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Theme

Epidemiological study of *Escherichia coli* distribution in the Mila region, correlation with meteorological parameters

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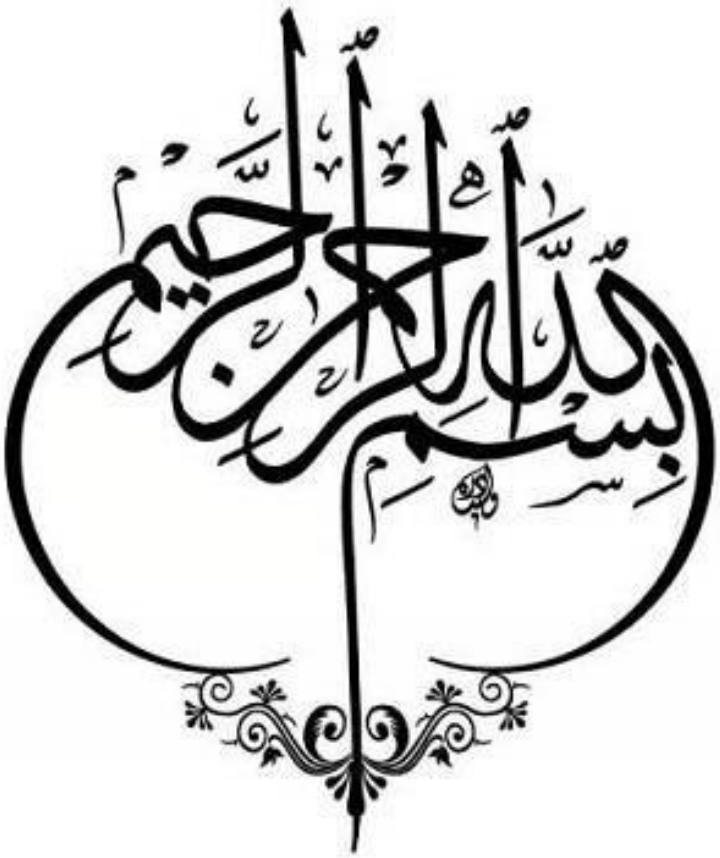
بسم الله الرحمن الرحيم

Thanks

At the end of this work,
we would like to express our thanks and our deep gratitude above all to God who gives us the strength, the courage, and the will to develop this modest work.
*We would like to thank Dr. "**KHAREIF NACEREDDINE Saliha**" for having agreed to judge this work and to chair the jury of our defense.*
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We would like to thank the entire medical team of the bacteriology laboratory of the Public Hospitalier Brother Maghlaoui Mila establishment.

Fairouz and Hasna

Dedications

Praise be to God, Lord of the worlds, and prayers and peace be upon our master Muhammad, the most honorable of the messengers, and upon his family and companions.

Years of hard work and vigil, we conclude with lines, even if they are many, they will not narrate the joy, sadness, fear, and hope we experienced... We have arrived, you who said you will not pray, so praise be to God who shamed us with His mercy, compassion, and grace.

*To the one who suffered from fatigue, and endured the hardships of life for us, to the one who said to me on the day that you will not be miserable as long as I live, to my support, my support, my strength and my reclining, my dear father "**Abd el Hamid**".*

*To the warm heart and the merciful chest, to the one who gave me without question and pushed me to move forward despite the difficulties, to the one whose eyelids left sleep to comfort me, to the one who drew the slogan of success on my heart and made it a medal on my chest, to the one who owes her abundant and great credit for what she has reached, to Qara My dear mother's eyes "**Fatima**"*

*To those who are closer to me than my soul, to my support and consolation in this life to my brothers "**Sumaya, Amina, Suhaila, Fella, Abla**".*

*To my sweet heart and supporter, to my warm haven my friend "**Khadidja**".*

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To all my family and everyone who has supported and encouraged me in my life and given me a push forward.

To everyone who helped me from near or far, even with a kind word.

To all the people I met in my life and had beautiful situations with them.

To all of the breadth of my heart and did not satisfy my paper.

*Finally, I dedicate this humble work to the one who supported me and took my steps with me and eased the difficulties for me, to the one who lit the lamps of knowledge and knowledge in my heart, to the one who preceded me in everything and advised me before anything happened, to the one who granted without waiting and forgave without excuses to my dear teacher."**Tayaa Hakima**"*

To all of you... I dedicate this work.

Fairouz

The trip was not short and it should not have been, neither the dream was near nor the road was fraught with facilities, but I did it

To everyone who has enlightened the mind of others with his knowledge or guided with the correct answer, he showed by His grace the humility of the scholars and by His grace the eminence of the knowledgeable.

*To my first example, to the one whose name I bear with pride, to the one who harvested thorns from my path to pave the path of Science for me, to the one who gave me and continues to give me infinitely, to the one who raised my head high with pride in him, "**dear father, may God preserve me**."*

*To the one whose heart saw me before her eyes, to my tree that does not wither, to the shadow to which I Shelter all the time "**beloved mother, God save her**."*

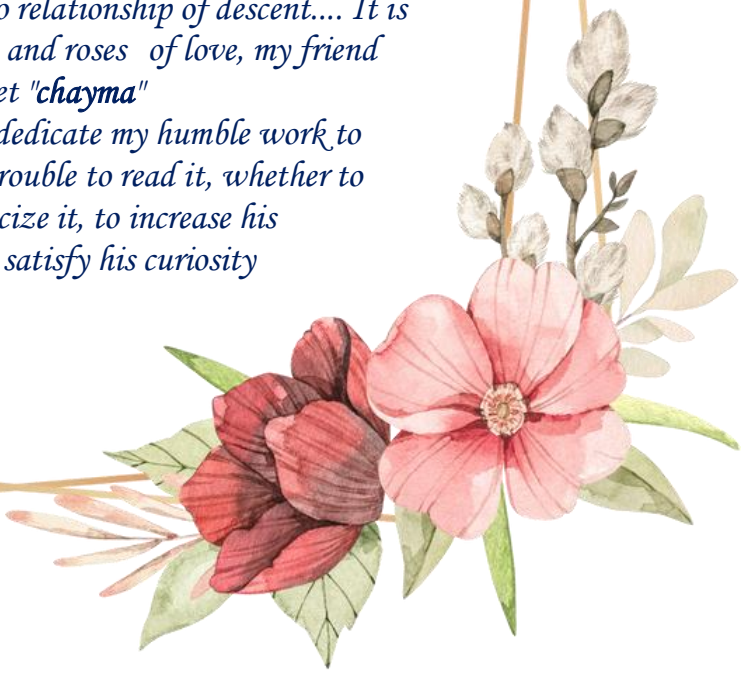
*To the candles that illuminate my path, "**my sister chaima**", "**my brothers Yasser, Munsif, Hudaifa**" they encouraged me and continued to give without getting bored.*

*To my two little sisters, the Daughters of my only cousin "**Yousra" Khadija**, who support me in everything.*

*To the companion of the successful journey and who accompanied me during the study, my friend, sister and only "**Fairouz**"*

*To those with whom I had no relationship of descent.... It is the fragrance of friendship and roses of love, my friend and sweet "**chayma**"*

And last but not least, I dedicate my humble work to everyone who takes the trouble to read it, whether to evaluate it, to criticize it, to increase his knowledge or to satisfy his curiosity



Hasna

Abstract

In order to determine the epidemiological and clinical characteristics of human *Escherichia coli* in the district of Mila, we developed a retrospective study during a period extending from January (2012 to December 2022), and the other prospective during three-months (January-March 2023). We collected the data at the level of Bacteriology laboratory of the General Hospital Maglaoui Brothers Mila, for the retrospective descriptive analytical study we treated 14596 examinations, 495 were positive, an infestation rate of 3.39%.

-Of the positive cases, 66.87% were female and 33.13% were male.

- Patients aged (20-44 years) are the most exposed to *E. coli*.

-The years 2019 and 2020 had the highest infection rates, 4.45% and 4.45% respectively.

-The spread of these bacteria was observed during the fall season.

-A high rate of this bacterium was noted during the months of November, December.

-The climatic conditions of Mila region, like the ambient temperature increase the dissemination of the bacteria. Sunshine, average humidity, precipitation, and average monthly wind speed cause a decrease in the bacterial index of *Escherichia coli*.

The results obtained during the three months of our prospective study (January-March 2023), confirmed what we deduced from the results above (The retrospective study from 2012 to 2022).

Keywords: *Escherichia coli*, epidemiology, prevalence, correlation, meteorological parameters, Mila.

Résumé

Afin de déterminer les caractéristiques épidémiologiques et cliniques d'*Escherichia coli* humain dans le district de Mila, nous avons élaboré une étude rétrospective durant une période s'étendant de Janvier 2012 à Décembre 2022, et l'autre prospective de trois mois (Janvier-Mars 2023). Nous avons collecté les données au niveau de laboratoire de Bactériologie de l'Hôpital Général Frères Maglaoui Mila, pour l'étude analytique descriptive rétrospective nous avons traité 14596 examens, 495 étaient positifs, un taux d'infestation de 3,39 %.

- Parmi les cas positifs, 66,87 % étaient des femmes et 33,13 % étaient des hommes.
- Les patients âgés (20-44 ans) sont les plus exposés à *E. coli.*
-Les années 2019 et 2020 ont enregistré les taux d'infection les plus élevés, respectivement 4,45% et 4,45%.
-La propagation de ces bactéries a été observée durant la saison automnale.
-Un taux élevé de cette bactérie a été notée durant les mois de Novembre, Décembre.

-Les conditions climatiques de la région de Mila, comme la température ambiante augmentent la dissémination de la bactérie. La durée d'insolation, l'humidité moyenne, les précipitations et la vitesse mensuelle moyenne du vent provoquent une diminution de l'indice bactérien d'*Escherichia coli*.

Les résultats obtenus au cours des trois mois de notre étude prospective (Janvier-Mars 2023) ont confirmé ce que nous avions déduit des résultats ci-dessus (l'étude rétrospective de 2012 à 2022).

Mots clés : *Escherichia coli*, épidémiologie, prévalence, corrélation, paramètres météorologiques, Mila.

الملخص

من أجل تحديد الخصائص الوبائية والسريرية للإشريكية القولونية البشرية في منطقة ميلة ، قمنا بتطوير دراسة بأثر رجعي خلال فترة تمتد من يناير 2012 إلى ديسمبر 2022 والأخرى المرتقبة لمدة ثلاثة أشهر (يناير - مارس 2023) . جمعنا البيانات على مستوى معمل الجراثيم بـالمستشفى العام الإخوة م غلاوة ميلة ، للدراسة التحليلية الوصفية بأثر رجعي عالجنا 14596 فحصًا ، 495 منها كانت إيجابية ، ومعدل الإصابة 3.39٪.

-من بين الحالات الإيجابية 66,87% إناث و 33,13% ذكور.

-الأشخاص الذين تتراوح أعمارهم بين (20-44 سنة) هم الأكثر تعرضاً لداء الاشريكية القولونية.

-سجلت الأعوام 2019 و 2020 أعلى معدلات إصابة 4,45% و 4,45٪ على التوالي.

-لوحظ انتشار هذه البكتيريا خلال فصل الخريف.

- لوحظ ارتفاع معدل هذه البكتيريا خلال شهري نوفمبر وديسمبر.

إن الظروف المناخية لمدينة ميلة ، مثل درجة الحرارة المحيطة تزيد من انتشار البكتيريا .تسبب أشعة الشمس ، ومتوسط الرطوبة ، والتساقط ، ومتوسط سرعة الرياح الشهرية انخفاضًا في المؤشر البكتيري للإشريكية القولونية .

أكدت النتائج التي تم الحصول عليها خلال الأشهر الثلاثة من دراستنا المرتقبة (يناير - مارس 2023) ما استخلصناه من النتائج أعلاه (الدراسة بأثر رجعي من 2012 إلى 2022).

الكلمات المفتاحية: الاشريكية القولونية ، علم الأوبئة ، انتشار، الارتباط، العوامل المناخية، ميلة.

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ABBREVIATIONS LIST

ANDI	National Investment Development Agency
A.N.I.R.E.F	National Agency for Land Intermediation and Regulation
C°	Degree Celsius
CFU	Colony Forming Unit
DNA	Deoxyribonucleic Acid
D.S.P.M	Mila Public Health Department
E	*Escherichia*
E. Coli	*Escherichia Coli*
EHS	Specialized Hospital
EPH	Public Hospitals
EMB	Eosin Methylene Blue
g	Gramme
km³	Cubic Kilometer
m	Meter
PH	Hydrogen Potential
R	Correlation Coefficient
SPSS	Statistical Package for the Social Sciences
TSA	Trypticase-Casein- Soy
%	Percentage
Fig	Figure
ANOVA	Analysis of variance
&	And
SBI	The Simple Bacterial Index

SUMMARY

INTRODUCTION

INTRODUCTION

On any possible, reasonable or fair criterion, bacteria are – and always have been – the dominant form of life on earth. And that is how Gould's piercing comment introduces us to the importance of bacteria to nature and, in particular, to humans **(Gould and House, 1996).**

Homo sapiens, for example, is a multitude of such ecosystems, colonized by an unfathomable variety of bacteria. Some of them, called commensals, can be advantageous to humans either by producing useful metabolites or fighting off harmful infectious agents. Others, called pathogens, can damage their host which results in the development of a particular disease. Being a pathogen or commensal is only of value to the colonized organism, and transition to a new niche of its host can often provide a commensal bacterium with pathogenic potential **(Connell *et al.*, 1996).** Some of these bacteria are pathogenic to humans or animals, such as *Salmonella spp*, *Campylobacter spp*, *Yersinia spp*, *Listeria spp*, and pathogenic coli bacteria. **(Johnson and Russo, 2005)**

Escherichia coli are one of the microorganisms most frequently studied worldwide. They are Gram-negative bacilli, facultative anaerobic, rod-shaped bacterium, which are most often found in the gastrointestinal tract as a normal coloniser of warm blood organisms (mainly in mammals, but are also present in birds, reptiles and fish). Taxonomically, *E. coli* belong to the Enterobacteriaceae family and are an important component of the intestinal microbiota, being involved in some essential metabolic processes such as the production of vitamin K and vitamin B12 (**Bentley and Meganathan, 1982)**. *E. coli* also help to maintain the anaerobic environment needed for most of the microbiota by consuming oxygen that enters the gut and competitively exclude pathogens from the lower intestine of their hosts **(Blount, 2015)**. *E. coli* can also adapt to life in an external environment outside hosts, such as soil, water, plants and food, due to their hardiness and metabolic flexibility **(Savageau , 1983)**.Thus, *E. coli* is a species in which we find both commensal strains, colonizing healthy individuals, and strains that have acquired virulence factors. The latter are capable of inducing different clinical signs and are grouped into pathovars. A pathovar is defined as a set of strains having a common virulence phenotype (same adhesion profile on cell culture, same symptoms induced in the host...). **(Johnson and Russo, 2005)**

Escherichia coli is the best studied bacterium and the experimental microorganism of choice for many microbiologists. This major bacterium of human colon and warm-blooded animals is very useful for the analysis of fecal contamination and is the bacterium most frequently involved in urinary tract infections. It can also cause diarrhea by various mechanisms, as well as various community or nosocomial infections **(Bourjilat, 2009)**.

Despite establishing symbiotic relationships, this well-known microorganism can also have an important pathogenic role within their hosts, especially from a human point of view. It is able to cause a wide variety of infections that can reach a high prevalence and even cause significant morbidity and mortality **(Russo and Johnson, 2003)**. In addition, certain pathogenic strains of *Escherichia coli* are known to doctors as causative agents of infantile gastroenteritis or the famous "traveller's diarrhea", often of waterborne origin, it is also an indisputable pathogen for humans. and the animal, responsible for acute diarrhea of the choleraform (turista) type, hemorrhagic dysenteryform, urinary tract infections: cystitis, pyelonephritis, septicemia, meningitis **(Stewart *et al.*, 20015)**.

Understanding the complexity of interactions between pathogens, environmental parameters, social factors and diarrheal diseases therefore represents an important societal challenge for the forth coming years (**Robert *et al.*, 2021**) . Climate change induced heavy rainfall and flooding indeed has caused epidemics of waterborne diseases like diarrhoea (**Delpla *et al.*, 2009; Funari *et al.*, 2012; Zhang *et al.*, 2012**).In addition, weather climate can have an impact on the presence of *E. coli* bacteria. Climate change is anticipated to have a strong impact on both the quantity and quality of water resources, potentially boosting the presence, dissemination and transmission of pathogens [**(Elsas *et al.*, 2011 ;Carlton *et al.*, 2014 ,2016)**]. Environmental variables influence Faecal Indicator Bacteria (FIB) in surface water. Understanding that influence is important, because presence of FIB, which are an indication of faecal contamination, means that harmful pathogens could be present that could also be influenced by environmental variables. Although some recent studies have focused on this topic (**Majedul Islam *et al.*, 2017**).

For this purpose, we have oriented our work in the direction of the statistical evaluation of the current state of the prevalence of *E. coli* in Algeria, more precisely in the region of Mila and make a retrospective epidemiological study on cases of human *E. Coli* collected at the level of public hospital Brothers Maghlaoui - Mila during the period (2012 to

2022). Also this study aims to detect the influence of the climatic changes on the dissemination and transmission of this bacteria by following the correlation between the spread of *E. coli* and climatic parameters changes.

The main objectives of this study are:

A. List existing cases of tuberculosis human *E. Coli* in the wilaya of Mila.

B. Determine certain parameters that can influence the proportions of affected subjects (age, sex, months, seasons and years).

C. Assess the correlation between the spread of *Escherichia coli* and meteorological parameters (temperature, precipitation, wind speed, humidity and sunshine duration).

Apart from the introduction and the conclusion, as well as some suggestions and recommendations, this work is divided into four chapters:

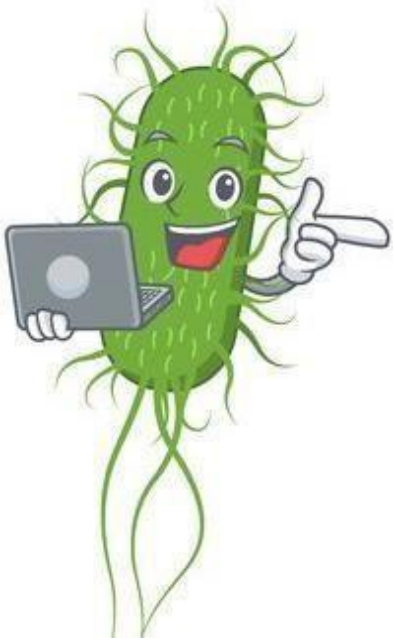
* The first will deal with a bibliographical synthesis on the human *E.coli* and the pathogen related to this bacteriosis;

* The second will deal with material and methods, related to the various biological techniques used at the level of the laboratory of bacteria during the realization of this work;

*The third will have to present and interpret the results;

* The fourth will be devoted to the discussion of the obtained results in relation to the data of the scientific literature.

PRESENTATION OF BIOLOGICAL MODEL



1. PRESENTATION OF BIOLOGICAL MODEL

1.1. History

It is difficult to imagine today the excitement in the days of Louis Pasteur (1822-1895) and Robert Koch (1843-1910), when doctors and microbiologists were chasing the agent responsible for every disease. It was in this climate that a young German pediatrician, Theodor Escherich (1857-1911), described in 1885 the species Bacterium coli comune, isolated from the stool of babies fed exclusively on breast milk (**Richard and Guennadi, 2008)**.

At that time, infant mortality in Germany was high and diarrhea epidemics were frequent. In an original approach, Escherich examined the bacteria found in the intestines of both healthy and sick newborns. He found that the intestinal tract, sterile at birth, was rapidly invaded by many species of bacteria. Later, but before the child was weaned, this fauna disappeared and was replaced by a bacillus that he named Bacterium coli comune. Escherich observed that this bacillus was found in all individuals, healthy and sick, and that it was harmless. However, it was later shown that there were pathogenic variants, one of which was an agent of diarrhea. Escherich noted that B. coli was an easy bacillus to grow in the laboratory, where it grew well on complex organic media and on inorganic media with a sugar addition. In the third edition of their Manual of Tropical Diseases, published in 1919, Aldo Castellani and Albert J. Chalmers renamed Escherich's bacillus *Escherichia coli* in honor of its discoverer (**Richard and Guennadi, 2008)**.

1.2. General information on *Escherichia coli*

Escherichia coli (E. coli) are a genomically and phenotypically highly heterogeneous group of facultatively anaerobic, Gram-negative bacilli within the family *Enterobacteriaceae* **(Nataro and Kaper, 1998)**. They were first described in 1885 by Theodor Escherich as Bacterium coli commune (the common colon bacterium) during his studies on neonatal and infant fecal microbiota **(Neill *et al.*, 1994)**. Most of its members are typically motile, non-pathogenic commensal inhabitants of the gastrointestinal tract of humans and animals. *E. coli* colonisation of the gastrointestinal tract occurs within hours or few days following birth. In humans, *E. coli* typically colonise the gastrointestinal tract of infants within 40 hours of birth and is the predominant facultative anaerobe found in the human colonic flora **(Todar, 2005b)**. Once established in the gastrointestinal tract, strains of *E. coli* may persist for months or years.

Physiologically, *E. coli* are versatile bacteria well adapted to their characteristic habitats. They are capable of utilising a wide variety of substrates for growth. Glucose is the preferred carbon source of *E. coli* and growth can occur in media with glucose as the sole organic constituent **(Martinez *et al.*, 2012)**. Wild-type *E. coli* have no growth factor requirements, and are capable of synthesising all essential growth factors such as purines, pyrimidines, amino acids and vitamins from the carbon sources being utilised **(Todar, 2005a)**. As facultative anaerobes, *E. coli* utilise fermentation or anaerobic respiration for growth in the absence of oxygen. During fermentation *E. coli* employs the mixed acid fermentative pathway that produces alternative acidic end products such as lactate, acetate, formate and gases such as carbon dioxide and hydrogen in variable amounts **(Sumbali and Mehrotra, 2009)**. In addition, *E. coli* growth is also supported by anaerobic respiration, which generates the majority of its cellular energy under anaerobic conditions **(Sumbali and Mehrotra, 2009)**. The ability of *E. coli* to grow under aerobic and anaerobic conditions has enabled their adaptation to intestinal (anaerobic) and extra-intestinal (aerobic or anaerobic) environments in humans and animals.

E. coli cells respond to a multitude of environmental stimuli including temperature and pH. Optimal growth of *E. coli* occurs at 37°C although it can still grow and multiply within the temperature range of 7–8°C to 46°C **(International Commission on Microbiological Specifications for Foods, 1996)**. Some strains of *E. coli* have been shown to grow at temperatures of 48–50°C and survive well in chilled (0–4°C) and frozen food at −20°C **(Ministry for Primary Industries − New Zealand, 2001)**. The growth pH of *E. coli* ranges between 4.4–10.0, with an optimum of 6–7 **(Desmarchelier and Fegan, 2003)**. Some *E. coli* strains have been shown to survive at pH 2.5–3.0 for over four hours or more **(Molina *et al.*, 2003)**. Prior exposure to acidic conditions increases survival of some *E. coli* strains under very low pH conditions such as that encountered in the human stomach (pH 1.5–3.5) for periods of more than three hours, the time generally required to clear an average meal **(Ministry for Primary Industries − New Zealand, 2001)**. Shifts of temperature or pH within the normal growth range of *E. coli* generally result in quick, transient adjustments in the level of cell components and enzyme activity. However, deviations in temperature and pH above or below the normal growth range may lead to stress conditions in *E. coli* resulting in either reduced growth or survival potential. Under stress conditions, *E. coli* induces a general stress response providing cross protection against multiple stresses as well as a specific

response towards the primary stress **(Jones, 2012)**. The stress response serves as an immediate and long term strategy of adaption of *E. coli* cells to various stresses through changes in cellular physiology and morphology as well as gene expression.

Although *E. coli* are a highly diverse group of organisms, serotype analysis has facilitated their differentiation to a great extent **(Fig.1)**. *E. coli* serotyping is based on their surface antigen profiles, namely somatic (O), flagellar (H), and capsular (K) **(Nataro and Kaper, 1998)**, which was initially proposed by Kauffman **(Kauffman, 1947)**. The O antigen of *E. coli* defines the serogroup while a specific combination of O and H antigens defines the serotype. Currently, more than 700 serotypes of *E. coli* have been identified **(Griffin and Tauxe, 1991)**. In *E. coli*, the serologic antigens themselves do not confer virulence but rather serve as readily identifiable chromosomal markers that correlate with specific virulent clones.

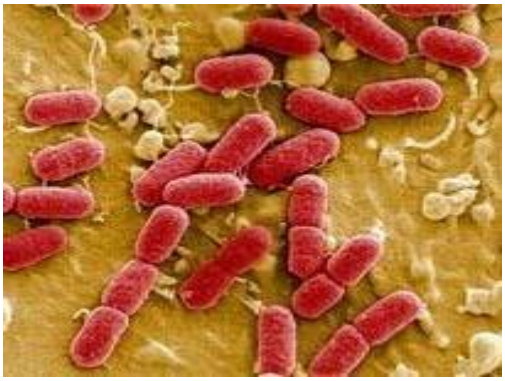


Fig. 1: *Escherichia coli.* **(Blount, 2015).**

1.3. Taxonomy

The species *Escherichia coli* is part of the Enterobacteriaceae family **(table 1)**. It has been characterized on the phenotypic, biochemical and physiological levels. Today, techniques based on the use of DNA allow a genetic study of populations and the characterization of different strains of Escherichia coli. **(Stewart, 2015)**.

Table 01: Classification of *Escherichia coli* **(Stewart, 2015).**

Reign	Prokaryotes
Domain	Bacteria
Branch	Proteobacteria
Class	Gammaproteobacteria
Order	Enterobacteriale
Family	Enterobacteriaceae
Gender	Escherichia
Species	*Escherichia coli*

1.4. *E coli* habitat

- **Primary habitat**

E. coli is a normal host in the digestive tract of humans and animals. In humans it is present at a rate of 10^7 to 10^9 bacteria per gram of those. However, this number is much lower than those of the anaerobes which constitute the dominant flora. The digestive tract is its primary habitat. This bacterium is present mainly in the colon and caecum at concentrations approximately > 10^6 CFU (colony forming unit)/g of intestinal content. *E. coli* nests more particularly in the mucus covering the epithelial cells of the wall of the digestive tract, which constitutes an ecological niche conducive to its development due to its conditions of temperature, humidity and nutrient availability. **(Smati, 2015).**

- **Secondary habitat**

E. coli is released into the environment via faeces at a concentration of approximately 10^8 CFU/g of faeces. It ends up in environmental waters through effluents, such as wastewater, slurry or manure from farm animals or through animal droppings farmed or wild animals. The presence of *E. Coli* in the environment is evidence of faecal contamination. This is why we systematically detect it in food or bathing waters (colimetry). (**Smati, 2015).**

The environment is the secondary habitat of *E. coli*. It is, unlike the primary habitat, rather unfavorable to their survival **(Darcan *et al*., 2009) (Table 2).** In the environment, the *E. coli* bacterium is subjected to several types of pressure, biotic (predation and flora competition) **(Pommepuy *et al*., 1996)** and abiotic (light, temperature, oligotrophy and salinity) **(Lil *et al*.,**

2014). *E.coli* is able to grow on ordinary media such as TSA (Trypticase-Casein- Soy) medium and EMB (Eosin Methylene Blue) medium. (**Fig. 2 and 3**).

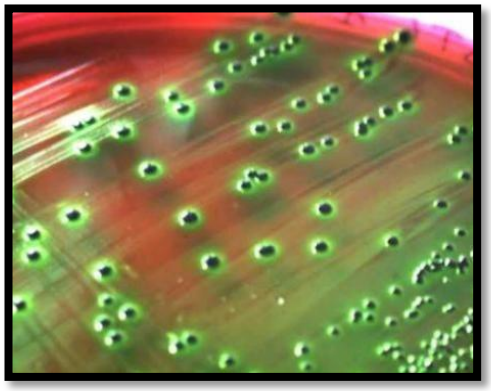


Fig. 02: *Escherichia Coli* on **EMB** Medium **(Smati *et al.*, 2017)**

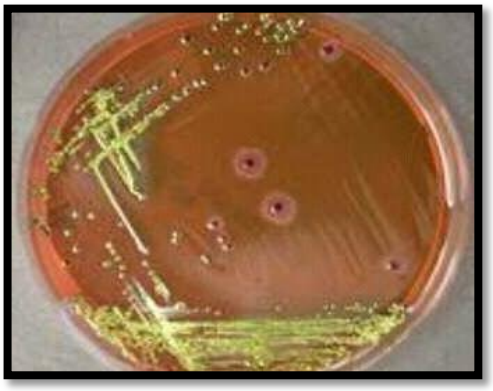


Fig. 03: *Escherichia Coli* on **TSA** Medium **(Denis *et al.*, 2007)**

Table 02: Main parameters influencing the survival of bacteria in the environment. **(Baliere, 2016).**

Factors	Description
Biotics	
Predation	Exogenous enteric bacteria used as a nutrient source by indigenous populations
Competition	Competition for access to nutrients with indigenous populations.
Oligotrophic	Accessibility to nutrient sources, sedimentation
Abiotics	
Solar radiation	Dependent on the seasons and the geographical area. Level of penetration of the rays in the water conditioned by the parameters of diffusion and absorption of the fluid
Temperature	Positive or negative variation on the survival of certain microorganisms Dependent on the seasons
Rainfall	Conditions the humidity level. Impacts river flows, leads to the dilution of drained pollutants, participated in the resuspension of organic matter.
Soil Texture	Sandy, muddy, dry, wet, hard or soft
pH	Acid environment due to composting, fermentation or chemical treatments
Salinity	Responsible for osmotic shock, especially observed in seawater

The population of *E. coli* in the secondary habitat is renewed by the addition of bacteria from the primary habitat. A minority of *E. coli* is able to colonize and persist in the environment outside its host **(Walk *et al.*, 2007)**. This population of *E. coli* known as environmental colonizers is qualified as a naturalized population **(Ishii *et al.*, 2006)** or environmental microbiota coliforms **(Walk *et al.*, 2007)** it becomes a new autochthonous microbial community.

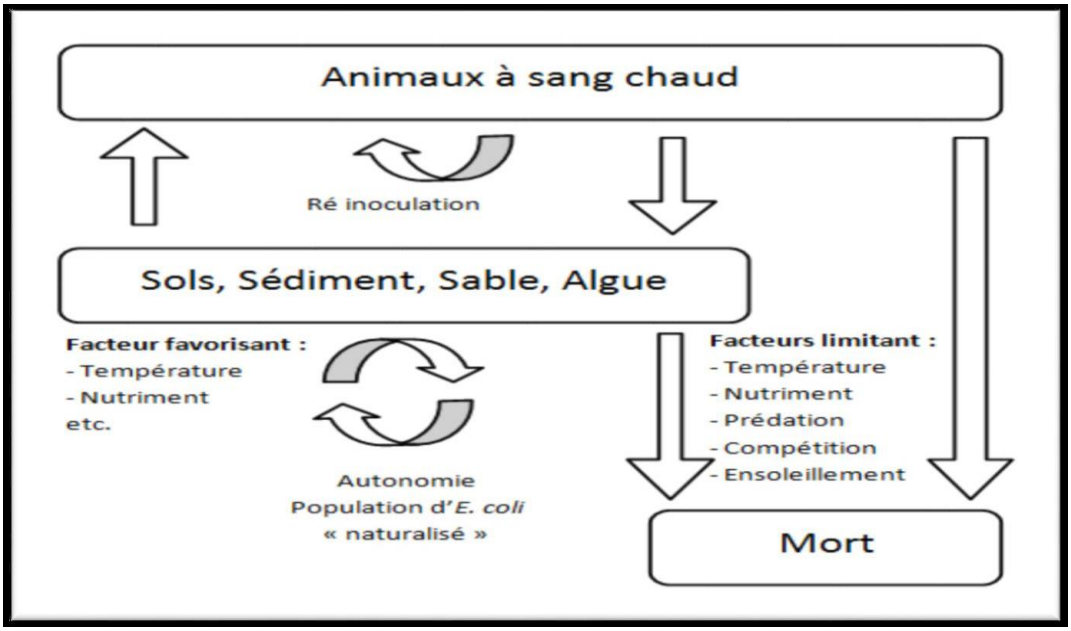


Fig. 04: Life cycle of *E. coli* **(Darcan, 2009)**

This process of adaptation or naturalization in the secondary environment has been observed at the level of environmental faecal coliforms with the identification of *E. coli* having developed the capacity to produce a capsule to protect itself from external aggressions **(Power *et al.*, 2005)**. The significant presence of genes associated with the mechanism of biofilm formation in environmental *E. coli* strains versus fecal strains also suggests an adaptation of *E. coli* strains for better survival in the environment **(Tymensen *et al.*, 2015)** . Similarly, to resist the pressure exerted by the lack of water in certain soils and the osmotic shock caused by the presence of salt in seawater, *E. coli* strains have developed an ability to produce trehalose-type organic solutes. to resist desiccation and salinity **(Zhang and Van, 2012)**.

1.5. Bacteriological characters

1.5.1. Morphological and structural characters

Escherichia coli or coli bacillus is an asporulated bacterium measuring 2 to 4 µ long by 0.4 to 0.6 µ wide. It is a fine and elongated bacterium with rounded ends, mobile thanks to a peritrichous ciliature (**fig. 5**) This non-demanding germ, on ordinary agar, gives smooth, shiny and homogeneous colonies **(Lobril, 1998)**.

-A- **-B-**

Fig. 05: *Escherichia coli* under electron microscope, **-A-** Magnification x 1000 and **-B-** Magnification x 15000. **(Thorene, 1994).**

1.5.2. Cultural characters

E. coli is a facultative anaerobe, it grows easily on ordinary media at 37°C and pH 7.5. **(O. G, 2007.**

- On nutrient agar: appeared as rounded colonies, moist, shiny and whitish or slightly yellowish smooth (S) or rough (R) or sometimes mucous **(Ploy *et al*., 2007).**

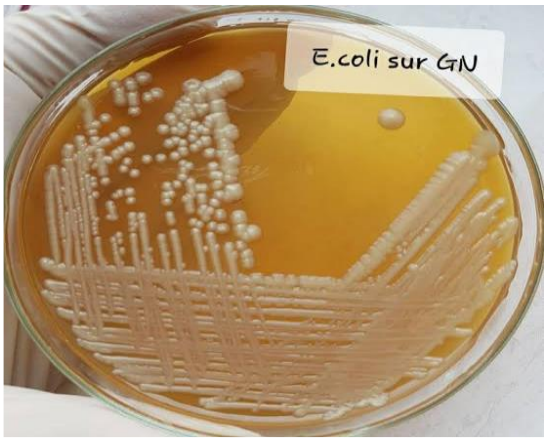


Fig. 06: Appearance of *E.coli* on nutrient agar **(Ploy *et al.*, 2007)**

- On EMB (methylene eosin blue) medium, it gives dark purple colonies with a greenish metallic luster.

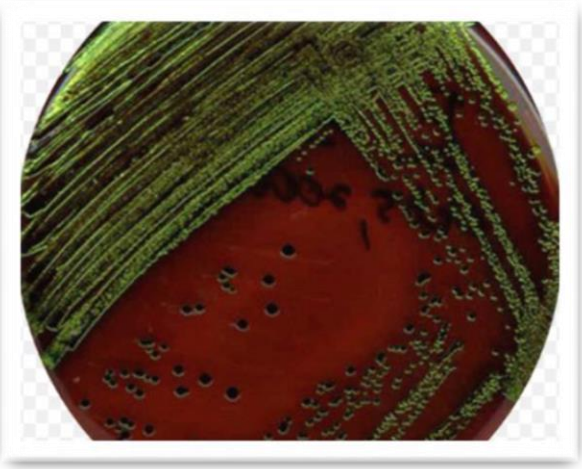


Fig. 07: Appearance of *E.coli* on EMB medium **(Ploy *et al.*, 2007)**

- On Hektoen medium of salmon colonies. **(Denis , 2007).**

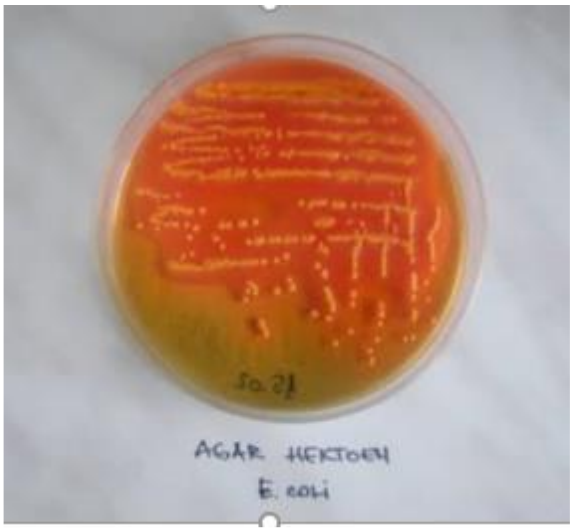


Fig. 08: Appearance of *E.coli* on Hektoen medium**(Denis , 2007).**

- On blood agar: round, translucent, sometimes haemolytic colonies. **(Denis , 2007).**

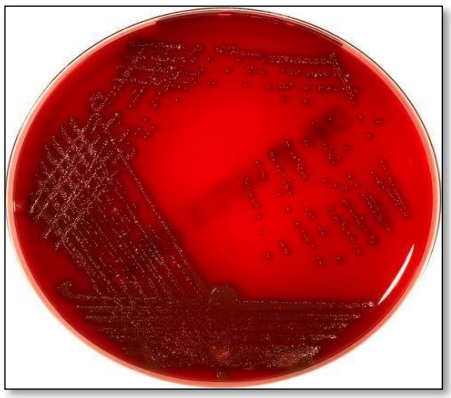


Fig. 09: Appearance of *E.coli* on blood agar. **(Denis , 2007).**

-On Mac conkey, red colonies surrounded by an opaque halo of the same color.

Fig. 10: Appearance of *E. coli* on Mac Conkey medium **(Denis , 2007).**

1.5.3. Biochemical characters

According to the enterobacteriaceae identification key of **Le Minor and Viron (1982)**, *E.coli* has a set of discriminating biochemical characters, which allows it to be differentiated from a heterogeneous microbial population.

E.coli has the ability to ferment various sugars (glucose, lactose, mannitol and sucrose for certain strains) with the production of organic acids (**table 3**). During the fermentation of glucose, gas is produced. One of the distinguishing characteristics of *E.coli* is the production of indole from tryptophan. It is facultative aero-anaerobic, urease negative, tryptophan deaminase negative, does not produce acetoin (Voges-Proskauer reaction negative) and does not use citrate as a carbon source. It reduces nitrates to nitrites, has no oxidase but has catalase **(Joly and Reynaud, 2002).**

Table 03: The biochemical characters of *Escherichia coli* (**Abraham,and Diassana, 2018**).

Tests	Results
Glucose	+
Lactose	+
Hydrogen Sulfide	-
Voges-Proskauer	-
Urease	-
Indole	+
Simmons Citrate	-
Orthonitrophenyl-β-D Galactopyranoside	+
Arginine dihydrolase	+/-
Gelatinase	-
Malonate	-
Phenylalanine Deaminase	-
Lysine Decarboxylase	+
Ornithine Decarboxylase	+
Tryptophan Deaminase	-
Nitrate Reductase	+

+: positive character

-: Negative character

+/-: Inconstant character

1.5.4. Molecular characters

From the molecular point of view, the identification of *Escherichia coli* in the samples is based on the detection of certain virulence genes which are characteristic of the different strains (**Abraham and Diassana, 2018**).

1.6. *E.Coli* antigens

1.6.1. Somatic Antigens O

Somatic O antigens are lipopolysaccharide (LPS) in nature composed of more than 150 complex lipopolysaccharides and are located on the outer membrane (an integral part of the thin cell wall) of Gram-negative bacteria. It contains a large number of repeating units of 3-6 sugar oligosaccharides, the combination of which determines the diversity of O antigens. The genes encoding the enzymes involved in the synthesis of the O antigen are grouped in the rfb gene group; a profile noted "R" can be obtained by electrophoresis corresponding to a serogroup of *Escherichia coli* **(Suveillane, 1997)**.

Currently, some medical analysis laboratories use agglutination with sera to determine the serogroup, but this technique is limited by the increasing number of sera to be produced, by the presence of cross-agglutination between the O antigens of Escherichia coli, Shigella and those of Salmonella, and by the transition from the creamy consistency of the colony to a rough consistency resulting in the absence of synthesis of the O antigen. It is for this reason that a molecular serotyping technique has been developed. **(Suveillane, 1997)**.

1.6.2. Surface or envelope antigens K

There are 3 types of K antigen designated by the letters L, A or B.

- The L antigen: is the most common but is thermolabile (it is destroyed in half an hour at 100°C), so heating causes a loss of antigenic power, the power to bind antibodies and the power to mask the O antigen **(Posl *et al.*, 1998)**.

- **Antigen A:** is rare, it is a capsular antigen (encapsulated *Escherichia coli* are relatively common in urinary tract infections). Ag A is very thermostable (it needs autoclaving to destroy it) **(Posl *et al.*, 1998)**.

- **Antigen B:** is always present in enteropathogenic Escherichia coli of infantile gastroenteritis. It has intermediate thermolability: after half an hour at 100°C there is still

antigen O can come into contact with the serum by "perforating" the envelope, antibody binding is still positive but the antigenic power is gradually lost (depending on the duration of heating) **(Posl *et al.*, 1998)**.

1.6.3.Flagellar antigens H

The flagellar antigen H (AgH) is of protein nature and is involved in the construction of the flagellum allowing the mobility of the bacteria. However, some strains lose their motility and are classified as non-motile (NM or H-) and are not used for identification of pathogenic E. coli but are of great interest from the point of view. The diversity of H antigens is due to the different types of flagellin composing the flagellum structure. Typing is also done by serum agglutination, but is only developed in very few laboratories in the world **(Reid, 1999).** The H antigen is encoded by the fliC gene. Immobile E. coli also possess the fliC gene but are unable to synthesize a functional flagellum **(Machado, 1998)**.

1.7. *Escherichia coli* genome

The genetic makeup of the non-pathogenic laboratory strain *Escherichia coli* were fully sequenced in 1997. Their genome consists of 4.6 million base pairs encoding about 4,200 proteins. In 2001, the genome of an entero-hemorrhagic *Escherichia coli* strain was sequenced. It contains 5.5 million base pairs encoding 5,400 proteins. The following year, the genome of an *Escherichia coli* strain causing urinary tract infections and neonatal meningitis was sequenced. It contains 5.2 million base pairs encoding 5,300 proteins. Comparison of the genomes of these three Escherichia coli strains reveals that only 40% of their genes are common compared to 99% of the genes of humans and great apes. This testifies to the remarkable evolutionary potential and versatility of this bacterial taxon. Indeed, pathogenic Escherichia coli strains have acquired during evolution a repertoire of virulence genes, which allow them to colonize new ecological niches by bypassing the host's defense mechanisms. The expression of a specific repertoire of virulence factors is correlated with a particular pathology and allows defining different phatovars **(Dobrint, 2005)**.

1.7.1. Genomic plasticity of *Escherichia coli*

Over the past 10 years, 250 bacterial genomes, mostly of medical interest, have been deciphered and several hundred more are being sequenced and annotated. In silico comparison of commensal and pathogenic *Escherichia coli* revealed that chromosomal genes are ordered in a relatively conserved manner from one strain to another. The other striking fact of this comparative genomic analysis is the presence in pathovars of additional genetic

material (10 to 30%) **(Dobrint, 2005)**. The plasticity of the *Escherichia coli* genome is at the basis of this process. The complete genome sequence of several *Escherichia coli* strains shows the presence of numerous insertion sequences (IS), bacteriophagic sequences, as well as other unusual sequence ranges that testify to the extraordinary genome plasticity of this bacterial genus. The clinical isolates of *Escherichia coli* have the largest genomes, whereas the non-pathogenic laboratory *Escherichia coli* genome is 4.63 Mb. Thus, it appears that the gap between commensal and pathogenic *Escherichia coli* is due to the acquisition of repertoires of virulence genes **(Machado *et al.*, 1998)**.

It is possible that the acquisition of these genes is facilitated by an important ability to mutate. Indeed, more than 1% of *Escherichia coli* or *Salmonella* isolates involved in foodborne infections are "mutators" with a strong tendency to mutate, a phenomenon correlated with a deficiency in certain DNA repair systems. Virulence genes are most often located on transmissible genetic elements such as transposons, plasmids or bacteriophages. In addition, they can be clustered on large blocks of chromosomal DNA called "virulence islands". **(Machado *et al.*, 1998)**.

1.7.2. Comparison between the genome of pathogenic and commensal strains

Comparing the genomes of pathogenic and commensal strains, there is a difference in size and organization. Thus, the evolution of the different pathovars has been done through the acquisition of new sequences, inserted on the chromosome or on plasmids. These sequences are mostly found on PAIs, conferring the virulence of the pathovars. However, the insertion of smaller gene clusters has also been demonstrated. These gene clusters confer advantages to the pathogenic bacteria, and allow their survival. Thus, to conventional modeling of the bacterial genome divided into a backbone of genes necessary for the life of bacteria **(Mokady *et al.*, 2005)**.

1.7.3. Genetic diversity

E. coli is the most studied bacterial species to date. It is within this species that the most genomes are available, fully sequenced and annotated: approximately 3,690 genomes of *E. coli* were available in GenBank as of November 2015. **(Blattner *et al.*, 1997).** Their genome consists of 4.6 million base pairs encoding about 4,200 proteins. This testifies to the remarkable evolutionary potential and versatility of this bacterial taxon **(fig.11)**. Indeed, pathogenic *Escherichia coli* strains have acquired during evolution a repertoire of virulence

genes, which allow them to colonize new ecological niches by bypassing the host's defense mechanisms **(Dobrint, 2005)**.

The set of genes of the *E. coli* species constitutes the pan-genome of this bacterial species where currently more than 18,000 genes have been listed **(Elsaset, 2011)**.The *E. coli* pan-genome consists of three parts:

- **The core-genome** or the universal genome which groups the genes common to all strains, it is the stable part of the genome. The core-genome encodes the vital functions of the cell. This part includes about 2,000 genes and represents about 11% of the *E. coli* genome **(Dobrint, 2005)**.
- **The single genome** contains the genes specific to a strain. It is a variable part, not common to all strains, which encodes proteins involved in the improvement of the "fitness" of the bacterium, in the mechanisms of colonization and adaptation of the bacterium to the different environmental conditions encountered without obvious function and is conditioned by the presence of mobile genetic elements such as plasmids, bacteriophages or islands of pathogenicity. These elements are integrated into the bacterial chromosome or replicated independently thanks to the cellular machinery. This part groups together approximately 63% of the genome **(Dobrint, 2010)**.
- **The peripheral** or volatile genome includes all the genes present only in a subset of strains. This part is used to distinguish strains from each other and carries information about serotypes. The function of most of the genes in this category is not well known. This part represents about 26% of the pan-genome **(Dobrint, 2010)**.

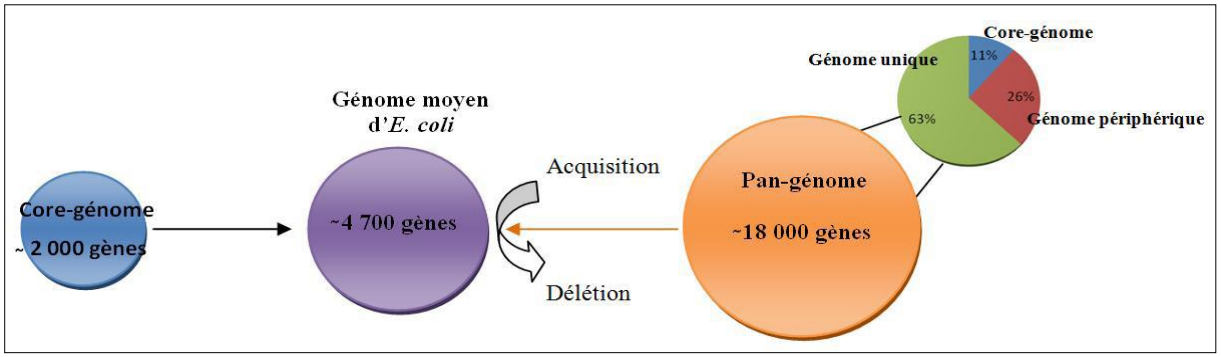


Fig. 11: Representation of the composition of the average *E. coli* genome **(Ferens and Hovde, 2011).**

1.7.4. The mobile elements of pathogenicity

The genetic elements thus exchanged can be grouped into 4 categories: plasmids, transposons, phages and islands of pathogenicity **(Mainil, 2003)**.

- **Plasmids**

They are defined as double-stranded, circular DNA structures that are autonomous from the bacterial chromosome in terms of their control and replication. Their size varies from a few kilo bases to a few hundred kilo bases. They can be transferred horizontally by conjugation or mobilization, between bacteria of the same species or not. They are the most mobile elements of the genome **(Mainil, 2003)**.

- **Transposons**

Transposons are DNA sequences which can be transferred with or without replications from a chromosome to one or more plasmids. They are not autonomous **(Guiraud, 1993)**.

- **Phages**

Phages, incorporated into the chromosome or plasmid, bring additional genetic information to their host bacteria **(Guiraud, 1993)**.

- **Genomic and/or pathogenicity islands**

The genomic and/or pathogenicity islands group together genes which confer on their host bacteria traits of versatility and adaptability as well as virulence. These islets are characterized by a G+C content that differs from the rest of the genome, by repeated sequence motifs that flank their ends, and by a chromosomal insertion at the level of transfer RNA genes. These characteristics bear witness to their acquisition through a trans horizontal process (**fig. 12**).

They comprise approximately between 10 and 200 kb, which represents large parts of the genome **(Hacker, 2000)**. They were initially identified in uro-pathogenic *Escherichia coli*. **(Dobrindt *et al.*, 2002).** The islets therefore confer on the host bacterium additional functions or capacity for adaptation to the environment, resistance to antibiotics, metabolic capacities (the capture of iron), capacity for symbiosis or virulence, for example most of the genes coding for production of virulence factors in *Escherichia coli* found in islands of pathogenicity **(Hacker, 2000)**.

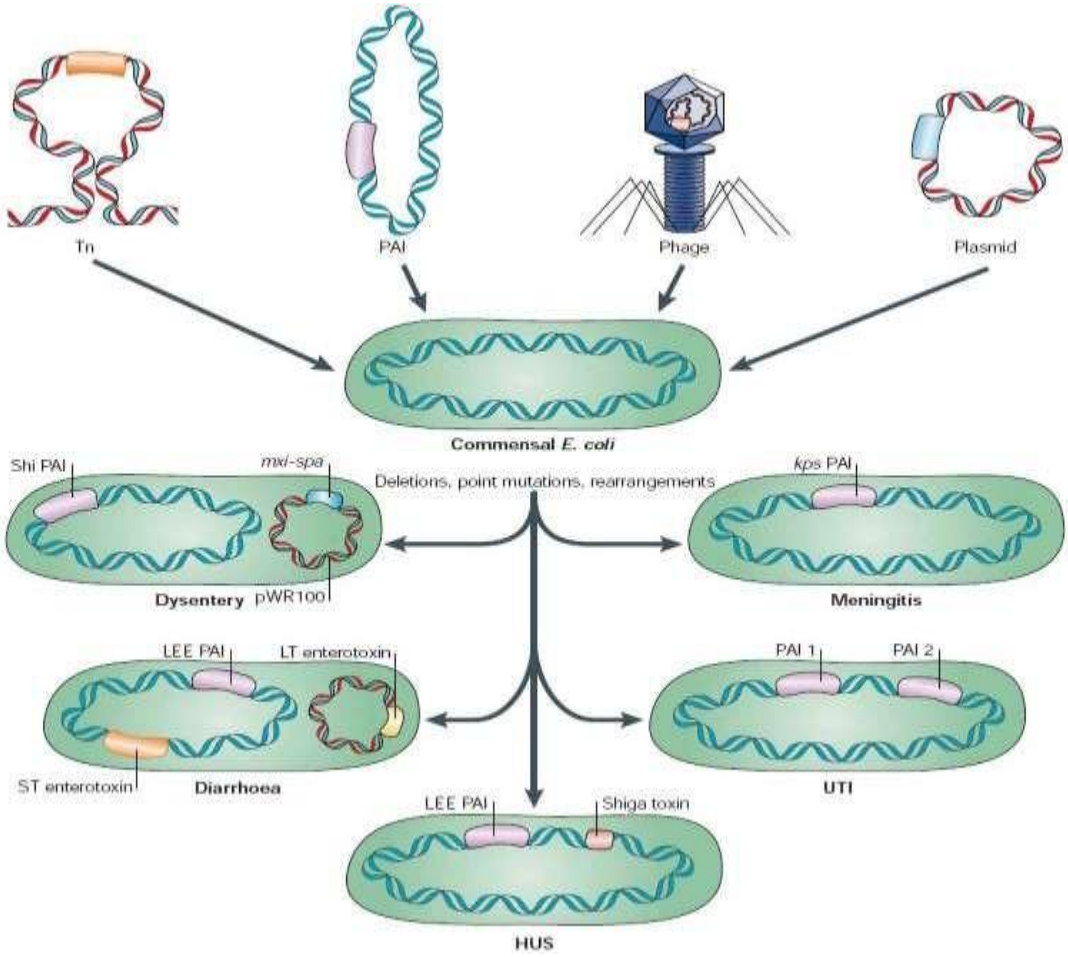


Fig. 12: Mobile genetic elements **(Kaper *et al.*, 2004)**.

1.8. Pathogenicity

1.8.1. Infections

E. coli is a commensal species that interacts with its host in a mutualistic relationship. However, *E.coli* can also be an opportunistic pathogen or an obligate pathogen due to the expression of specific virulence factors. Opportunistic or obligate pathogenic strains have developed different modes of interaction with their host resulting in various clinical signs. They are conventionally separated into two groups: intestinal pathogenic *E. coli* (InPEC) responsible for gastroenteritis and extra-intestinal pathogenic *E. coli* (ExPEC) responsible for urinary tract infections, peritonitis, nosocomial pneumonia, meningitis or more sepsis. Based on the modes of host/bacteria interaction and clinical signs of infection, strains are classified into “pathovars” or “pathotypes” which group together strains of specific serotypes **(Croxen and Finlay, 2010)**.

1.8.2. Infections extra-intestinal

Extra-intestinal pathogenic *E. coli* (ExPEC), are not associated with infections when these strains are found in the intestinal tract. However, when they colonize tissues outside the intestine, they can caused significant infections **(Johnson and Russo, 2002).**ExPECs can be grouped into four categories:

- Uropathogenic *E. coli* (UPEC)
- *E. coli* associated with sepsis (SEPEC)
- *E. coli* associated with neonatal meningitis (NMEC)

ExPECs possess a wide range of virulence factors. ExPECs appear to have been created by the accumulation of certain virulence factors in *E. coli* strains belonging to the B2 and D phylogenetic groups **(Johnson and Russo, 2002)**.

➢ **Uropathogenic *E. coli* (UPEC)**

They are responsible for the majority (90%) of infections occurring in the normal urinary tract: cystitis, pyelonephritis. Their pathogenicity is characterized by adhesion to uro-epithelial cells thanks to several types of adhesins and other factors such as alpha hemolysin and siderophores **(Söderström *et al.*, 2008)**.

➢ ***E. coli* associated with sepsis (SEPEC)**

Certain serotypes of *Escherichia coli* (K1 in particular) are capable of inducing serious neonatal infections. These are sepsis possibly complicated with meningitis **(Ségolène, 2016)**.

➢ ***E. coli* associated with neonatal meningitis (NMEC)**

Neonatal meningitis-associated *E. coli* (NMEC) causes meningitis in newborns, with 15-40% of affected newborns dying. A significant number of survivors suffer severe neurological defects. Bacteria are carried by the hematogenous (blood) route **(Kaper *et al.*, 2004)**.

1.8.3. Intestinal infection

Pathogenic strains of *Escherichia coli* are recognized as agents responsible for diarrheal syndromes of food or water origin **(Andrade *et al.*, 1989)**. Four main pathotype groups or intestinal pathovars are described according to the clinical signs **(Lavigne, 2004)**.

- Enterotoxigenic *E. coli* (ETEC)
- Enteropathogenic *E. coli* (EPEC)
- Enteroinvasive *E. coli* (EIEC)
- Enterohemorrhagic *E. coli* (EHEC)

- **Enterotoxigenic *E. coli* (ETEC)**

These strains trigger acute "cholera-like" diarrhea in children under two years of age, especially in developing countries, and are also considered responsible for a significant number of "turista" travelers' diarrhea **(Haslay and Leclerc , 1993).**

These strains are pathogenic by secretion of enterotoxin, which is encoded by a plasmid. The toxin is most often a heat-labile toxin or LT (very similar to that of vibrio cholerae), but sometimes thermostable or ST. LT causes, after a cascade of biochemical events, intestinal hypersecretion of water and chlorides, intestinal hyperperistalsis and explosive diarrhea, which last 1 to 3 days. The plasmids involved also carry genes responsible for the production of pili or fimbriae (adhesins) that allow the attachment of *E.coli* to the intestinal mucosa **(Berche *et al.*, 1991).**

- **Enteropathogenic *E. coli* (EPEC)**

At the origin of epidemic enteritis previously also called infantile gastroenteritis (GEI). These *Escherichia coli* were a major cause of diarrhea in infants who were rampant in maternity wards and nurseries. EPECs colonize the intestinal mucosa by strongly adhering to intestinal enterocytes, produce attachment and effacement lesions characterized by the localized destruction of microvilli of the brush border and by inducing alterations in the cytoskeleton of epithelial cells **(Riley *et al.*, 1983)**.

- **Enteroinvasive *E. coli* (EIEC)**

These strains cause acute diarrhea or dysenteric syndromes ("dysentery-like") with the presence of mucus, blood, and leukocytes in the stool. EIECs have the property of penetrating into cells by invasion of the colonic mucosa. The virulence of these strains is linked to the presence of a plasmid very similar to that known in Shigella. Some of these strains would produce a cytotoxin like Shigella **(Berche *et al.*, 1991).**

- **Enterohaemorrhagic *E. coli* (EHEC)**

The O157:H7 serotype is the most common. These strains cause bloody diarrhea, without pus or fever, and hemorrhagic colitis.After fixing on the surface of the cells of the mucosa, abrasion of the border in brush of intestinal villi, they produce powerful cytotoxins, known as virotoxins and called SLT (Shiga-like) because they resemble the toxin of Shigella dysenteriae. They are produced under the control of chromosomal (integrated) converting bacteriophages. SLTs disseminate through blood and inhibit protein synthesis. EHEC harbor a plasmid encoding an adhesin **(Berche *et al.*, 1991).**

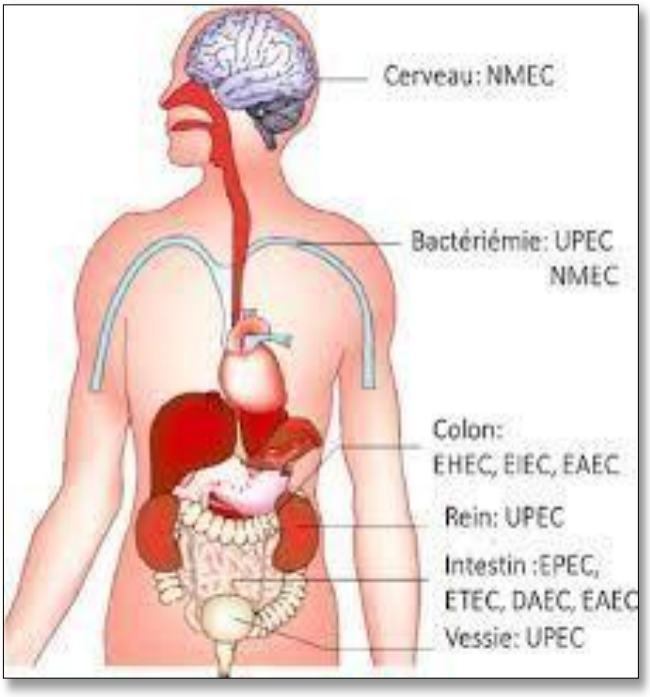


Fig. 13: Infection sites of different *Escherichia coli* pathovars **(Croxen *et al.*, 2010)**.

1.8.4. The main stages of *Escherichia coli* infection

After ingestion, *Escherichia coli* are able to resist gastric acidity. They pass through the small intestine and reach the colon.

- The EHEC would then be able to adhere and colonize the colonic mucosa.

-The Stxs toxins, then secreted by the bacteria, would cross the intestinal epithelium by transcytosis, would reach the circulatory system and could thus reach the specific cellular receptors (Gb3) present on the surface of the endothelial cells, mainly intestinal, renal and cerebral **(Grimont *et al.*, 2003).**

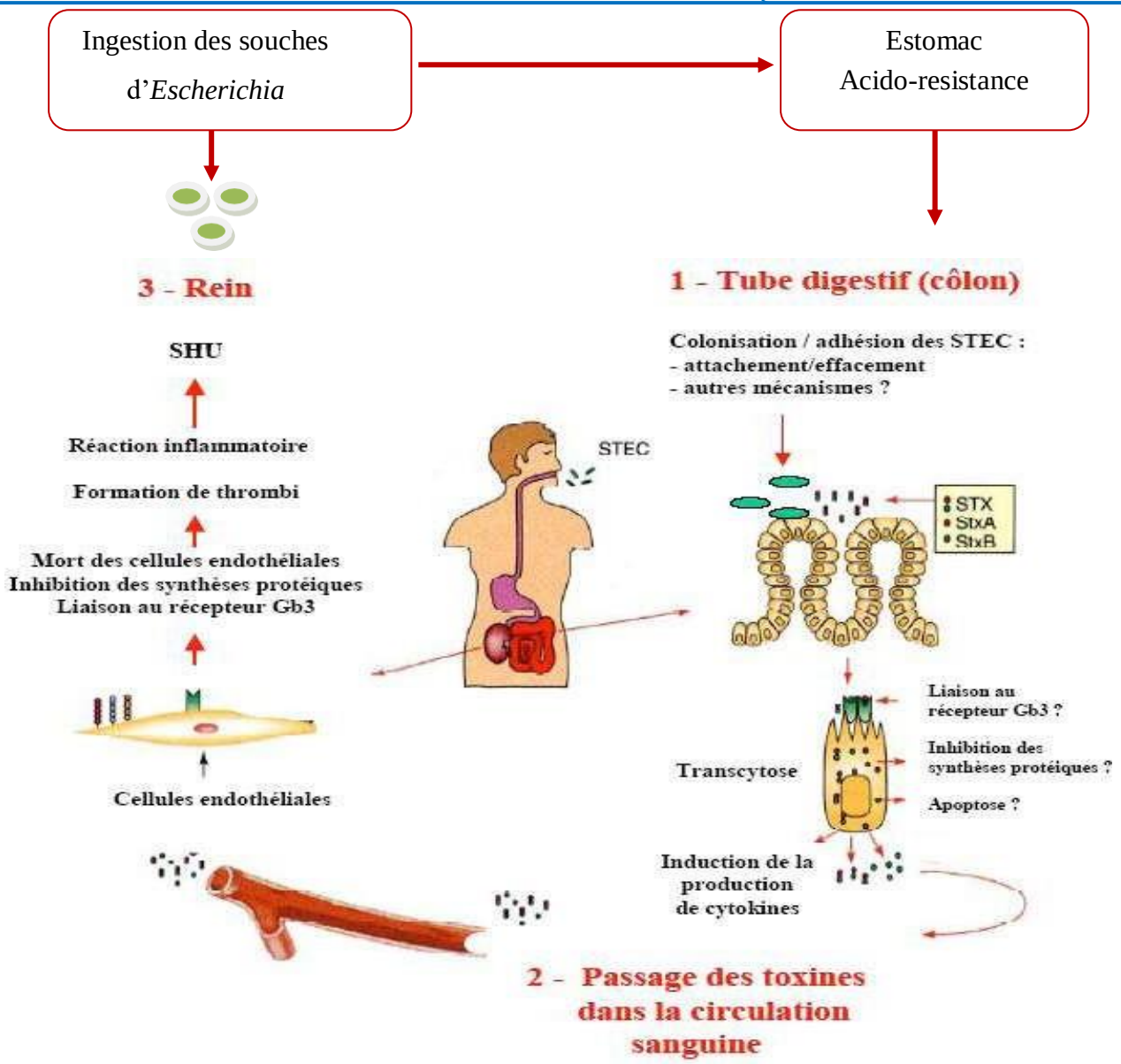


Figure 14: Main steps of the infectious process of *Escherichia coli* **(Grimont *et al.*, 2003).**

1.8.5. Clinical aspect of *Escherichia coli* infections

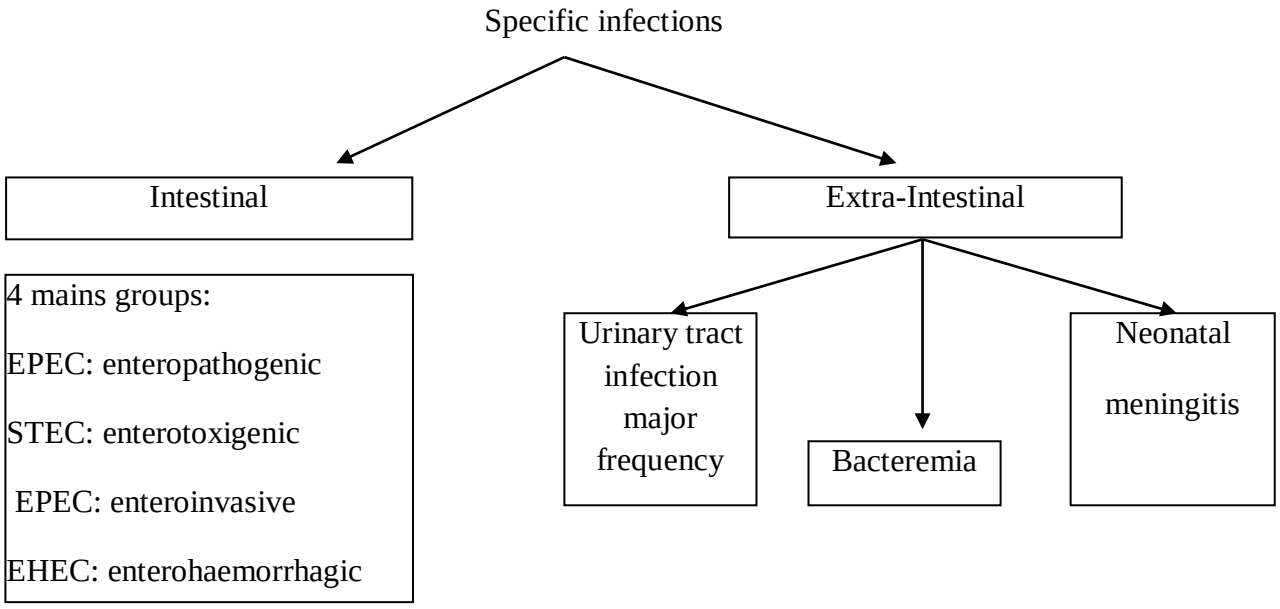


Fig. 15: Clinical aspects of *Escherichia coli* infections **(Philippe *et al.*, 2004)**.

1.8.6. Concept of pathogenicity of *Escherichia coli*

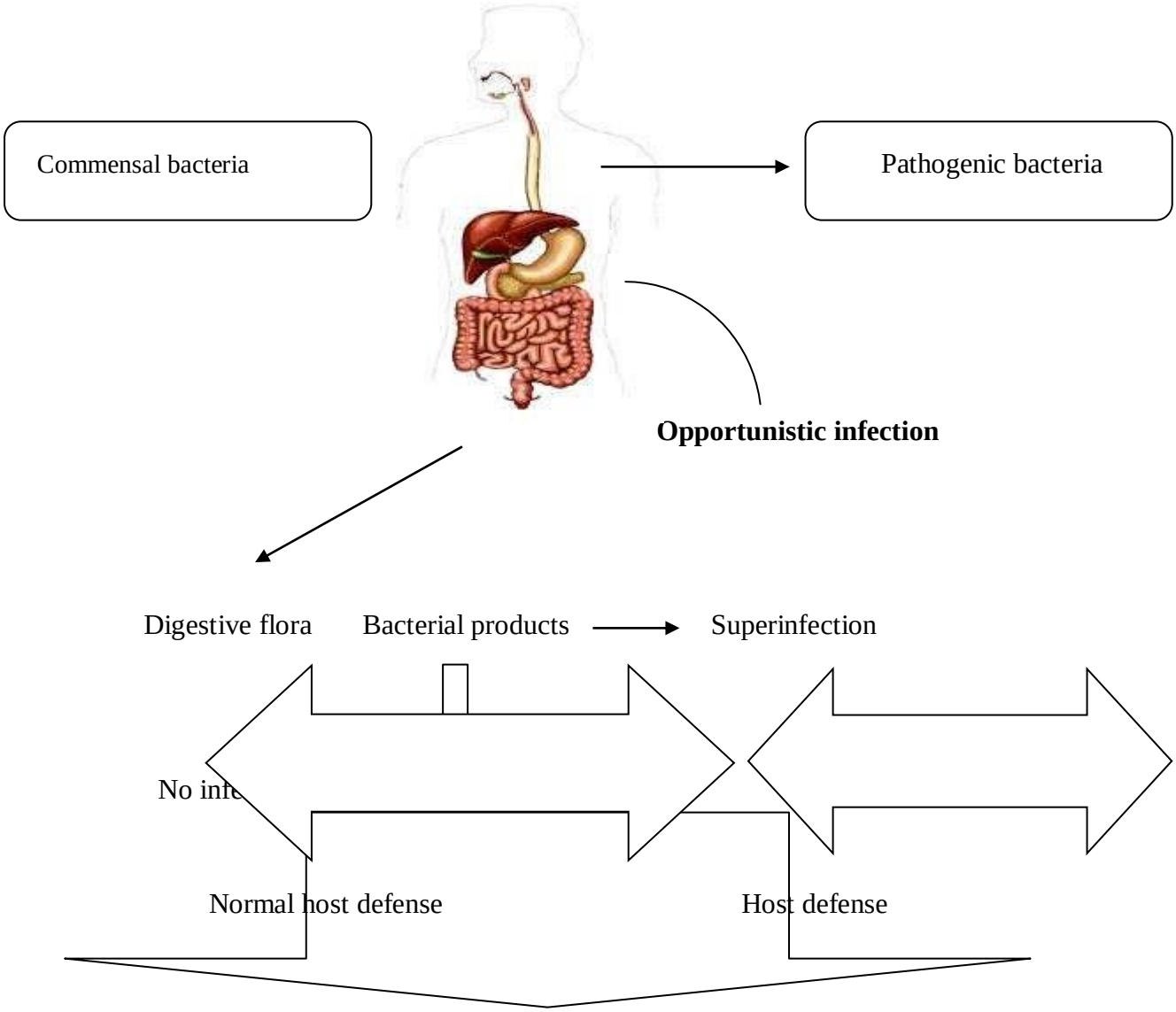


Fig. 16: The concepts of pathogenicity of *Escherichia coli* **(Philippe *et al.*, 2004).**

1.9. Virulence factors

Defining virulence factors and understanding the mechanisms involved in the pathogenicity of *Escherichia coli* strains are essential prerequisites for assessing the public health risk associated with the existence of these pathogens. Thus, the combination of virulence factors involved in the pathogenicity of strains still remains to be determined. The study of the pathogenicity factors of *Escherichia coli* has shown that in the species there are many variants of the factors **(Escobar *et al.*, 2006)**.

1.10. Capsule

It is polysaccharide in nature. There are 80 different immunological varieties (K antigens). The capsule makes phagocytosis more difficult and inhibits the action of complement.

The K1 type capsule is not very immunogenic (it has the same structure as the group B meningococcal capsule). Type K1 *E. coli* are responsible for the majority of neonatal infections **(Nauciel *et al.*, 2007)**.

1.11. Adhesions

Adhesins give strains of *Escherichia coli* the property of attaching to epithelial cells, protein in nature; they are most often carried by common pili. Adherence is an essential step in the pathogenesis of enteric bacteria. The main ones are the factors involved in the development of enterocyte attach and efface (A/E) lesions, which is responsible for diarrhea, and are characterized by effacement of the microvilli of cells of the intestinal epithelium in the area contact between the bacterium and the target cell **(Mainil, 1999)**. Adhesins give the bacterium the ability to attach to specific host receptors **(Mainil, 1999).**

1.12. Toxins

Some groups of *E. coli* produce specific toxins. The enterohemolysins of enterohemorrhagic *E. coli*, the ST and LT enterotoxins, and the SLT1 and SLT2 cytoxins are toxins that alter the integrity of enterocytes **(Achri and Lalouatni, 2018).**

The mode of action of these toxins is in different ways: **(Achri and Lalouatni, 2018)**

- ✓ By facilitating tissue invasion.
- ✓ By reading host cells (hemolysin α).
- ✓ By blocking protein synthesis (Shiga toxins).

1.13. Iron capture systems

Iron is an essential metal for the survival of most micro-organisms, UPEC have developed small molecules called "Siderophores" which have a great affinity for iron which allows them to extract it from the ferroproteins of the host and colonize the urinary tract where iron is poorly available. **(Kathleen, 2013).**

1.14. Proteins

Outer membrane proteins and LPS give bacteria the ability to escape the bactericidal activity of host serum by opposing complement fixation **(Levine , 1987).**

1.15. Transmission mode

1.15.1. Food transmission

Meat products are the source of a large number of *Escherichia coli* infections **(Vernozy-Rozand *et al.*, 2005).**

Beef is the major source of contamination mainly due to undercooking **(Barany and Roberts, 1995)**. Meat from other meat animals or poultry has also been implicated **(Paton, 2001)**.

-Unpasteurized milk and milk products have also been the source of epidemics.

The current route of contamination of milk is contamination from bovine faeces during milking. Indeed, the conditions of milking and the environment in which it takes place play a major role in the contamination of milk **(Allerberger, 2001).**

- Raw fruits and vegetables (lettuce, radishes, spinach, onions...) can be directly contaminated by irrigation water, from contaminated soil following the spreading of livestock effluents or via the activity of soil fauna **(Baranyi and Roberts , 1995).**

1.15.2. Transmission through direct contact with farm animals and their environment

Transmission of *Escherichia coli* through direct or indirect contact with farm animals or their droppings has been described in sporadic cases **(O'brien *et al.*, 1982)**. In addition, the rate of healthy *Escherichia coli* carriers is higher in the population living in constant contact with animals **(Evans *et al.*, 2002)**. For example, among English farmers, *Escherichia coli* seroprevalence ranges from 1.6% to 5%.

Contaminated soil from farm animals has also been implicated in *Escherichia coli* outbreaks, particularly during outdoor events such as festivals or tourist camps on soil previously grazed by ruminants **(Ogden *et al.*, 2002)**.In Scotland, contaminated soil has been implicated in 11% of environmental *Escherichia coli* infections **(Strachan *et al.*, 2006)**.

1.15.3. Inter-human transmission

Most cases are caused by indirect contamination by people in contact with patients. Moreover, this transmission is more significant when general hygiene is poor and close contacts are present. Fecal-oral transmission is a real concern in foster homes **(Sugiyama *et al.*, 2005)**, day care centers, and psychiatric centers. This method of transmission is also responsible for the spread of infection within families and hospitals.

The median duration of trans is 13 days for enterohaemorrhagic *Escherichia coli* and 31 days for HUS **(Bielaszewska *et al.*, 1997)**.

1.15.4. Water transmission

Waterborne outbreaks are usually associated with drinking water or accidental ingestion of water while swimming. Between 1997 and 2004, the U.S. surveillance system reported that 6% of waterborne outbreaks were related to EHEC **(Dziuban *et al.*, 2006)**.

- Consumption of untreated well water, private spring water, and tap water has resulted in isolated cases of *Escherichia coli* infection and outbreaks **(Jackson *et al.*, 1998).**
- Accidental ingestion of water while swimming in a lake or other natural water body **(Jackson *et al.*, 1998)**.

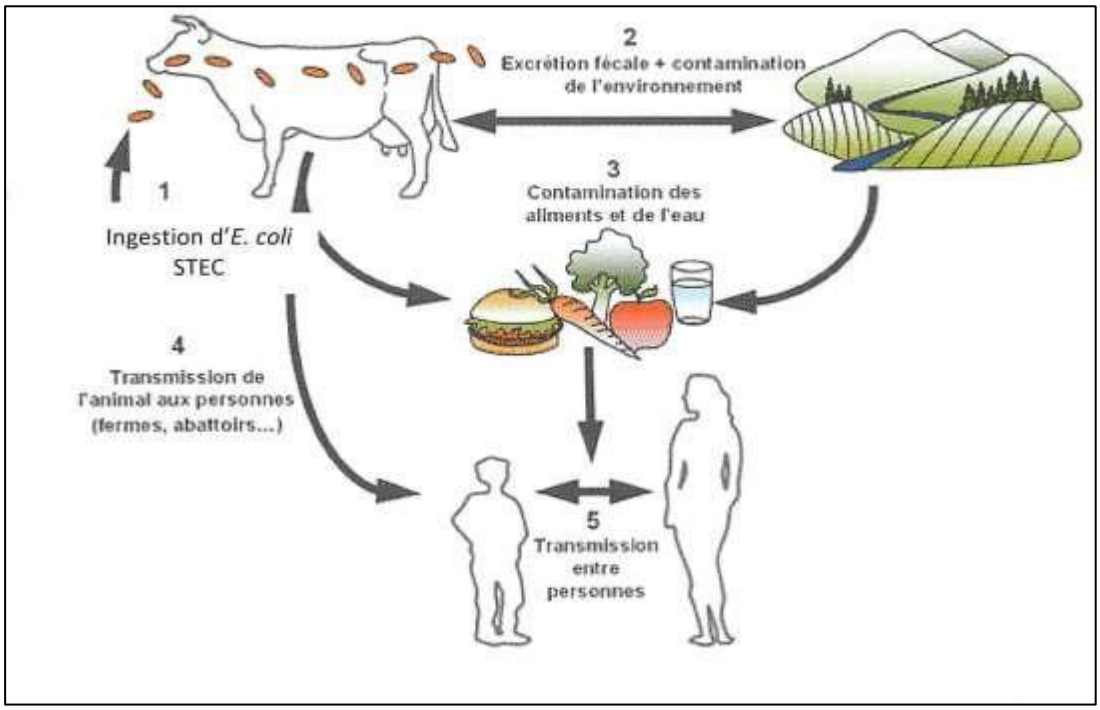


Fig. 17: *Escherichia coli* contamination routes **(Monnet, 2000)**

1.16. *Escherichia coli* infection diagnose

To diagnose *Escherichia coli* infection in a laboratory, several tests can be performed depending on the type of sample and the symptoms presented by the patient.

Some of the most common tests are:

Stool culture: This involves collecting a stool sample and incubating it in a laboratory to check for the presence of *E. coli* bacteria. This test can also help identify the specific strain of *E. coli* causing the infection.

Blood culture: This test involves drawing a blood sample and incubating it in a laboratory to check for the presence of *E. coli* bacteria in the bloodstream.

Urine culture: This test involves collecting a urine sample and incubating it in a laboratory to check for the presence of E. coli bacteria in the urinary tract.

Enzyme-linked immunosorbent assay (ELISA): This test detects antibodies to *E. coli* in blood, urine, or stool samples. ELISA can also be used to detect toxins produced by some strains of *E. coli*.

Polymerase chain reaction (PCR): This test can detect the DNA of *E. coli* bacteria in samples such as stool, urine, or blood.

It is important to note that the laboratory diagnosis of *E. coli* infection requires careful attention to sample collection and handling, as well as the use of appropriate culture media and testing techniques.

1.16.1. Sampling conditions

***Collecting the sample:** The sample can be collected from a variety of sources, such as fecal matter, sewage, or contaminated food. It's important to use sterile collection methods to avoid contamination from other bacteria.

***Transporting the sample:** The sample should be transported to the laboratory as quickly as possible to prevent any changes in the bacterial population. If transport is delayed, the sample should be stored at a temperature of 4°C or lower.

* **Preparing the sample:** The sample should be prepared by diluting it in a sterile buffer solution to achieve a suitable bacterial concentration for analysis.

* **Growing the bacteria:** *E. coli* can be grown on a variety of media, including MacConkey agar, EMB agar, and nutrient agar. The plates should be incubated at a temperature of 37°C for 24-48 hours.

***Identifying the bacteria:** *E. coli* can be identified by its characteristic colony morphology and biochemical reactions, such as lactose fermentation and indole production.

***Testing for antibiotic resistance:** If necessary, the *E. coli* isolate can be tested for antibiotic resistance using standard methods such as the Kirby-Bauer disc diffusion method or broth microdilution.

It's important to note that specific sampling conditions may vary depending on the purpose of the analysis and the requirements of the laboratory.

✓ **Sampling**

Sampling *E. coli* in a laboratory is a common practice to detect and quantify the presence of this bacterium in various samples, such as water, food, fecal matter, etc.

Here are some general steps for performing *E. coli* sampling in a laboratory:

Sample collection: Collect the sample from the presumed source of contamination. For example, if you want to detect the presence of *E. coli* in water, you need to collect a water sample.

Sample preparation: Depending on the type of sample collected, it is often necessary to prepare it before testing for the presence of *E. coli*. For example, if you collect a water sample, you may need to filter or concentrate it to increase the amount of *E. coli* present in the sample.

Bacterial culture: Bacterial culture is a key step in detecting *E. coli*. You can grow the collected sample on a specific selective culture medium for E. coli. After incubation, E. coli colonies appear on the culture medium.

Confirmation: To confirm that the bacterial colonies are indeed *E. coli*, it is often necessary to perform specific biochemical and molecular tests. For example, you can use tests to detect the presence of the beta-glucuronidase enzyme that is specific to *E. coli*.

It is important to follow appropriate protocols and safety standards to avoid cross-contamination or exposure to bacteria during sampling and bacterial culture.

1.16.2. Macroscopic examination of *E.coli*

Macroscopic examination of *E. coli* refers to the observation of the bacterium with the naked eye or with the help of a magnifying glass. *E. coli* is a rod-shaped, gram-negative bacterium that is commonly found in the lower intestine of warm-blooded animals, including humans.

Here are some characteristics of *E. coli* that can be observed macroscopically:

*** Size and shape:** *E. coli* cells are rod-shaped and measure approximately 1-3 micrometers in length and 0.5-1 micrometer in diameter.

***Color:** *E. coli* colonies grown on agar plates are usually smooth, round, and slightly raised, with a slightly glossy appearance. They are often described as a pale yellow or beige color.

***Texture:** *E. coli* colonies are generally smooth and slightly slimy in texture.

***Growth rate:** *E. coli* is a fast-growing bacterium, with a doubling time of approximately 20 minutes under ideal laboratory conditions.

*** Odor:** *E. coli* has no characteristic odor.

*** Motility:** *E. coli* is a motile bacterium and can move around using its flagella.

In summary, macroscopic examination of *E. coli* can reveal its size, shape, color, texture, growth rate, and motility. These observations can provide valuable information for identifying and studying this important bacterium

1.16.3. Microscopic examination of *E. coli*

E. coli is a type of bacteria commonly found in the intestines of humans and animals. It is a gram-negative, rod-shaped bacterium that is often used as a model organism in microbiology research.

Microscopic examination of *E. coli* typically involves the use of a compound light microscope. To prepare a sample for observation, a small amount of bacterial culture is placed on a microscope slide and stained with a suitable dye, such as crystal violet or safranin. The staining process helps to increase the contrast between the bacterial cells and the surrounding medium, making it easier to see the individual cells.

Under the microscope, *E. coli* appears as a small, rod-shaped bacterium with a diameter of around 0.5 micrometers and a length of 2-3 micrometers. The cell wall of *E. coli* is made up of a thin layer of peptidoglycan, surrounded by an outer membrane containing lipopolysaccharides. The lipopolysaccharides are responsible for the endotoxin activity of *E. coli*, which can cause inflammation and other health problems.

1.16.4. Gram staining

This is a technique that makes it possible to distinguish bacteria according to the structure of their cell wall.

1.16.4.1. Biochemical tests

The microscopic appearance and the characteristics of the colonies are not sufficient to accurately identify the bacteria. It is necessary to look for other characters, mainly biochemical or metabolic characters. When the germ is obtained in pure culture, it is re-seeded on various identification media which make it possible to study its enzymatic equipment.

The identification allows first of all recognizing the colonies of *E. coli* according to their morphological and cultural characteristics, and then biochemical tests are carried out on an API 20E gallery.

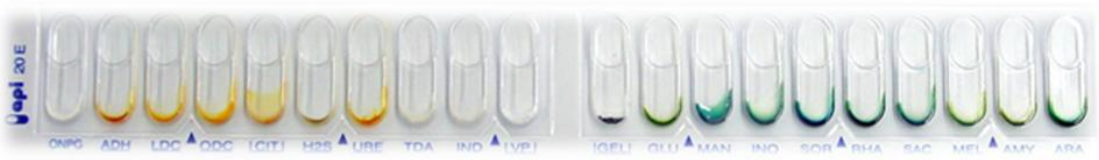
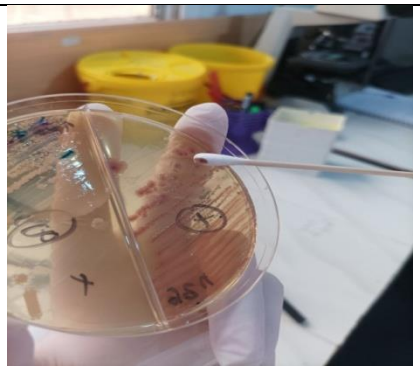
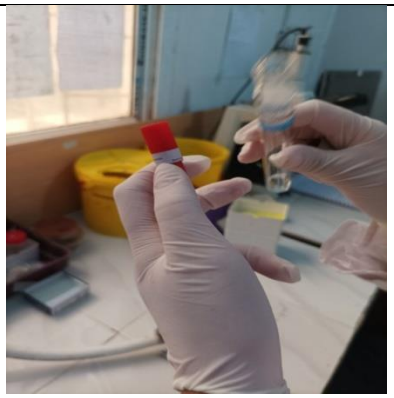
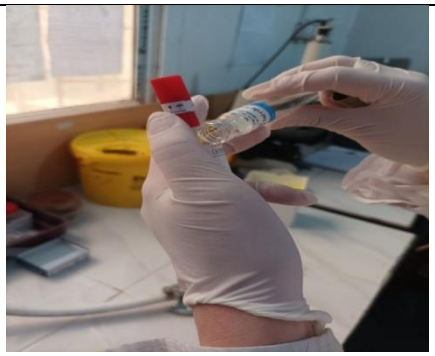
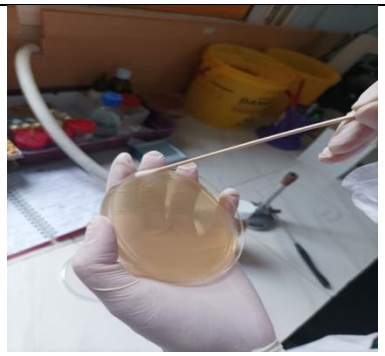
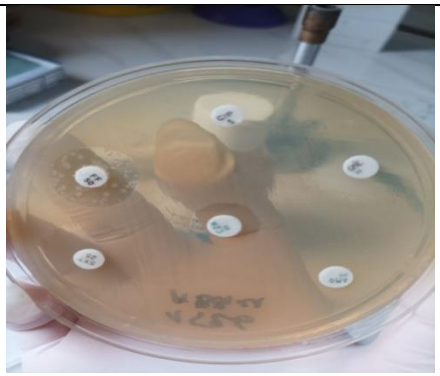
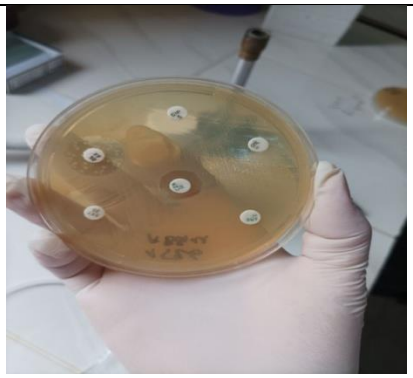


Fig. 18: An API 20E gallery before seeding.

A stock suspension is prepared by removing, using a sterile Pasteur pipette, a few well-isolated colonies of *E. coli* from a positive Petri dish; then inoculated into a sterile screw tube containing sterile physiological water.The well-homogenized bacterial suspension must have an opacity equivalent to 0.5 Mac Ferland (MF).(**Table 04**).

A positive reaction results in a change in the color of the medium or in the presence of a cloudiness either spontaneously after incubation, or after addition of one or more reagents. The reactions are read using the reading table (see Appendix) and identification is obtained using an analytical catalog or identification software. (**Fig. 19**).

Table 04: Stages Biochemical tests **(Personal photos, 2023).**

 Remove a few well-isolated colonies of *Escherichia coli* from a positive Petri dish	 They are seeded in a sterile spiral tube containing sterile physiological water.
 Leave a time	 Replant in a petri dish
 Add reagents	

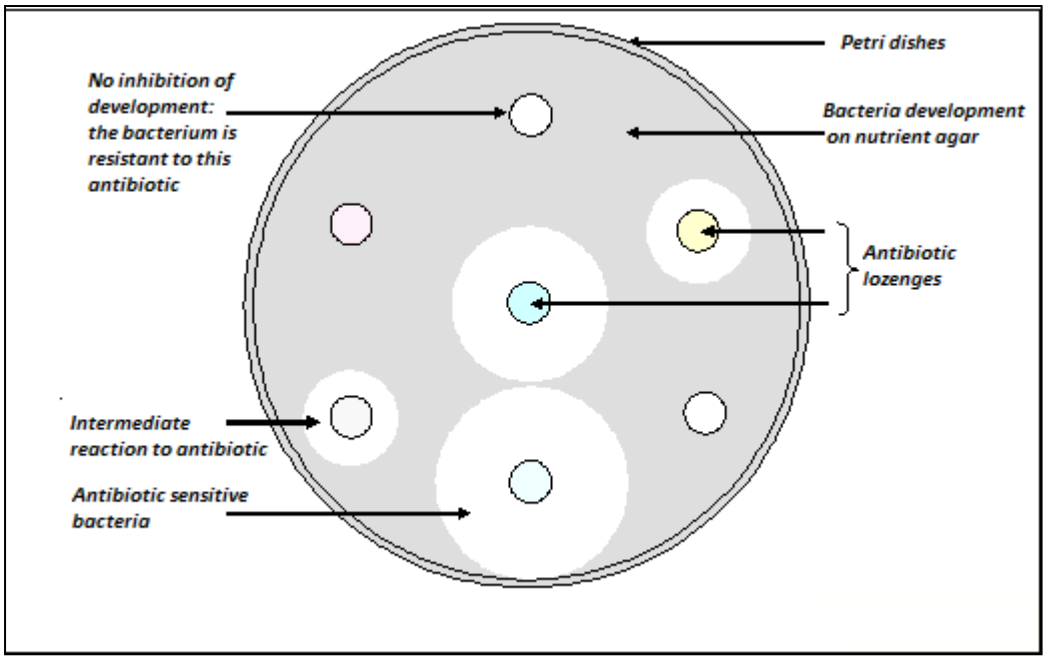


Fig. 19: Example of an antibiogram result (**Boukhemis and Boutersa, 2015)**

1.17. Treatment against *E. coli*

1.17.1. Curative treatment

The treatment of urinary and biliary infections is based on antibiotic therapy and the correction of favourable factors (anatomical, stones...). The curative treatment of acute diarrhea caused by coli bacteria is based on rehydration. The treatment of peritoneal infections is based on drainage and antibiotic therapy **(Croxen, 2010)**.

Escherichia coli is sensitive to all beta-lactam antibiotics despite the production of a non-inducible chromosomal cephalosporinase of the AmpC type which can lead to a reduction in sensitivity to aminopenicillins, their combinations with clavulanate and/or C1G in certain strains **(Croxen, 2010).**

1.17.2. Preventive treatment

Preventive treatment is mainly based on general hygiene measures, especially food hygiene, and individual hygiene measures **(Croxen, 2010)**.

Vaccine: At present, no effective vaccine is available on the veterinary market. However, although a number of vaccine trials have been conducted using attenuated strains in experimental models and have been successful with homologous strains, they are still ineffective against infections with hetero-logous field strains **(Stordeur, 2002)**.

Researchers from the University of Michigan (USA) published on September 18, 2009 in the journal PLOS Pathogens promising results obtained on mice that create a strong immune resistance against *Escherichia coli* bacteria.

1.18. Prevention of *Escherichia coli* infection

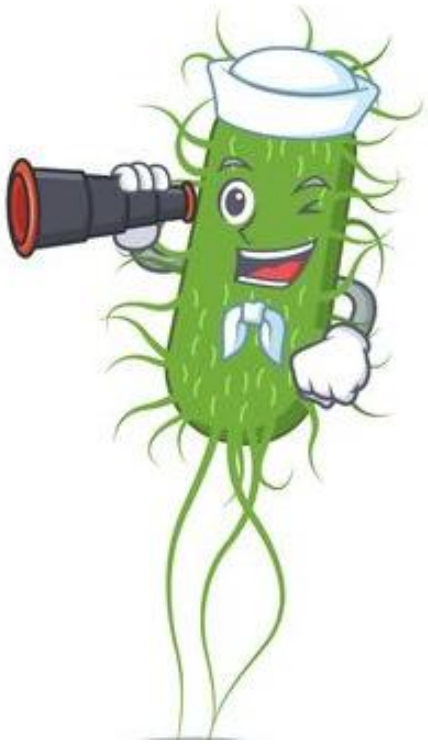
-Observe good personal hygiene:

Wash hands thoroughly with soap and water after using the toilet or changing a diaper, after touching animals or coming into contact with their feces, after handling raw meat or poultry, and before preparing or eating food **(Chapman *et al.*, 1997).**

-Take food safety precautions Wash raw fruits and vegetables before eating. Cook all meats (meat, poultry and seafood) thoroughly. Avoid contact between cooked foods and poultry or other raw meat. Consume only pasteurized dairy products (milk, cheese, yogurt and ice cream) **(Bonardi *et al.*, 1999).**

-Drink properly treated water Avoid drinking water from recreational water sources, such as swimming pools and hot tubs. Do not drink untreated surface water from lakes or streams. Boil water for one minute to kill all known pathogens such as Cryptospridium and *Escherichia coli*. Test your private well water twice a year for bacteria **(Blanco *et al.*, 1993).**

PRESENTATION OF THE STUDY SITE



2. PRESENTATION OF THE STUDY AREA

2.1. Geographic location

The wilaya of Mila is located in northeastern Algeria at an altitude of 464 m, and 70 km from the Mediterranean Sea. It is also in the eastern part of the Tell Atlas, a mountain range that extends from west to east across the northern territory of the country **(A.N.D.I., 2013).** The wilaya of Mila is limited:

- To the northwest by the wilaya of Jijel.
- In the North East by the wilaya of Constantine.
- In the West by the wilaya of Sétif.
- In the East by the wilayas of Constantine and Skikda.
- In the South East by the wilaya of Oum El Bouaghi.
- In the South by the wilaya of Batna.

The wilaya of Mila covers an area of 9373km² **(A.N.I.R.E.F., 2011).**

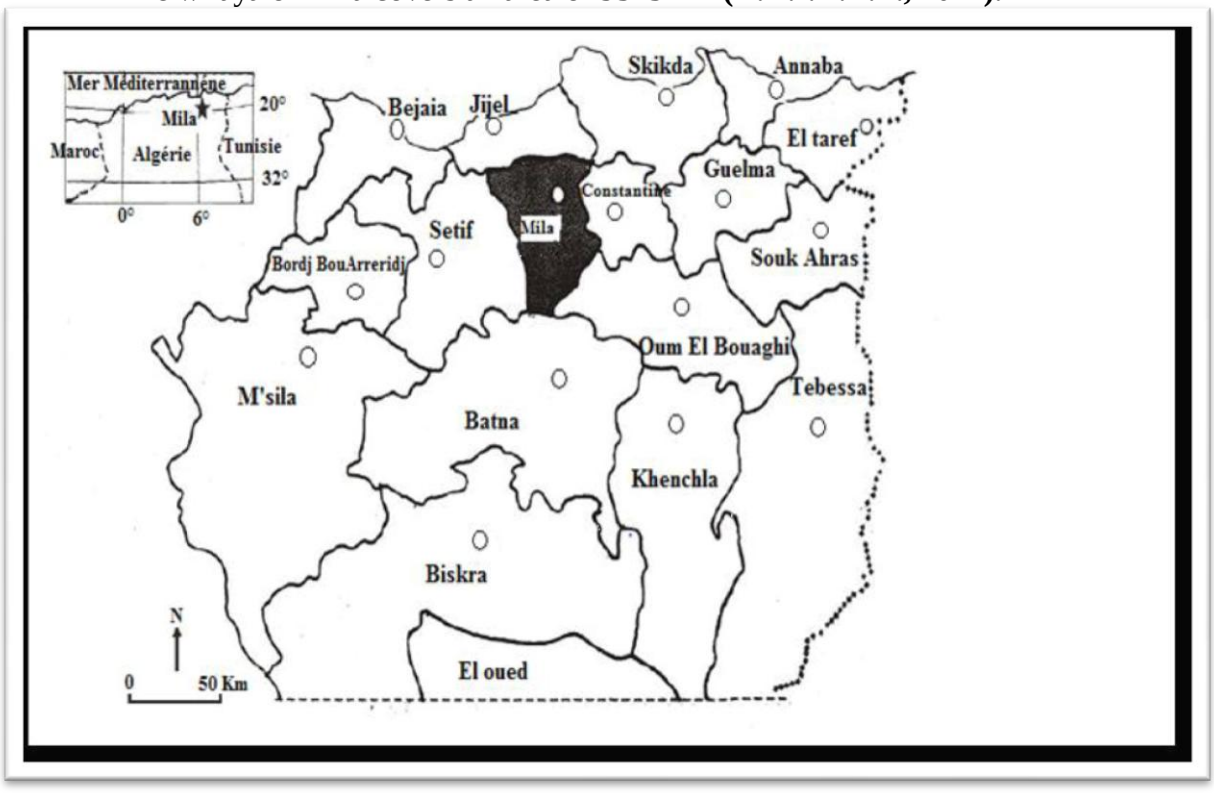


Fig. 20: Geographic location of the Mila region **(Doula and Ferhat, 2014)**.

2.2. Demographic situation

The wilaya of Mila is divided into 13 daïras. It covers an area of 3,480 km² with a population of 865,370 inhabitants, i.e. a density of 248.7 inhabitants/Km^2. This density varies

from one commune to another due to multiple regional specificities of an economic (agriculture, industry and trade), geomorphological (nature and relief of the land) and administrative (area allocated to each commune during the administrative division) nature. **(Abid, 2014).** Number of women is slightly higher than that of men, 408604 for 401766.

The population of the wilaya is relatively young, more than 50% is located in the age range of 1 to 24 years, or 420887 inhabitants, for a total of 810370 inhabitants. The population is largely rural and suburban. It is generally made up of workers on the land, both on the high plains and in the mountainous regions. The urban population, concentrated in the large cities, is still imbued with the values of rurality **(Seddiki *et al*., 2013)**.

2.3. Administrative Aspects

The wilaya of Mila was created during the last Algerian administrative division of 1984, with the city of Mila as the capital of wilaya 43 **(A.N.D.I, 2013)**.The wilaya of Mila has 13 daïras comprising 32 municipalities (**table 05)**.

Table 05: The administrative division of the Mila region **(Boularas and Kadjoudj, 2016)**.

Dairas	Municipalities
Mila	Mila- Ain Tine- Sidi Kkhlifa
Grarem Gouga	Grarem Gouga- Hamala
Sidi Merouan	Sidi. Mérouane- Chigara
Oued Endja	Oued Endja- Zeghaia- A. Rachdi
Rouached	Rouached- Tiberguent
Terrai Beinen	Terrai Beinen- AmiraArres- Tassala Lamtai
Ferdjioua	Ferdjioua- Y. B. Guecha
Tassadane.H	ZarzaTassadane Hadda- Minar
Bouhatem	Bouhatem- D. Bousselah
Ain Baidah H	Ain B.Ahrich- AyadiBerbes
Teleghma	Telaghma- OuedSeguen - El M'chira
Chelghoum Laid	Chelgoum El Aid- O. Atmania- AinMelouk
Tadjenanet	Tadjnanet- Ben Yahia A- OuledKhlouf

2.4. Vegetation

The plant cover is not very important; it consists mainly of cereal crops and wild grasses **(Remmache, 2006)**.

2.4.1. Agricultural activities

The total agricultural area is important in the wilaya of Mila, it covers more than 90% of the territory of the wilaya (about 315,745 ha). It also evolved positively between 1999 and 2010 (+12.8%). The usable agricultural area is also important, it has certainly changed little over the last ten years, but it has remained quite significant, of the order of 2370557 ha. This shows that we are in an essentially agricultural region. On the other hand, the irrigated area-even if it has increased slightly in 10 years (+5.8%)-is considered to be very low, and this is explained by the ban on the use of water from the two dams (Beni Haroun and Grouz). The rest of the land is made up of rangeland, maquis forest and unproductive land **(Metaai and Beldi, 2011).**

2.4.2. Forest heritage

The forest area in the wilaya of Mila covers 33870 ha or 9.7% of the total area of the wilaya. The Aleppo pine represents the dominant species of the forests of the wilaya, it occupies about 48.57% of the total forest area it is generally found in the forests of Ferdjioua, Ain Beida, Bouhatem, Mila, Chelghoum-Laid and Tadjnanet (**fig. 21**).

- ✓ The cork oak occupies about 16.73% which are generally found in the forests of Grarem, Sid-Merouane, Tassadane and Tarai-Beinen.

Other forest species such as oak zeen, pinion pine, ash and eucalyptus occupy small areas respectively about: 1.29%, 1.77%, 0.59%, 0.29% of the total forest area (**Metaai and Beldi, 2011).**

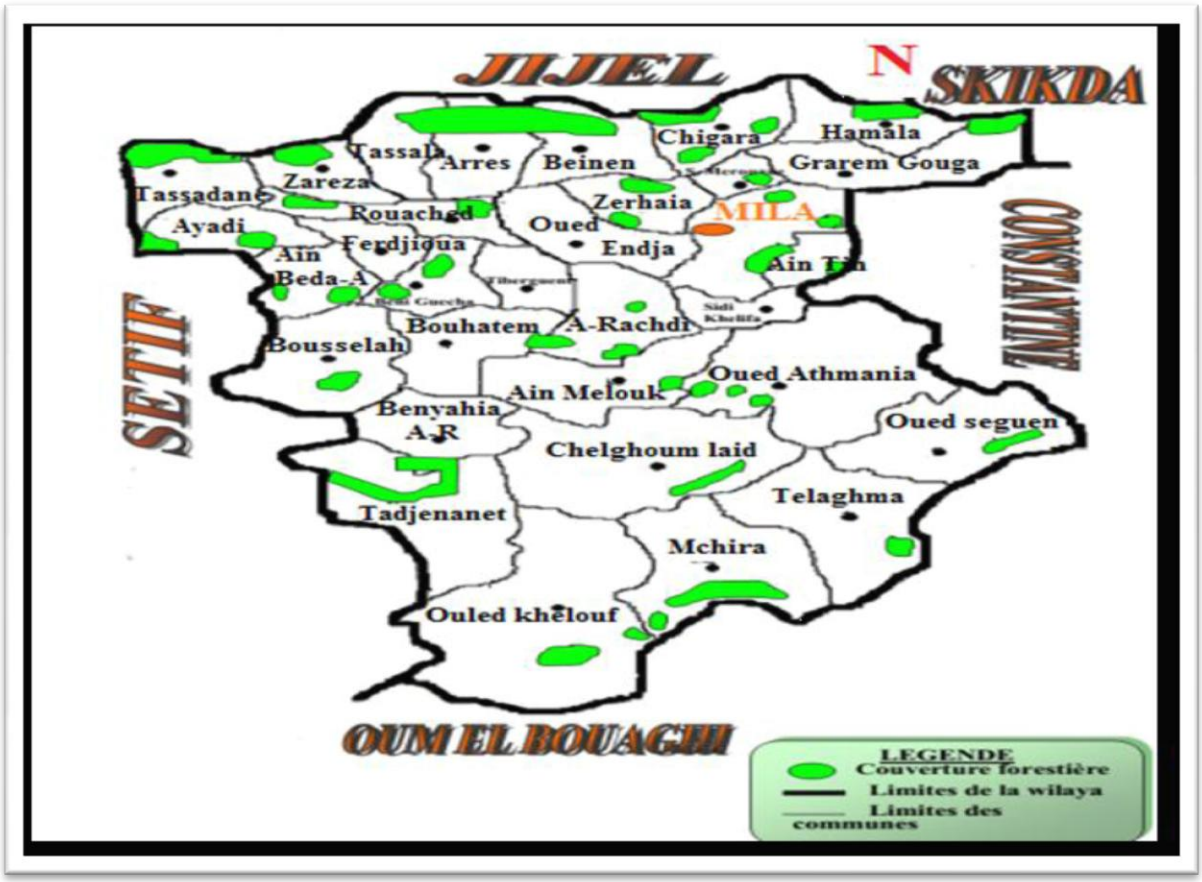


Fig. 21: Map of the forest cover of the wilaya of Mila **(Doula and Ferhat, 2014)**.

2.5. Geology

The study region in the Alpine chain of North Africa whose complex geological framework is characterized by the presence of thrust sheets. These aquifers constitute vast sets of terrains from the Antecambrian to Lower Miocene age which moved (in the form of "thick" scales) horizontally over distances of several kilometers and were deposited according to varied and complex methods **(Boularas and Kadjoudj, 2016)**.

2.6. Pedology

In general, the Mila region is covered by vertic light brown soils **(Berkal and Elouaere, 2014)**. These soils are characterized by a clayey structure, medium to fine on the surface and finer in depth. They are rich in exchangeable potassium, calcareous and poor in assimilable phosphorus. As well as this type of soil has high water retention and is characterized by the appearance of shrinkage cracks in dry periods **(Berkal and Elouaere, 2014).**

2.7. Relief

The wilaya of Mila is entirely surrounded by mountain ranges belonging to different paleogeographic domains:

-In the north, a set of high mountains, characterized by very high altitudes and excessively marked slopes, such as: M'Cid Aicha and Sidi Driss.

-In the south, a set of high plains (plains and hills), such as: Djebel Osman and Grouz. Djebel Lakhal, Chettaba and Kheneg from the east, and Djebel Boucherf and Oukissene by the west **(A.N.D.I, 2013; Merghadi *et al.*, 2018).**

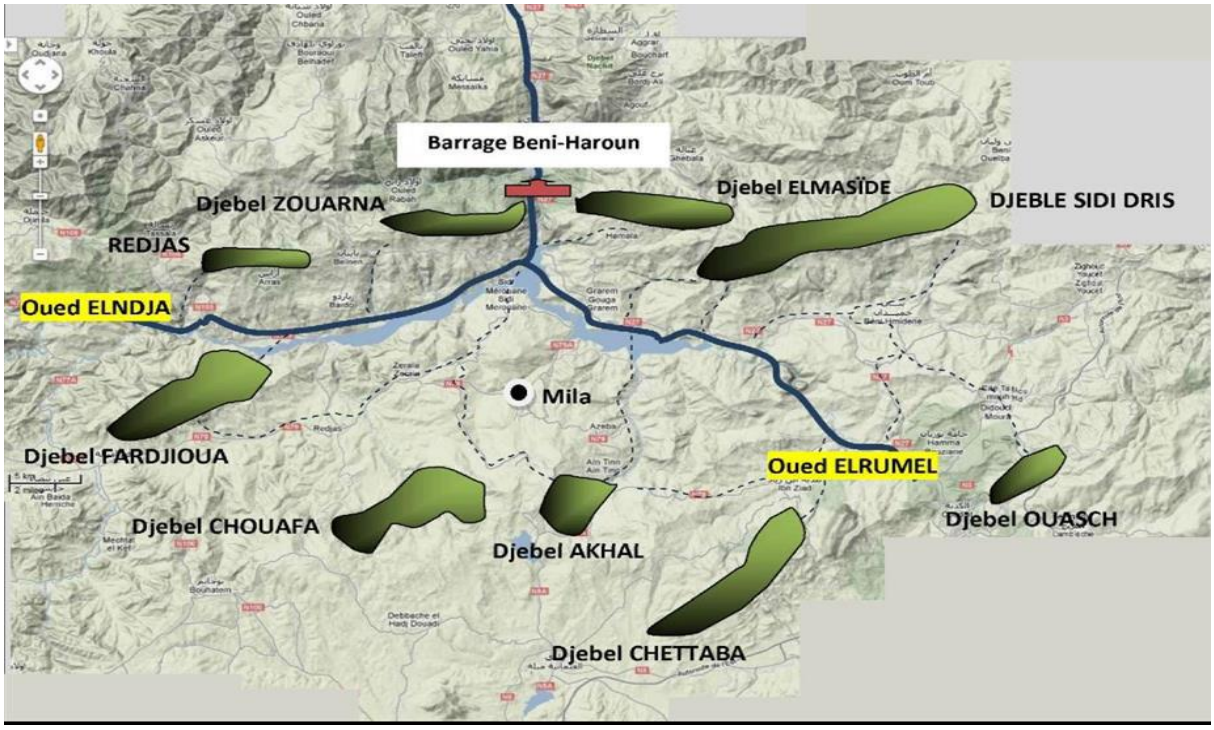


Fig. 22: Map of the major relief features of the Mila basin **(Barik, 2012)**

2.8. Hydrographic network

The wilaya is home to an important hydrographic network composed of rivers and dams: the largest water dam at the national level, the Beni Haroun dam, which supplies a large part of eastern Algeria with drinking water and irrigation water, as well as the Oued Athmania dam, and the Oued Seguène dam. The Oueds Rhumel and Oued Endja (Oued El Kebir) are the main sources of supply for the Beni Haroun dam **(Abid, 2014)**.

There are 415 water sources in the wilaya; 57 wells and 87 boreholes located in the southern part of the wilaya **(Soukehal and Cherrad , 2011)**.

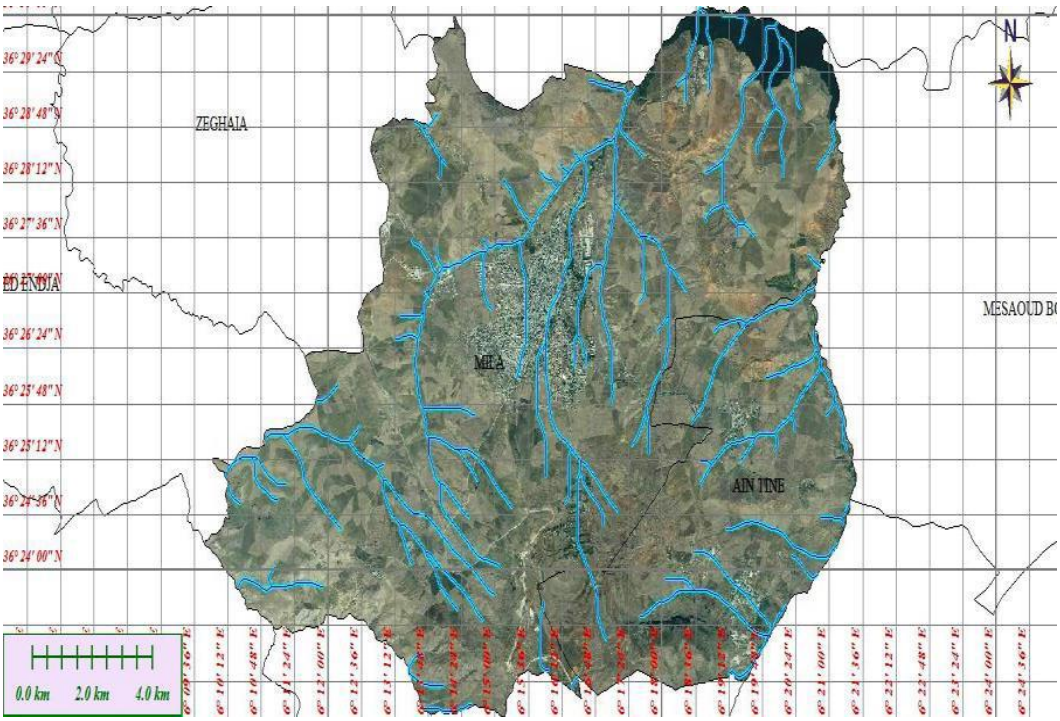


Fig. 23: Hydrographic network of the Mila region **(Soukehal and Cherrad ,2011)**

The Beni Haroun dam located at the heart of a huge hydraulic complex, with a storage capacity of 960 million cubic meters, and a height of 120 meters **(Seddiki *et al.*, 2013).** It is the largest Artificial Reservoir in Algeria and the second largest on the African continent (after the Al Sad El Alli dam in Egypt) with a reserve of 1 billion m3 of water reached in February 2012 (i.e. 40 Million m3 beyond its target capacity), distributed over 3,900 hectares. Located on Wadi el Kebir, it is fed by two main branches, with the wadis Rhumel and Endja **(Seddiki *et al.*, 2013)**.

Fig. 24: The Beni Haroun dam **(Abid, 2014).**

2.9. Climatology

Climatology is the set of meteorological characteristics of a given region. However that, climate is the set of meteorological phenomena that characterize the average state of the atmosphere at a point on the earth's surface (**Soukehal, 2009**). The most important environmental factor is certainly the climate. It has a direct influence on the fauna and flora. It demonstrates an impact on migratory birds: shifting of migration periods, modification in the reproduction and survival of species, displacement of breeding and wintering areas.

The climate of the wilaya of Mila is a typical Mediterranean climate. It is characterized by :

- A wet and rainy season (winter) extending from November to April.
- And a long hot and dry summer period from May to October **(Zouaidia, 2006)**.

➢ **Temperature**

Temperature is an essential and fundamental ecological climatic factor for the life of living beings. Temperature can affect organisms directly or indirectly because thermal conditions affect other organisms to which an individual is ecologically related, although these relationships could be complex. It acts directly on the reaction rate of individuals, on their abundance and their growth **(Faurie *et al.*, 1980)** and it explains that living beings can only carry out their activities in a range of temperatures ranging from 0 at 35°C. A moderate mediterranean temperature during the months of autumn, winter and spring. During the summer, the temperature increases rapidly, especially inside the wilaya. In any case, the temperature is favorable for crops both in summer and winter **(Soukehal, 2011)**.

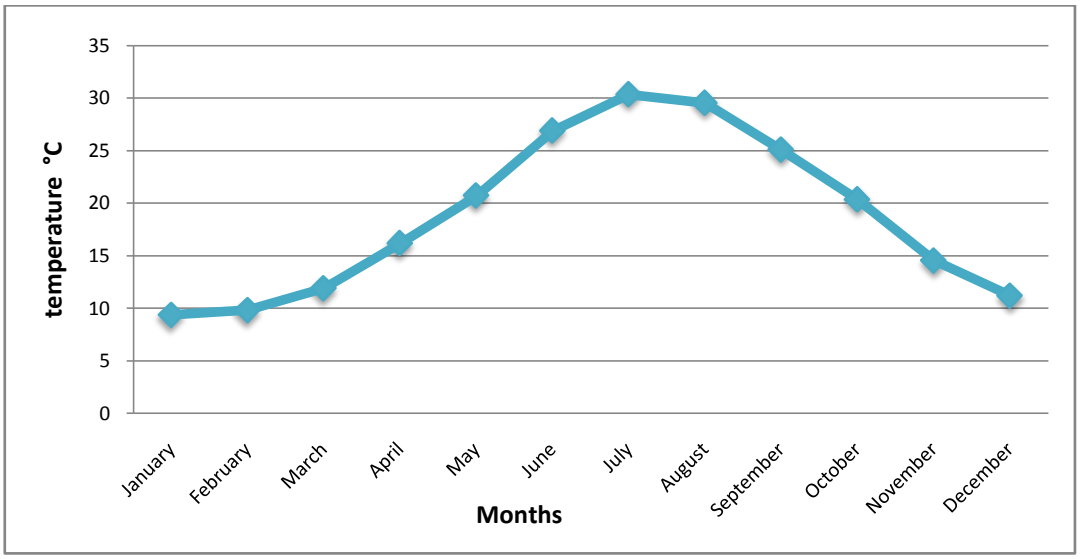


Fig. 25: Average monthly temperature of the Mila region **(Mila meteorological station, 2012 to 2022).**

According to **fig. 25**, which gives the average monthly temperature changes for our region, we note that the maximum temperature is recorded during the month of July when it reaches 30.36 degrees Celsius, while the month of January is characterized by cold degrees, with a temperature not less than 9.36 Celsius.

- **Precipitation**

Precipitation refers to any type of water that falls from the sky, in liquid or solid form **(Dajoz, 2000)**. It represents an essential climatic factor with regard to the ecological cycle, the hydrographic regime and agricultural activity. The variation of annual precipitation is the striking fact in this wilaya. Rainfall in Mila is unevenly distributed across the months of the year and precipitation is, naturally, confined to the cool semester which begins in November and ends in March. The lack or abundance of precipitation has a significant effect on water reserves; quantities mobilized and quantities exploited. The drought acts directly on the behavior of the population in this area **(Soukehal, 2011)**.

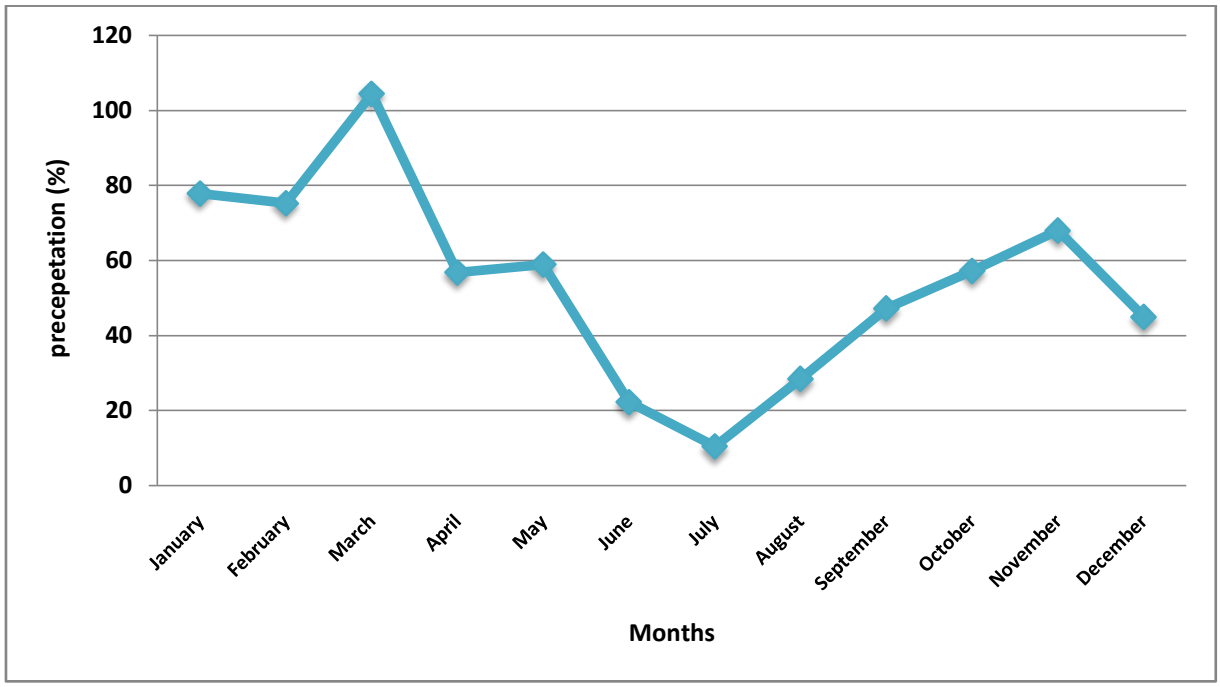


Fig. 26: Average monthly precipitation in the Mila region **(Mila meteorological station, 2012 to 2022).**

The study area is considered one of the wettest areas. From **the fig. 26** above, we see that March is the month with the most rain, as it experienced an excess of 104.36 mm, and on the

contrary, July experienced a deficit of 10.36 mm, which is the driest month and the annual average of precipitation.

***Seasonal pattern of precipitation**

The rainfall year has been divided into four conventional seasons. The seasonal regime of our study region during the period (2012 - 2022) is of the S. W. A. S type (Spring, Winter, Autumn, Summer). The existence of a summer drought period is one of the essential factors that explain the characteristics of Mediterranean forests.

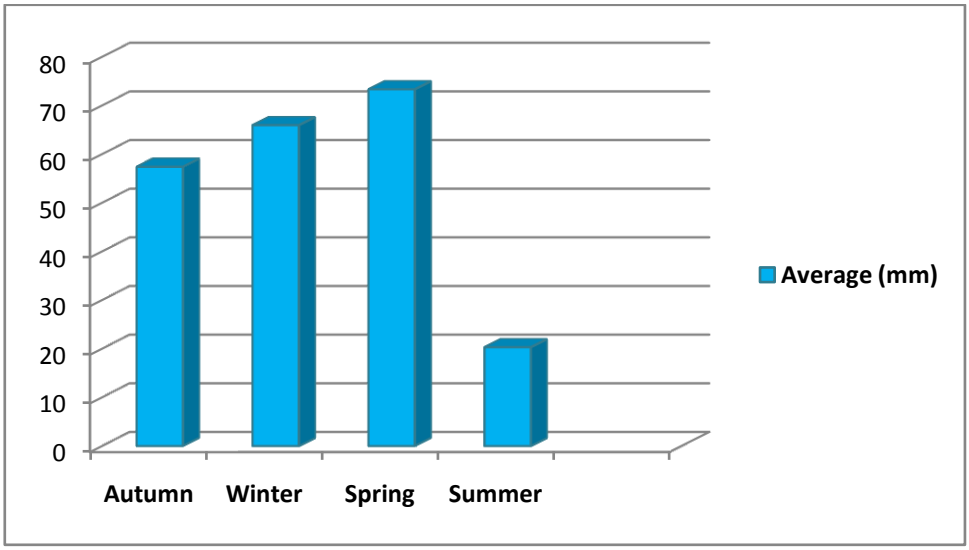


Fig. 27: Seasonal rainfall diagram of the Mila region (2012 - 2022).

This diagram (**Fig. 27**) shows that the spring season is the wettest with an average of 73.33 mm/month, which produces groundwater recharge, while summer is dry with a low recharge of 20, 30 mm/month, which produces evaporation.

- **Humidity**

It is the ratio between the quantity of water vapor in a given volume of air and the quantity possible in the same volume at the same temperature **(Villemeuve, 1974)**. It depends on several climatic factors such as rainfall, temperature and wind **(Faurie *et al.*, 1980)**.

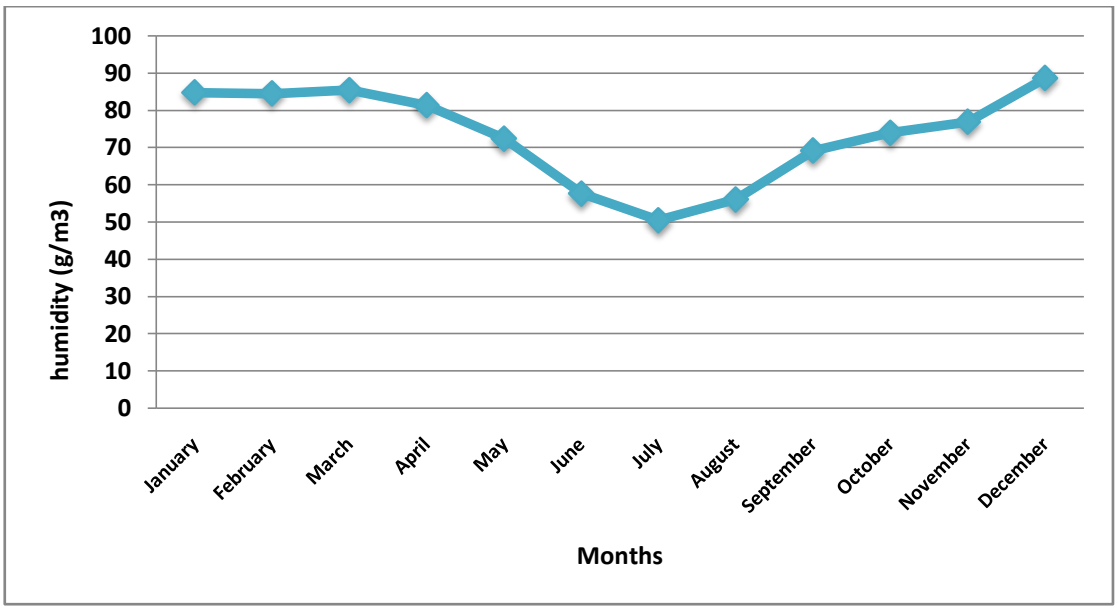


Fig. 28: Average monthly humidity variations in the Mila region **(Mila Meteorological Station, 2012 to 2022).**

According to **fig. 28**, the month with the highest humidity is December with 88.66% and the month with the lowest value is July with 50.45%.

- **Wind speed**

The wind is one of the most characteristic elements of the climate. It acts by activating the precipitation which can induce a drought **(Seltzer, 1946).**

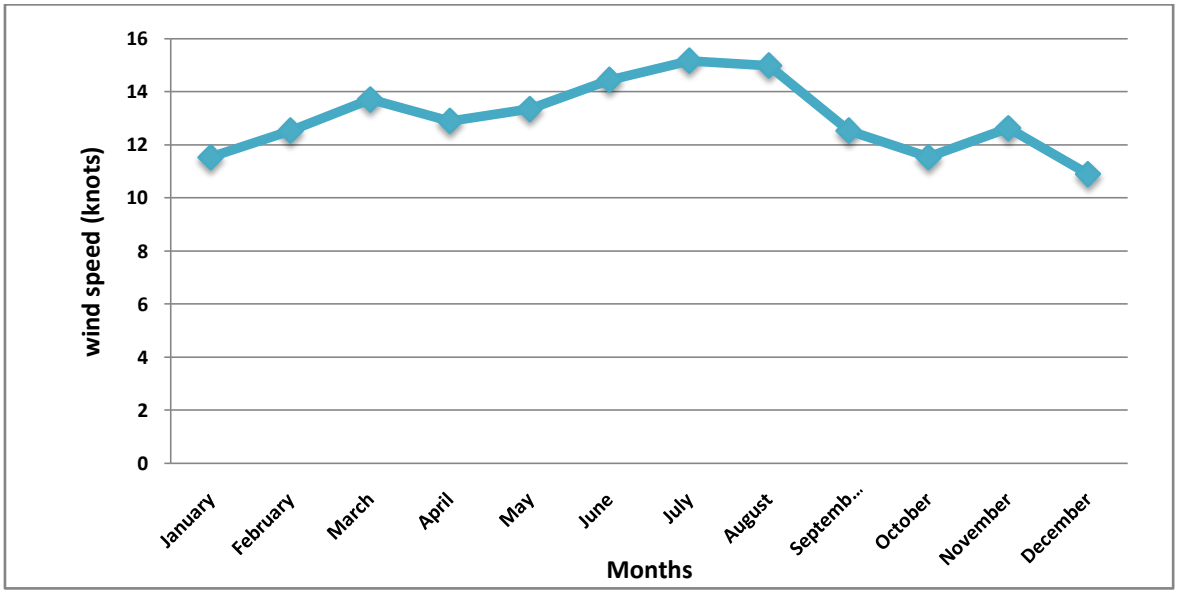


Fig. 29: Average monthly wind speed variations in the Mila region **(Mila meteorological station, 2012 to 2022).**

Fig. 29 shows that the maximum wind speed is recorded in the month of July with a maximum value of 15.18 knots, and the minimum speed is in the month of December with a value of 10.9 knots.

***Climate summary**

The establishment of a synthesis of climatic factors, namely rainfall and temperature, requires the study of the following two parameters:

- ✓ The Bagnouls and Gaussen rainfall diagram.
- ✓ The Emberger rainfall quotient.

According to Bagnouls and Gaussen, a dry period is due to the intersections of temperature and precipitation curves. This relation makes it possible to establish a pluviometric histogram on which the temperatures are brought to a double scale of the precipitations. The analysis of the diagram (**Fig. 30**) shows that the dry period is about 03 months. It extends from June to August, while the wet period extends from September to May. The determination of this period is of great importance for the knowledge of the water deficit period.

***Emberger Rainfall Quotient**

This index helps us to define the 5 types of Mediterranean climate from the most arid to high mountain (Emberger, 1955). It is based on the rainfall and temperature regime.

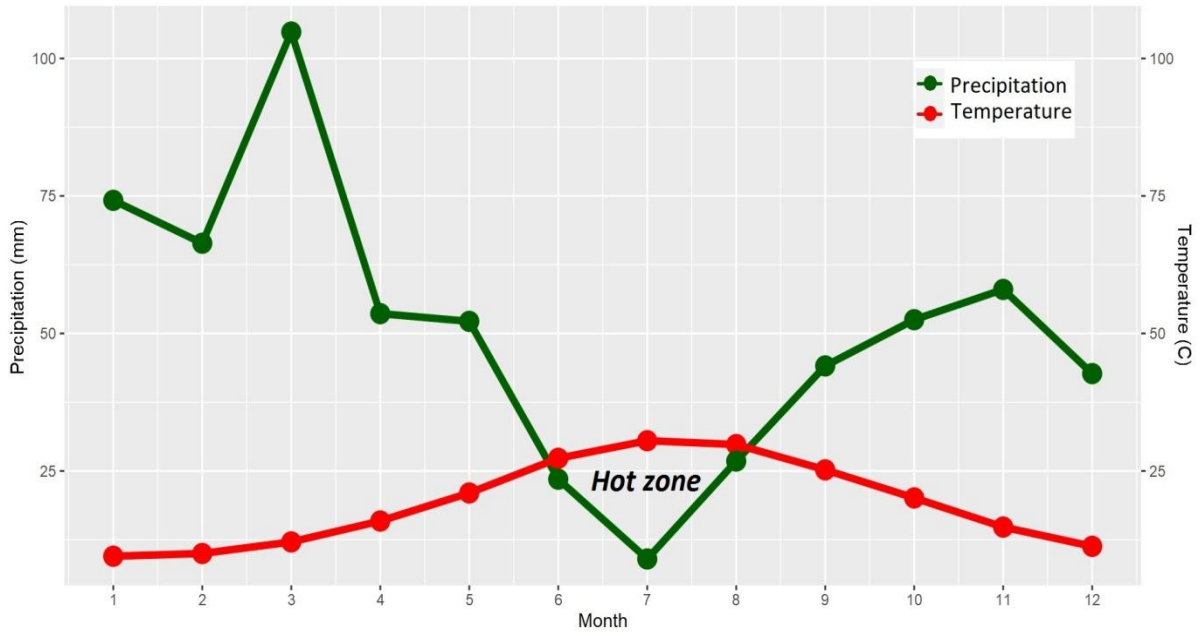


Fig. 30: Bagnouls and Gaussen rainfall diagram of the Mila region (2012 - 2022).

According to the climatic data and the value of Q index of Emberger's climagram, we deduce that the region of Mila where the perimeter of our study is located is classified in the bioclimatic stage of subhumid vegetation with warm winter**. (Fig. 31)**

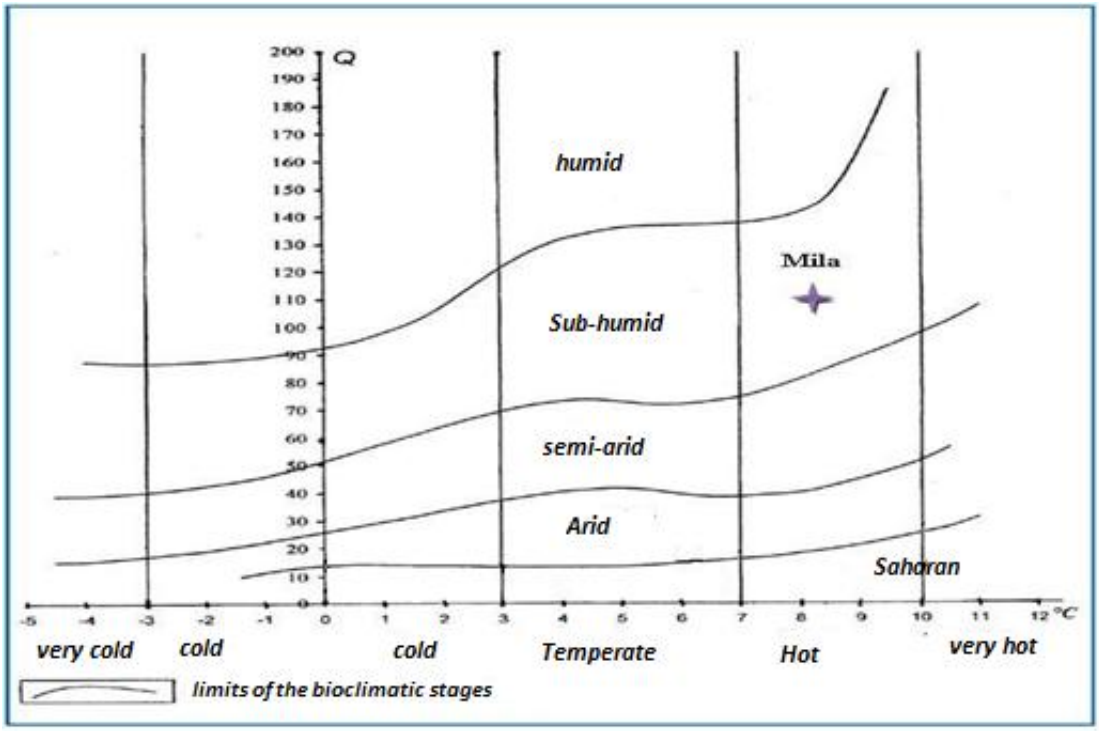


Fig. 31: Situation of the Mila region in the Emberger climagram (**Chahlet and Kerdoud, 2018**).

2.10. Health structure

Epidemiologically, the wilaya records several hundred cases of notifiable diseases each year, with tuberculosis occupying 1st place followed by meningitis and a few dozen cases of zoonoses (leishmaniasis and brucellosis) as well as some cases of viral hepatitis B and C **(Abid , 2014)**. The wilaya of Mila is organized around five (05) Public Hospitals Establishments **(DSPM, 2014)**.

Table 06: Public Hospital Establishments (EPH) **(DSPM, 2014)**.

Denomination	Number of services
Public Hospital Establishments Brothers Maghlaoui Mila.	7
Public Establishments Hospitaliers Brothers Tobal Mila.	9
Public Hospital Establishments Brothers Boukchem Oued El Athmania.	8
Public Hospital Establishments Ferdjioua.	11
Public Hospital Establishments Chelghoum Laid	12

Table 07: Local public health establishments **(DSPM, 2014)**.

Denomination	Number of polyclinics	Number of treatment rooms	Communes covered
Mila	14	34	08
Ferdjioua	10	46	09
Chelghoum laid	09	29	06
Ain Beida ahrich	06	29	06
Tadjnanet	02	19	03
Total	40	157	32

With a specialized hospital establishment, the EHS in psychiatry of Oued Athmania, 38 polyclinics, 145 treatment rooms and 02 private clinics **(DSPM, 2014)**.

MATERIAL AND METHODS

3. MATERIALS AND METHOD

3.1. Epidemiological investigation

3.1.1. Location, Type and Duration of Study

This epidemiological study of bacteria (*Escherichia coli*) takes place at the level of central laboratory service, unit of bacteriology-medical of public hospital establishment Frères Maghlaoui - Mila.

The present study was conducted following the retrospective analytical descriptive method based on the documentary analysis of the registers during the study period which was carried out from January 2012 to December 2022 over a period of 10 years. We associated this part with another prospective study on human *E.coli* during the period (January -March 2023) at the same department.

3.1.2. The patients

This epidemiological study focused on all *E.coli* examinations of patients sent to the bacteria laboratory.

The patients in our study include adults and children hospitalized or consulting in the different departments of the Hospital, from very diverse origins both geographically (different municipalities in the Mila region), and socially (sick from the public and private sectors).

Our prospective study focused on 531 patients referred to the bacteriology laboratory during the first three months of 2023.

3.2. Bacteriological analysis (January-March 2023)

3.2.1. Material

The material used: (**fig. 32**)

- ✓ Bunsen burner
- ✓ Petri dishes
- ✓ Swab
- ✓ Optical microscope
- ✓ Blades and slats
- ✓ sampling bottles
- ✓ Platinum handle
- ✓ Sampling bottles

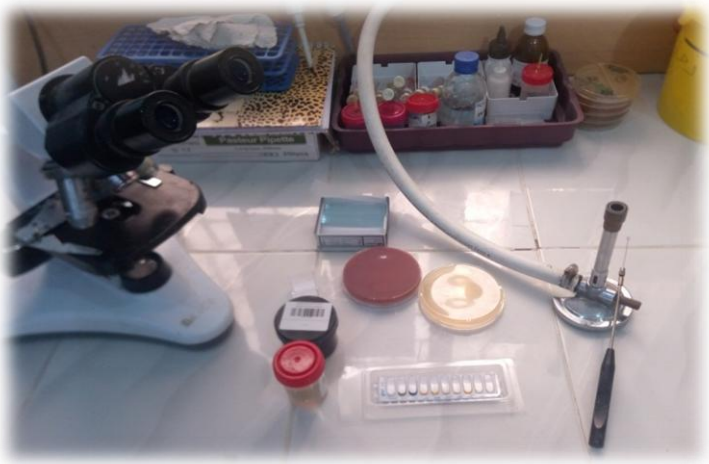


Fig. 32: Laboratory equipment used for the diagnosis of human *Escherichia coli*.

(Personal photo, 2023)

3.2.2. Reagents

- ✓ Chromagar clear orientation
- ✓ Physiological water

Fig. 33: Reagent used in the laboratory for *Escherichia coli* **(personal photo, 2023).**

3.2.3. Methods

The protocol followed for the isolation and identification of *E. coli* is described as follows.

3.2.4. Urine collection

The urinary sample is an essential part of the ECBU. It must be carried out with great care because it determines the quality of the analysis and its result. Usually, the urine is preferably

collected in the morning or after staying at least 3 hours in the bladder. If you have taken antibiotics, wait 3 days after stopping treatment.

After washing your hands with soap, clean yourself carefully with a wipe for intimate use. The pot is opened by placing the lid with the nozzle upwards; the first jet is eliminated, then the urine is collected directly in the pot. The sample is brought to the laboratory as soon as possible, otherwise stored at +4°C without exceeding 4 hours. Once the urine sample has arrived at the laboratory, a series of operations are carried out.

3.2.5. Diagnostic method

- **Cytobacteriological examination of urine (ECBU)**

It is a microbiological examination that allows both to diagnose a urinary tract infection by identifying the responsible germ and to help choose the best treatment.

It is the most requested examination in medical practice and its interpretation is relatively easy, in theory.

- **Macroscopic examination**

This examination can note if there are changes in the physical characteristics of the urine such as color, odor and appearance.

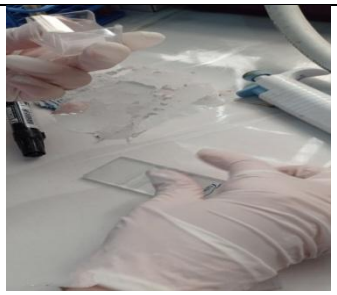
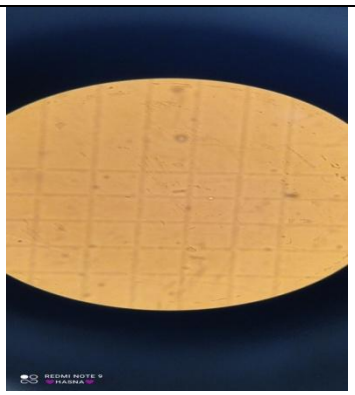
- **Microscopic examination**

The cytobacteriological examination of urine is a microbiological examination that allows us to diagnose a urinary infection by examining the form (cocci, bacillary) and mobility of germs (bacteriological examination), as well as determining the presence of leukocytes, red blood cells, crystals and yeasts (cytological examination). **(Table 08)**

- **Fresh state**

Direct examination is performed by homogenizing the urine sample, then placing two drops using a Pasteur pipette on a clean glass slide covered with a coverslip. Observation is made at 10 x 40 magnifications.

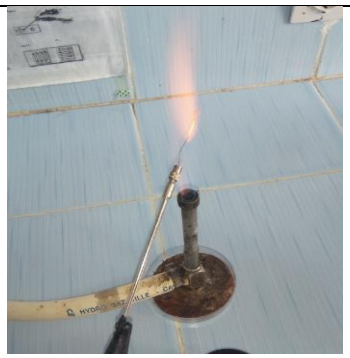
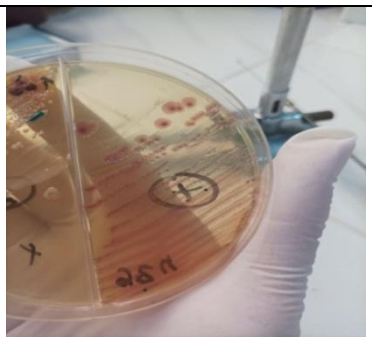
Table 08: Fresh state stages **(Personal photos, 2023).**

 Drop of urine	 Lamella deposition
 Observation with Gx10 and Gx40	 Observation of fresh urine under optical microscope (G×40)

- **Macroscopic examination after culture**

This test is used to visually determine the shape and appearance of colonies on a Petri dish. In this test, the clear chromagar orientation solution is applied to a Petri dish using a sterile pipette **(table 09).** We take a urine sample which is seeded in narrow lines on the medium, and then the seeded dishes are incubated at 37°C for 18 to 24 hours. Readings are taken by observing the boxes with the naked eye. **(Fig. 34).**

Table 09: Stages of macroscopic examination after culture. **(Personal photos, 2023).**

 Sterilization of application handle	 Taking a urine sample
 seeded the urine on the Petri dish	 Macroscopic appearance of *Escherichia coli* strains

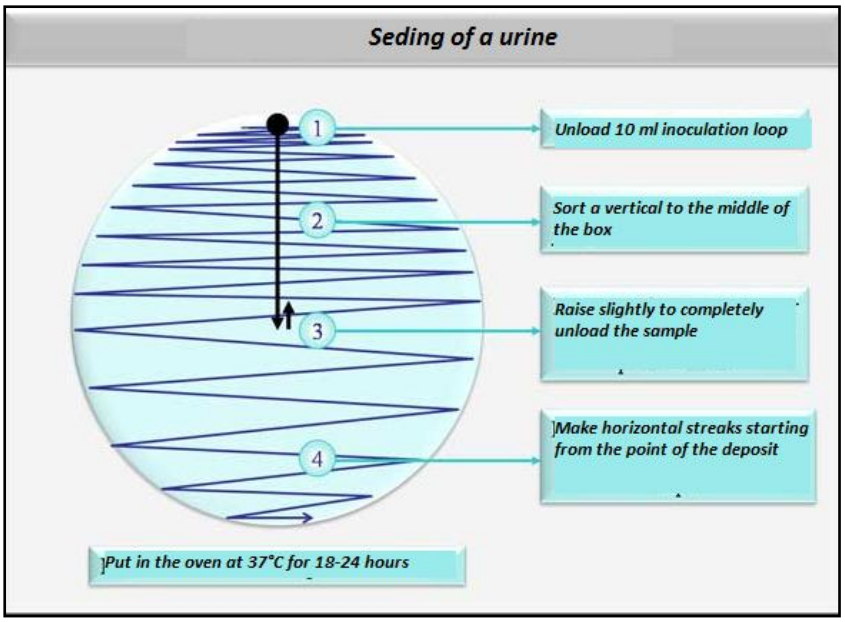


Fig. 34: Seeding technique **(Bouarroudj and Boutebza, 2015)**.

3.2.6. Data collection

The collection of information in the first part is made from the registers of the bacterium service, where the samples were taken, as well as the collection of clinical

information, farm sheets including: the identity of the patients (surname, first name , sex and age), the date of collection, the departments and the results of the macroscopic and microscopic examinations of the EPS. The data collected over a period of 10 years, from January 2012 to December 2022 were recorded on a Windows Excel file.

3.3. Meteorological data

The data necessary for the realization of this study were provided from the meteorological station of Ain Tin, It is the meteorological data relating to the wilaya of Mila concerning five climatic parameters which are:

- ✓ The average annual temperature.
- ✓ The average annual sunshine.
- ✓ The average annual humidity.
- ✓ The average annual wind speed.
- ✓ The average annual precipitation.

3.5. Statistical analysis of data

Data were entered into Excel software and processed using SPSS [(Statistical Package for the Social Sciences) V 26] and the software R. Descriptive statistics of these variables for sex and age slices, years, seasons, months were represented in boxplots using the package {ggplot2} of the software R that was employed in all statistical tests and graphics. The variation each parameter following sex , age slices,months seasens years and the interaction (sex × ages slices) was tested usingone way and two-way analysis of variance (ANOVA). Then student's t test were conducted to distinguish the variability in each group of parameters. Pearson correlation tests were applied between all *E.coli* dissemination parameters measured (age, sex, months, seasons, years and meterologecal parameters) in order to understand the behavior and relationships between *E. coli* dissemination combined under the meterologecal parameters conditions in the Mila region.

The resulting correlation matrix was plotted in an interactive correlation diagram using the package {corrplot} in R. Using the package {nlme} in R, we implanted generalized linear mixed models (GLMMs) with to test the following relationships: effects of T, Sun, P, WS and H on *E. coli* dissemination variation.The statistical significance of all tests was set at $p < 0.05$ and 95% of the confidence interval.

RESULTS

4. RESULTS

4.1. Retrospective analysis of the study population

This survey reveals cases diagnosed at the bacteria analysis laboratory levels of the wilaya the Mila during the period (2012-2022). According to the prescription of the attending physician, patients with *E.coli* are referred for testing of blood, urine, stool, or any material with infection. During this period 14596 patients are testing.

***Simple Bacterial Indicator (SBI)**

The Simple Bacterial Index is the percentage of people infected with bacteria out of the total number of subjects examined.

$$SBI = \frac{\text{Number of positive cases}}{\text{Total number of subjects examined}} \times 100$$

4.1.1. Distribution of patients according to infestation rate during the descriptive study

Fig.35 and **table 10** have show that from 14596 subjects screened for the *E. coli* bacterium, 495 are infected, the infection rate is (3.39%) during the study period.

Table 10: Distribution of patients according to infestation rate during the period (2012-2022).

	Effective	Frequency
Positive cases	495	3,39%
Negative cases	14101	96,60%

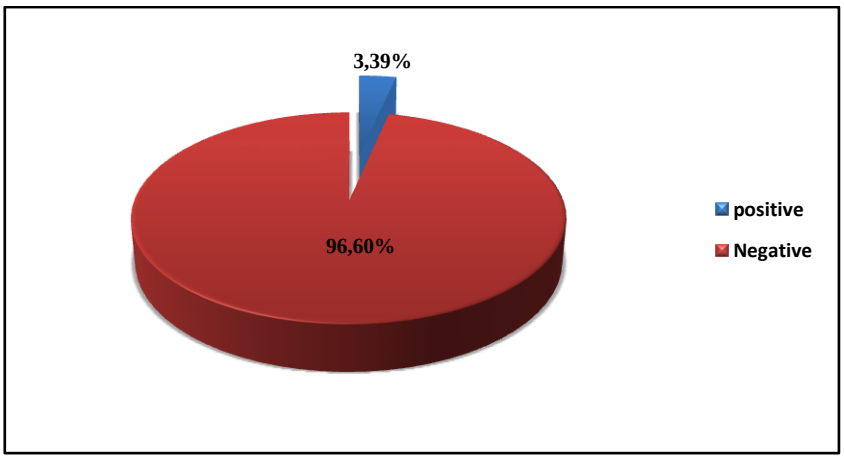


Fig. 35: The distribution of patients according to the infestation rate during the study period.

4.1.2. Distribution of infected patients according to sex ratio during the study period

Our study showed those women are more exposed to *Escherichia coli* by 66.87% in the state of Mila (**table 11. Fig. 36**), the graphs represented in the **fig. 37, 38, 39** shows that women and men were more infected during the years (2015, 2019). Significantly higher rates of infection were recorded in the autumn and winter seasons for both sexes. Also, the months of November and December have seen an increase in the number of cases for females and for male's sex the highest rates of infection have been noted in October and November months. The results obtained showed statistically significant differences ($p < 0.001$) between sex ratio for *E. coli* infection**.** One-way ANOVA reveals a not significant sex effect ($p > 0.05$).

Table 11: Distribution of infected patients according to the sex ratio during the study period.

Sex	Effective	SBI%
woman	331	66,87%
Male	164	33,13%

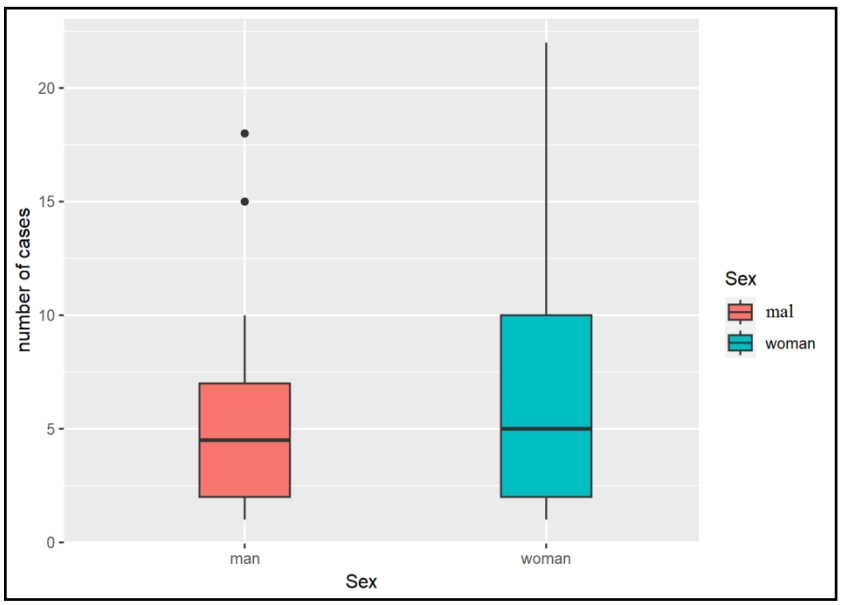


Fig. 36. Boxplots displaying the distribution of infected patients according to sex ratio during the period (2012-2022).

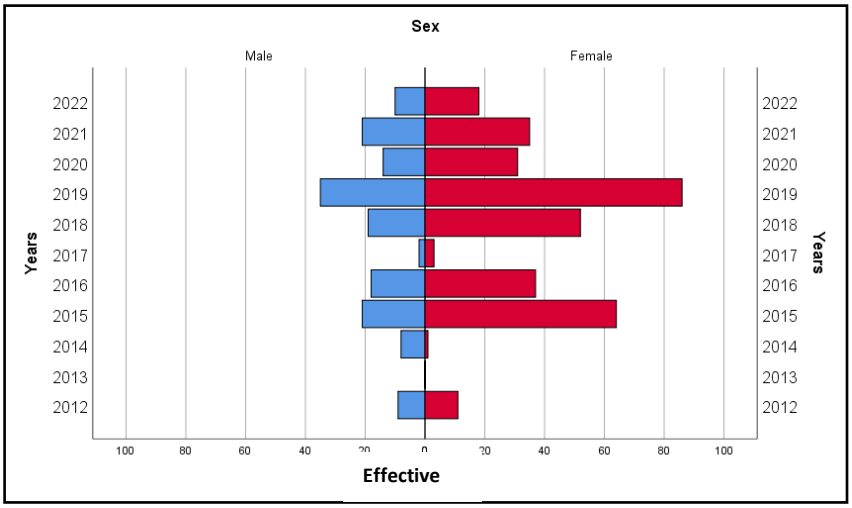


Fig. 37: Distribution of patients according to the sex ratio during the study period according to years.

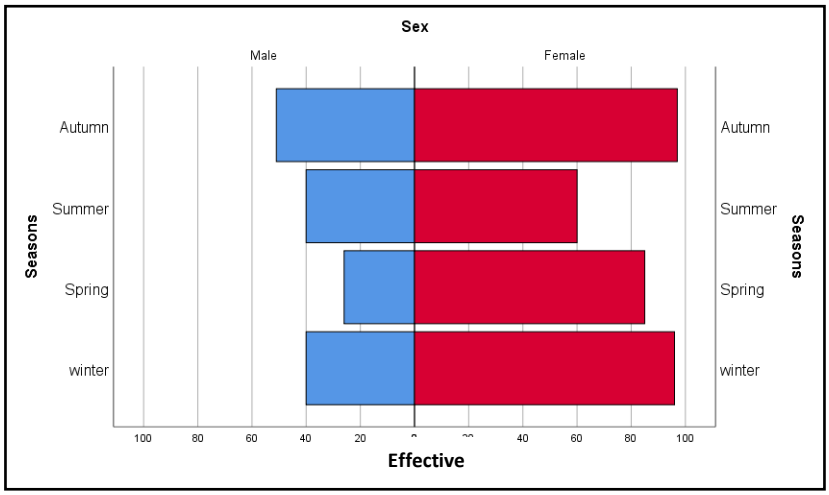


Fig. 38: Distribution of patients according to the sex ratio during the retrospective study period according to seasons.

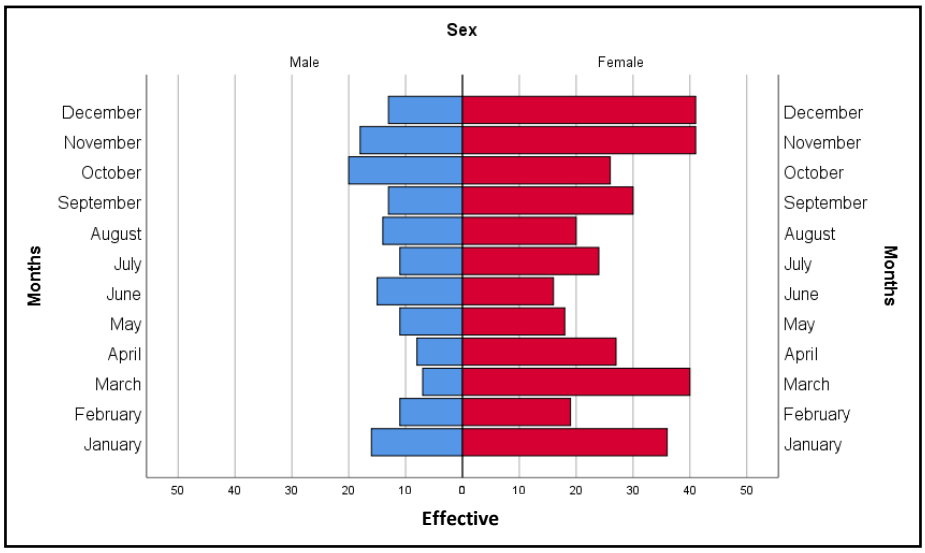


Fig. 39: Distribution of patients according to the sex ratio during the study period according to months.

4.1.3. Distribution of infected patients according to age slices during the period (2012-2022)

According to the data presented in the **table 12** and **fig. 40** we note that the most affected age group is [20-44] years with 172 cases and a rate of 34.74%, followed by the category [45-65] years by 121 cases and a rate of 24.44%. The category [10-14] years are the less representative group by 21 cases and a rate of 4.21% over the study period. The **figures 41, 42** have shown that the age category most threatened with this bacterium is wider in males, especially during the autumn season**.** The analysis of the data showed a statistically significant differences ($p< 0.001$) between ages slices for *E. coli* infection**.** One-way ANOVA reveals a significant age effect ($p< 0.05$).ANOVA bivariate analysis show a very highly significant sex-age interaction ($p < 0.001$) (**table in annex**).

Table 12: Distribution of infected patients according to age during the period (2012-2022).

Age slices	Effective	SBI%
[0-1]	53	10,71%
[2-4]	26	5,25%
[5-9]	27	5,45%
[10-14]	21	4,24%
[15-19]	27	5,45%
[20-44]	172	34,74%
[45-65]	121	24,44%
≥65	48	9,70%

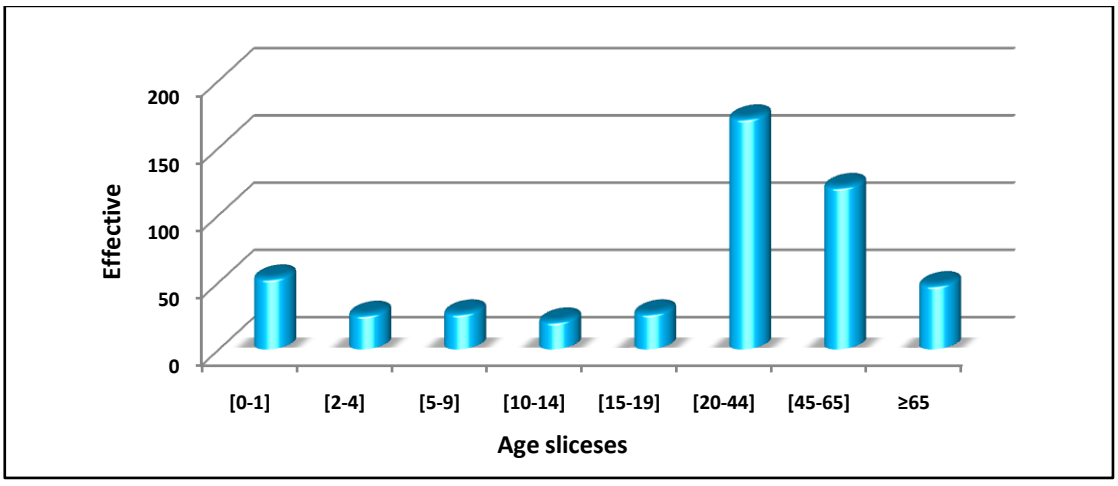


Fig. 40: Distribution of infected patients according to age slices during the period (2012-2022).

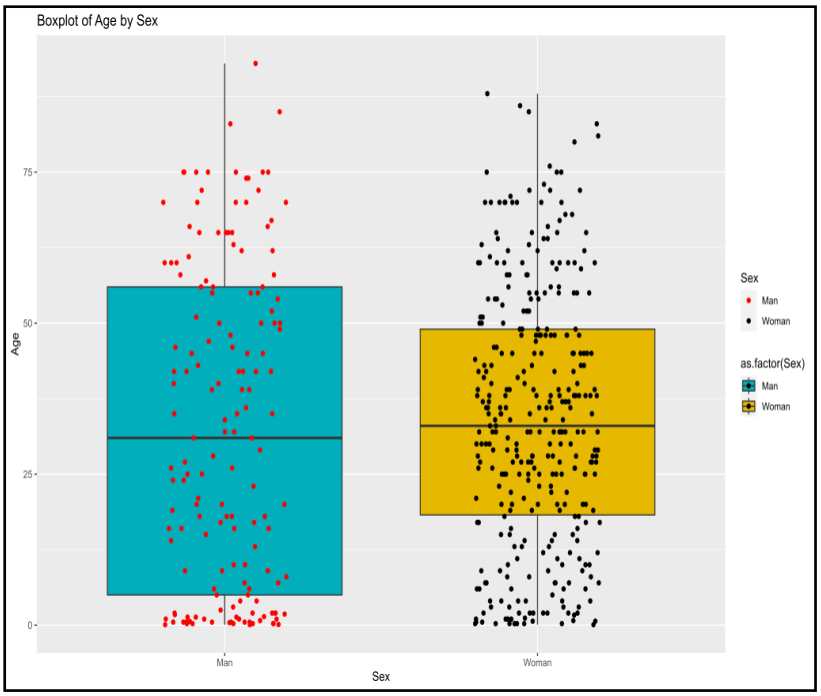


Fig. 41: Boxplots displaying the distribution of age slices of infected patients according to sex ratio during the period (2012-2022).

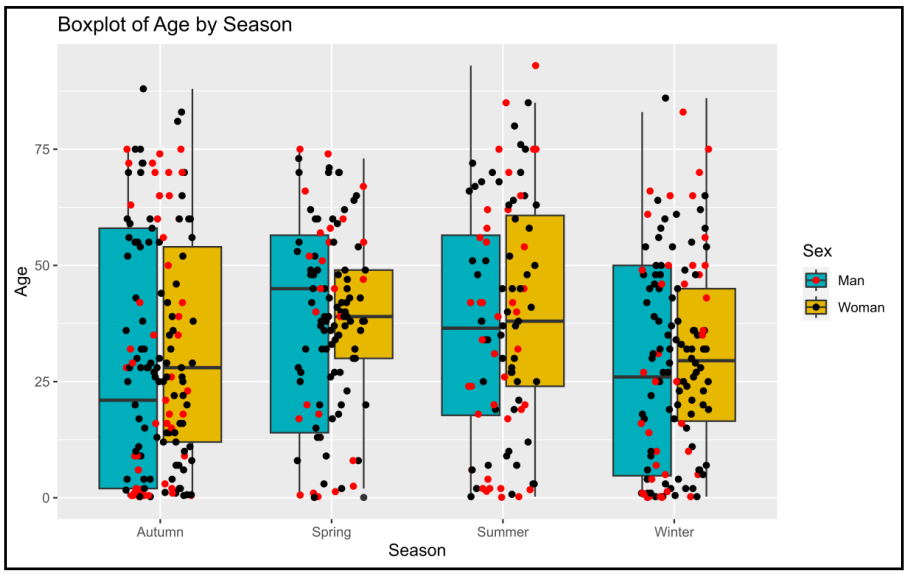


Fig. 42: Boxplots displaying the distribution of age slices of infected patients according to seasons during the period (2012-2022).

4.1.4. Distribution of infected patients according to the months during the study period

The **table 13** and **fig.43** show that the highest number of bacterial cases was recorded during the months of November, December recorded at 11.92%, 10.91%, the lowest percentage was detected in the months of May and February at a rate of 5.86%, 6.06% over the study period. The results obtained showed statistically significant differences ($p < 0.001$)

between months for *E. coli* infection**.** One-way ANOVA reveals a very highly significant month's effect ($p > 0.001$).

Table 13: Distribution of infected patients by months during the period (2012-2022).

Months	Effective	SBI%
January	52	10,50%
February	30	6,06%
March	47	9,49%
April	35	7,07%
May	29	5,86%
June	31	6,26%
July	35	7,07%
August	34	6,87%
September	43	8,69%
October	46	9,29%
November	59	11,92%
December	54	10,91%

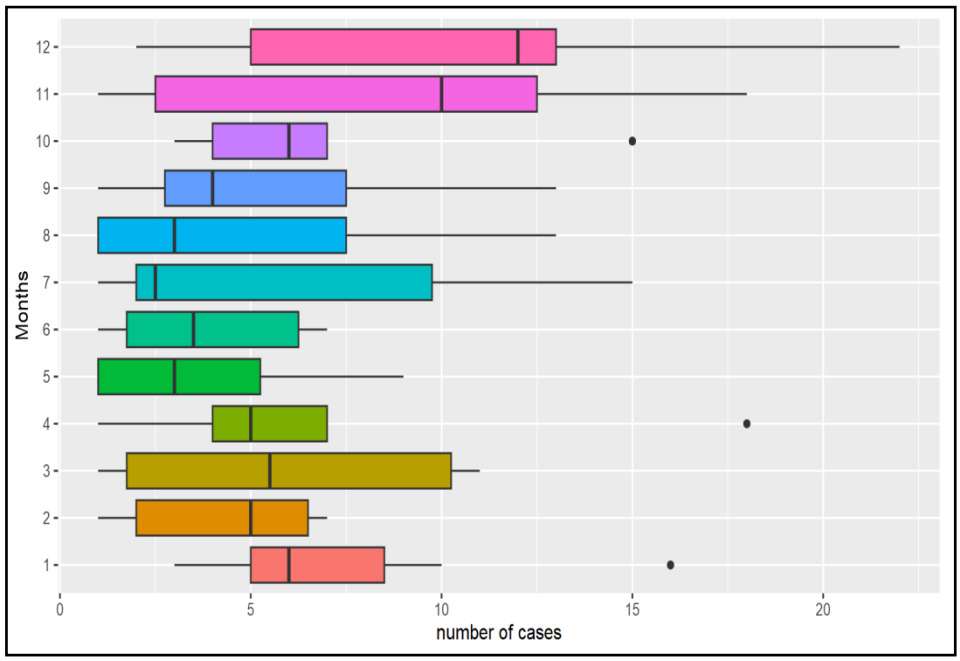


Fig. 43: Boxplots displaying the distribution of infected patients by months during the period (2012-2022).

4.1.5. Distribution of infected patients according to the seasons during the period (2012-2022)

The **table 14, fig. 44** and **45** have shown that the highest number of infected cases was observed during the fall season, followed by the winter season especially during the years 2015.2019 and 2020, while the lowest cases were recorded during the summer season. The results obtained showed statistically significant differences ($p < 0.001$) between seasons for *E. coli* infection**.** One-way ANOVA reveals a very highly significant season's effect ($p > 0.001$).

Table 14: Distribution of infected patients according to the seasons during the period (2012-2022).

Seasons	Effective	SBI%
Winter	136	27,47%
Spring	111	22,42%
Summer	100	20,20%
Autumn	148	29,89%

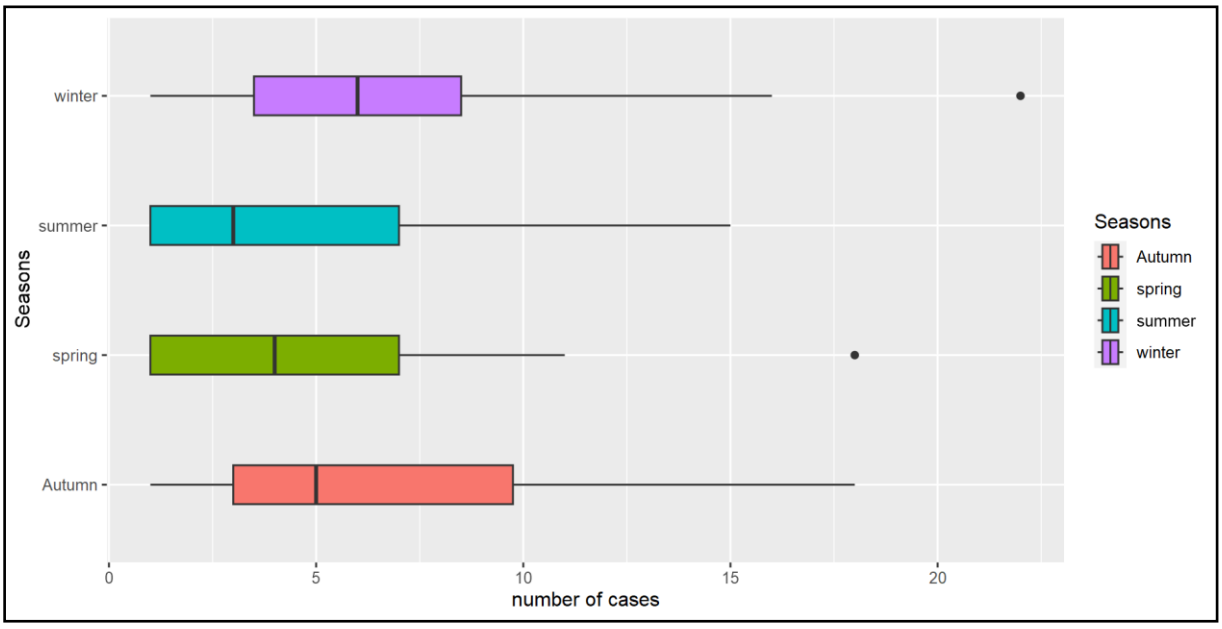


Fig. 44: Boxplots displaying the distribution of infected patients according to the seasons during the period (2012-2022).

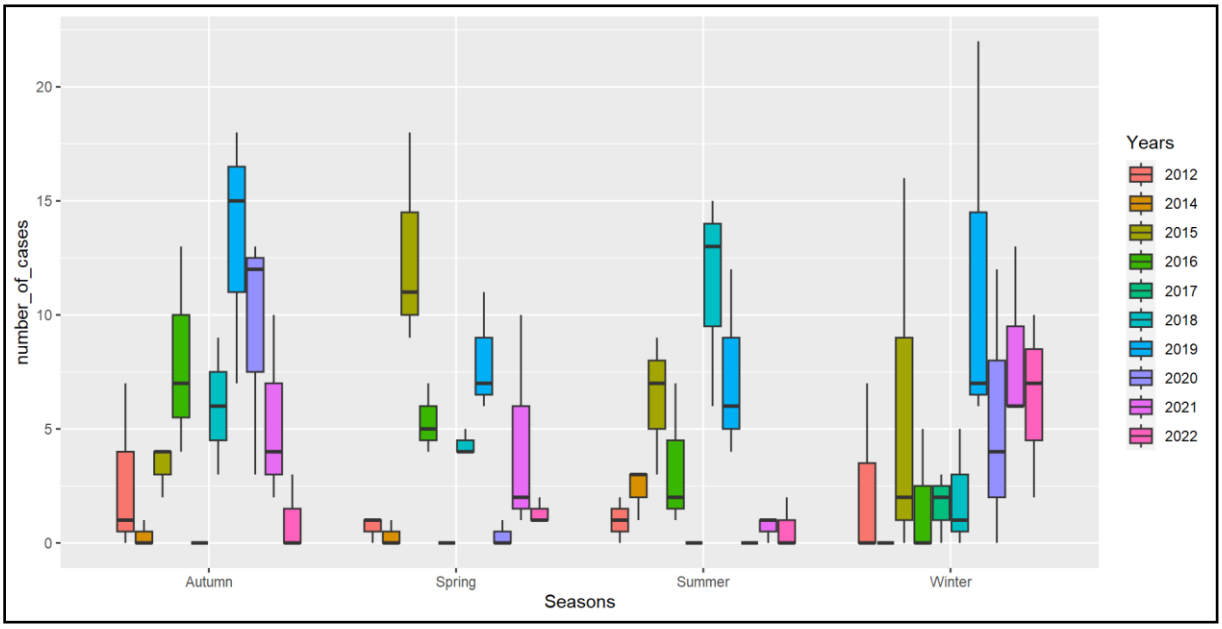


Fig. 45: Boxplots displaying the seasonal distribution of infected patients according to the years during the period (2012-2022).

4.1.6. Distribution of infected patients according to the years during the period (2012-2022)

The years 2019, 2015 and 2018 recorded the highest rates of *E. coli* infection, 4.45%, 4.15% and 3.52%, respectively, compared to other years (**table 15,fig. 46**), the rate of *E.coli* ranged from 0% to 4.45% over the period (2012-2022). The results obtained showed statistically significant differences ($p < 0.001$) between years for *E. coli* infection**.** One-way ANOVA reveals a very highly significant year's effect ($p > 0.001$).

Table 15: Distribution of infected patients according to the years during the period (2012-2022).

Years	Effective	Positive cases (+)	SBI%
2012	970	20	2,06%
2013	64	0	0%
2014	851	9	1,06%
2015	2048	85	4,15%
2016	1479	55	3,72%
2017	384	5	1,30%
2018	2016	71	3,52%
2019	2720	121	4,45%
2020	1023	45	4,39%
2021	1761	56	3,18%
2022	1271	28	2,20%

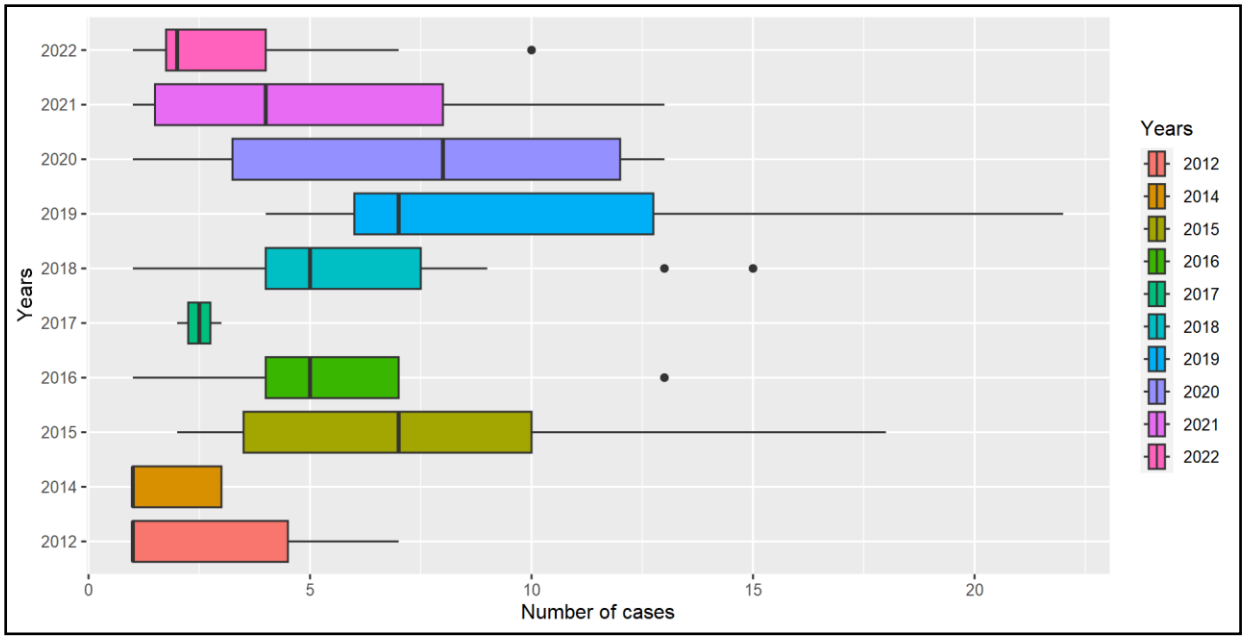


Fig. 46: Boxplots displaying the distribution of infected patients according to the years during the period (2012-2022).

4.2. Overall prevalence of *Escherichia coli* during the prospective study

Our prospective study was interested in 531 patients who were referred to a bacteriology laboratory during the first three months of 2023 and allows us to perform microscopic examination of a sample in its fresh condition and can detect pathogenic *E. coli*.

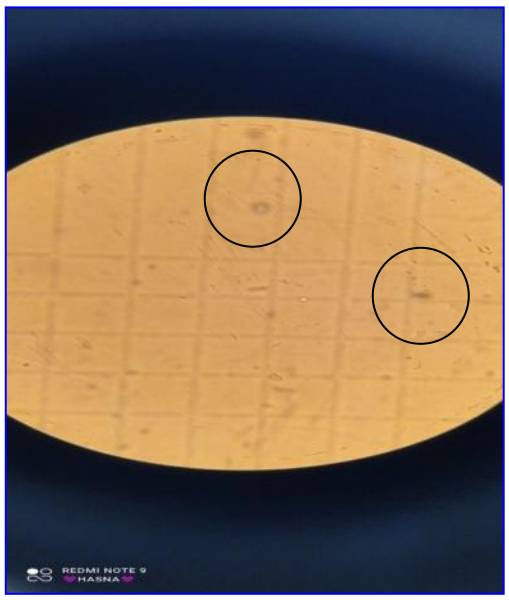


Fig. 47: Observation of *Escherichia coli* detected in fresh urine under optical microscope (G×40) **(Personal photo, 2023).**

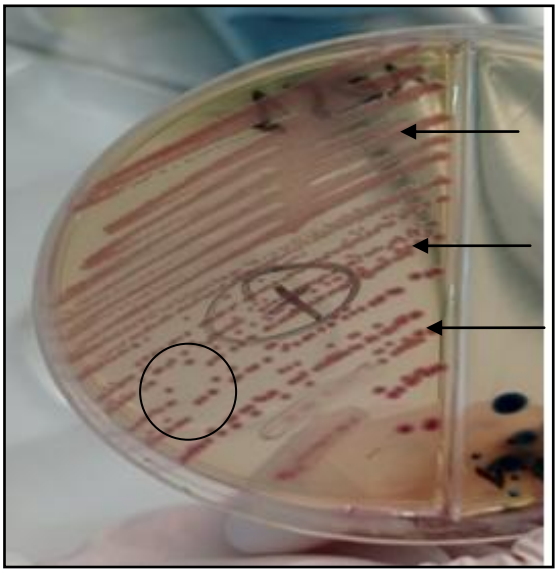


Fig. 48: Detection of *Escherichia coli* strains **(Personal photo, 2023).**

4.2.1. Distribution of patients according to infestation rate during the period (January-March 2023)

Table 16 and **figure 49** have shown that from 522 subjects examined for human *E. coli* bacteria, 9 were found infected, from January-March 2023 with an infestation rate of (1.69%).

Table 16: Distribution of patients according to infestation rate during the period (January-March 2023)

	Effective	Frequency
positive cases	09	1,69%
negative cases	522	98,30%

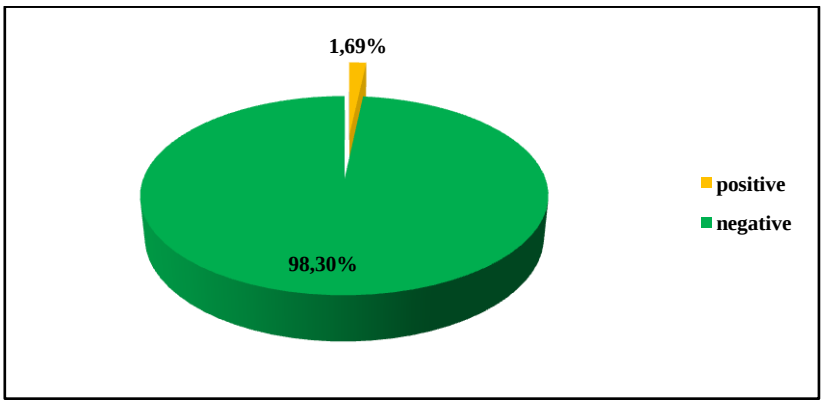


Fig. 49: Distribution of patients by infestation rate over the prospective study period.

4.2.2. Distribution of infected patients by sex ratio during the prospective study period

Table 17 and **figure 50** reveal that the most of infected cases were women (88.88%).

Table 17: Distribution of infected patients according to the sex ratio (January-March 2023):

Sex	Effective	SBI%
Woman	08	88,88%
Male	01	11,11%

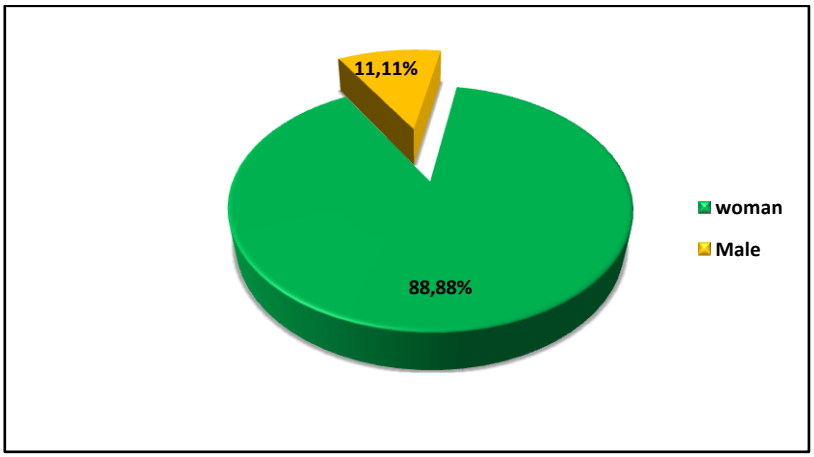


Fig. 50: The distribution of infected patients according to the sex ratio during the period (January-March 2023)

4.2.3. Distribution of patients by age group during the prospective study period

We notice that the age group ≥65 years is the most affected by *E. coli* with 5.57% followed by the age groups included between [45-65] years with a percentage of 0.38% (**table 18, figure 51)**.

Table 18: Distribution of infected patients according to age (January-March 2023):

Age slices	Effective	SBI%
[0-1]	1	0,19%
[2-4]	1	0,19%
[5-9]	1	0,19%
[10-14]	0	0%
[15-19]	0	0%
[20-44]	1	0,19%
[45-65]	2	0,38%
≥65	3	5,57%

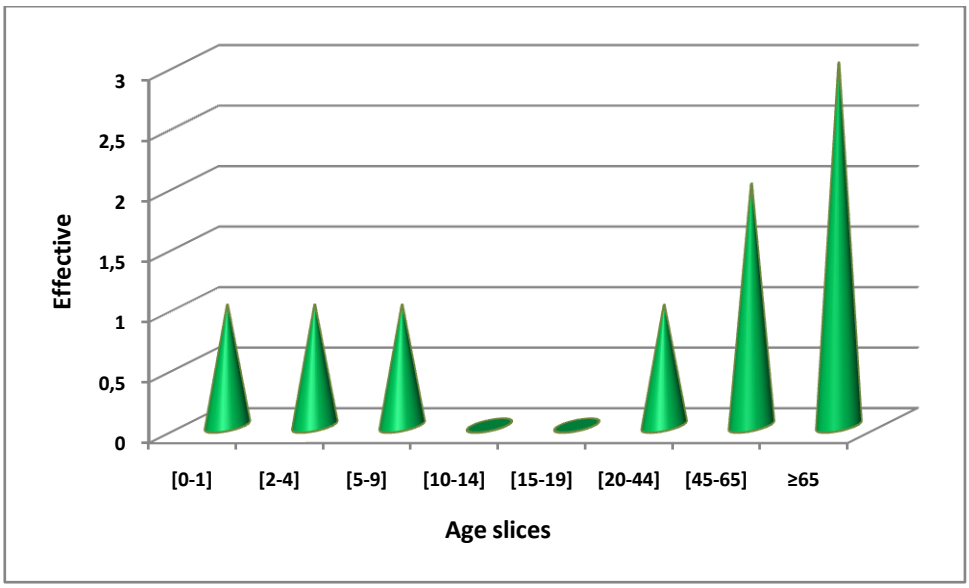


Fig. 51: Distribution of patients according to age groups during the period (January-March 2023)

4.2.4. Distribution of infected patients by months during the period (January-March 2023)

Table 19 and **figure 52** shown that the highest number of bacteria *E. coli* cases was noted during the month of February 0.95%, followed by the months of January and March with 0.38% and 0.19% during the period (January-March 2023).

Table 19: Distribution of infected patients by month during the period (January-March 2023).

Months	Effective	SBI%
January	02	0,38%
February	05	0,95%
March	01	0,19%

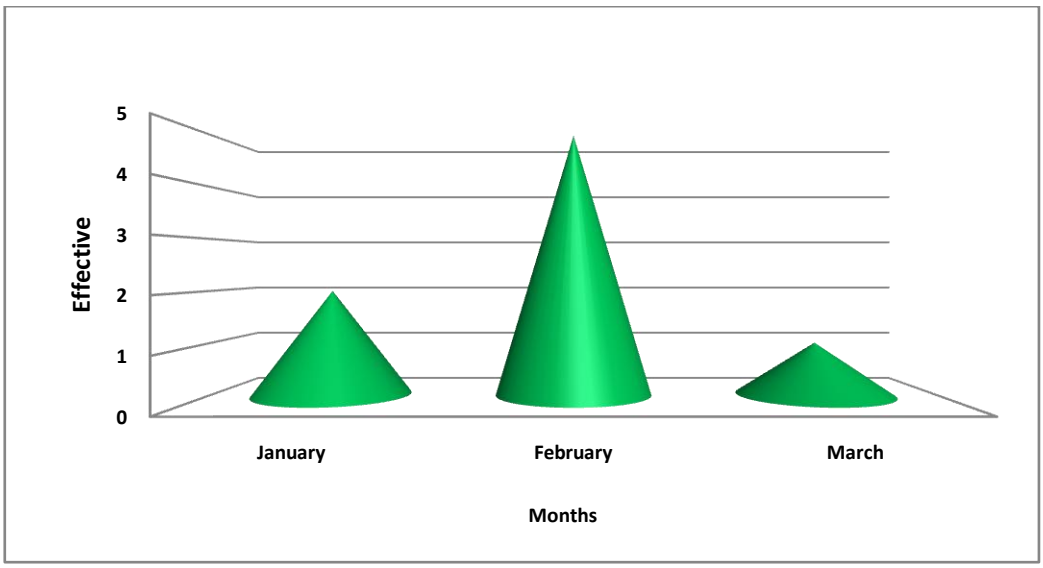


Fig. 52: Distribution of infected patients by months during the period (January-March 2023).

4.3. Correlation between the variation of meteorological parameters and the propagation of *Escherichia coli* during the period (2012-2022)

To identify the relationship between meteorological parameters and the propagation of human *Escherichia coli* in the Mila region, we have used the model of diagram using the package {corrplot} in R. Using the package {nlme} in R, we implanted generalized linear mixed models (GLMMs) to following the relationships: effects of T, Sun, P, WS and H on *E. coli* dissemination variation. Pearson correlation will be used to clarify the correlation between the dissemination of human *E. coli* and the variation of different meteorological parameters.

4.3.1. The relationship between the variation of the average temperature and the number of infected cases during the period (2012-2022)

Linear regression and Pearson correlation (**figure 53,58**) showed that the number of infected cases increases progressively with the increase in the average temperature so there is a very strong positive correlation between the variation of the average temperature (°C) and the number of infected cases during the period (2012-2022).

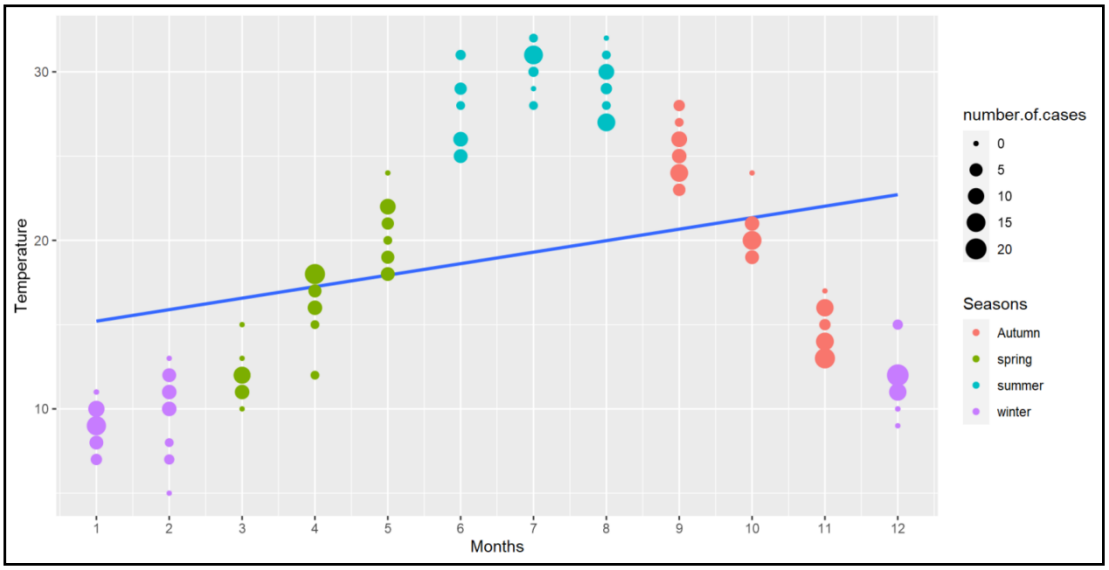


Fig. 53: The correlation between the average temperature (°C) and the number of infected cases during the period (2012-2022).

4.3.2. The relationship between the variation of the average precipitation and the number of infected cases during the period (2012-2022)

Linear regression and Pearson correlation (**Figure 54,58**) showed that the number of infected cases decreases progressively with the increase in mean precipitation so there is a very strong negative correlation between the variation in mean precipitation (mm) and the number of infected cases during the period (2012-2022).

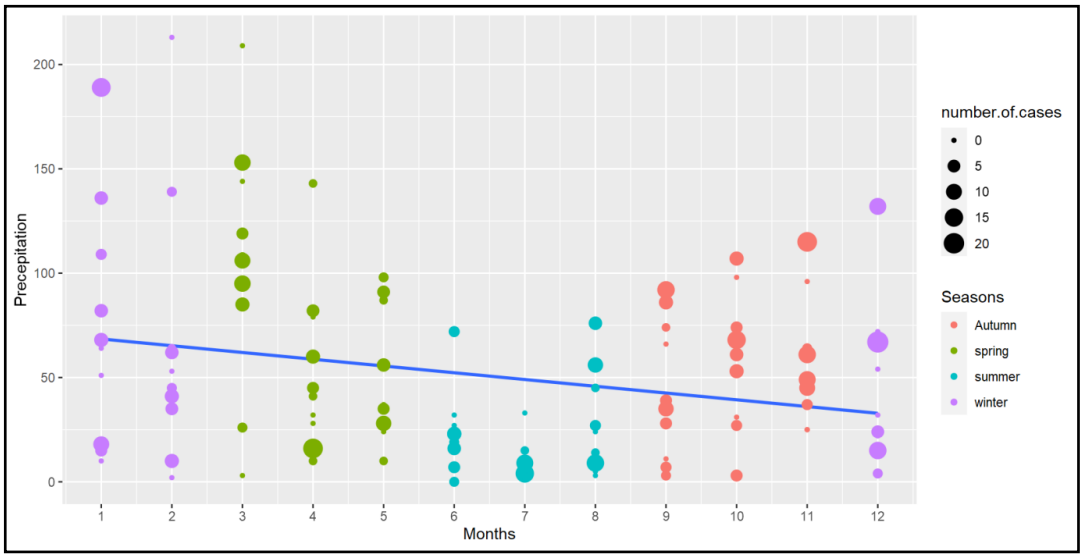


Fig. 54: The correlation between the average wind speed (knots) and the number of infected cases during the period (2012-2022).

4.3.3. The relationship between the variation of the average wind speed and the number of infected cases during the period (2012-2022)

Linear regression and Pearson correlation (**Figure 55,58**) showed that the number of infected cases decreases progressively with the increase in mean wind speed so there is a very strong negative correlation between the variation in mean wind speed (knots) and the number of infected cases during the period (2012-2022).

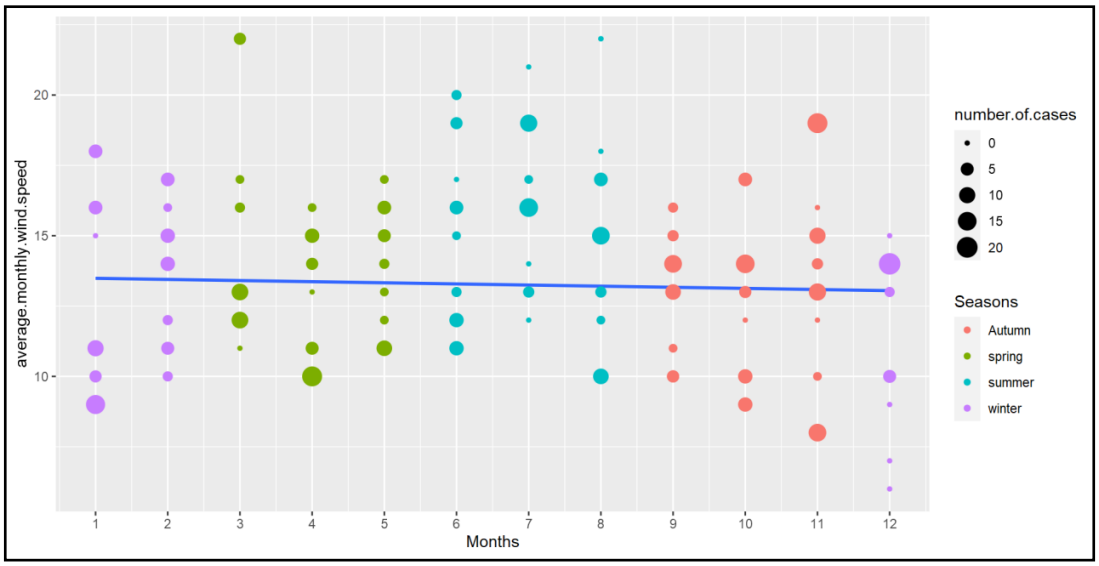


Fig. 55: The correlation between the average wind speed (knots) and the number of infected cases during the period (2012-2022).

4.3.4. The relationship between the variation of the average humidity and the number of infected cases during the period (2012-2022)

Linear regression and Pearson correlation (**figure 56,58**) revealed that the number of infected cases decreases progressively with increasing humidity, so there is a very strong negative correlation between the average humidity (g/m^3) and the number of cases infected during the period (2012-2022).

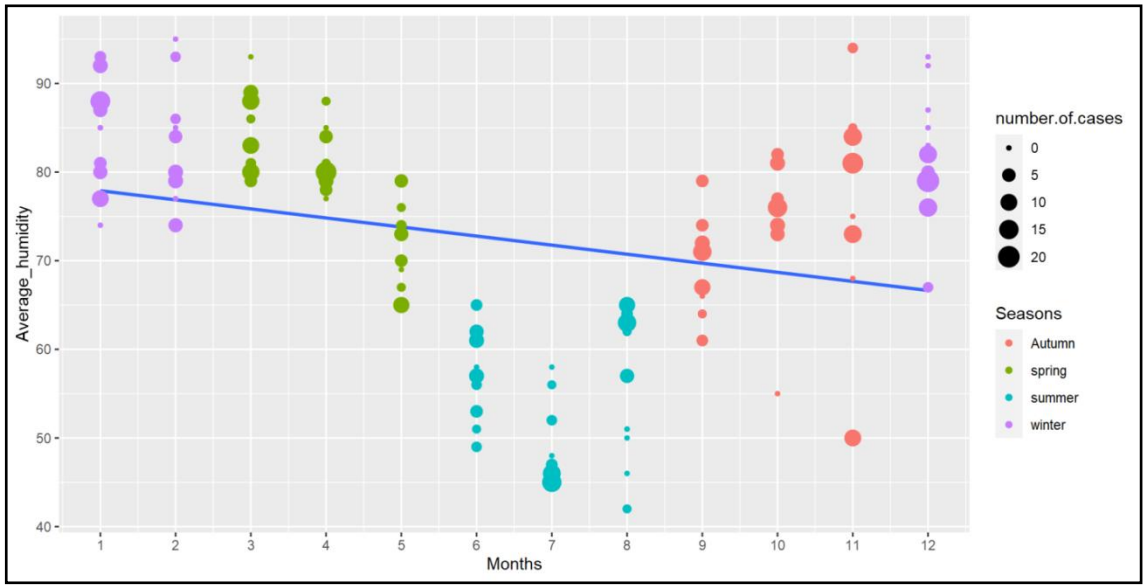


Fig. 56: The correlation between the average humidity (g/m^3) and the number of infected cases during the period (2012-2022).

4.3.5. The relationship between the variation of the average sunshine duration and the number of infected cases during the period (2012-2022)

Linear regression and Pearson correlation (**figure 57,58**) showed that the number of infected cases decreases progressively with the increase in the average duration of sunshine (hours) so there is a very strong negative correlation between the average duration of sunshine (hours) and the number of infected cases at during the descriptive study period.

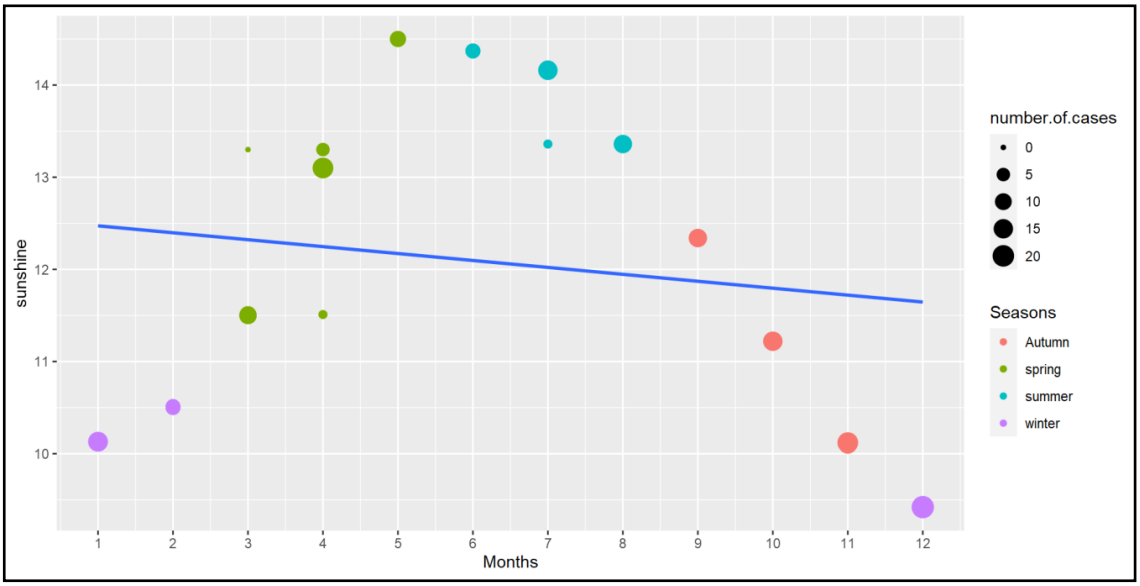


Fig. 57: The correlation between the average sunshine duration (hours) and the number of infected cases during the period (2012-2022).

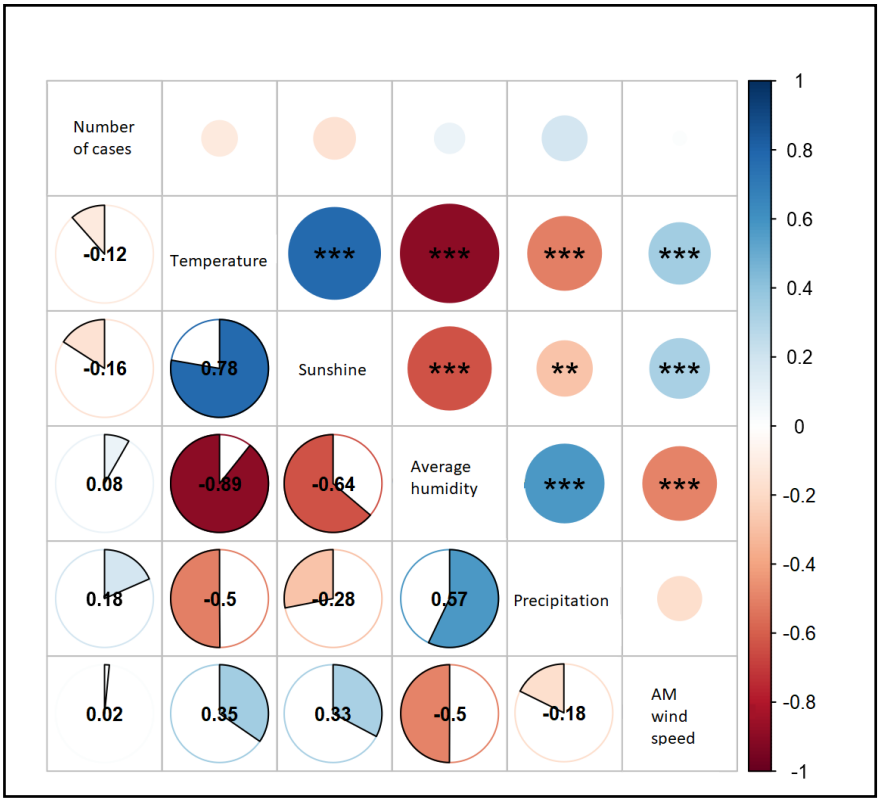


Fig. 58. Correlation matrix applied between meteorological parameters and number of cases of human *E. coli* bacteria. Pearson correlation tests are given as correlation coefficients (values below the diagonal, color shading, pie chart and square sizes) and the *p*-value (values above the diagonal). Significant correlations ($p \leq 0.05$)

Discussion

5. DISCUSSION

Escherichia organisms are rod-shaped, Gram negative bacilli that commonly inhabit the large intestine (*E.coli*) and are naturally excreted in stools. The urinary tract is the most common site of *E. coli* infection and more than 90% of all uncomplicated urinary tract infections (UTIs) are caused by *E. coli* infection **(Madappa, 2014)**, particularly in women because of the proximity of the urethra to the anus **(Gould, 2010)**. *E. coli* UTIs are caused by uropathogenic strains of *E. coli*.

This work aimed to determine the prevalence of *Escherichia coli* diagnosed in the bacteriology department of Maghlaoui-Mila Hospital over a period (January 2012 to December 2022).

In this study, the comparison of our results with the results of the scientific literature allowed us to identify human *E. coli* and to highlight the relationships of intestinal bacteria with different criteria such as age and sex of patients, years, months, and weather parameters.

We treated 14596 cases of which 495 were positive, which corresponds to a total infection rate of 3.39% during the period (January 2012 to December 2022) which is comparable to the results found in other studies conducted in hospitals in northwestern Algeria (CHU of Tlemcen, Sidi Bel Abbes and Oran) where they found a 240 cases of positive samples **(Ayad; 2016).**

Our results were also lower and higher than those of our Moroccan neighbors who observed a prevalence 32% in 2008 **(Tajdid *et al*., 2008)** and Nepal 28% in 2014 **(Bakour *et al*., 2014)**. Also recorded in Morocco 9.9% in 2015 **(Moutachakkir *et al*., 2015)**.The latter values probably reflect an unstable health condition favorable to the mode of infection by water or food contaminated with the bacteria.

On the other hand in England in 2013 was 53.1% **(Public Health England, 2014**) noted a higher prevalence than our result This difference in the rate can be explained by the improvement of sanitary conditions and the rise in the standard of living of the population.

This significant difference in these studies may be due to the fact that contaminated water is an important source of infection in humans, either through direct consumption or through its use in food processing.

Our study showed that women are more likely to be infected with *Escherichia coli* by 66.87% compared to men and this phenomenon is explained by the fact that women are more likely to develop urinary tract infections than men. Our results are consistent with other studies that found rates of 75% and 25% in women and men, respectively **(Ben Abdallah *et al.*, 2006; Bourjilat *et al.*, 2009)**. This is due to the following reasons: the anatomy of the reproductive system in women due to urinary tract infections, and they have a very short urethra, which facilitates the access of bacteria; The proximity between the anus and the urinary meatus facilitates access to the urethra for intestinal bacteria from the rectum, in addition, the man has good protection of the genitals; longer than the urethra. In some women, increased sexual activity can trigger symptoms of a UTI **(Lemaite *et al.*, 2005)**. Pregnant women are particularly at risk due to the pressure of the fetus on the urinary system, hormonal changes **(Pecher and Jacobs, 1994)**, and therefore pregnancy dilates the excretory tract.

Our results regarding the effect of age for the age groups included in this study ranged from newborns to ≥65 years, and the age group [20-44] was the most affected by *E. coli* with an infection rate of 34.74% followed by age [45-65] This agrees with the results of the study done in Meknes **(Moukrad, Filali and Makoudi, 2012)**. There is therefore a relationship between infectious risk and age. *E. coli* urinary tract infection is frequent in elderly patients these can be explained in women with two peaks, one at the beginning of sexual activity and the other in the postmenopausal period. In men, these infections are less frequent but can be increased after 50 years of age, in relation to prostatic pathology **(Ben abdalah *et al.*, 2006)**. In children this situation can be explained by the immaturity of the immune system, especially in the newborn **(Valerie *et al.*, 2012)**.

The results obtained show that the infestation rate during the period (2012-2022) was very high in 2019, 4.45% compared to other years where we notice that the prevalence of infection by *E. coli* have been fluctuating from one year to another. This remarkable growth in 2019 shows that the collective and individual prevention measures as well as the hygiene rules applicable to the risks related to water and food, must always be maintained to fight against this bacterium.

On the other hand we observed a slight increase in positive cases during the years 2015-2018 with SBI vary between 4.15% and 3.52% this can be explained by a better control of clinical and biological diagnosis by health personnel which would lead to the detection of more cases.

The analysis of the distribution according to seasons showed that the majority cases of *E. coli* bacteria indicate a fall prevalence of 29.89%, followed by a slight increase in winter and spring of 27.47% and 22.42%, while the presence of bacteria decreased during the hot summer, which is an inappropriate environment for the growth of bacteria, which is not consistent with what they have pointed out **(Duncan *et al*., 2000).** In Mozambique, observed an increase in *E. coli* and thermotolerant coliforms respectively, at the onset of the rainy season. (**Musa *et al*.,** The significant presence of *E. coli* bacteria during the summer and spring seasons due to the appropriate temperature for the growth of this bacteria in the United States of America, and its presence is low and non-existent in the cold months when temperatures fall.

We note that the rate of this bacteria by month is high during the months of November 11.92%, December 10.91% and January 10.50%. The other increase is recorded during the month of March 9, 49% that confirms the winter predominance. *E. coli* bacteria rise towards the optimal temperature that allows their growth **(Elsas *et al*., 2011)**. *E. coli* thrives at low temperatures. Prolonged exposure to cold temperatures such as 11°C or 7°C can reduce the density of *E. coli* bacteria in the medium **(Campos *et al*., 2013; Elsas *et al*., 2011)**.

Climate change induced heavy rainfall and flooding indeed has caused epidemics of waterborne diseases (**Delpla *et al*., 2009; Funari *et al*., 2012; Zhang *et al*., 2012**). waterborne diseases are related to the,concentration of waterborne pathogens in surface water (**Freeman *et al*, 2009**).Environmental variables influence Faecal Indicator Bacteria (FIB) in surface water. Understanding that influence is important, because presence of FIB, which are an indication of faecal contamination, means that harmful pathogens could be present that could also be influenced by environmental variables (**Majedul Islam *et al*.,2017**).

Escherichia coli are affected by temperature in winter. This result is consistent with the fact that winter temperatures are generally close to freezing, whereas the optimum temperature for *Escherichia coli* is 37°C, the average body temperature of mammals. Thus, temperature is a limiting factor for the reproduction and survival of *E. coli* bacteria in the environment during the winter period. This conclusion is the same as that reached by **Stanford *et al* in 2016**. In this case, we can say that the association becomes positive and leads to a higher bacterial index, and for this reason we have noticed the increase in bacterial cases in winter over the past 10 years.

Bacteria are more sensitive to changes in precipitation than fungi, particularly to decreases in precipitation (**Maestre *et al*.,2015**). **Li *et al*.,2022** show that changes in precipitation influenced bacterial communities increased precipitation may decrease the pathogen concentration of surface water due to dilution (**Lucas *et al*., 2014**).

Ventilation and air movement can reduce bacterial growth because moisture evaporation increases significantly as air movement increases, which dries the surfaces of substrates and inhibits bacterial growth. The evaporative heat transfer coefficient increases exponentially as the air movement increases (**Rabia,2021).** increasing the ventilation speed was much more effective at reducing the bacterial growth than reducing the humidity of the air, which even at a low level could effectively reduce the bacterial growth (**Yujia *et al*.,2022)**

Our prospective study during the first 3 months of 2023 in the Bacteriology and Bacteria laboratory at the Frères Maglaoui-Mila public hospital confirms our retrospective analytical study, as we obtained the same results concerning the effect of sex winter predominance on the distribution of *Escherichia coli*. Older people are the most infected > 65 years. Elderly people experience enhanced susceptibility to viral infections and subsequent superimposed bacterial infections. Altered immune responses in the elderly are responsible for many diseases, as well as for increased susceptibility to infections and cancer (**Daniel, 2010).** The risk of infection with Gram-negative bacilli is increased in the elderly, and *Escherichia coli* represents the primary cause of community-acquired bacteraemia in patients aged > 65 years **(Martin *et al*.,2006; Crane *al*.,2007; Diekema *et al*., 2002).**

CONCLUSION

CONCLUSION

E. coli is a truly resourceful microorganism possessing many facets. It is known for its fast-growing rate in chemically defined media and its adaptability, for ease of handling. So, *E. coli* is the most studied and well-understood organism on the planet.

Escherichia coli strains are a very efficient tool in laboratories, but it can also be a formidable pathogen that can cause severe illnesses, not only in the intestinal tract but also in other systems such as the urinary, nervous or blood systems (extra-intestinal infections).

Our data go in the direction of a better consideration of the local epidemiology of *Escherichia coli* at the level of the region of Mila, this retrospective analytical descriptive study takes place at the level of central laboratory service, medical bacteriology unit of public hospital establishment Brothers Maghlaoui – Mila (January 2012- December 2022) and a prospective study lasting three months (from January to March 2023).

The results obtained showed that 3.39% of the subjects were carriers of *Escherichia coli* among the positive cases, and the percentage of women reached 66.87%, followed by man 33.13%. There may be biological or behavioral factors that make women more susceptible to *E. coli* infection than men, including differences in sex hormones or social and environmental factors related to lifestyle.

Age also plays an important role in the spread of these bacteria. The age group [20-44] is the most exposed to the bacteria. This may be due to lifestyle and behavioral factors in this age group. This may include diet, habits or exposure to potential pathogens in the work or living environment.

According to our results, fall and winter are the seasons most associated with increased *E. coli* cases .Environmental factors associated with winter and fall, such as colder temperatures and changes in humidity, may have an effect on the spread and growth of germs.

In conclusion, it should be noted that this study represents an important contribution to our understanding of the relationship between *E. coli* and weather parameters and provides a basis for future research in this area. We hope that the results of this study will contribute to the development of prevention and control strategies for *E. coli* and thus improve the health of individuals and society in general. More efforts and research should be directed in this area to

better understand the transmission and spread pathways of this disease and develop effective preventive measures.

It is important to understand that weather and environmental factors are not the only factors contributing to the spread of *E. coli* and the relevant health authorities must work together to implement comprehensive preventive measures targeting all factors affecting the prevalence of this condition.

Bibliographic Reference

BIBLIOGRAPHIC REFERENCE

A.N.D.I (Agence Nationale de Développement de l'Investissement). 2013. La spectaculaire chut de Tamda près Ahmed Rachedi. *Rapport technique*.4p

A.N.I.R.E.F (Agence Nationale d'Intermédiation et de Régulation Foncière). 2011. Les zones industrielles et le développement local. *Séminaire régional de Bejaia*.

Abid L. 2014. La couverture sanitaire dans la wilaya de Mila. Sur le site : http://www.santemaghreb.com/algerie/documentations_pdf/docu_36.pdf

Abraham M.DIASSANA. 2018. Identification des souches *d'Escherichia coli* dans les selles en rapport avec la malnutrition à dioro. Bamako : s.n., 2018.

Achri Sarah., Lalouatni Borhane. 2018. Etude phénotypique des souches *Escherichia coli* multi-résistantes. Constantine : s.n., 2018.

Allerberger F., Wagner M., Schweiger P., Rammer HP., Resch A., Dierich MP. 2001. *Escherichia coli* O157 infections and unpasteurised milk. *Euro Surveill* 2001 ; 6 :147-151.

Andrade, J. R. C., DaVeiga, V. F& Suassuna, I. 1989. An endocytic process in HEp-2 cells induced by enteropathogenic *Escherichia coli*. *Journal of medical microbiology*, 28(1), 49-57.

Ayad A. 2016. Etude des mécanismes de résistance aux antibiotiques chez *Escherichia coli* au niveau des hôpitaux de l'Ouest algérien.

Bakour S., Touati A., Bachiri T., Sahli F., Tiouit D., Naim M. 2014. First report of 16S rRNA methylase ArmA-producing Acinetobacter baumannii and rapid spread of metallo-β-lactamase NDM-1 in Algerian hospitals. *J Infect Chemother* 20: 696–701.

Baliere C. 2016. Les *Escherichia coli* potentiellement pathogènes dans l'environnement littoral : cas des STEC et des EPEC .*Université de Bretagne Occidentale*; 2016. Disponible sur: http://archimer.ifremer.fr/doc/00312/42322/

Baranyi J., Roberts TA. 1995. Mathematics of predictive food microbiology. *Int J Food Microbiol.* 1995 ; 26 :199-218.

Barik D. 2012. Rapport du stage de Mila. Master 01 : *Géomorphologie appliqué. Faculté des sciences de la terre et de la géographie et de l'aménagement du territoire*. p 35.

Ben Abdallah H., Sahnoun O., Beb Romdhane F., Loussaief C., Noomen S., Bouzouaia N., Bouguessa R N., Ibadene H., Messai Y., Ammari H., Lounes S., Bakour R &Amp Guillaume A. 2006. Dissemination of ESBL, and Qnr determinations in *Enterobacter Cloacae* in Algeria. Algiers; 44 (12): 4584-4586.

Bentley R., Meganathan R. 1982. Biosynthesis of vitamin K (menaquinone) in bacteria. *Microbiol Rev*.;46(3):241–80.

Berche P.,Gaillard J. L., Simonet M. 1991. Bactériologie : Les bactéries des infections humaines. *Médecine- Sciences Flammarion, 3eme édition, France*, pp.77-601.

Berkal K & Elouaere F. 2014. Inventaire et écologie des oiseaux d`eau au niveau du Barrage de Béni Haroun (Wilaya de Mila) : saison d'hivernage 2013/2014. *Mémoire Master II, Centre Universitaire de Mila.* 85p.

Bielaszewska M., JaNnda J., Blahova K., MinAarikova H., Jikova E., MA. 1997. Human *Esherichia coli* O157:H7 infection associated with the consumption of unpasteurized goat's milk. *Epidemiol Infect 1997*; 119: 299-305.

Blanco M., J. Blanco, J. E. Blanco and J. Ramos. 1993. Enterotoxigenic, verotoxigenic,and necrotoxigenic *Escherichia coli* isolated from cattle in Spain. *Am J Vet Res* **54:**1446-1451.

Blattner Frederick R., Guy Plunkett., Craig A., Bloch Nicole., T Perna., Valerie Burland., Monica Riley., Julio Collado-Vides., Jeremy D., Glasner Christopher., K Rode and Ying Shao 1997. The complete genome sequence of *Escherichia coli* K-12.Science. 277 (5331):1453-1462.

Blount ZD. 2015. The unexhausted potential of *E. coli*. *Elife*. 2015;4:1–12.

Bonardi S., E. Maggi., A. Bottarelli., M. L. Pacciarini., A. Ansuini., G. Vellini., S.Morabito and A. Caprioli. 1999. Isolation of Verocytotoxin-producing *Escherichia coli* O157:H7 from cattle at slaughter in Italy. *Vet Microbiol* **67:**203-211.

Bouarroudj Yousra., Boutebza Fatima Zohra. 2015. *Les infections urinaires.* Constantine : s.n., 2015.

Boukhemis Amina., Boutersa Amina. 2015. Identification et antibiorésistance de souches d'*Escherichia coli* et de *Klebsiella pneumoniae* des infections urinaires à l'aide des moyens classiques et des moyens automatisés. Constantine : s.n., 2015**.**

Boularas H ., Kadjoudj N. 2016. Climat, environnement et maladies à transmission vectorielle : Cas de la leishmaniose cutanée dans la wilaya de Mlia. 43 p.

Bourjilat F. 2009. Etude prospective de la résistance chez *E. coli* dans l'hôpital de Meknès. Maroc. Eline. *Microbial. Rev.* 22, p 120-123.

Bourjilat F., Dersi, N., Bouchrif, B., Amarouch H & Amp Timinouni M. 2009. Profil de résistance aux antibiotiques des *Escherichia coli* uropathogènes communautaire au Maroc. *Eur J Sci Res*,38 (1), 57- 62.

C

Chahlat Fella., Kerdoud Karima. 2018. Etude de comportement d'installation chez l'Hirondelle de fenêtre (*Delichon urbica*) dans les étages climatiques de la wilaya de Mila. *Master protection des écosystèmes. Centre universitaire de Mila*.

Carlton E.J, Woster A.P, DeWitt P, Goldstein R.S, Levy K. 2016. A Systematic Review and Meta-Analysis of Ambient Temperature and Diarrhoeal Diseases. *International Journal of Epidemiology*. 2016; 45, 1: 117-30. https://doi.org/10.1093/ije/dyv296.

Carlton EJ., Eisenberg J.N.S., Goldstick J., Cevallos W., Trostle J., Levy K. 2014. Heavy Rainfall Events and Diarrhea Incidence: The Role of Social and Environmental Factors. *American Journal of Epidemiology*.2014;179,3:344-52. https://doi.org/10.1093/aje/kwt279.

Chapman P.A., Siddons C.A., Gerdan Malo A.T. 1997. A 1-year study of *E. coli* O157 in cattle, sheep, pigs and poultry. *Epidemiol Infect* 1997;**119**:245-250.

Connell, I. W Agace., P Klemm and C Svanborg. 1996. Type 1 fimbrial expression enhances *Escherichia coli* virulence for the urinary tract. *Proc Natl Acad Sci USA* 93, 9827-9832.

Crane SJ., Uslan DZ., Baddour LM. 2007. Bloodstream infections in a geriatric cohort: a population-based study.*American Journal of Medicine.* 2007; 120: 1078–1083.

Croxen M., Finlay B. 2010. Molecular mechanisms of *Escherichia coli*. *pathogenicity.Nat Rev Microbiol* 8:26-38.

D

D.S.P.M (Direction de Santé Publique de Mila). 2014. Structures sanitaires de la Wilaya de Mila. Sur le site : http://www.dsp-mila.dz/index.php/structures-sanitaires.

Dajoz R. 2000. Précis d'écologie: cours et exercices résolus. *7 ième édition. dunod*, paris. 613 p.

Daniel R. Goldstein.2010. Aging, imbalanced inflammation and viral infection. *Virulence* 1:4, 295-298; *July/August* 2010; © 2010 Landes Bioscience www.landesbioscience.com.

Darcan C., Ozkanca R., Idil O., Flint KP. 2009. Viable but non-culturable state (VBNC) of *Escherichia coli* related to EnvZ under the effect of pH, starvation and osmotic stress in sea water. *Pol J Microbiol*. 2009;58(4):307-17.

Delpla I., Jung A-V., Baures E., Clement M., Thomas O. 2009. Impacts of climate change on surface water quality in relation to drinking water production. Environ Int 35:1225–1233.

Denis F., Ploy M-C., Martin C., Bingin É and Quentin R. 2007. Bactériologie médicale .*édition Masson*.

Desmarchelier P & Fegan N. 2003. Enteropathogenic *Escherichia coli*. *In:* HOCHING, A. D. (ed.) Foodborne microorganisms of public health significance. *6th ed. Sydney*: *Austrtalian Institute of Food Science and Technology* (NSW Branch).

Diekema DJ., Pfaller MA., Jones RN.2002. Age-related trends in pathogen frequency and antimicrobial susceptibility of bloodstream isolates in North America: SENTRY Antimicrobial Surveillance Program, 1997–2000. *International Journal of Antimicrobial Agents* .2002; 20: 412–418.

Dobrindt Ulrich., Gabriele Blum-Oehler., Gabor Nagy., György Schneider., André Johann., Gerhard Gottschalk., Jörg Hacker .2002. Genetic structure and distribution of four pathogenicity islands (PAI I(536) to PAI IV(536)) of uropathogenic *Escherichia coli* strain 536. *Infect Immun* 70,6365-72 .

Dobrint U. 2005. (Path-) genomics of *Escherichia coli .Int J .Med Microbiol*. 2005; 295:357-378.

Dobrint U. ,M. Geddam Chowdary., G. Krumbholz & J. Hacker. 2010. Genome dynamics and its impact on evolution of *Escherichia coli*. *MedicalMicrobiology and Immunology*. 199 (3):145-154.

Doula H., Ferhat R. 2014. Entomo faune de l'olivier dans la région de Mila. *Mémoire présenté en vue de l'obtention du diplôme de Master.Université Constantine 1. Faculté des Science de la Nature et de la Vie*.52-60.

Duncan S.H., Booth I.R., Flint H.J and Stewart C.S. 2000. The potential for the control of *Escherichia coli* 0157 in farm animals. *Joumal of Applied Microbiology Symposium Supplement*, 88, 1578-1658.

Dziuban EJ., Liang JL., Craun GF., Hill V., Yupa., Painter J. 2006. Surveillance for waterborne disease and outbreaks associated. *MMWR Surveill Summ*. 2006 Dec . Vol. 57 / SS-9 22;55(12):1-30.

E

Elsas J.D.V, Semenov A.V, Costa R, Trevors J.T. 2011. Survival of *Escherichia Coli* in the Environment: Fundamental and Public Health Aspects. *The ISME Journal*. 2011; 5, 2: 173-83. https://doi.org/10.1038/ismej.2010.80.

Elsaset a. v. 2011. Survival of *Escherichia coli* in the environment: fundamental and publichealth aspects. *The Isme Journal*. 5 (2):173-183.

Escobar Páramo P., Le Menac H A., Le Gall T., Amorin C., Gouriou S., Picard B & Denamur E. 2006. Identification of forces shaping the commensal *Escherichia coli* genetic structure by comparing animal and human isolates. Environmental microbiology, *8*(11), 1975-1984.

Evans J., Chalmers R.,Chart H., Salmon R.L, Kench SM., Coleman TJ. 2002. Evidence of persisting serum antibodies to *Escherichia coli* O157 lipopoysacchride and Verocytoxin in members of rural communities in England.*Eur J Epidemiol* 2002 ;16 :885-889.

F

Faurie C. Ferra C and Medori P. 1980. Ecologie. *Edition. J. B. Baillère.* Paris. 168p.

Ferens W. A and C. J. Hovde. 2011. *Escherichia coli* O157:H7: animal reservoir and sources of human infection. *Foodborne Pathogens Disease*. 8 (4):465-487.

Freeman J., Anderson D., Sexton D. 2009. Seasonal peaks in Escherichia coli infections: possible explanations and implications. *Clin Microbiol Infect* .15:951–953.

Funari E., Manganelli M., Sinisi L. 2012. Impact of climate change on waterborne diseases. *Ann Ist Super Sanità.* 48:473–487.

G

Gould D. 2010. Causes, prevention and treatment of *Escherichia coli* infections. *Nurs Stand* 24(31): 50–6. doi: 10.7748/ns2010.04.24.31.50.c7692

Gould D. 2011. *Escherichia coli* recognition and prevention. *Primary Health Care* 21(8): 32 9. doi: 10.7748/ phc2011.10.21.8.32.c8738

Gould S. J. 1996. Full House: The Spread of Excellence from Plato to Darwin 175- 192.

Griffin P. M & Tauxe R. V. 1991. The epidemiology of infections caused by *Escherichia coli* O157:H7, other enterohemorrhagic *E. coli*, and the associated hemolytic uremic syndrome. *Epidemiol Rev*, 13**,** 60-98.

Grimont G., V Livrelli, P., Mariani-Kurkdjan., N. Pradel, and E. Oswald. 2003. Physiopathologie des maladies dues aux STEC, p. 41-59. *In* AFSSA (ed.), Bilan des connaissances relatives aux *Escherichia coli* producteurs de Shiga-toxines (STEC). AFSSA, Maisons-Alfort.

Guiraud J. P. 1993. Génétique Génétique microbienne, Bases théroriques et introduction aux applications pratiques. Paris : Technique et docuentation-Lavoisier, 1993 ;chap 2 et 3, pages 83-151.

H

Hacker J. 2000. Pathogenicity islands and the evolution of microbes.Annu Rev Microbiol 54, 641-79.

Haslay C., Leclerc H. 1993. Microbiologie des eaux d'alimentation. Technique & Documentation, Lavoisier, 495p.

I

International Commission On Microbiological Specifications For Foods. 1996. Intestinally pathogenic *Escherichia coli*. Microorganisms in food 5: Microbiological specifications of food pathogens. London: Blackie Academic and Professional.

Ishii S., W. B. Ksoll R.. E. Hicks, and M. J. Sadowsky. 2006. Presence and growth of naturalized *Escherichia coli* in temperate soils from Lake Superior watersheds. *Applied and Environmental Microbiology*. 72 (1):612-621.

J

Jackson SG., Goodbrand RB., Johnson RP.,Odorico VG., Alves D., Rahn K.. 1998. *Escherichia coli* O157 :H7 diarrhoea associated with well water and infected cattle on an Ontario farm.*Epidemiol Infect* 1998 ; 120: 17-20.

Jean-Philippe Lavigne. 2004. – DFGMS2 'Infectieux.

Johnson J. R & Russo T. A. 2002. Extra intestinal pathogenic *Escherichia coli*: "the other bad *E coli*". *Journal of Laboratory and Clinical Medicine*, *139*(3), 155-162.

Johnson JR, Russo TA. 2005. Molecular epidemiology of extraintestinal pathogenic (uropathogenic) *E. coli*. *Int J Med Microbiol.* 2005 Oct;**295**(6-7):383-404. Revue.

Joly B and Reynaud A. 2002. Entérobactéries systématique et méthodes de diagnostic. Edition TEC & DOC.

Jones T. H. 2012. Response of *Escherichia coli* to Environmental Stress. *In:* WONG, H. C. (ed.) Stress Response of Foodborne Microorganisms. New York: Nova Science Publishers Inc.

K

Kaper J.B., Nataro J.P & Mobley H.L. 2004. Pathogenic *Escherichia coli*. *Nat Rev Microbiol* 2, 123-40.

Kathleen C. 2013. Étude des mutations de résistance des *Escherichia coli* uropathogènes résistants à l'antibiotique fosfomycine. *Mémoire présenté pour l'obtention du grade de Maître ès science : microbiologie appliquée* ,105p.

Kauffmann F. 1947. The serology of the coli group. *J Immunol*, 57**,** 71-100.

L

Lavigne, J. P. 2004. *Escherichia coli* – DFGMS2 'Infectieux.p7.

Lemaite L.,Puech P., Fauque L., Delomez J., Leroy C., Fantoni J-C and Biserte J. 2005. Apport de l'imagerie dans la prise en charge des infections de l'appareil urinaire. *Ann Urol* ;39 :170-196.

Levine M. 1987. *Escherichia coli* that Cause Diarrhea: Enterotoxigenic, Enteropathogenic, Enteroinvasive, Enterohemorrhagic, and Enteroadherent. *Journal of Infectious Diseases*, 155(3), 377–389.

Li J., Benti G., Wang D., Yang Z and Xiao R. 2022. Effect of Alteration in Precipitation Amount on Soil Microbial Community in a Semi-Arid Grassland. Frontiers in Microbiology.March2022.Volume13.Article842446.https://www.frontiersin.org/articles/10.3389/fmicb.2022.842446/full.

Lil Mendis N., Trigui H., Oliver JD., Faucher SP. 2014. The importance of the viable but non-culturable state in human bacterial pathogens. *Front Microbiol*. 2014;5:258.

Lobril JR. 1998. Réévaluation du modèle de croissance de Monod : effets des antibiotiques sur l'énergie de maintenance. *Thèse de l'université de Lyon I France* 1998 : 42-77.

Lucas FS., Therial C., Gonçalves A., Servais P., Rocher V., Mouchel J-M . 2014. Variation of raw wastewater microbiological quality in dry and wet weather conditions. *Environ Sci Pollut Res* 21:5318–5328

M

M. M. Majedul Islam ., Nynke Hofstra and Md. Atikul Islam. 2017. The impact of environmental variables on faecal indicator bacteria in the Betna River Basin, Bangladesh Environ. Process. (2017) 4:319–332 .DOI 10.1007/s40710-017-0239-6

Majedul Islam, M.M. Nynke Hofstra, Md. Atikul Islam. 2017. The Impact of Environmental Variables on Faecal Indicator Bacteria in the Betna River Basin, Bangladesh. Environ. Process. (2017) 4:319–332. DOI 10.1007/s40710-017-0239-6.

Madappa T. 2014. *Escherichia coli* infections. http://tinyurl . com/qfxuocb.

Maestre F. T., Delgado-Baquerizo M., Jeffries T. C., Eldridge D. J and Singh B. K. 2015. Increasing aridity reduces soil microbial diversity and abundance in global drylands. *Proc. Natl. Acad. Sci. U.S.A.* 112, 15684–15689. doi: 10.1073/ pnas.1516684112.

Mainil J. 2003. Facteurs de virulence et propriétés spécifiques des souches invasives *d'E.coli* : les adhésines et facteurs de colonisation.*Ann. Méd. Vét.*, 147, 105-126.

Mainil j. 1999. Verocytotoxins and Shiga verotoxigenic *Escherichia coli* in animals. *Vet. Res.*, 1999,30, 235-257.

Martin GS., Mannino DM., Moss M. 2006. The effect of age on the development and outcome of adult sepsis. *Critical Care Medicine* 2006; 34: 15–21.

Martinez-Gomez K., Flores N., Castaneda H. M., Martinez-Batallar G., Hernandez Chavez G., Ramirez O. T., Gosset G., Encarnacion S. & Bolivar F. 2012. New insights into *Escherichia coli* metabolism: carbon scavenging, acetate metabolism and carbon recycling responses during growth on glycerol. *Microb Cell Fact*, 11**,** 46.

Merghadi A., Abderrahmane B., Tien Bui D. 2018. Landslide susceptibility assessment at Mila Basin (Algeria): a comparative assessment of prediction capability of advanced machine learning methods. ISPRS. *International Journal of Geo-Information*, 7(7), 268.

Metaai. S., beldi. H. 2011. Evaluation du degré de la contamination par les pesticides des eaux et des sédiments du barrage de béni haroun (mila). *Mémoire d fin d`études. Université de jijel*. 23 p.

Ministry For Primary Industries – New Zealand. 2001. *Escherichia coli* O157: H7. Available:http://www.foodsafety.govt.nz/elibrary/industry/*Escherichia_Coli*.Organism_ Invades. pdf [Accessed 22 Aug 2015].

Mokady D., Gophna U., Ronez. 2005. Extensive gene diversity in septicemic *E. coli* strains. *J Clin Microbiol.* 2005 Jan;**43**(1):66-73.

Molina P. M., Parma A. E. & Sanz M. E. 2003. Survival in acidic and alcoholic medium of Shiga toxin-producing *Escherichia coli* O157:H7 and non-O157:H7 isolated in Argentina. *BMC Microbiol*, 3**,** 17.

Monnet D. 2000. Antibiotic use and bacterial resistance. *Ann Fr Anesth Reanim* 19: 409 17.

Moukrad N., Filali F. R., & Amp Makoudi, Y. 2012. Prévalence de la multi-résistance bactérienne aux antibiotiques des infections urinaires dans la ville de Meknès (Maroc) et son évolution dans le temps. *Sciences Lib ED. Mercenne*, V. 4 N° 121105 (2012), ISSN 2111- 4706.

Moutachakkir M., Chinbo M., Elkhoudri N and Soraa N. 2015. Antibiotic resistance of uropathogenic *Enterobacteriaceae* in pediatric wards at the University Hospital of Marrakech. *J Ped Puericult* 28: 16-22.

N

Nataro J. P. & Kaper, J. B. 1998. Diarrheagenic *Escherichia coli*. *Clin Microbiol Rev*, 11**,** 142-201.

Nauciel C., Vildé J.L. 2007. Bactériologie médicale. Elsevier Masson SAS .p 122-123.

Neill M. A., Tarr P. I., Taylor D. N. & Trofa A. F. 1994. *Escherichia coli*. *In:* HUI, Y. H., GORHAM, J. R., MURELL, K. D. & CLIVER, D. O. (eds.) Foodborne Disease Handbook. New York: Marcel Decker Inc.

O

O. G. 2007. «Utilisation des méthodes biométriques dans l'identification de quelques bacteries à gram négatif thése de DOCTORAT en pharmacie,» Mali, 2007.

O'brien AD, Laveck GD,Thompson MR., Formal S.B.1982. production of Shigella dysenteriae type 1-like cytotoxin by *Escherichia coli*. J Infect Dis 1982 ;146 :763-769.

Ogden ID., Hepburn NF., Macrae M., Strachan NJ., Fenlon DR., Rusbridge SM., Pennington TH. 2002. Long-tem survival of *Escherichia coli* O157 on pasture following an outbreak associated with sheep at a scout camp.Lett Appl Microbiol 2002 ; 34 :100-104.

P

Paton AW.,Srimanote P.,Woodrow MC., Paton JC . 2001. Characterization of Saa, a novel autoagglutinating adhesin produced by locus of enterocyte effacement-negative Shiga toxigenic *Escherichia coli* strains that are virulent for humans.*infect Immun* 2001 ;69 :6999- 7009.

Pechere. J-C., Girard. J-F. 1994. Les infections. *3 éme édition, Maloine*, Canada.

Public Health England. 2014. Results from the mandatory surveillance of *Escherichia Coli*. http://tinyurl.com/ mst8d7b.

Ploy M-C., Denis F., Martin C., Bingin É and Quentin R. 2007. Bactériologie médicale édition Masson.

Pommepuy M ., M Butin., A Derrien., M Gourmelon., R R Colwell., M Cormier . 1996. Retention of enteropathogenicity by viable but nonculturable *Escherichia coli* exposed to seawater and sunlight.*Applied and Environmental Microbiology*. 62 (12):4621-4626.

Posl P., Linermas P ., Mainil J and Deprez P .1998. Production des vérocytotoxine par *Escherichia coli* du porc. Annales de médicine vétérinaire. p 133.31-38.

Power M. L., J. Littlefield-Wyer., D. M. Gordon., D. A. Veal., and M. B. Slade. 2005. Phenotypic and genotypic characterization of encapsulated *Escherichia coli* isolated from blooms in two Australian lakes. *Environmental Microbiology*. 7 (5):631-640.

Public Health England .2014. Results from the mandatory surveillance of *Escherichia Coli*. http://tinyurl.com/ mst8d7b.

R

Rabia S. 2021. ASHRAE Handbook Fundamentals (I P); Section 9.3; *ASHARE: Peachtree* Corners, GA, USA, 2021.

Reid S. D. 1999. Sequence diversity of flagellin (fliC) allelesin pathogenic *Escherichia coli*. *J Bacteriol* 181, 153–160.

Remmache.I. 2006. potentiel en substances utiles non métalliques (gypse et sel gemme) du bassin de mila (algérie nord orientale). *Thèse de magistère en géologie. Université mentouri, constantine*. 2 p.

Richard d'Ari., Guennadi Sezonov. 2008. Biologie et génétique *d'Escherichia coli :* les organismes modèles. *Paris : Belin éducation*, 2008.

Riley L. W., Remis R. S., Helgerson S. D., McGee H. B., Wells J. G., Davis B. R & Blake P. A. 1983. Hemorrhagic colitis associated with a rare *Escherichia coli* serotype. *New England Journal of Medicine*, 308(12), 681-685.

Robert Elodie ., Manuela Grippa., Dayangnéwendé Edwige Nikiema., Laurent Kergoat., Hamidou Koudougou., Yves Auda., Emma Rochelle-Newall. 2021. Environmental determinants of *E. coli*, link with the diarrheal diseases, and indication of vulnerability criteria in tropical area (Kapore, Burkina Faso). *Plos neglected tropical diseases*. August 17, 2021.

Russo TA., Johnson JR. 2003. Medical and economic impact of extraintestinal infections due to *Escherichia coli*: Focus on an increasingly important endemic problem. *Microbes Infect*. 2003;5(5):449–56.

§

Savageau MA. 1983. *Escherichia coli* habitats, cell types, and molecular mechanisms of gene control. *Am Nat*. 1983;122(6):732–44.

Seddiki H., Chaalal M., Stambouli I. 2013. Mila la wilaya. Spectaculaire chut de Tamda près Ahmed Rachedi. *Rapport technique.Ed, Albayazin.* 101p.

Ségolène M. 2016. Caractérisation de souches d'*Escherichia coli* pathogènes urinaires provenant de guadeloupe : portrait de la diversité des facteurs de virulence présents. *Mémoire présentée pour l'obtention du grade de Maître ès sciences (M.Sc.) en Microbiologie Appliquée. Université du Québec*.

Seltzer A. 1946. Le climat de l'Algérie. Inst. Météo. Phys. glob. Université. Alger.219p.

Smati A. 2015. Quantitative analysis of commensal *Escherichia coli* populations reveals host-specific enterotypes at the intra-species level. *Microbiology Open*. 4 (4):604-615.

Söderström A., Österberg P., Lindqvist A., Jönsson B., Lindberg A., Blide Ulander S & Kühlmann-Berenzon S. 2008. A large *Escherichia coli* O157 outbreak in Sweden associated with locally produced lettuce. *Food borne pathogens and disease*, *5*(3), 339-349.

Soukehal B., Cherrad S. 2011. Les ressources en eau dans la wilaya de Mila mobilisation, consommation et comportement de ménages. *Science et technologie* D -N°34.

Soukehal B. 2009. La wilaya de Mila: villes, villages et problématique de l'alimentation en eau potable. *Th. Doc. Univ. Mentouri-Constantine. Algérie* .135p.

Stanford K., Johnson R. P., Alexander T. W., Mc Allister T. A., Reuter T. 2016. Influence of season and feedlot location on prevalence and virulence factors of seven serogroups of *Escherichia coli* in feces of western-Canadian slaughter cattle. *PLoS One*, *volume11*.Repéré à https://journals.plos.org/plosone/article?id=10.1371/journal. pone. 0159866

Stewart . 2015. Plos Biologiy February 2015.

Stordeur P M. J. 2002. « La colibacillose aviaire ». *Ann. Méd. Vet.*146.

Strachan NJ., Dunn GM., Locking ME., Reid TM., Ogden ID. 2006. *Escherichia coli* O 157 : burger bug or environmental pathogen .*Int J Food Mmicrobiol* 2006 ; 112 :129-137.

Sugiyama A., Iwade Y.,Akachi S.,Nakano Y.,Matsuno Y.,Yano T. 2005. An outbreak of Shigatoxin – producing *Escherichia coli* O157:H7 in a nursery school in Mie Perfecture. *Jpn J Infect Dis* 2005 ; 58 :398-400.

Sumbali G & Mehrotra R. S. 2009. Principles of Microbiology, New Delhi, Tata McGraw Hill Education Private Limited.

Surveillane E. 1997. Surveillance des infections à *E. coli* entérohémorragiques (EHEC) et du syndrome hémolytique et urémique (SHU) en Europe. DGV de la commission des communautés européennes 1997 ; p : 12.

T

Tajdid M.R., Boumhil L., Iken M., Adnaoui M and Benouda A. 2008. Resistance to fluoroquinolones and third generation cephalosporin of *Escherichia coli* from urines. *Med Mal Infect* 40: 70-3.

Thorene G. 1994. Hurmonal immune responses to shiga-like toxins and *Escherichia coli* ; p43.

Todar K. 2005. Pathogenic *E. coli*. Todar's Online Textbook of Bacteriology [Online]. Available: http://textbookofbacteriology.net/tuberculosis.html.

Tymensen, L. D., F. Pyrdok, D. Coles, W. Koning, T. A. McAllister, C. C. Jokinen, S. E. Dowd, and N. F. Neumann. 2015. Comparative accessory gene fingerprinting of surface water *Escherichia coli* reveals genetically diverse naturalized population. *Journal of Applied Microbiology*. 119 (1):263-277.

V

Valérie B., Sophie S., Yannick A. 2012. Particularité des infections noocomiales chez l'enfant fragile.Spécificités en néonatologie. *Université Paris 7.Diderot,sorbonne* Paris cité, France.p :41- 50.

Vernozy-Rozand C., Montet MP.,Berardin M. 2005. Isolation and characterization of Shiga toxin-producing *Escherichia coli* strains from raw milk cheeses in France.*Lett Appl Microbiol* 2005 ;41 : 235-241.

Villemeuve O. 1974. Glossaire de météorologie et de climatologie. Les presses l'Université, Laval. Imprimé au Canada. 560 p.

W

Walk, S. T., E. W. Alm., L. M. Calhoun., J. M. Mladonicky and T. S. Whittam. 2007. Genetic diversity and population structure of *Escherichia coli* isolated from freshwater beaches. *Environmental Microbiology*. 9 (9): 2274-2288.

Y

Yujia Qiu ., Yan Zhou ., Yanfen Chang ., Xinyue Liang ., Hui Zhang ., Xiaorui Lin ., Ke Qing ., Xiaojie Zhou and Ziqiang Luo.2022. The Effects of Ventilation, Humidity, and Temperature on Bacterial Growth and Bacterial Genera Distribution. *Int. J. Environ. Res. Public Health* 2022, 19, 15345.

Z

Zhang L., Seagren EA., Davis AP., Karns JS. 2012. Effects of temperature on bacterial transport and destruction in bioretention media: field and laboratory evaluations. *Water Environ Res* .84:485–496.

Zhang, Q., and T. Van. 2012. Correlation of Intracellular Trehalose Concentration with Desiccation Resistance of Soil *Escherichia coli* Populations. *Applied and Environmental Microbiology*. 78 (20):7407-7413.

Zouidia. H. 2006. in Benacha & Benaskeur 2015. Bilan des incendies de forets dans l'Est algérien cas de Mila, Constantine, Guelma et Souk-Ahras. *Thème de magistère en écologie et Environnement. Université Mentouri, Constantine.* 126 P.

Annex

Annex

One-Sample Test

	Test Value = 0					
					95% Confidence Interval of the Difference	
	t	df	Sig. (2-tailed)	Mean Difference	Lower	Upper
Sex	80,371	494	,000	1,683	1,64	1,72

ANOVA one way

Sex

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	3,931	11	,357	1,671	,077
Within Groups	103,273	483	,214		
Total	107,204	494			

One-Sample Test

	Test Value = 0					
					95% Confidence Interval of the Difference	
	t	df	Sig. (2-tailed)	Mean Difference	Lower	Upper
Age	56,906	494	,000	5,404	5,22	5,59

ANOVA one way

Age

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	94,580	11	8,598	1,968	,030
Within Groups	2110,612	483	4,370		
Total	2205,192	494			

ANOVA one way

Sex

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	3,931	11	,357	1,671	,077
Within Groups	103,273	483	,214		
Total	107,204	494			

ANOVA bivariate Table

			Sum of Squares	df	Mean Square	F	Sig.
Sex * Age	Between Groups	(Combined)	8,651	7	1,236	6,107	,000
		Linearity	2,001	1	2,001	9,890	,002
		Deviation from Linearity	6,649	6	1,108	5,476	,000
	Within Groups		98,553	487	,202		
	Total		107,204	494			

One-Sample Test

Test Value = 0

	t	df	Sig. (2-tailed)	Mean Difference	95% Confidence Interval of the Difference	
					Lower	Upper
Months	41,199	494	,000	6,838	6,51	7,16

ANOVA one way

Months

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1284,569	9	142,730	12,696	,000
Within Groups	5452,502	485	11,242		
Total	6737,071	494			

One-Sample Test

Test Value = 0

	t	df	Sig. (2-tailed)	Mean Difference	95% Confidence Interval of the Difference	
					Lower	Upper
Seasons	47,489	494	,000	2,525	2,42	2,63

ANOVA one way

Seasons

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	76,258	9	8,473	6,680	,000
Within Groups	615,176	485	1,268		
Total	691,434	494			

One-Sample Test

Test Value = 0

	t	df	Sig. (2-tailed)	Mean Difference	95% Confidence Interval of the Difference	
					Lower	Upper

Years	61,497	494	,000	6,929	6,71	7,15

ANOVA one way

Years

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	499,633	11	45,421	8,422	,000
Within Groups	2604,892	483	5,393		
Total	3104,525	494			

Field: Nature and Life Sciences	**Sector:** Ecology and environment	**Specialty:** Protection of Ecosystems

Abstract

In order to determine the epidemiological and clinical characteristics of human *Escherichia coli* in the district of Mila, we developed a retrospective study during a period extending from January (2012 to December 2022), and the other prospective during three-months (January-March 2023). We collected the data at the level of Bacteriology laboratory of the General Hospital Maglaoui Brothers Mila, for the retrospective descriptive analytical study we treated 14596 examinations, 495 were positive, an infestation rate of 3.39%.

-Of the positive cases, 66.87% were female and 33.13% were male.

- Patients aged (20-44 years) are the most exposed to *E. coli*.

-The years 2019 and 2020 had the highest infection rates, 4.45% and 4.45% respectively.

-The spread of these bacteria was observed during the fall season.

-A high rate of this bacterium was noted during the months of November, December.

-The climatic conditions of Mila region, like the ambient temperature increase the dissemination of the bacteria. Sunshine, average humidity, precipitation, and average monthly wind speed cause a decrease in the bacterial index of *Escherichia coli*.

The results obtained during the three months of our prospective study (January-March 2023), confirmed what we deduced from the results above (The retrospective study from 2012 to 2022).

Keywords: *Escherichia coli*, epidemiology, prevalence, correlation, meteorological parameters, Mila.

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