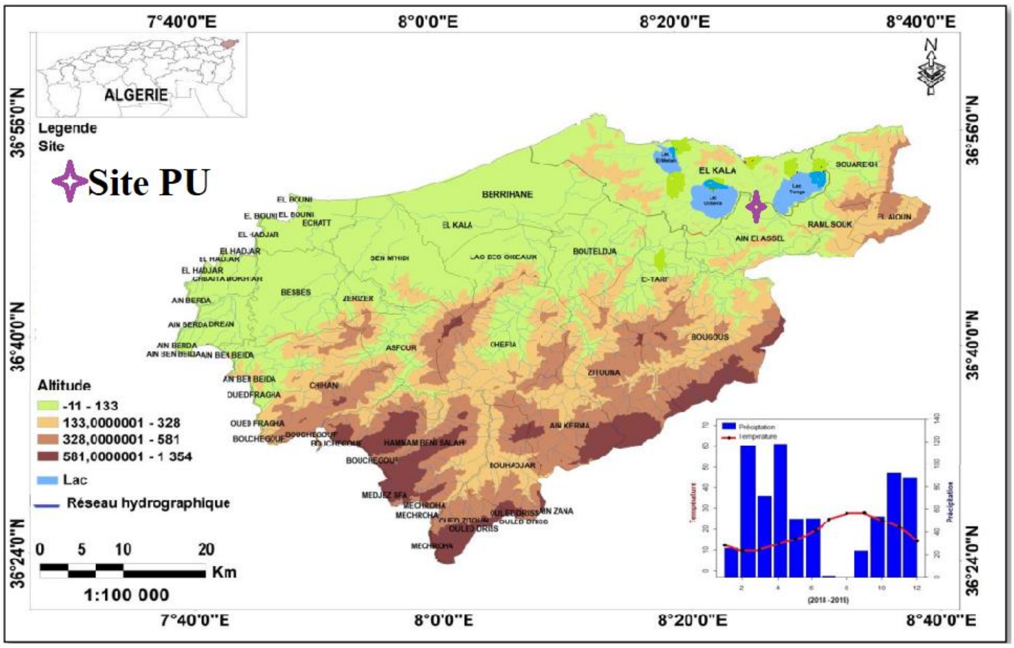


Figure 26 : Geographic Map Illustrating the Distribution of PU Sites in the El Kala Region, Algeria (Alayat, 2019).

2-2 Characteristics:

2-2-1 Topography:

The region exhibits diverse topography, characterized by successive depressions, some of which are filled with water to form lakes or marshes. Elevations such as domes and cliffs are common, often densely covered with vegetation (Hammouda & Taher, 2012).

2-2-2 Water Resources:

The El Kala National Park is surrounded by a significant hydrographic network, including lakes such as Lake El-Melah and Lake Oubeira, in addition to Lake Tonga and Lake Mexa. These water bodies are fed by natural springs located in the southeastern part of the region (Benyacoub et al., 1998).

2-2-3 Climate:

Located in the extreme northeastern part of Algeria, the PNEK lies at the interface between the Mediterranean Sea to the north and mountainous barriers to the south. This geographical setting contributes to climatic irregularity in the area. The study region experiences a Mediterranean climate marked by strong seasonal contrasts. A mild, rainy season occurs between October and April, during which temperatures never drop below 0°C, and annual precipitation exceeds 800 mm in wet years. Summers are hot and dry from June to September, while autumn and spring are relatively short transitional seasons

❖Climate Characterization:

For a specific station, climate characterization involves identifying dry and wet periods using the Gaussen ombrothermic diagram, along with positioning the station within the Elmagram to determine its bioclimatic phase (Djamai, 2020).

❖Gambesombrothermic and Gaussen Pluviometric Diagrams

The Gaussen ombrothermic diagram utilizes monthly precipitation and temperature data to delineate wet and dry periods in any region (Dajoz, 2003).

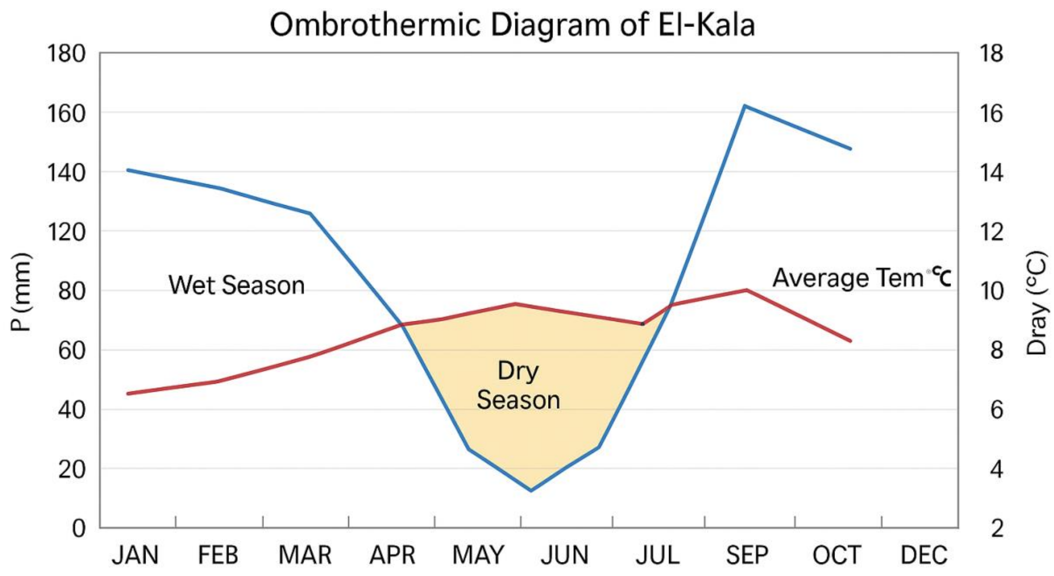


Figure 27: Image Gaussen Ombrothermic Diagram of the El-Kala Region OverFor twenty years.(1988-2018).

Climatic Characterization Using Gaussen and Emberger Methods

The results obtained from the GAUSSEN thermic diagram revealed two distinct seasons in P.N.E.K:

- A **dry season** (May to September) characterized by precipitation deficits relative to evapotranspiration rates
- A **humid/rainy season** (September to May) with sufficient precipitation

Emberger Climatic Classification System:

The Emberger climagram (Dajoz, 2003) enables classification of Mediterranean climate types, with particular relevance in regions showing increased precipitation due to humid conditions.

Key Climate Indicator:

The **Emberger pluviothermal quotient (Q_2)** (Emberger, 1955) serves as:

- A robust Mediterranean climate index reflecting north-south gradients
- A synthetic bioclimatic parameter incorporating:
    - Annual precipitation (P)
    - Temperature extremes:
        - Mean minimum of coldest month (m)
        - Mean maximum of hottest month (M)

Calculation Methodology (Djamia, 2020):

The Emberger quotient is calculated as:

$$Q_2 = (2000 \times P) / (M^2 - m^2)$$

Where:

- P = Mean annual precipitation (mm)
- M = Mean maximum temperature of hottest month (°C)
- m = Mean minimum temperature of coldest month (°C)

Scientific Value:

1. Precise bioclimatic zoning of Mediterranean ecosystems
2. Quantitative assessment of climate gradients
3. Foundation for ecological and hydrological studies
4. Essential tool for climate change impact evaluation in protected areas

Table 06: Values of the Emberger Pluviometric Coefficient in the El Kala Region (1988-2018 Period)Meteorological Station (Lac des Oiseaux Weather Station**).**

Humid to Sub-humid	Bioclimatic Stage
98.80	Q_2
7.20	m (°C)
32.50	M (°C)
723	P(mm)

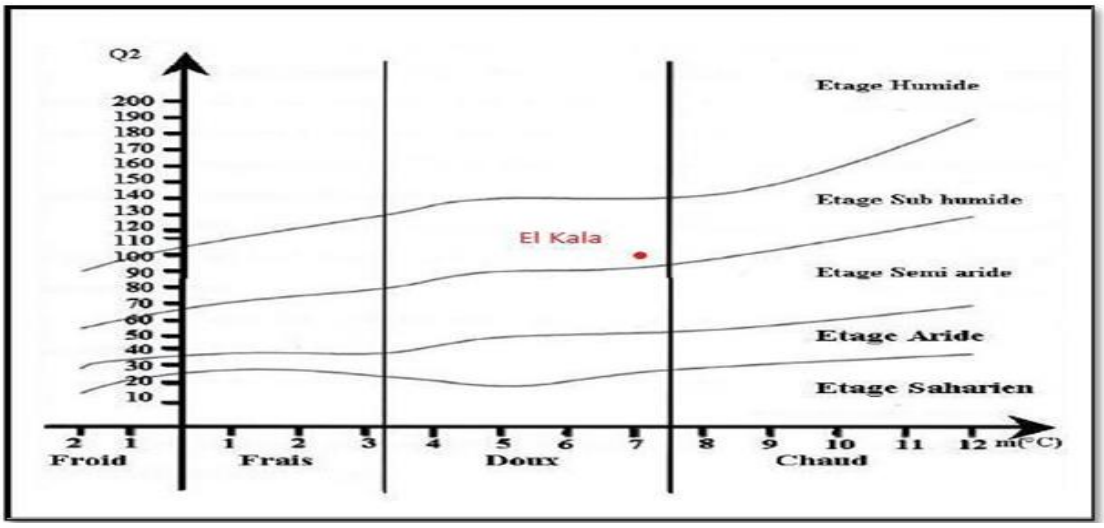


Figure 28 : Bioclimatic zone of the El-Kala region according to Emberger's climagram (1988-2018 period; Lac des Oiseaux meteorological station).

Biotic Factors:

The study conducted in El Kala National Park on sustainable development and biodiversity revealed the following:

Overview of Plant Diversity in El Kala:

The vegetation cover comprises **1,590 species**, including:

- **27 protected species**
- **80 endemic species**
- **20 species listed on the IUCN Red List**
- **175–177 bryophyte species,** of which **52 are protected**

Additionally, the park hosts a reserve containing **148 medicinal plants** belonging to **55 families** and **118 genera** (Sarri, 2017).

Overview of Faunal Diversity in El Kala:

The park harbors **718 species**, including:

- **17 mammal species**
- **3 reptile species**
- **13 insect species**
- **87 bird species**
- **9 marine fish species** (Sarri, 2017).
-

3.Study on the Effect of Essential Oils and on Mosquito Larvae:

3-1 Biological Materials

Larval Collection: Aedes caspuis (pallas,1771) larvae were sampled from multiple breeding habitats across the study area.

Plant Material: Medicinal plants endemic to the region were selected for extraction**. Hydrodistillation** and **standard extraction methods** were employed to obtain plant extracts and essential oils.

3-2- Animal Subjects:

3-2-1. Collection of Mosquito Specimens:

For this study, the *A.*mosquito species was selected based on the following characteristics:

- **High prevalence** in the study region.
- **Disease transmission capacity**, particularly as a vector for blood-borne epidemics.
- **Rapid reproduction rate**, which promotes the emergence of pesticide-resistant mutant generations.

Female Aedes caspius mosquitoes are known for their particularly painful bites and are considered a significant nuisance, especially on rocky beaches. Beyond their direct impact, they can also serve as vectors for various pathogens, including animal filarial worms and arboviruses such as Tahyna virus and myxomatosis virus. They may be naturally infected with West Nile virus and are involved in the transmission of tularemia (Joubert et al., 1967; Brunhes et al., 2001; Becker et al., 2003). Additionally, studies conducted in southern Spain have detected the presence of avian Plasmodium in this species (Ferraguti et al., 2013; WHO, 2023).

The systematic classification of *Aedes caspius* mosquitoes was first established by Pallas (1771) as follows:

Kingdom: Animalia

Subkingdom: Metazoa

Phylum: Arthropoda

Subphylum: Hexapoda

Class: Insecta

Subclass: Pterygota

Infraclass: Neoptera

Superorder: Holometabola

Order: Diptera

Suborder: Nematocera

Family: Culicidae

Genus: *Aedes*

Species: *Aedes caspius* (Pallas, 1771)

3-2-2 Mosquito Sampling Protocol:

Specimens were collected based on mosquito abundance, species diversity, and site accessibility. Sampling sites included Lake Souk Libya and other locations within the study area.

Figure 29: Image and location of the area from which we took the mosquitoes(Personal image, 2025**)**

The samples were collected using a dipper (Graff, 1965; dipping method) (Rioux et al.), the standard method for obtaining mosquito larvae. Various types of collection dippers, either plastic or metal, with a capacity of 1 liter, are employed (Croset et al., 1975; Subra, 1976).

Figure 30 :**Image** Depiction Automated-size dipper (Personal image, 2025)

Mounting and Identification:

Only larvae that have reached the fourth stage are subject to reliable identification. The mounting process itself aims to observe specimens under an optical microscope. Larvae at this stage, collected from the same breeding site, undergo 48-hour baths in a 10% NaOH solution or are placed in alcohol to render the chitin transparent and destroy tissues attached to the cuticles, thereby facilitating microscopic observation. Subsequently, the larvae are rinsed three times with distilled water and mounted on slides and coverslips in a drop of glycerol, fixed with transparent nail polish for systematic identification (Bendali-Saoudi, 1989).

Figure 31: Image Depiction of fourth-instar mosquito larvae (Personal image, 2025).

4- Second Biological Materials:

Two medicinal plants, *Origanum majorana* (marjoram) and *Thymus serpyllum* were selected.

Plant Selection Criteria

The plants were chosen based on the following criteria:

1. Their application in **alternative medicine**.
2. Their **traditional uses** by local communities.
3. Their absence near **mosquito breeding sites** (to minimize ecological interference).
4. Their utilization in the **cosmetic industry** (e.g., perfumes, natural oils, and pharmaceutical preparations).
5. Their status as **naturally growing species** with high abundance in their native harvesting locations.

Figure 32: Image Illustration depicting the desiccation phase of plant *Origanum majorana* (marjoram) (Personal image, 2025),

Figure 33: Image Illustration depicting the desiccation phase of plant *Thymus serpyllum*. (Personal image, 2025)

5- Study Phases:

The study aimed to evaluate the efficacy of essential oils from two medicinal plants,*Thymus serpyllum* and *Origanum majorana* (marjoram), harvested in the regions of El Kala and Oued Souf. The study spanned approximately two to three months and was divided into three phases as follows:

5-1. Phase 1: Harvesting, Drying, and Extraction of Essential Oils

The selected medicinal plants were harvested, dried, and subjected to essential oil extraction. This phase was conducted in the biology laboratories of the University of El Oued - Martyr Hamma Lakhdar.

5-2. Phase 2: Mosquito Sample Preparation

Following the near-completion of oil extraction, mosquito larvae were collected and prepared for the final phase of the study: **biological control testing**.

5-3. Phase 3: Testing Phase 1 Products on Mosquito Larvae

The extracted essential oils were tested against a species of mosquito larvae to assess their insecticidal potential.

Preparation of Essential Oil Extracts:

The essential oils from (Thymus serpyllum) and *Origanum majorana* (marjoram) were extracted separately using **hydrodistillation**. For each plant, 100 g of dried powdered plant material was immersed in 600 mL of distilled water within a distillation flask. The mixture was heated to boiling using a **Clevenger apparatus**. The distillation process was conducted for 3–4 hours at a rate of 3 mL/min. The first distillate droplets were collected, and the resulting essential oil was stored in amber vials under dark conditions at 4°C to prevent degradation.

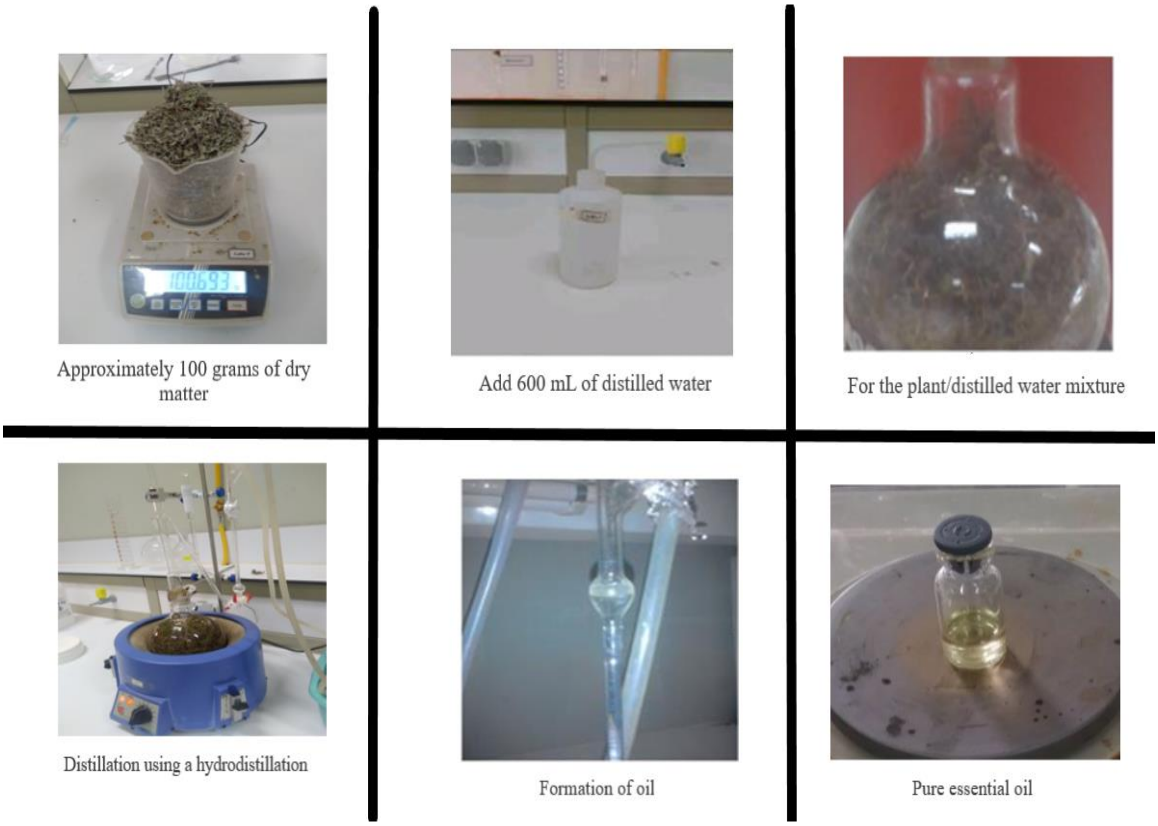


Figure 34: Essential oil extraction method (personal photo;2025)

6- Essential Oil Yield Percentage:

The essential oil yield (Rd%) was calculated relative to the dry plant material (expressed as mL/50 g of dry matter). The yield is defined as the ratio of the extracted oil mass (M′*M*′) to the initial dry sample mass (M*M*), expressed as a percentage using the following formula (adapted from [Author], 2010):

Rd%=(M′M)×100Rd%=(*MM*′)×100

Where:

- **Rd%**: Essential oil yield percentage.
- **M′*M*′**: Mass of extracted essential oil (g).
- **M*M***: Mass of dry plant sample (g).

This procedure was applied to *both and Origanum* majorana (Marjoram). The extracted essential oils were stored in airtight amber glass vials, further wrapped in aluminum foil to prevent photodegradation or volatilization under light exposure.

Evaluating the efficacy of extracted oil in biological control of mosquito larvae:

- Mosquito Control Using Essential Oils of Marjoram and Thyme:

In this experiment, 16 containers (90 mm diameter) were prepared, each containing 10 fourth-instar mosquito larvae. Larvae were transferred using a plastic pipette. Each container was filled with 100 mL of distilled water, along with food provisions and an emulsified essential oil solution (prepared with Tween 80). The tested concentrations were: 25 µL/L, 125 µL/L, 250 µL/L, 350 µL/L, and 500 µL/L. For the control group, larvae were placed in containers with a 1% Tween 80 solution. Three replicates were prepared for each concentration and the control. Mortality rates were assessed at 24, 48, and 72 hours post-exposure, with hourly recordings of dead larvae.

Figure 35 : Image illustrating the application of essential oils (marjoram and thyme) and Tween emulsifier to mosquito larvae in experimental bioassays (Personal image, 2025).

Table 07 : Essential Oil Concentrations Used in Scientific Experiments:

Concentration (µL/mL)	**Group**
0.000 (Tween 80 only)	**Control**
0.025	C1
0.125	C2
0.250	C3
0.375	C4
0.500	C5

After a designated time interval, the number of dead and live larvae is recorded. The efficacy percentage against larvae, relative to the control group, is calculated using the following formula:

Efficacy (%) = (Number of dead larvae / Total number of larvae) × 100

7- Histological study:

Histological Processing of Mosquito Larvae:

- **Test Organism:** 4th instar larvae of *Aedes caspius*.
- **Treatment:** Larvae were exposed of thyme (*Thymus sp.*) and marjoram (*Origanum majorana*) essential oils for 4, 6, 12, and 24 hours.

o Post-exposure, surviving larvae were fixed and dehydrated using an automated tissue processor (Leica TP1020) through graded ethanol baths (70%–100%) over 48 hours .

o Tissues were paraffin-embedded at 60°C , sectioned at 4 µm thickness using a microtome and stained with H&E .

o Slides were analyzed under a Leica DM1000 light microscope (1000x magnification; to evaluate physiological alterations in midgut cells (Bancroft & Stevens, 1996).

-1 Sample Preparation:

o Prior to histological sectioning, larval heads and tracheal siphons were excised using a scalpel.

o Samples were dehydrated to remove intracellular and extracellular water through sequential immersion in seven ethanol baths (70% to 100% ethanol).

o Tissues were cleared in three xylene baths to eliminate ethanol residues and enhance transparency.

Figure 36 : Image illustrating the embedding cassette (for tissue samples) and the Automatic Tissue Processor Unit

2. **Paraffin Embedding:**

o Dehydrated samples were embedded in paraffin wax (melting point: 58–60°C) using a histological oven. Paraffin infiltration ensures homogeneous tissue support for sectioning.

Figure 37 : Imageillustrating the embedding cassette (for tissue samples) and the Automatic Tissue Processor Unit (Personal image, 2025)

3. **Sectioning and Staining:**

- Embedded tissues were sectioned at 5 µm thickness using a microtome.
- Sections were stained with hematoxylin and eosin (H&E) for cellular visualization, mounted on slides, and examined under a light microscope (400x magnification) to assess midgut epithelial damage in mosquito larvae.

Figure 38 : Images illustrating the cutting process (Boudebia Ouafa ,2024)

Figure 39 : Images illustrating **Staining: Hematoxylin and Eosin (H&E)**

Coloration(Personal image, 2025)

-After the slides are prepared, they can be observed under a Light Microscop**e** for analysis.

Figure 40 : Images showing mosquito samples finally under the microscope(Personal image, 2025)

All Materials and Reagents :

1. **10% Formalin** (Formaldehyde solution)
2. **Ethanol**
3. **95% Ethanol**
4. **Xylene or Toluene**
5. **Paraffin Wax**
6. **Embedding Cassettes**
7. **Microtome**
8. **Glass Slides**
9. **Slide Warmer**
10. **Hematoxylin**
11. **Eosin**
12. **Distilled Water**
13. **Mosquito larvae/pupae** (Aedes/species).

CONCLUSION

For centuries, humanity has suffered from the adverse effects of mosquitoes - diminutive yet formidable vectors of deadly diseases including malaria, dengue fever, West Nile virus, Zika, and other viral and parasitic infections. Their remarkable adaptive capabilities and global distribution have established them as a persistent threat to both public health and ecological equilibrium.

These threats have compelled the development of diverse control strategies, ranging from chemical pesticides (initially effective yet ultimately ecologically damaging) to physical interventions like larval habitat elimination, which similarly incurred biodiversity costs. These repeated failures in establishing sustainable solutions have driven researchers toward more ecologically conscious approaches: environmentally-friendly biological control.

Within this framework, medicinal and aromatic plants have emerged as natural alternatives, containing bioactive compounds capable of targeting larval populations while preserving ecosystem integrity. Leveraging Algeria's rich phytodiversity, this investigation focused on wild thyme (*Thymus serpyllum)* and marjoram (*Origanum majorana*), extracting their essential oils and evaluating larvicidal efficacy. Results demonstrated significant larval mortality rates, confirming their potential as promising candidates for developing effective, eco-friendly biopesticides.

The essential oils of *Thymus serpyllum* and *Origanum majorana* demonstrated significant larvicidal activity against *Aedes caspius* after 24, 48, and 72 hours of exposure. Among the two, thyme essential oil proved to be more effective, inducing total larval mortality as early as 48 hours, even at low concentrations. In contrast, marjoram oil required up to 72 hours to achieve comparable results, particularly at lower doses.

Toxicological assessments confirmed a dose-dependent mortality response, with median lethal concentrations (LC_{50}) of 39.62 µL/L for *T.serpylum* and 175 µL/L for *O. majorana,* indicating the greater potency of thyme. This effectiveness is characterized not only by a faster action but also by a more consistent biological response at lower concentrations.

Furthermore, histological analyses of longitudinal sections of the midgut and caeca in 4th instar larvae revealed marked cellular alterations, particularly in larvae treated with *T. serpyllum* . These digestive tissue damages confirm the direct toxic effect of the oil on larval midgut cells and support its potential as a larvicidal agent.

In conclusion, *Thymus serpyllum* stands out as a strong candidate for biological mosquito control strategies, especially when rapid action is required. The use of such plant-based extracts offers a promising alternative to chemical insecticides, combining high larvicidal efficacy with reduced environmental impact

These findings open promising avenues for the development and use of plant-based bioinsecticides, particularly in:

- ✧ Integrated mosquito control programs, as natural alternatives or complements to synthetic chemical insecticides.
- ✧ Eco-friendly formulations, such as ready-to-use larvicidal sprays, gels, or diffusers based on essential oils.
- ✧ Sustainable vector management strategies, especially in regions facing resistance to conventional larvicides.

To maximize the value of these essential oils, the following recommendations are proposed:

Recommendations for Future Research and Implementation

Based on the findings of this study regarding the larvicidal effects of Thymus serpyllum and Origanum majorana essential oils, the following scientific and practical recommendations are proposed:

1-Biological and Ecological Research:

- ✧ Extend testing to other mosquito species (Culex, Anopheles) to determine broad-spectrum efficacy.
- ✧ Evaluate larvicidal impact on all life stages (eggs, pupae, adults) for a comprehensive insecticidal profile.
- ✧ Assess the environmental safety of the oils on non-target organisms (e.g., aquatic life, pollinators).
- ✧ Investigate degradation rates of the oils in aquatic ecosystems to evaluate persistence and stability.

2-Agronomic and Community Development:

- ✧ Encourage local cultivation of T. serpyllum and O. majorana in rural/arid areas like El Oued as a sustainable source of bioinsecticides.
- ✧ Integrate these plants into public health initiatives to reduce dependency on synthetic chemicals.

3-Monitoring and Field Validation :

- ✧ Conduct field trials in natural mosquito breeding sites to validate laboratory results under real-world conditions.

- Establish resistance-monitoring systems to ensure long-term effectiveness and guide future vector control strategies.

References

References in Arabic :

1) ادحيه مصطفى النباتات الطبية في مناطق الجلف ، بوسعادة والمسيل . . دراسة نبات القزحPituranthosأنواعه التركيب الكيميائي والنشاطية البيولوجية للزيوت الطيارة للسيقان شهادة دكتورة جامعة فرحات عباس سطيف 2009 .

2) الشماع علي عبد الحسين (1989) . العقاقير وكيمياء النباتات الطبية دار الكتب للطباعة والنشر بيروت العراق.

3) أمقران الصادق استخلاص وتحليل الزيت الطيار لنبات Ciste de Montpellierبواسطة الكروماتوغرافيا الغازية لمرتبطة بمطيافية الكتلة مذكرة لنيل شهادة أستاذ التعليم الثانوي المدرسة العليا للاساتدة القبة القديمة الجزائر 2015 .

4) المليحان، م. ع (2024). دراسة مورفولوجية لأوراق نبات البردقوش (Origanum majorana) واستخداماته الطبية في علاج قشرة الرأس والصناعية كمادة حافظة في اللحوم. الواحة الطلابية للجامعة الوطنية الخاصة.

5) بوخبتي حبيبة النباتات الطبية المتداولة في المنطقة الشمالية لولاية سطيف دراسة تشريحية لنوعين من جنس Mentheوالنشاطية ضد البكتيرية لزيوتهما الأساسية مذكرة الماجستير جامعة فرحات عباس 2010.

6) جميلة شحاته سليم, محمد سمير و حافظ محمد متولى بعوض جنس الكيولكس Culexوعلاقته بنقل مسبب مرض داء الفيل 20/07/2020 .

7)زردوم سليمان دراسة تشريحية النبات Artemisia campestriفي منطقة أريس ودراسة النشاطية ضد بكترية والضد التأكسدية لزيتها الأساسي جامعة فرحات عباس سطيف مذكرة ماجستير في بيولوجيا وفيزيولوجيا 2015.

8) مخدمي نور الهدى استعمال المستخلصات المائية لنبتتي Matricaria pubscensو Pituranthos chloranthosكمعطرات طبيعية للجبن "أمير ، ودراسة النشاطية ضد البكتيريا لزيوتهما العطرية مذكرة الماجستير جامعة فرحات عباس سطيف 2014 .

9) موسوعة النباتات الطبية في دولة الإمارات العربية المتحدة مركز زايد لبحوث الأعشاب الجزء الأول 2005156ص

Foreign references :

1) Adam M., Dobiáš P., Pavlíková P., Ventura K. (2009)- Comparison of solid-phase and single-drop microextractions for headspace analysis of herbal essential oils. Cent. Eur. J. Chem, 7(3): 303-311.

2) A. D. Raina (2011). Essential oil composition of origanum majorana and organum Vutgar. Hirturm growing in India .chem.Net comp.47: 1015-1017 .

3) Alayat, M. S. (2024–2025). Systematic and bio-ecological study of Culicidian fauna in two different regions (Humid & Arid) of Algeria: Biological control against two mosquito species [Unpublished doctoral dissertation]. Algerian.

4) Alcaraz, F., & Delgado, M. J. (1999). Thyme-brushwood communities (tomillares) of semiarid South-eastern Spain.

5) ALLANE T.(2009)- Etude de pouvoir antioxydante et antibactérienne de quelques espèces végétales locales alimentaires et non alimentaires., mémoire de magister.Université Badji Mokhtar – Annaba..

6) Al-Mekhlafi, F. A. (2017). Larvicidal, ovicidal activities and histopathological alterations induced by *Carum copticum* (Apiaceae) extract against *Culex pipiens* (Diptera: Culicidae). Saudi Journal of Biological Sciences. https://doi.org/10.1016/j.sjbs.2017.02.010

7) Alves, R., et al. (2010). Mosquito vectors in tropical urban areas. *Journal of Medical Entomology, 47*(3), 45-53.

8) AL-Sheibany., Ikbal, S., Kadeem, K.H and A.S. Abdulla. (2005). Isolation and Identification of Volatile oils from Iraq Thyme (Thymus spicata) and study the antimicrobial activity Babylon University- Journal of Chemistry-volume 18, 289-298.

9) A.N.D.I. (2013). Agence Nationale de Développement de l'Investissement – Algeria. Retrieved from http://www.andi.dz

10) A.O.A.C(2006). Official methods of analysis. Horwitz, W. L., George, W. and Latimer, Jr.(18th ed). Washtington, De: Assoctiation of official Agricultural chemists.

11) Aouir, F., Chaibi, R., Merabti, B., Benhissen, S., Sifi, I., & Gouzi, H. (2025). Volatile oils of Thymus serpyllum and Artemisia absinthium: GC-MS analysis and insecticidal activity against Culiseta longiareolata. [Journal Name], [Volume(Issue)], [page range]. https://doi.org/[DOI-if-available]

12) Arafa Nermeen M, Amer Ahmed M, Helmy Naiera M, El-Shenawy R. Callus culture, phytochemical analysis and antioxidant and cytotoxic effects of marjoram (Origanum majorana). Egypt Pharma J 2023; 22: 545-550.

13) Arulselvan, P., et al. (2016) 'Role of antioxidants and natural products in inflammation, Oxidative Medicine and Cellular Longevity, 2016, 5276130. doi: 10.1155/2016/5276130.

14) Aziz, E. E.; Hendawi, S. T.; Ezz El Din, A. and Omer, E. A. (2008). Effect of soil type and irrigation intervals on plant growth, essential oil yield and constituents of Thymus vulgar-is plant. American-Eurasian J Agric. & Environ. Sci., 4(4): 443-450.

15) Baba Aissa. F., 2011., Encyclopédie des plantes utiles.lére édition par Elmaarifa.p218.

16) Bakkali F., Averbeck S., Averbeck D., Idaomar M. (2008)- Biological effects of essential oils. - Review- Food and Chemical Toxicology, 46, 446-475.

17) Bancroft, J. D., & Stevens, A. (1996). Theory and Practice of Histological Techniques (4th ed.). Churchill Livingstone.

18) Becker, N., Petric, D., Zgomba, M., Boase, C., Madon, M., Dahl, C., & Kaiser, A. (2003). Mosquitoes and Their Control. Kluwer Academic/Plenum Publishers.

19) Beloued A., 2009. plantes médicinal d'Algérie 5éme édition. office des publication universitaire. P 284.

20) Bhatt, S., et al. (2013). The global distribution and burden of dengue*Nature* -. Fontenille, D., & Jupp, P. G. (2010). *Anopheles* species and malaria transmission in Africa.

21) Bina, F. and Rahimi, R. (2016) 'Sweet marjoram: a review of ethnopharmacology, phytochemistry, and biological activities', Journal of Evidence-Based Complementary & Alternative Medicine, 22(1), pp. 175-185. doi: 10.1177/2156587216650793.

22) Brunhes, J., et al. (1999). Les moustiques de l'Afrique méditerranéenne: Identification, biologie, écologie. IRD Éditions.

23) Bruneton J. (1999)- Pharmacognosie, phytochimie, plantes médicinales. 3ème édition, éd.TEC et DOC, Paris.

24) Burt, S., 2004., Essential oils: their antibacterial properties and potential applications in foods a review. Int. J. Food Microbiol. 94, 223-253. essentielles d'origans (Lamiaceae) cultives issus de graines d'origine méditerranéenne., thèse doctorat.. Université Blaise Pascal .

25) Centers for Disease Control and Prevention (CDC). *(2024). Mosquito-borne diseases surveillance report* .

26) Chickoune A., 2007., Huiles essentielles de thym et d'Origan étude de composition,de l'activité antioxydant antimicrobienne mémoire de magister. Institut nationale agronomique El Harrach- Alger.

27) Chikh, S., & Djema, K. (2020). Literature review on mosquitoes (Diptera: Culicidae) in Algeria and control methods used [Unpublished master's thesis]. University of Mouloud Mammeri of Tizi-Ouzou, Faculty of Biological Sciences and Agronomic Sciences.

28) Da Porto C., Decorti D. (2009)-Ultrasound-assisted extraction coupled with under vacuum distillation of flavour compounds from spearmint (carvone-rich) plants: Comparison with conventional hydrodistillation. Ultrasonics Sonochemistry, 16, 795-799.

29) De Oliveira, A.A. et al. (2021) Larvicidal, adulticidal and repellent activities against *Aedes aegypti L.* of two commonly used spices, O*riganum vulgare L.* and *Thymus vulgaris L.*, South African Journal of Botany, 140, pp. 17–24.

30) Derabla Cherifa Zamouche Amel master université Guelma étude del'activité antibactérienne des extraits alcooliques d'ail (allium sativum)et de cannelle (cinnamomum zelanicum).2016p10 .

31) Djamai, K. (2020). Climatic characterization of northeastern Algeria using Gaussen diagrams. Climatic Methods Review .

32) Dris, R. (2019). Étude des vecteurs de maladies en Algérie [Thèse de doctorat, Université d'Alger].

33) DUBOIS J., MITTERAND H., D DAUZAT A., 2006., Dictionnaire étymologique et historique du français – Larousse.

34) El-Akhal, F., Guemmouh, R., Maniar, S., Taghzouti, K., & El-Ouali Lalami, A. (2016). Larvicidal activity of essential oils of Thymus vulgaris and Origanum majorana (Lamiaceae) against the malaria vector Anopheles labranchiae (Diptera: Culicidae). International Journal of Pharmacy and Pharmaceutical Sciences, 8 (3), 1–6.

35) Elshafie, H. S., & Camele, I. (2017). An overview of the biological effects of some Mediterranean essential oils on human health. BioMed Research International, 2017, Article 9268468. https://doi.org/10.1155/2017/9268468

36) Emberger, L. (1955). Une classification biogéographique des climats. CNRS.

37) Erenler, R., Sen, O., Aksit, H., Demirtas, I., Yaglioglu, A. S., Elmastas, M., & Telci, I. (2016). Isolation and identification of chemical constituents from Origanum majorana and investigation of antiproliferative and antioxidant activities. Journal of the Science of Food and Agriculture, 96 (3), 822-836. https://doi.org/10.1002/jsfa.7155

38) FATHY M., SOLIMAN., MIRIAM F., YOUSIF., SOUMAYA S. ZAGHLOUL., 2009.,seasonal variation in the essential oil composition of origanum majorana 1. cultivated in Egypt., naturforsch., (64)., 611-614p.

39) Ferraguti, M., Martínez-de la Puente, J., Roiz, D., Ruiz, S., Soriguer, R., & Figuerola, J. (2013). Avian Plasmodium in mosquitoes in southern Spain. Parasites & Vectors, 6(1), 1–8.

40) Figueiredo, A. C., Barroso, J. G., Pedro, L. G., & Scheffer, J. J. C. (2008). Factors affecting secondary metabolite production in plants: Essential oils. Flavour and Fragrance Journal, 23(4), 213–226.

41) FURIA, T. E., 8c Bellanca, N., 1971., Fenaroli's handbook of favor ingredients, The Chemical Rubber Co., Cleveland, OH.

42) Galovičová, L., et al. (2021). Essential oil composition and bioactivity of Thymus serpyllum from Central Europe. Molecules, 26(9), 2761.

43) GBIF Secretariat . (2023). Thymus serpyllum L. in GBIF backbone taxonomy. Available at: 10.15468/39omei (Accessed December 13, 2023). [DOI]

44) Haj-Husein, I., Tukan, S., & Alkazaleh, F. (2016). The effect of marjoram (Origanum majorana) tea on the hormonal profile of women with polycystic ovary syndrome: A randomised controlled pilot study. Journal of Human Nutrition and Dietetics, 29 (1), 105-111. https://doi.org/10.1111/jhn.12290

45) Harbach, R. E. (1988). Mosquitoes of the world: A taxonomic catalogue. The Natural History Museum.

46) Hussain, A. I., Anwar, F., Sherazi, S. T. H., & Przybylski, R. (2010). Chemical composition, antioxidant and antimicrobial activities of basil (Ocimum basilicum) essential oils depends on seasonal variations. Food Chemistry, 123(3), 806–815.

47) Ietswaart, J.H., 1980. A Taxonomic Revision of the Genus Origanum (Labiatae): (Labiatae), Leiden Botanical Series. Springer Netherlands.

48) Jinu, U., Rajakumaran, S., Senthil-Nathan, S., et al. (2017). Potential larvicidal activity of silver nanohybrids synthesized using leaf extracts of *Cleistanthus collinus* (Roxb.) Benth. ex Hook. f. and *Strychnos nux-vomica L.* nux-vomica against dengue, Chikungunya and Zika vectors. Physiological and Molecular Plant Pathology. doi:10.1016/j.pmpp.2017.05.003

49) Jordan, M. C., et al. (2013). Genetic diversity and chemotypic variation in Thymus vulgaris. Phytochemistry Reviews, 12(3), 501–510.

50) Jukic, M and M. Milos. (2005). Catalytic oxidation and antioxidant properties of Thyme essential oils (Thymus vulgaris L.). CROATICA CHEMICAL ACTA, CCACAA.78(1), 105-110. ISS-0011-1643., CCA-2985.

51) KAHOULI I., 2010.,effet antioxydant d'extraits de plantes (Laurus nobilis L., Rosmarinus officinalis, Origanum majorana, Oléa Europea L.) dans l'huile de canola chauffée.. mémoire de magister. Université Laval.

52) Kalimuthu, K., Panneerselvam, C., Chou, C., et al. (2017). Control of dengue and Zika virus vector *Aedes aegypti* using the predatory copepod Megacyclops formosanus: Synergy with Hedychium coronarium-synthesized silver nanoparticles and related histological changes in targeted mosquitoes. Process Safety and Environmental Protection, 109, 82–96.

53) Kovaaevia N. Folk Medicine. 1st ed. Faculty of Pharmacy, University of Belgrade: Belgrade, Serbia, 200.

54) Leal et al., (2017). Thymus Plants: A Review-Micropropagation, Molecular and Antifungal Activity. 10.5772/66623.

55) Le Berre, M. (1989). Faune du Sahara: vertébrés. Editions Recherche sur les Systèmes Écologiques.

56) Le Berre, M. (1990). Reptiles du Sahara. Editions Recherche sur les Systèmes Écologiques.

57) Lecointre, G., & Le Guyader, H. (2001). Classification phylogénétique du vivant (3rd ed.). Belin.

58) Mabberley, D. J. 1997. The Plant-Book: A Portable Dictionary of the Vascular Plants, 2nd ed. Cambridge University Press. Cambridge.

59) Madani, S., Djamel, S., Noui, H., Boudjelal, A., 2012. Ethnobotanical study of therapeutic plants used to treat arterial hypertension in the Hodna region of Algeria. Glob. J. Res. Med. Plants Indig. Med. 1 (9), 411.

60) MECHERGUI K., JAOUADI W., COELHO J., SERRA M.C., KHOUJA M.L.(2015)-biological activities and oil properties of origanum glandulosum desf., lavoisier.,2-8p.

61) National Office of Meteorology (ONM). (2023–2024). Climatic data of El Oued Province. Retrieved from https://www.tutiempo.net

62) Nunes, F. C., Leite, J. A., Oliveira, L. H., et al. (2015). The larvicidal activity of *Agave sisalana* against L4 larvae of *Aedes aegypti* is mediated by internal necrosis and inhibition of nitric oxide production. Parasitology Research, 114(2), 543–549.

63) Ouelbani, R., Bensari, S., Mouas, T.N., Khelifi, D., 2016. Ethnobotanical investigations on plants used in folk medicine in the regions of Constantine and Mila (North-East of Algeria). J. Ethnopharmacol. 194, 196–218.

64) Ozenda, P. (1991). La végétation du Sahara. CNRS Éditions.

65) Prasanth Reddy, V., Ravi Vital, K., Varsha, P. V., & Satyam, S. (2014).Review on Thymus vulgaris traditional uses and pharmacological properties. Med Aromat Plants, 3(164), 2167-0412.

66) Premrov Bajuk, B., Prem, L., Vake, T., Žnidaršič, N., & Snoj, T. (2022).The effect of thymol on acetylcholine-induced contractions of the rat ileum and uterus under ex vivo conditions. Frontiers in Pharmacology, 13, 990654.

67) Prerna, Vasudeva, N., 2015. *Origanum majorana* L. -Phyto-pharmacological review. IJNPR 6 (4) [December 2015].

68) Renta NW. Herb yield and chemical common oregano vulgare essential oil according to the plant development stage. Kert Polotea 2012; 55:3.

69) Ricordi, C., Garcia-Contreras, M. and Farnetti, S. (2015) 'Diet and inflammation: possible effects on immunity, chronic diseases, and life span', Journal of the American College of Nutrition, 34(Suppl 1), pp. 10-13. doi: 10.1080/07315724.2015.1080101.

70) Rubin M. (2004). Guide pratique de phytothérapie et d'aromathérapie. Edition Ellipses, Pris,pp,1-71.

71) SAIMI A . (2014)-contribution a l'evaluation de la sensibilite d'Escherichia coli isolés d'infections urinaires communautaires aux quinolones et au extrait d' Origanum glandulosum et cynoglossum cheirifolium., mémoire de magister., Université Aboubekr Belkaid Tlemcen.

72) Sari, M., Sarri, D., Hendel, N., Boudjelal, A., 2012. Ethnobotanical study of therapeutic plants used to treat arterial hypertension in the Hodna region of Algeria. Glob. J. Res. Med. Plants Indig. Med. 1, 411.

73) SCHAAL S.,2010.,Les plantes médicinales des pelouses calcaires de la réserve naturelle de Montenach (57) .,thèse doctorat., Université Henri Poincare - Nancy 1.

74) Schaffner, F., et al. (2001). Larval anatomy of Culex pipiens [Diagram]. European Mosquito Bulletin.

75) Seddik Miloud magister UNIVERSITE d'Oran analyse physico-chimique chromatographique et spectroscopique de l'huile essentielle 2010 p 7-8.

76) Service, M. W. (1993). Mosquito ecology: Field sampling methods (2nd ed.). Chapman & Hall.

77) SKOULA M., HARBORNE J.B.,2002., The taxonomy and chemistry of *Origanum, Oregano*: The genera of Origanum and Lippia, 67-109. In Medicinal and aromatic plants - industrial profils; (25); ISBN 0-415-36943-6

78) Sundararajan, B., & Kumari, B. R. (2017). Novel synthesis of gold nanoparticles using *Artemisia vulgaris* L. leaf extract and their efficacy of larvicidal activity against dengue fever vector *Aedes aegypti L*. Journal of Trace Elements in Medicine and Biology, 43, 187–196.

79) Talei, G.R and M.H. Meshkatalsadat. (2007) . Antibacterial activity and chemical constitution of essential oils of *Thymus eriocalyx* from west of Iran. Pakistan Journal of biological science.10(21), 3923-3926.

80) Tea, K., Verica,D.V and M. Miaden. (2006) Antioxidant activity of Aqueous Tea infusion prepared from Oregano, Thyme and wild Thyme .food Technol. Biotechnol. 44 (4), 485-592.

81) Tepe, B. and D. Dimitra. (2004). In Vitro Antimicrobial and Antioxidant Activities of the Essential Oils and Various Extracts of Thymus. eigii M. Zohary et P.H. Davis J. Agric. Food Chem, 52, 1132-1137.

82) Triantaphyllouk., Blekas G. & Boskou D.,2001., Antioxidative properties of water extracts obtained from herbs of the species Lamiaceae. International Journal of Food Sciences and Nutrition 52(4), 313-317.

83) UNESCO. (1972). Hydrogeological Section Across the Sahara. United Nations Educational, Scientific and Cultural Organization.

84) VERA R.R.., CHANE-MING J.,1999., Chemical composition of the essential oil of marjoram (Origanum majorana L.) from Reunion Island. Food Chemistry 66 (1999) 143_145.

85) Voisin, J. F. (2004). Les oiseaux de l'Algérie. Société d'Études Ornithologiques.

86) Wesołowska, A., & Grzeszczuk, M. (2015). Chemical composition of Thymus serpyllum essential oil. Acta Scientiarum Polonorum Hortorum Cultus, 14(1), 125–136.

87) WHO. (2023). Vector-borne diseases: Global distribution of Aedes mosquitoes. World Health Organization. Retrieved from https://www.who.int

88) Wikiwand. (2024). El Oued province. Retrieved from https://www.wikiwand.com

89) WordPress – D. Abbass. (n.d.). Geographic coordinates of Oued Souf. Retrieved from https://wordpress.com

90) World Health Organization (WHO). (2021). World malaria report 2021.

91) Zeljković, S. Ć. and Maksimović, M. (2015). Chemical composition and bioactivity of essential oil from Thymus species in Balkan Peninsula. Phytochemistry Reviews, 14(3): 335-352.

92) Zeroug, S. (2018). Evaluation of the larvicidal effect of Origanum majorana extract on Culex pipiens. University of Constantine Thesis.

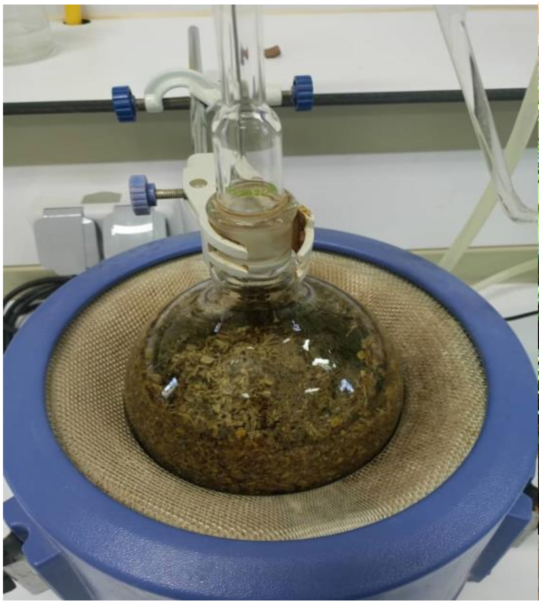
93) Anonyme 1 : https://garden-n.com/ar/بذور-البردقوش/p92323937

94) Anonyme 2 : https://www.mysticgardenseeds.net/product/الزعتر-الزاحف/

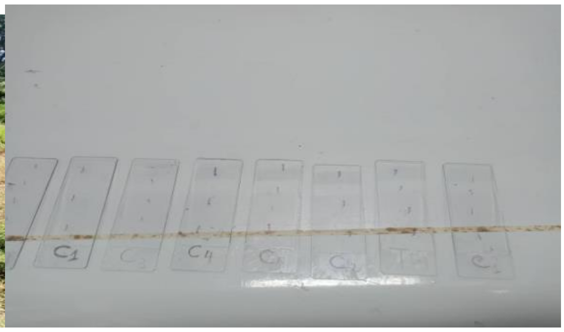
95) Anonyme 3 : www.mawdoo3.com

96) Anonyme 4 : https://kunoozalteeb.com/blog/هل-الزيوت-العطرية-الأساسية-مفيدة-للصح

ANNEXES



Titre des photos ???????

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