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Title:

EFFECT OF PLASTIC POLLUTION WASTE ON THE SOIL ORGANISMS: THE CASE OF EARTHWORMS (Aporrectodea trapezoids **and** *Aporrectodea rosea.)*

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Table of Contents

Chapter III: General information about plastic pollution.

Chapter IV: Materials and Methods

Chapter V: Results and Discussions.

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With the help of Almighty God, who traced my way of my life, I was able to carry out this work which I dedicate:

To my dear parents

To my brother

To all my teachers from primary school to university

To you …

Abstract

Plastic waste and microplastic pollution have negative effect for the soil organisms. The objective of our study is to see the effect of pollution by microplastic and plastic waste on earthworms which live in soil of production. To achieve our goal we did small experiment in ecology laboratory at University Constantine 01, faculty of nature and life science. Two samples were tested; polluted sample (MP) and non-polluted (MNP). by using earthworms. Two different species were in our experiment *Aporrectodea trapezoids* and *Aperrectodea rosea*.

Results obtained revealed that earthworms which are in polluted incubation media had low biomass compared to those in non-polluted jars (BM/P = 0.69g ± 0.28 ; BM/NP = 5.61g ± 049). It seems that the pH values of plastic waste was lowered than the non-polluted ones (pH/P = 7.33 ± 0.02 ; pH/NP = 7.40 ± 0.06) . Also, the results show that the EC values are high in polluted soils compared to unpolluted ones (EC/P = 1015 µS/cm ± 30.21 ; EC/NP = 856.6 µS/cm ± 138.92).

We can conclude that pollution by plastic waste can have negative effects on soil organisms, biodiversity and human.

Key words : Pollution by plastic waste, microplastic , Soil pollution , Pedofauna ,Earthworms.

Resume

La pollution par les déchets plastiques pourra avoir un impact négatif sur les organismes du sol. L'objectif de notre étude est de voir l'effet des déchet du plastique et microplastique sur les lombriciens. Pour atteindre notre but nous avons réalisé une simple expérimentation au laboratoire d'écologie à la faculté des sciences de la nature et de la vie de l'université des frères Mentouri, Constantine1, en utilisant les espèces lombriciennes suivantes *Aporrectodea trapezoids* et *Aperrectodea rosea*.

Les résultats obtenus révèlent que les vers de terre des milieux d'incubation pollués ont une biomasse faible par rapport aux milieux non pollués (BM/P = 0.69g ± 0.28 ; BM/NP = 5.61g ± 049). Il semble que les valeurs de pH des milieux pollués tendent abaissé que celles des milieux d'incubation non pollués pH/P = 7.33 ± 0.02 ; pH/NP = 7.40 ± 0.06). Aussi, les valeurs de la CE sont plus élevées dans les sols pollués comparés à ceux non pollués (EC/P = 1015 µS/cm ± 30.21 ; EC/NP = 856.6 µS/cm ± 138.92).

Nous pouvons conclure que la pollution par les déchets plastiques pourra avoir des effets négatifs sur les organismes du sol, la biodiversité et l'Homme.

Mots clés : pollution par les déchets plastique, microplastiques, pollution des sols, pédofaune, lombriciens.

List of abbreviation.

Stm– system

NP – Not Polluted

P – Polluted

MP – medium polluted

MNP – Medium non polluted

EC – Electrical conductivity

Ap – Aporrectodea

Ew – Earthworm

Nb – Number

BMP – Biomass of media pollutant

BMNP – Biomass of media non pollutant

MC – Mean square

SC – Sum of squares

PCA – Principal Component Analysis

List of tables

List of figures

List of photo

General introduction

Problematic:

Microscopic bits of plastic have been showing up throughout the environment. Most measurements of it have been made in water. But this pollution can taint the air and soil, too. This study finds that microplastics in soil can stunt the growth of earthworms. And that's worrying, because earthworms tend to help make soil good for growing plants.

Worms create little tunnels as they move through the ground. These tunnels allow air and water to get to plant roots. Without worm tunnels, soil can dry out and become too compacted for plants to grow well.

Plastics made from fossil fuels are just over a century old. Production and development of thousands of new plastic products accelerated after World War II, so transforming the modern age that life without plastics would be unrecognizable today. Plastics revolutionized medicine with life saving devices, made space travel possible, lightened cars and jets saving fuel and pollution and saved lives with helmets, incubators, and equipment for clean drinking water.

The conveniences plastics offer, however, led to a throw-away culture that reveals the material's dark side: today, single-use plastics account for 40 percent of the plastic produced every year. Many of these products, such as plastic bags and food wrappers, have a lifespan of mere minutes to hours, yet they may persist in the environment for hundreds of years.

How plastics move around the world

Most of the plastic trash in the oceans, Earth's last sink, flows from land. Trash is also carried to sea by major rivers, which act as conveyor belts, picking up more and more trash as they move downstream. Once at sea, much of the plastic trash remains in coastal waters. But once caught up in ocean currents, it can be transported around the world.

The researchers conclude that, fragments of plastic are present practically all over the world and can trigger many kinds of adverse effects.

The study estimates that one third of all plastic waste ends up in soils or freshwater. Most of this plastic disintegrates into particles smaller than five millimeters, known as microplastics, and these break down further into nanoparticles (less than 0.1 micrometer in size). The problem is that these particles are entering the food chain.

Importance of soil fauna for the soil durability

Soil fauna play an essential role in soil functions as they are involved in processes such as the decomposition of organic matter, the formation of humus and the nutrient cycling of many elements (nitrogen, sulphur, carbon). Moreover, edaphic fauna affect the porosity and aeration of soil as well as the infiltration and distribution of organic matter within soil horizons. The ecosystem services provided by soil fauna are one of the most powerful arguments for the conservation of edaphic biodiversity. Decomposition of organic matter by soil organisms is crucial for the functioning of an ecosystem because of its substantial role in providing ecosystem services for plant growth and primary productivity.

The activity of earthworms produces a significant effect, not just on the structure, but also on the chemical composition of the soil, since a large part of the organic matter ingested by earthworms is returned to the soil in a form easily used by plants. While they are feeding, earthworms also ingest large quantities of mineral substances (minimally so in the case of the epigeic), that are then mixed with the organic matter ingested and, after having been cemented with a little mucous protein, are expelled in piles called worm casts.

The soil fauna, in particularly mollusks and earthworms, also has an effect on the soil through the secretion of cutaneous mucous, that have a cementing effect on the particles in the ground, assisting the stability and structure of the soil and making it less vulnerable to processes of erosion. The mucous secretions, the feces (especially those of earthworms) and the bodies themselves of the animals (when they die) influence in large measure the concentration of nutrients present in the soil particularly potassium, phosphorous and nitrogen

Earthworms significantly affect plant growth through their effects on microorganisms, aggregation of soil, and nutrient supply (Sabrina *et al.,* 2009). surface, consuming soil along the way. Coiled soil masses known as casts are excreted from the worm's digestive system, making the soil more fertile. The earthworm's burrowing action continually moves mineral-rich soil to the surface, which improves plant growth.

Earthworms contribute to soil turnover, structure and formation and serve as a fertility enhancer in various ways. Earthworms and their casts are useful in land improvement, reclamation and in organic waste management (Edwards and Baker, 1992; Lavelle and, Martin, 1992; Johnson, 1997; Villenave *et al.,* 1999). Soil productivity can be improved by manipulating the community of earthworms in the soil (Brown *et al.,* 1999).

Earthworms have important roles in soil physical, chemical and biological properties (Edwards, 2004). Earthworms eat soil organic matter and litter and increase availability of plant nutrients in their casts (Brown *et al*., 2004). The nutrients can increase plant growth and yield of crops as a result (Edwards and Bohlen, 1996). These are good indicators that the earthworm activities and behavior interact strongly with physical, chemical and biological properties of the soil (Pattana and Pongthep, 2009).

Earthworms are considered as key ecological mediators that have the capacity to affect soil functions and microbial activities (Binet *et al*., 1998; Lavelle *et al*., 2016), by producing an energy-rich mucus that activates microorganisms through a priming effect (Jenkinson, 1966) and signal molecules that have hormone-like effects and influence plant gene expression (Puga-Freitas and Blouin, 2015). The mutualistic interaction existing between earthworms and the soil microbiota has been named the "Sleeping Beauty Paradox" (Lavelle *et al*., 1995; Brown et al., 2000), where *dormant* soil microorganisms, awaiting suitable environmental conditions are activated by the *kiss* of the earthworm made of easily assimilable glycoproteins present in the drilosphere in the form of intestinal or cutaneous mucus as already mentioned. This triggers the acceleration of microbial processes for a short period of time ("hot moment") and in a limited soil space ("hot spot"), at the microscale of a biopore or aggregate (Kuzyakov and Blagodatskaya, 2015) which reverberates on a larger scale, at the drilosphere and soil levels (Brown et al., 2000; Hoang *et al.*, 2016;Lipiec *et al.*, 2016).
Earthworms have a direct and important effect on the soil microbiota through their nutrition. This effect may depend on their food preference, selection, food ingestion rate, digestion and assimilation, as mentioned by Curry and Schmidt (2007). Earthworms can digest microorganisms (Brown, 1995; Chapuis-Lardy *et al.*, 2010) thereby decreasing microbial biomass, especially that of fungi (Shan *et al.*, 2013). They may also select or stimulate soil microbes (Khomyakov *et al*., 2007; Nechitaylo et al., 2010) which help them digest the soil organic matter, since the earthworm gut often lacks the sufficient enzymes to do so (Lattaud *et al.*, 1997, 1998; Fujii *et al.*, 2012). This process may enrich the soil in certain bacterial taxa, for example in bacteria able to decompose the organic matter that earthworms feed on or in denitrifying bacteria able to survive in the reduced oxygen conditions of the earthworm gut (Drake and Horn, 2007;Hong *et al.*, 2011).

The effect of earthworms on soil microbial communities is critical as they are one of the most important fauna group in soils, in terms of number and biomass (Blouin et al., 2013). Besides, earthworms can have a very high rate of substrate or soil ingestion. Epigeic earthworms can ingest 3–50 mg (dry matter) of dung or any other kind of litter per gram of earthworm per day and the geophagias worms 200–6,700 mg (dry matter) of soil per gram of earthworm per day (Curry and Schmidt, 2007). In this section, we will synthesize the available information regarding how earthworms influence the abundance or activity of soil microorganisms, depending on their functional groups.

Earthworms create macro pores, which positively affect water infiltration and root growth. Their castings improve microbial growth, nutrient content and soil structure. Earthworm casts contain nitrate, phosphorous, magnesium, potassium and calcium.

Earthworms use a lot of water, since they produce 60 percent of their body weight in urine every day. Their urine is nitrogen-rich and provides an excellent fertilizer. Field worms easily produce about 50 lbs. of nitrogen/acre.

The organic material bound to earthworms is about one ton/acre. This is released gradually as the worms die in the dry summer, providing a great nutrient reservoir for our plants.

The life of an earthworm is hard. Their bodies are about 70 percent protein, rich food for many predators. Their major enemies are insect-eating birds, like robins. If you watch a robin hunt, it pauses, cocks its head and then hops.

However, tilling the soil does reduce the earthworm population; not so much from killing them, but because tilling aerates the soil.

Aeration in turn reduces the organic matter that the earthworm uses as food. Adding manure, green manure or compost will help provide food to earthworms and replenish what is lost from tilling.

Soil type can also effect earthworm populations. Clay to loamy soils have less temperature and moisture change and a larger food source than sandier soils; and therefore have higher earthworm populations.

The population of adult earthworms is highest in the spring, and decreases in the dry summer months. In the hot dry months of summer, you often don’t find many earthworms. In the cooler, wetter fall there is an increase in young worms. To start the spring with a high number of earthworms, it’s important to protect the young and the eggs over winter.

Earthworms can freeze solid and still live if the freeze is slow and they don’t thaw out and refreeze often. Any form of ground cover, cover crops or residue allows more earthworms to survive the winter. Fields that are plowed and left bare are almost devoid of earthworms in the spring. Luckily, earthworms have a high reproduction rate.

II.1: Historic : In the 19thcentury earthworms were considered a soil pest. Even though this view has changed, earthworms receive little attention in agricultural practice. Very few farmers only actively promote them. Increasingly heavy machines, intensive tillage and intensive use of pesticides have in many places eliminated earth- worms in fields. In contrast to this scenario, in the healthy soil of one hectare of grassland one to three million earthworms can be found.

Number and diversity of earthworms in a soil are considered an important criterion of soil fertility, because earthworms contribute in many ways to healthy and biologically active soils and better adaptation of farming systems to climate change, thus providing key soil functions that favor many positive ecosystem services. Due to their numerous services that increase sustainability of agroecosystems, earthworms should receive more attention in sustainable farming systems.

II.1. The Earthworms : An earthworm is a segmented worm; a terrestrial invertebrate belonging to the phylum Annelida. They are the common inhabitants of moist soil and feed on organic matter.

Earthworms are commonly called as farmer's friend. This is because the worm casting (faecal deposit) increases the fertility and burrowing helps in proper aeration of the soil. (Bazri*et al.*, 2010; Bazri*et al.*, 2013 (a); Bouché, 1972).

Earthworms are hermaphrodites and develop slowly, with the exception of the leaf litter dwellers. Only one generation with a maximum of 8 to 12 cocoons (eggs) is produced per year. Earthworms live 2 to 8 years, depending on the species. Sexually mature worms can be identified by the "genital belt" (clitellum) encircling the body.

II.2. Morphology : Earthworms have a tube-like arrangement or cylindrical shaped and reddish-brown segmented body. The body is divided into small segments. The dorsal side is characterized by a dark line of blood vessel sand ventral side is characterized by the genital openings. The mouth and the prostomium (an organ helps in burrowing) distinguish the anterior end.(Lee, 1985)

The segments 14-16 of a matured earthworm consist of a glandular tissue called clitellum which helps us to distinguish the mouth and tail ends. The body is divided into three segments with respect to clitellum- preclitellar, clitellar and post clitellar. (Bouché, 1972).

Earthworms are hermaphrodites i.e., they carry both male and female sex organs. Segments 5-9 accommodate four pairs of spermathecal apertures. The female genital pore is situated at the 14^{th}segment and a pair of male genital pores is situated at the 18^{th}segment. The body consists of S-shaped setae, which help in locomotion in the earthworm. Setae are present in each segment except in the first, last and clitellum segments (Bachelier,1978).

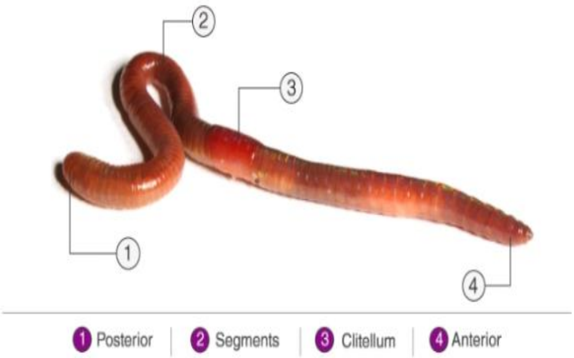

Figure 1: The diagram given represents the morphological features of an earthworm. (Source :https://byjus.com/biology/earthworm-morphology-anatomy)/)

II.3. External morphological features

Size-A fully grown, mature worm measures bout 3-5 mm in width and 150 mm in length.

Shape- Pheretimaposthuma is long, elongated, cylindrical and narrow in shape. Its body shape is well suited for burrowing habit. It is bisymmetric animal. Its anterior end is slightly pointed whereas the posterior end is blunt. A little behind the anterior end it is thickest.

Color- The dorsal surface of the body is dark brown in color due to the presence of the pigment called porphyrin. This pigment protects the animal from harmful UV rays. The dorsal surface also carries a dark colored median line which is due to the presence of dorsal blood vessel which is seen through the integument.

Body segmentation- The body of Pheretimaposthuma is soft and naked. It is divided prostomium, trunk and pygidium. Prostomium is fleshy lobe which overhangs mouth, trunk has 100-120 similar segments called as metameres or somite's and pygidium bears anus. The segments are separated externally by intersegmental grooves and internally by corresponding intersegmental septa. The external segmentation corresponds with the internal segmentation (Bouché, 1978).

II.4:External openings

- Mouth is situated at the anterior side of the first segment. It is surrounded by peristomium and overhung by prostomium (Lee and Foster, 1991).

- Along the mid dorsal line, in the intersegmental grooves a series of minute openings called dorsal pores are present. Coelomic fluid flows out through these pores and keeps the skin slippery and moist. The first pore lies in the groove between segments 12-13.

- **Anus** is the terminal opening present in the posterior terminus of pygidium.

II.4. Anatomy : Externally, a thin non-cellular cuticle covers the body wall of the earthworm. Underneath this cuticle, a layer of the epidermis, followed by two muscle layers and coelomic epithelium (inner layer) is sheathed. The epithelium consists of a single layer of glandular columnar epithelium (Bachelier, 1978).

II.5. Digestive System : The alimentary canal is a long tube running from first to the last segment of the body. The food of earthworms is the leaves and decaying organic matters which are mixed with soil.According to the diet, the parts of the alimentary canal and their secretion differ from other organisms. The alimentary canal begins at the mouth (buccal or oral cavity) (1-3 segments), passes through the pharynx, esophagus (5-7 segments), muscular gizzards (8-9 segments), stomach (9-14 segments), intestines, and finally ends at the anus. The food particles get digested gradually as they travel through various compartments of the alimentary canal

II.6. Excretory System : Nephridium is coiled tubules that regulate the volume and composition of the body fluids and thus, act as the excretory organ in earthworms. Nephridia are arranged in three segments- septal (15-last segments), integumentary (3-last segments) and pharyngeal nephridia (4-6 segments). A funnel that is connected to nephridia delivers wastes and excess fluid and is excreted out via the digestive tube.(Clive A. Edward. 1996)

II.6.1.Sensory System : Although earthworms lack eyes they have specialized receptor cells to recognize the changes around them. Specialized sensory organs and chemoreceptors help them to respond to stimuli perfectly. The sensory system of the earthworms is present in the anterior portion of the body.

II.7. Reproductive System : Earthworms are bisexual. Hence, each individual carries both male and female reproductive systems in them.

The male reproductive system consists of two pairs of testes (10-11 segments), vasa deferential (till 18thsegment), and two pairs of accessory glands (17thand 19thsegments). The prostate and spermatic ducts open by a pair of male genital pores (18thsegment). The spermatozoa are stored in the four pairs of spermathecae (6-9 segments).

The female reproductive system consists of one pair of ovaries and oviduct. Ovaries open into an ovarian funnel running below the ovaries and join the oviduct and open at female genital pore (14thsegment).

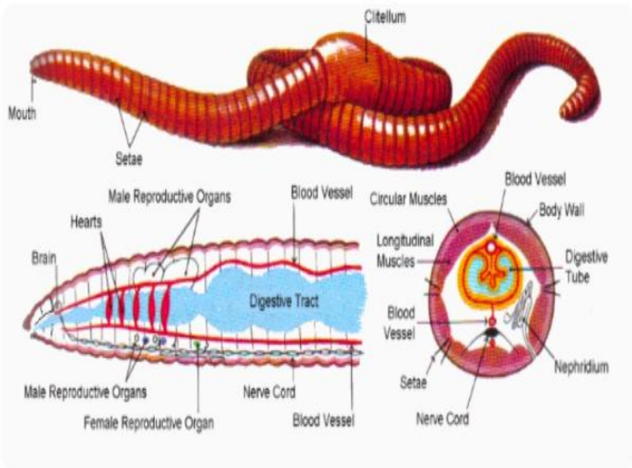

Figure.2. Reproductive stm (Source :https://www.toppr.com/content/concept/reproductive-system-of-earthworm-200667/)

II.8. Life cycle. After earthworms mate, their fertilized eggs are held in a protective cocoon. The baby worms (hatching) emerge and burrow into the soil, where they grow into juvenile then mature worms.

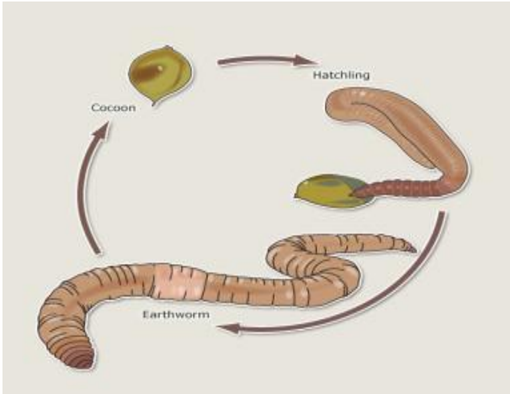

figure 3: life cycle. (source https://teara.govt.nz/en/diagram/15491/earthworm-life-cycle)

II.9. Earthworm ecology : Earthworms can be divided into **four groups,** called **ecotypes**, each of which describes a different ecological grouping based on its behavior.

II.9.1. Compost earthworms : As their name would suggest, these are most likely to be found in compost, or areas very rich in rotting vegetation. They prefer warm and moist environments with a ready supply of fresh compost material. They can very rapidly consume this material and also reproduce very quickly. Compost earthworms tend to be bright red in color and stripy- some people call the stripy species 'tiger worms'. Compost worms are often used to help dispose of waste as they can also remove contaminants from soil.

Compost earthworm species include *Eiseniafetida* and *Dendrobaenaveneta*

Figure 4: *Dendrobaenaveneta*, a compost earthworm (Source https://www.fwi.co.uk/arable/3-ways-measure-earthworm-numbers

Figure 5 : *Eiseniafetida* a compost earthworm (Source https://www.fwi.co.uk/arable/3-ways-measure-earthworm-numbers

II.9.2. Epigeic earthworms : Epigeic earthworms live on the surface of the soil in leaf litter. These species tend not to make burrows but live in and feed on the leaf litter. Epigeic earthworms are also often bright red or reddy-brown, but they are not stripy. Epigeic earthworm speicesinclude *Dendrobaenaoctaedra*, *Dendrobaenaattemsi*, *Dendrodrilusrubidus*, *Eiseniellatetraedra*…

II.9.3. Endogeic earthworms : Endogeic earthworms live in and feed on the soil. They make horizontal burrows through the soil to move around and to feed and they will reuse these burrows to a certain extent. Endogeic earthworms are often pale colors, grey, pale pink, green or blue. Some can burrow very deeply in the soil. Endogeic earthworm species include *Allolobophora Sp.*

II.9.4. Anecic earthworms : Anecic earthworms make permanent vertical burrows in soil. They feed on leaves on the soil surface that they drag into their burrows, as Anecic A*porrectodearosea, Octodriluscomplanatus and Aporrectodea trapezoids* the later *can be considered as Anecic or Endogeic*(Bazri*et al.*, 2013 a,b).

II.10. Classification of earthworms

Scientific classification	
Kingdom	Animalia
Phylum	Annelida
Class	Clitellata
Order	Opisthopora

Figure 6:classification of earthworms

In eastern Algeria, Bazri et al., (2013) a and b, identified 18 earthworm species represented by 3 families: Lumbricidae (with 8 genera; Aporrectodea, Allolobophora, Octodrilus, Eisenia, Dendrobaena, Eiseniella, Proctodrilus and Octolasion) , Megascolecidae (only one genus Microscolex) and Hormogastridae (with the genus Hormogaster). The percentage of combined dominance (PDC) reveals that the most frequent species are Ap. Trapezoides (PDC = 50.37%), Allolobophoramolleri (15.27%), Aporrectodearosea (14.75%) and OctodrilusComplanatus (3.96%).

Abstract: Plastic pollution in the environment is currently receiving worldwide attention. Improper dumping of disused or abandoned plastic wastes leads to contamination of the environment. In particular, the disposal of municipal wastewater effluent, sewage sludge landfill, and plastic mulch from agricultural activities is a serious issue and of major concern regarding soil pollution. Compared to plastic pollution in the marine and freshwater ecosystems, that in the soil ecosystem has been relatively neglected. In this study, we discussed plastic pollution in the soil environment and investigated research on the effects of plastic wastes, especially microplastics, on the soil ecosystem. We found that earthworms have been predominantly used as the test species in investigating the effects of soil plastic pollution on organisms. Therefore, further research investigating the effects of plastic on other species models (invertebrates, plants, microorganisms, and insects) are required to understand the effects of plastic pollution on the overall soil ecosystem. In addition, we suggest other perspectives for future studies on plastic pollution and soil ecotoxicity of plastics wastes, providing a direction for such research.

III.1. Plastic pollution in the soil environment: While plastic has many valuable uses, we have become addicted to single-use or disposable plastic with severe environmental consequences. Around the world, one million plastic drinking bottles are purchased every minute, while up to 5 trillion single-use plastic bags are used worldwide every year. In total, half of all plastic produced is designed to be used only once and then thrown away.

Only 9% of all plastic waste ever produced has been recycled. About 12% has been incinerated, while the rest — 79% — has accumulated in landfills, dumps or the natural environment. Cigarette butts — whose filters contain tiny plastic fibrous were the most common type of plastic waste found in the environment in a recent global survey. Drink bottles, bottle caps, food wrappers, grocery bags, drink lids, straws and stirrers were the next most common items. Many of us use these products every day, without even thinking about where they might end.

The same properties that make plastics so useful (their durability and resistance to degradation) also make them nearly impossible for nature to completely break down. Most plastic items never fully disappear; they just get smaller and smaller. Many of these tiny plastic particles are swallowed by farm animals or fish who mistake them for food, and thus can find their way onto our dinner plates. They've also been found in a majority of the world's tap water. By clogging sewers and providing breeding grounds for mosquitoes and pests, plastic waste (especially plastic bags) can increase the transmission of vector-borne diseases like malaria.

Many organisms, including humans, depend on the soil for their survival, and therefore, soil pollution is critical factor, even affect affecting food safety for humans (Akhtar, 2015) as industrial development has accelerated and the manufacture and disposal of plastics have increased, concerns on plastic pollution are growing.

Recently after Rilig (2012) pointed out the problem of microplastic (MP) pollution in soil and terrestrial ecosystems, people were encouraged to focus this problem again. Researchers have paid attention to plastic wastes in the soil media and warned about the dangers of small plastics in the soil and terrestrial ecosystems (Liu *et al.,* 2014). Many researchers also pointed out the potential effects of widespread plastic contamination in the soil environment, emphasizing on the adverse effects of plastics and MPs in soils (Riling, 2012; *et al.,* 2014)

Nevertheless studies on the distribution fate and transformation of plastic wastes in soil environment are still lacking. Several studies have estimated the concentrations of MPs in dry sludge dumped in landfills after wastewater treatment (Nizzetto *et al.,* 2016) .The development of techniques for the extraction and analysis of small plastics such as MPs from soil media have only begun recently (Fuller and Gautam, 2016)

III.2. Plastics waste in soil environment: Although numerous studies reported the occurrence of microplastics in aquatic ecosystems, microplastics in terrestrial ecosystems have received relatively little attention. In the terrestrial ecosystem, soil is an interface among lithosphere, hydrosphere, atmosphere and biosphere. Once entry in the soil, microplastics may persist, accumulate, and eventually reach high levels that can affect organisms and biodiversity. Additionally, microplastics can also act as a vector for the transfer of pollutants, either plastic additives or other toxicants absorbed from soil matrices, to soil biota and thus pose a hazard.

For example, (Zhang *et al.*)found the high concentration level of organophosphorus esters and phthalic acid esters in microplastics collected from 28 coastal beach soils in north China. In fact, the terrestrial environments are the critical source of plastic rubbish in water column. Soils were theoretically speculated to be the major storages for microplastics, which is bigger store more than oceanic basins. Another study points out that the total value of microplastic contaminations on land might be 4 to 23 old larger than that in the ocean. In recent years, researchers are paying more attention on microplastic pollution in the soils.

In farmland, microplastics pollution mostly originates from the application of plastic in agricultural practice. Theoretically estimation of microplastics may be the largest originate from the application of plastic mulching and sewage sludge.

The main source of microplastics is inputs from agricultural practices including the utilization of sewage treatment plant sludge and plastic mulching. Despite a high removal rate of microplastics in sewage treatment systems, the most of microplastics still remain in sludge. For example, micro- and macro-plastics were widely detected in sludge with the concentrations ranged from 1500 to 24,000 items kg . Mahon et al. found up to 15,800 particles kg of microplastics in sludge, and showed that some approaches including lime stabilization, anaerobic digestion, and thermal drying are insufficient to remove microplastics from sludge.

Figure 7: Sources and fate of soil microplastics in terrestrial environments. n (a), the potential uptake by plants (b), uptake and transport by soil animals (c), and trophic transfer in terrestrial food chain (d). (source.https://www.researchgate.net/figure/Sources-and-fate-of-soil-microplastics-in-terrestrial-environments-Red-fibers-and_fig3_328269150)

Microplastics in the topsoil might be incorporated into deeper topsoil by tillage, and even into the plough layer along large cracks or by the turbation of soil biota. The typical physical process for microplastics in soils is leaching. Leaching is an important process driving contaminants with certain properties to groundwater. Micro or nano plastics have not yet been analyzed in groundwater samples, but transport through bio pores has been identified as a possible mechanism for groundwater contaminations.

III.3. Classification and migration of microplastics in soil : Microplastics can be divided into primary and secondary microplastics based on the original manufactured particle size. Primary microplastics mainly include plastic microbeads and nanoparticles directly used in a very in a variety of industrial detergents and cosmetics. In addition, they may enter soil from atmospheric deposition (Allen *et al.*.2019). Secondary microplastics originate from large plastic products that have broken down in situ (e.g. plastic film residues household garbage).This may occur at the surface in response to solar UV irradiation or within the soil profile due to physical abrasion (abiotic) and biological attack (Andrady, 2011; Cole *et al.*, 2011)

The types of microplastics can be divided into fibers, fragments, thin films, and particles depending on plastic shape. Depending on the source, fibers often represent the predominant from if they enter soil from biosolids or irrigation waters derived from municipal wastewater (Jabeen *et al.,* 2017). In contrast the breakdown of plastic mulch films leads to a predominance of heterogeneous fragments while plastic coated fertilizers leads to a predominance of thin films.

Microplastics are further divided into small microplastics (<1 mm) medium (1–3mm) and large microplastics (3–5mm) according to their particle size (Andrady, 2011).

Nanoplastics are typically referred to as 1–1000um in size while picoplastics are < 1um in size.

The reason that categorizing microplastics size is important is that it affects their potential for transporting in soil and their potential to be taken up by cells.

The main source of macro and microplastics entering agricultural soils includes plastic mulch films municipal waste (e.g. municipal solid waste compost) biosolids (sewage sludge and anaerobic digestate) plastic coated fertilizers and atmospheric deposit (Adrés Rodríguez Seijo, 2018).

III.4. Impact of microplastics on soil structure: A loss of soil structure commonly occurs when large amounts of macro plastics are present in the soil. This is deleterious as it reduced the infiltration of rainwater and irrigation water negatively affects the soil's water holding capacity and may induce anoxia (Liu *et al.,* 2014).

It has also been reported that residual plastic mulch film damages the structure of soil aggregates and residual and reduces soil aeration and water permeability, thereby reducing root growth and overall plant productivity (Jiang *et al.*, 2017; Zeng *et al.*, 2018).

In contrast to macro plastics there are relatively few reports on the relationship between microplastics and the soil structure and aggregates (Zang and Liu, 2018) and no studies have clearly shown the influence of microplastics on soil structure. Further studies are required to determine where microplastics are physically located in the soil matrix and how this affects their fate behavior.

III.5. Effect of microplastics on soil physical and chemical properties: Several studies reported that microplastics have negative impact on soil organic carbon (C) and nitrogen (N) cycling soil microbial activity and nutrient transfer (Cao *et al.*, 2017) showed that addition of microplastics can stimulate soil enzymes activities and the accumulation of soluble nutrients in soil.

In addition plastic mulch residues inadvertently contribute to increasing the size of the stable soil organic C pool.

At a typical plastic contamination level of 5 – 25 kg /ha this equates to a C addition rate of ca. 4 - 20 kg/ha it should be noted that this is low in comparison to rates of organic C loss from most intensive agricultural systems and therefore should not be viewed in positive light. Hodson *et al.* (2017) found that microplastics can improve the bioavailability of zinc as medium but little is known about the potential risk to earthworms. In addition the underpinning mechanisms responsible for this increase in micronutrient bioavailability remain unknown.

In agriculture measurements of soil physical and chemical quality indicators have been used as indicators to evaluate the advantages and disadvantages of agricultural plastics. In some cases plastic mulch films improve specific soil quality indicators whilst in others a decline is apparent.

III.6. Impact of microplastics on soil organisms: Based on the negative impacts of plastic pollution on marine organisms there is increasing focus on the danger of microplastics to soil organisms (Cao *et al.*, 2017; Chae and An, 2018). Mesofauna (e.g. earthworms, mites , collembola) are known to be vital in maintaining soil quality , however , intensive agricultural systems typically lead to a loss in mesofauna abundance (George *et al.*, 2017). Consequently a further loss of these keystone organisms by plastics could represent a major threat to long-term agroecosystem functioning. Huerta Lwanga *et al.* (2016) studied the survival and fitness of earthworms exposed to microplastics in litter at different concentrations.

After incubation for 60d, the earthworms in different higher concentration microplastic conditions in the litter had a higher mortality rate and significantly lower growth rate compared with those of the lowest concentration.

The research also confirmed the concentration (transport and size) selection mechanisms of microplastics in terrestrial ecosystems. It should be noted however that concentrations of plastic used in those studies were 1000 folds higher than seen in most plastic contaminated agricultural soils. Cao *et al*, (2017) stated that a low soil microplastic concentration (<0.5%) has little impact on earthworms but when the microplastics concentration rose to 1% and 2% it significantly inhibited the growth of earthworms and increased their mortality.

The adverse effects of microplastics on soil organisms may be mainly caused by the significant accumulation of microplastics in the gut and stomach of organisms which can be their immune systems and affect their feeding behavior and development.

Agricultural plants are known to take up a range of nanoparticle and consequently it is likely that microplastics may enter the food chain through this route (Jassby *et al.,* 2019) has reported that polystyrene microplastics (0.2nm) can be absorbed and enriched in the root of raw vegetables and migrate from root to shoots .

III.7. Influence of microplastics on the groundwater environment: Some studies have indicated that microplastics in marine environments originated from terrestrial ecosystem (Horton et *al.,* 2017; Luo *et al.,* 2018; Wagner *et al.,* 2014). Soil microplastics can be transported from land to the underground environment via long distance movement, such as animal disturbance surface runoff and water infiltration (Blasing and Amelung 2018; Brodhagen *et al.,* 2015; Hurley and Nizzeto, 2018).

Thereby affecting the underground aquatic environment and even disturbing the marine ecosystem .There are scarce reports on the effects of microplastics on the groundwater environment (Chae and An, 2018) even though studies on the marine environment widely exist (McCormick *et al.,* 204; Zettler *et al.,* 2013).

IV. 1. Soil sampling: samples are collected from a soil profile representative to the soil of the surrounding area that used for agriculture here at Constantine.

The samples were taken from wheat-producing soil in the Constantine region, which is often exposed to pollution from plastic bags due to wind activity.

The soil is taken from the ecology laboratory of the Faculty of Natural and Life Sciences to prepare the incubation medium for earthworms in plastic boxes.

Photo 01: Soil sample.

IV.2. Materials and Equipment required

1. Spade or auger (screw or tube or post hole type)
2. Trowel.
3. Core sampler
4. Sampling bags
5. Plastic tray or bucket
6. plastic tubes (to store and preserve earthworms)
7. Labels and pen/pencil (to label your samples and ensure they don't get mixed up)
8. Gloves (to keep your hands clean)

IV.3. Earthworm Collecting Procedures: The species of earthworms were collected from different part of Constantine including that from Hostel of Zoughi, they were taken to laboratory for experiments and they were determined by Dr-HDR BAZRI K.E.D (Mentouri University of Constantine) supervisor of my thesis.

All earthworms collected and preserved must have a collection label with them, this contains information about where, when, and by whom the earthworm was collected.

Photo 02: Plastic waste that collected to produce microplastics.

IV.4. Preparation of microplastics: A bunch of plastic bags, bottles and other plastics wastes were collected around the sewage system in different place in Constantine. These plastic pollutants were in state that can be grinded easily and form this tiny particles of plastic we call microplastics. Microplastics, small plastic pieces <5 mm intentionally produced to be used in experiment to mix up with soil that we collected from areas that is used for agriculture so that earthworm can feed in .

IV.5. Preparation of earthworm incubation media: the experiment was launched on 25-02-2020, at the ecology laboratory where we prepared six culture media in plastic containers with a volume of 3 liters including the soil of three media is mixed with microplastics. Then, we introduced into each culture medium three adult earthworms: two individuals of the species *Aporrectodea trapezoids* and one individual of the species *Aporrectodea rosea.*

Incubation media are kept in the dark at room temperature and moist soil. A food intake based on dry vegetable matter represented by kitchen peelings, is added weekly to the six culture media to feed our earthworms.

N.B / Food intake was disrupted due to Covid19 and lockdown.

Photo 03. Polluted sample

Photo 04. Non polluted sample

IV.6. Determination of biological parameters : We have determined the following earthworm parameters:

**** Abundance:** it is measured as the number of individuals found per sample.

**** Age class:** we have individuals juvenile, sub adults or adults according to criteria based on morphologic. Generally on the size and presence of clitellum.

**** Biomass**: the total quantity or weight of organisms in a given area or volume.

IV.7: Physical-chemical analysis of the soil: We considered the analysis of several parameters which did not take place because of the Covid19 and the quarantine which lasted more than three months. As soil texture, permeability and Organic matter.

Only two parameters are measured, namely pH and electrical conductivity.

IV.7.1:pH Measurement analysis: it is the electro metric measurement of the activity of H ions presents in the soil solution. The determination of pH was carried out by the aid of electrodes pH meters in which suspension of soil were formed from mixture of soil and distilled water in the ratio 10g and 25ml respectively followed by agitation for 2 hours and rest for 24h (HUBERT , 1978)

Interpretation values of pH was summarized under the following table (BAIZE, 1989).

Table1: Interpretation scale of pH.

pH	<3.5	3.5 — 5	5 — 6.5	6.5 — 7.5	7.5 — 8.7	>8.7
Soil	Hyper acidity	Very acidic	Acid	Neutral	Basic	Very basic

From our experiments pH for Non polluted sample were 7.48, 7.39 and 7.34 the average for this samples were 7.4 which according to the scale of interpretation this is neutral soil.
On the other hand polluted samples were 7.30 , 7.36 and 7.35 the average for this samples were 7.33 which according to the scale of interpretation falls under neutral type soil.

IV.7.2 : Electrical conductivity analysis. is a measurement that correlates with soil properties that affect soil texture, cation exchange capacity (CEC), drainage conditions, organic matter level, salinity, and subsoil characteristics. In our experiments table number 2 shows the value of EC which taken after the end of experiments .
Values of EC was ranged from 684 —1035µS/cm in case of EC the optimum levels for soil 110000 µS/cm — 570000 µS/cm in this experiment this samples has average of 856 µS/cm for non polluted soil and average of 1015 for polluted soil which all of the two sample are under the optimum level.

Photo 06: EC meter.

Photo 07:pH meter.

I. Determination of earthworms collected:

The earthworms collected for our experiment are expressed by two species that dominate Algerian soil, these are the *Aporrectodea trapezoids* and *Aporrectodea rosea* species.

II. Biological parameters of earthworms:

Table n° 02: Evolution of earthworm demoecology in incubation environments

	Incubation environments	*Ap. trapezoïdes*	*Ap. rosea*	Total	Presence of cocoons	Biomass
Beginning of experimentation 25/02/2020	Medium 1 (P)	2	1	3	0	0.111g
	Medium 2 (P)	2	1	3	0	1.225g
	Medium 3 (P)	2	1	3	0	0.12g
	Medium 1(NP)	2	1	3	0	4.67g
	Medium 2(NP)	2	1	3	0	4.703g
	Medium 3(NP)	2	1	3	0	2.4g
End of experimentation 8/6/2020	Medium 1 (P)	0	1	1	0	0.368g
	Medium 2 (P)	2	1	3	0	1.005g
	Medium 3 (P)	1	1	2	0	0.725g
	Medium 1(NP)	0	1	1	0	6.262g
	Medium 2(NP)	0	1	1	0	5.292g
	Medium 3(NP)	2	1	1	0	5.297g

From table 02, the results show that the number of earthworms decreased in all culture media except medium number 2(P) where the three individuals (2 *Ap. Trapezoids + 1 Ap. Rosea*) are still present at the end of the experiment, which proves that this reduction is not linked to pollution by plastic waste. It seems that the earthworms have left the incubation environment for lack of food, water stress or space; especially since we abandoned the monitoring of the experiment during the confinement period.

Also, we noted the absence of cocoons and juveniles, earthworms did not reproduce in polluted and unpolluted environments, certainly because of the conditions of the experiment which was interrupted for a certain period.

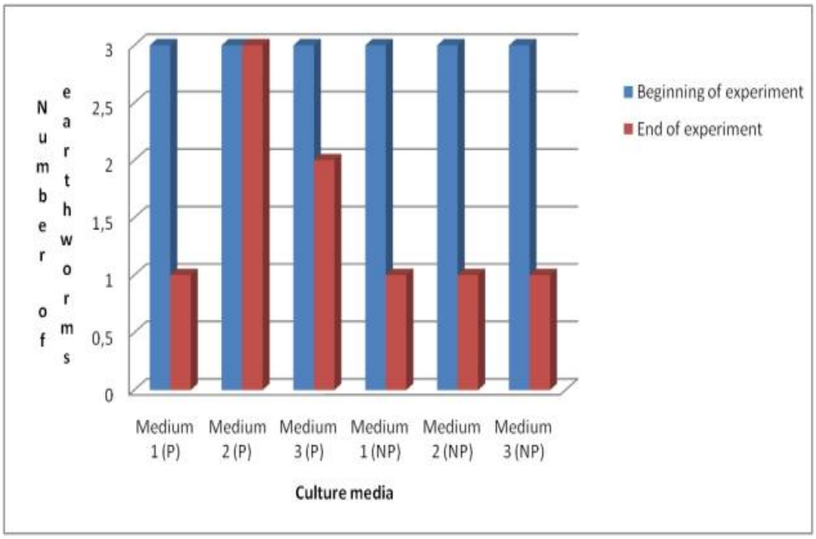

Figure.08: Evolution of the number of earthworms in incubation environments

Figure 08 shows that the biomass increases in the culture media of our experiment despite the loss and disappearance of some individuals of earthworms. But this increase is much greater in unpolluted environments. This explains why plastic pollution affects the ingestion and fattening of our earthworms. These results are consistent with the work of Cao et al, (2017), who indicates that ingestion of plastic by earthworms inhibits their growth and increases their mortality.

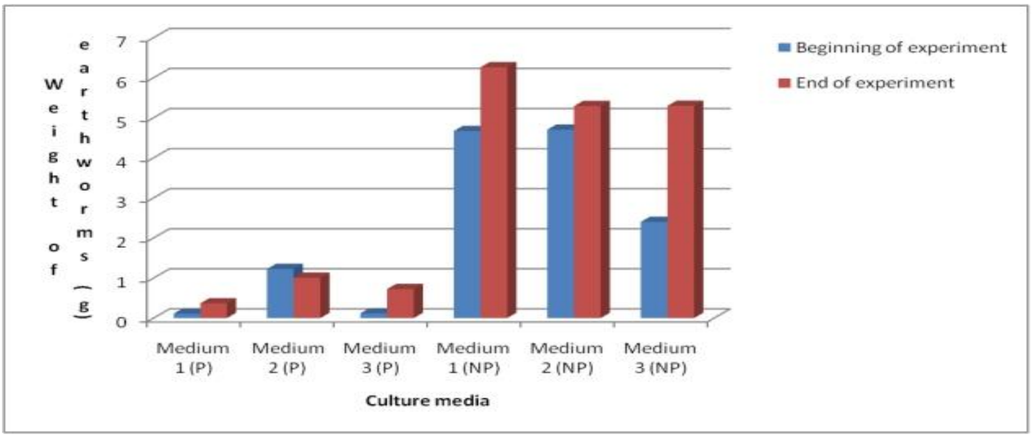

Figure.09: Evolution of earthworm biomass in incubation environmentThe differences between the incubation environments are very significant (F = 289.12; p <0.0000) (table ...).

Table 04 : Analysis of Variance (Significant effects marked at p <.05000)

	SC	Degree of freedom	MC	F	p
ord. origine	59844100	1	59844100	289.1289	0.000070
Media	36275168	1	36275168	175.2587	0.000188
Error	827923	4	206981		

The Newman-Keuls test shows two homogeneous groups a and b (Fig.….).

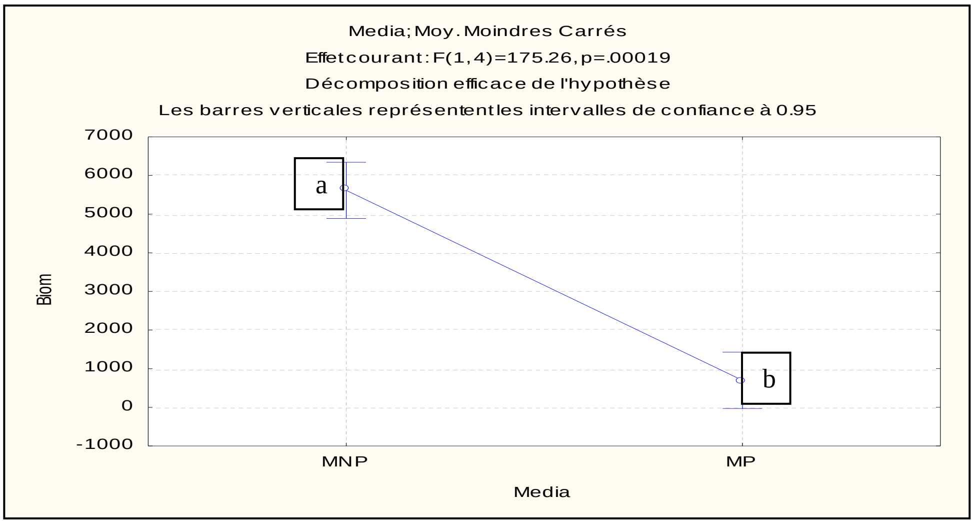

Figure .10: The homogeneous groups according to the Newman-Keuls test.

The works of Bouché, 1972; Diaz Cosín *et al.*, 2006 and Bazri, 2013; 2014, indicate that earthworms can withstand environmental stress through the diapause triggered by desiccation of the environment or a traumatic agent, such as the removal of a part of the body (Saussey, 1966). The worm does not eat any more, empties its intestine and settles in an individual spherical cubicle in depth where it is rolled up by excreting mucus in order to free itself from the external conditions. The exit from diapause can be controlled by neurosecretions from the worm; this is called compulsory diapause. can also be controlled by environmental conditions; it is an optional diapause (Saussey, 1966). This form of lethargy only modifies the water content of the worm very slightly, but can cause the animal to lose weight. Hibernation, caused by a low soil temperature, ends as soon as the soil warms up a few degrees.

III. The physical-chemical parameters of the soil:

Tableau n° 05: Evolution of pH and EC in incubation environments

End of experimentation	Date	pH	EC (µS/cm)
Medium 1(NP)	**8/6 /2020**	**7.48**	**985**
Medium 2 (NP)	**8/6 /2020**	**7.39**	**684**
Medium 3 (NP)	**8/6 /2020**	**7.34**	**901**
Medium 1(P)	**8/6 /2020**	**7.30**	**1034**
Medium 2(P)	**8/6 /2020**	**7.36**	**1035**
Medium 3(P)	**8/6 /2020**	**7.35**	**976**

The pH at the end of the experiment varies between 7.30 (Medium1 P) and 7.48 (Medium1 NP). The values are higher in the polluted environments. It seems that plastic waste increases the pH values (Fig. 11).

The graph below reveal that values of pH varies from 7.30 up to 7.48 was negligible to have any serious distress for earthworms for both samples polluted and non polluted. This means that microplastic pollution has impact on acidity and basicity of the soil. The average was 7.36 for sample contain microplastic while 7.40 for those non polluted samples . Even though in our experiments there is no high mortality rate for the samples which are polluted but still shows that microplastic has effect in soil pH.

It should be noted that earthworms prefer soils with a neutral to slightly alkaline pH (Bazri, 2013; 2014). In our experiment, pollution by plastic waste directs the soil towards an acid pH. And so ; towards an environment not conducive to the development of earthworms.

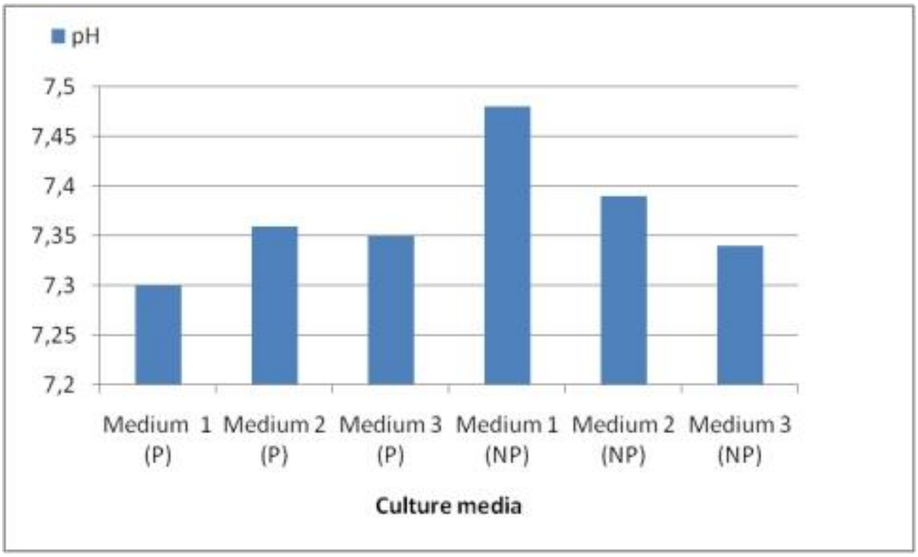

Figure.11: The pH values in the earthworm incubation environments at the end of the experiment.

According to Figure12, the EC values are high in polluted environments compared to unpolluted, it seems that reactions in the soil occur due to plastic debris which release molecules or cations in the solution of the ground ; which increase the values in polluted environments.

Values of EC was ranged from 684 —1035µS/cm in case of EC the optimum levels for soil 110000 µS/cm — 570000 µS/cm in this experiment this samples has average of 856 µS/cm for non polluted soil and average of 1015 for polluted soil.

This CE value found in this experiment is too low for non–polluted media compared to the polluted media this indicate the diversity between two medias and how microplastics has an effect on structure and properties of the soils. Changes in this properties of soil causes discomfort for fauna found within the soil which can cause death or growth stunt . As study shows that electricity conductivity of the soil it is important indicator of soil health .

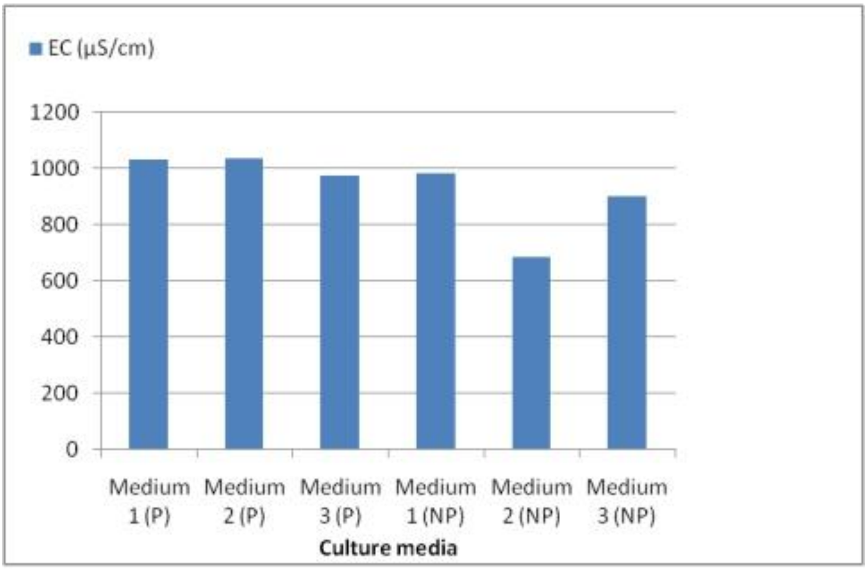

Figure.12: The values of the electrical conductivity in the earthworm incubation environments at the end of the experiment.

The analysis in principal components (PCA) with two factors and a quality of representation of 82.8%, shows that the two parameters CE and number of earthworms are positively correlated with the axis Fact1 but negatively with the axis Fact 2. However, they oppose the two pH parameters and the earthworm biomass which are negatively correlated with the Fact1 axis as well as the Fact2 axis. (Fig 13.)

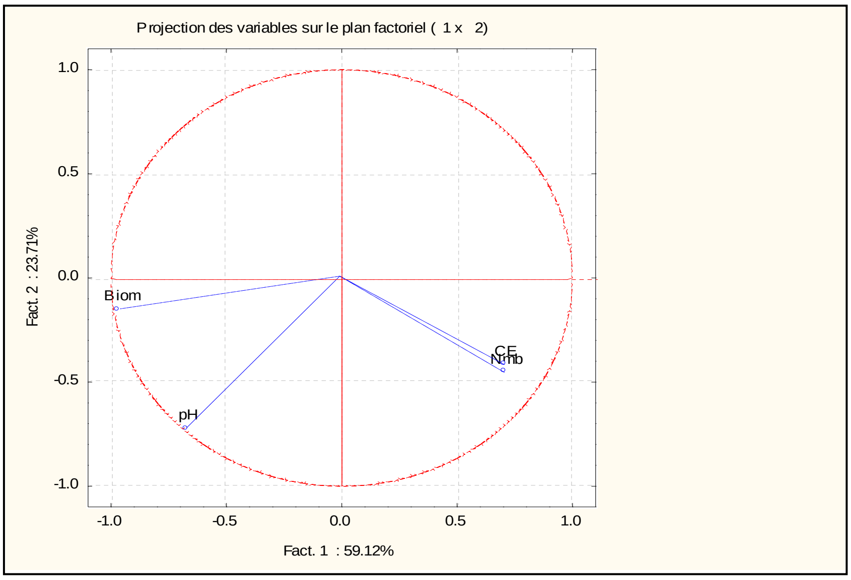

Figure.13: PCA graph according to the factors F1 x F2 at a quality of representation of 82.8%

The projection of the points confirms that the two incubation environments MP and MNP are different. The first polluted are positively correlated with the positive axis Fact1, and with the CE parameters and the number of earthworms, notably for MN2.

However, unpolluted media are negatively correlated with the Fact1 axis and well correlated with pH and biomass, particularly for MNP1. (Fig 14.).

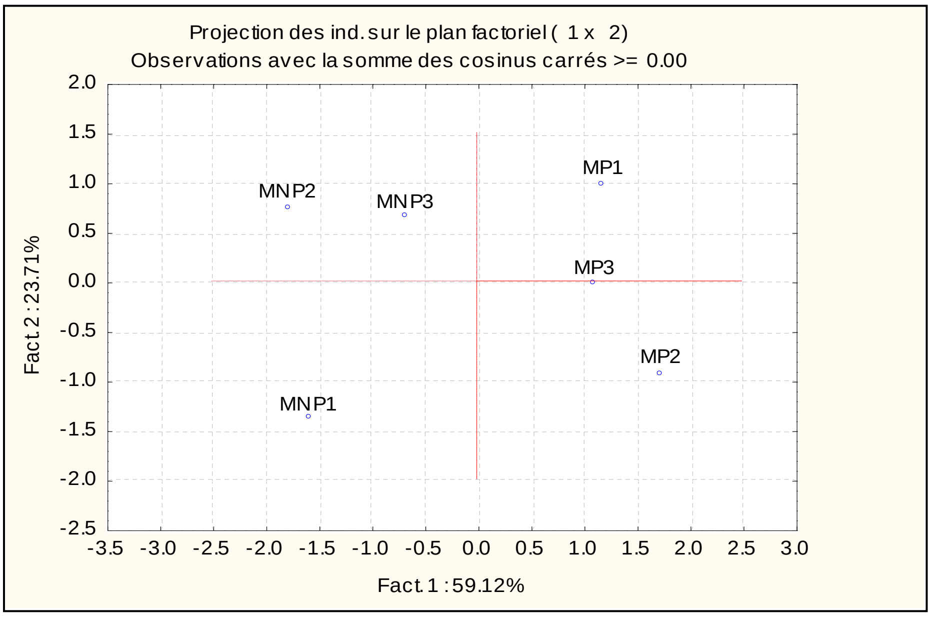

Figure.14: Graph of the projection of individuals from a PCA, according to the F1xF2 factors at a representation quality of 82.8%

These results show that earthworms cannot tolerate pollution from plastic waste; because the biomass of earthworms is very low in polluted environments but it is better in unpolluted environments. As for the result of the number of earthworms which appears high in polluted environments, this is explained by the escape of earthworms from incubation environments during the experimental period. Certainly, they left the jars for lack of food or insufficient space.

Conclusion and recommendation

Microplastic pollution considered to be one of the factors which has negative effects in soil and microorganisms (earthworms).

The activity of earthworms produces a significant effect, not just on the structure, but also on the chemical composition of the soil, since a large part of the organic matter ingested by earthworms is returned to the soil in a form easily used by plants. While they are feeding, earthworms also ingest large quantities of mineral substances (minimally so in the case of the epigeic), that are then mixed with the organic matter ingested and, after having been cemented with a little mucous protein, are expelled in piles called worm casts.

Several studies reported that microplastics have negative impact on soil organic carbon (C) and nitrogen (N) cycling soil microbial activity and nutrient transfer (Cao *et al.*, 2017) showed that addition of microplastics can stimulate soil enzymes activities and the accumulation of soluble nutrients in soil.

In our work we are going to focus on effects of microplastic on earthworms , in which the object of the experiment was to determine effects of microplastic pollution in soil of production by using microorganism (earthworms) which live in soil.

To reach our goal we did small experiment in ecology laboratory at University Constantine 01 faculty of nature and life science. Where by two samples were tested polluted sample (MP) and non-polluted (MNP).First in pollution samples 3 jars were filled with soil then microplastics while in the second non-polluted samples no microplastics were added.3 earthworms were added in each jars for both polluted and non-polluted samples .

Results shows that after 3 months few earthworms escaped the jars due to the lack of food and water during lockdown period caused by Covid19.While other earthworms which remained we did some experiments in them and we found that those which are in polluted jars had low biomass compared to those in non-polluted jars. Not only microplastics pollution had an effects on earthworms but also in soil as we observed that in those polluted sample there is an increase in soil pH.

We can conclude that polluted sample showed negative effect for the microorganism (earthworms) like loss of biomass .

For the best explanation and confirmation of the results it recommended that

- To use the smallest particle of microplastics which will be easy for earthworms to ingest during experimentation.

- To use different type of species in order to realize which species are most tolerant in microplastics pollution.

- To use earthworms of different age so that can comment about age tolerance in micropollution.

- To study other parameters like length etc.

- To pay attention during all time of experimentation

- But the most important thing is to protect the environment from plastic waste that threatens biodiversity and people through better management of plastic waste.

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Annex 1.

La matrice des données

	Media	Nmb	Biom	pH	CE
MP1	MP	1	368	7.3	1034
MP2	MP	3	1005	7.36	1035
MP3	MP	2	725	7.35	976
MNP1	MNP	1	6262	7.48	985
MNP2	MNP	1	5292	7.39	684
MNP3	MNP	1	5297	7.34	901

Annex 2. Anova à 1 facteur / Paramètre nombre des lombriciens

Tests Univariés de Significativité pour Nmb (Feuille de données1)

Paramétrisation sigma-restreinte

Décomposition efficace de l'hypothèse

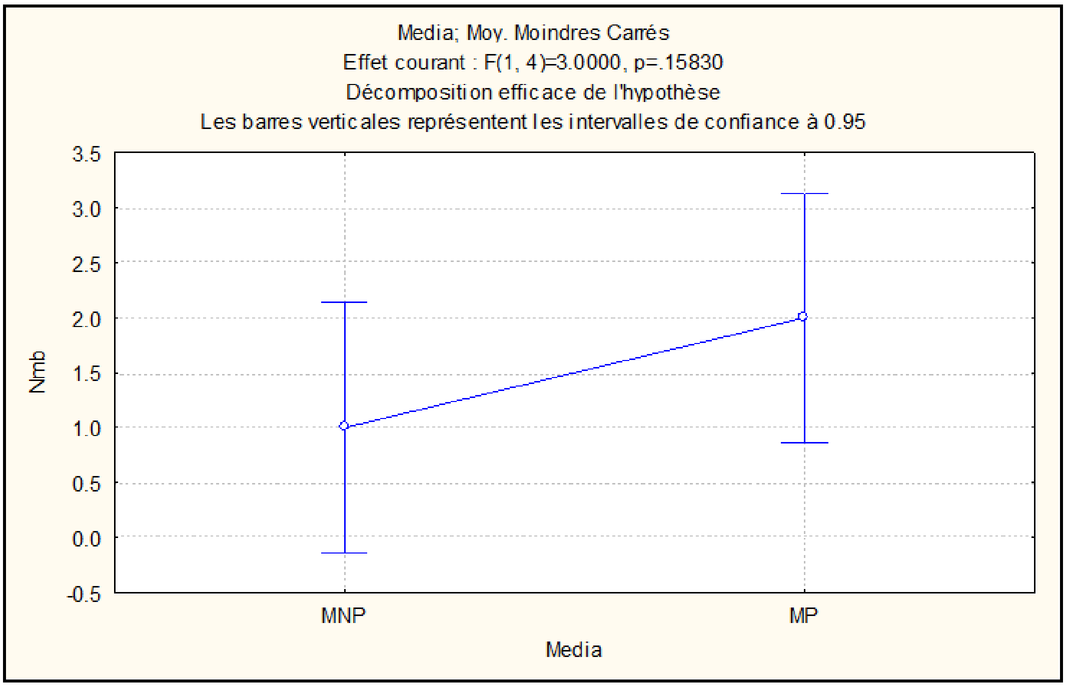
	SC	Degr. de Liberté	MC	F	p
ord. origine	13.50000	1	13.50000	27.00000	0.006533
Media	1.50000	1	1.50000	3.00000	0.158302
Erreur	2.00000	4	0.50000		

Test de Newman-Keuls ; variable Nmb (Feuille de données1)

Groupes Homogènes, alpha = .05000

Erreur : MC Inter = .50000, dl = 4.0000

	Media	Nmb Moyen.	1
1	MNP	1.000000	****
2	MP	2.000000	****



Annexe 3. Anova à 1 facteur / Paramètre biomasse des lombriciens

Tests Univariés de Significativité pour Biom (Feuille de données1)

Paramétrisation sigma-restreinte

Décomposition efficace de l'hypothèse

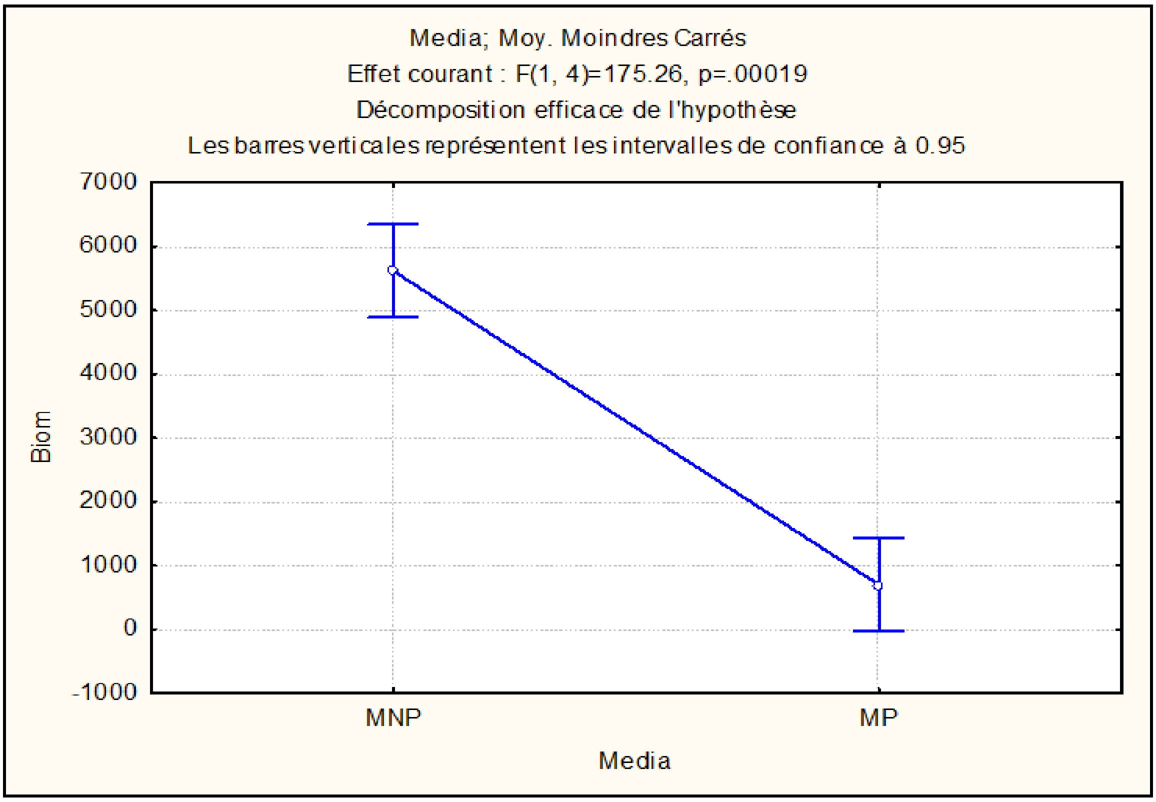
	SC	Degr. de Liberté	MC	F	p
ord. origine	59844100	1	59844100	289.1289	0.000070
Media	36275168	1	36275168	175.2587	0.000188
Erreur	827923	4	206981		

Test de Newman-Keuls ; variable Biom (Feuille de données1)

Groupes Homogènes, alpha = .05000

Erreur : MC Inter = 2070E2, dl = 4.0000

	Media	Biom Moyen.	1	2
2	MP	699.333	****	
1	MNP	5617.000		****



Annexe 4. Anova à 1 facteur / Paramètre pH des lombriciens

Tests Univariés de Significativité pour pH (Feuille de données1)

Paramétrisation sigma-restreinte

Décomposition efficace de l'hypothèse

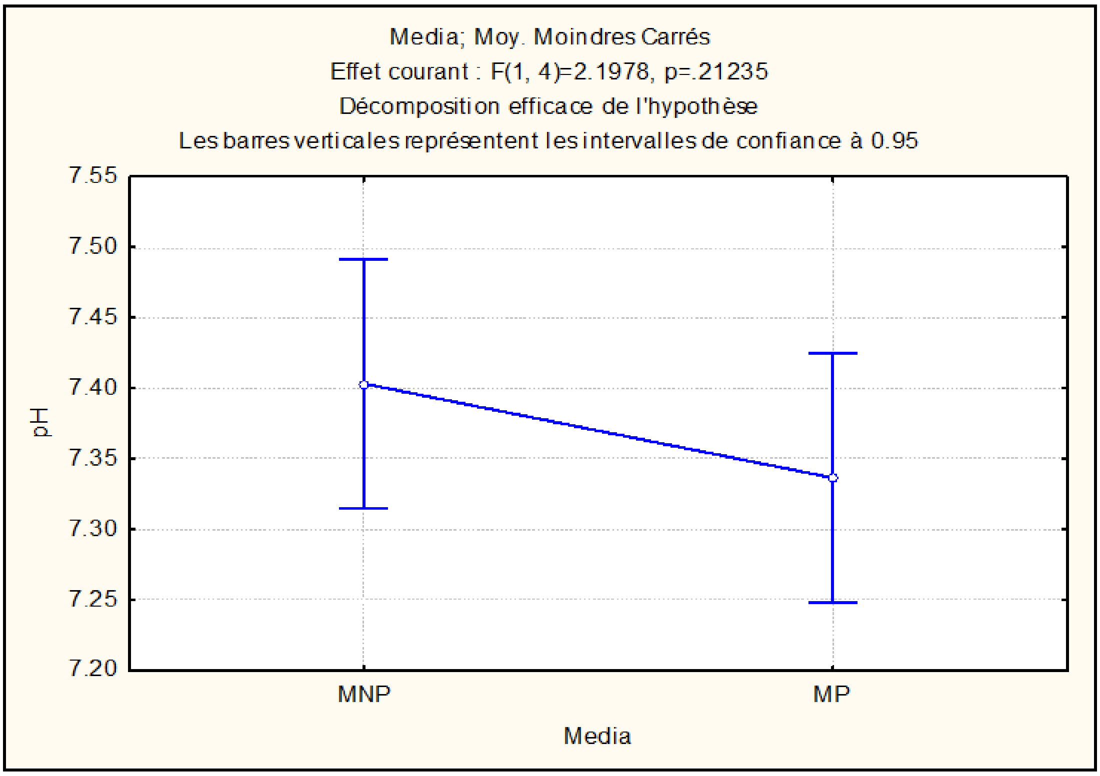
	SC	Degr. de Liberté	MC	F	p
ord. origine	325.9014	1	325.9014	107440.0	0.000000
Media	0.0067	1	0.0067	2.2	0.212347
Erreur	0.0121	4	0.0030		

Test de Newman-Keuls ; variable pH (Feuille de données1)

Groupes Homogènes, alpha = .05000

Erreur : MC Inter = .00303, dl = 4.0000

	Media	pH Moyen.	1
2	MP	7.336667	****
1	MNP	7.403333	****

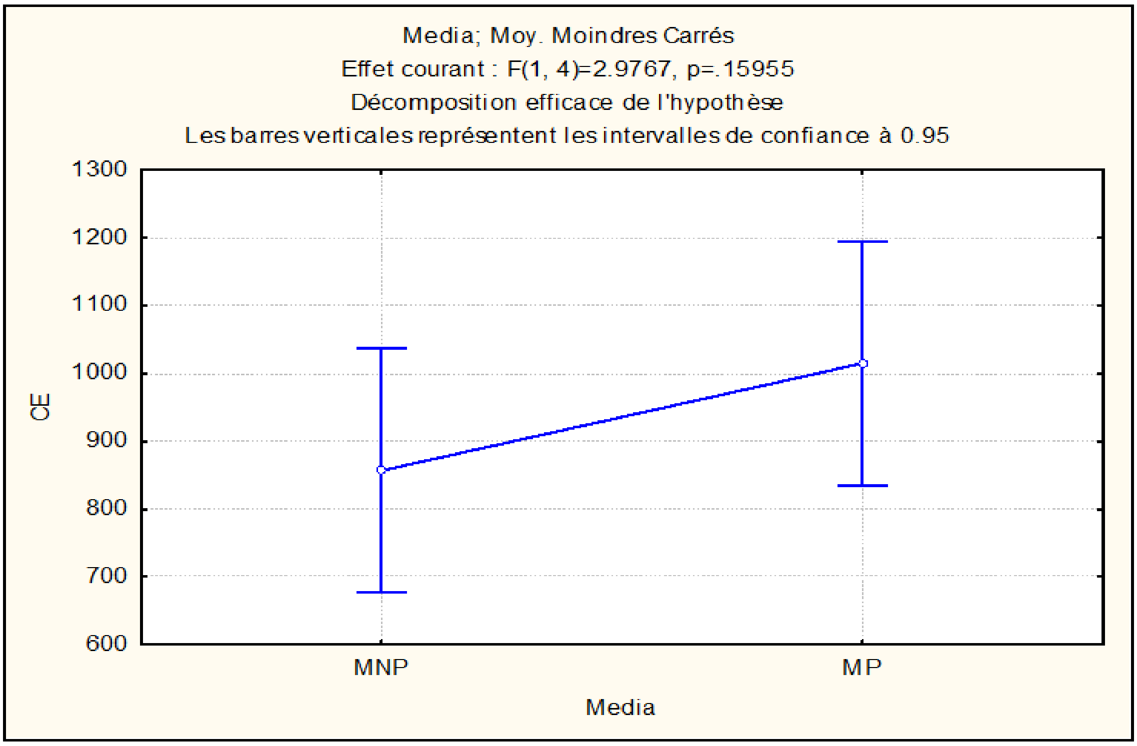


Annex 5. Anova with 1 factor / Parameter pH of earthworms
Univariate Significance Tests for CE (Datasheet1)
Sigma-restricted parameterization
Efficient decomposition of the hypothesis

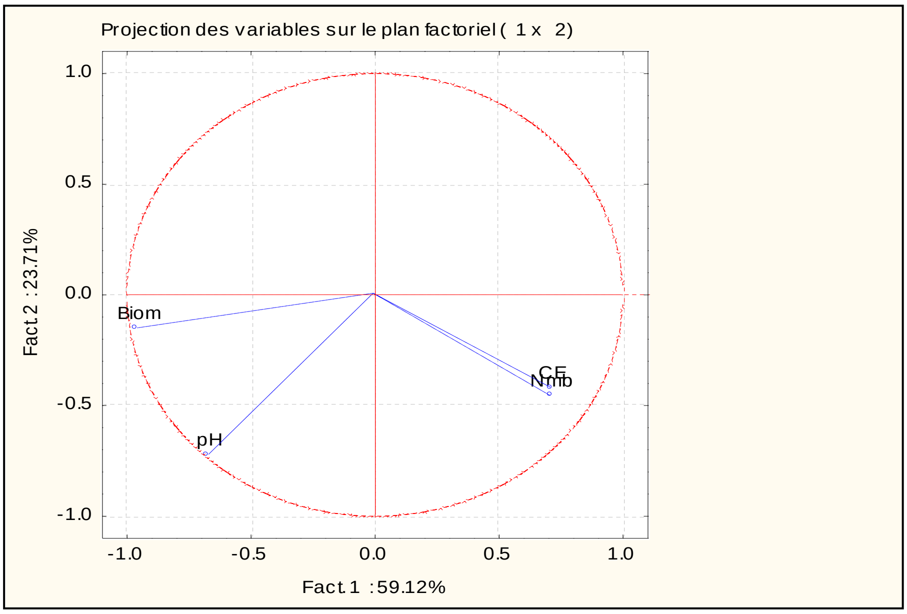
	SC	Degr. de Liberté	MC	F	p
ord. origine	5254704	1	5254704	415.9616	0.000034
Media	37604	1	37604	2.9767	0.159553
Erreur	50531	4	12633		

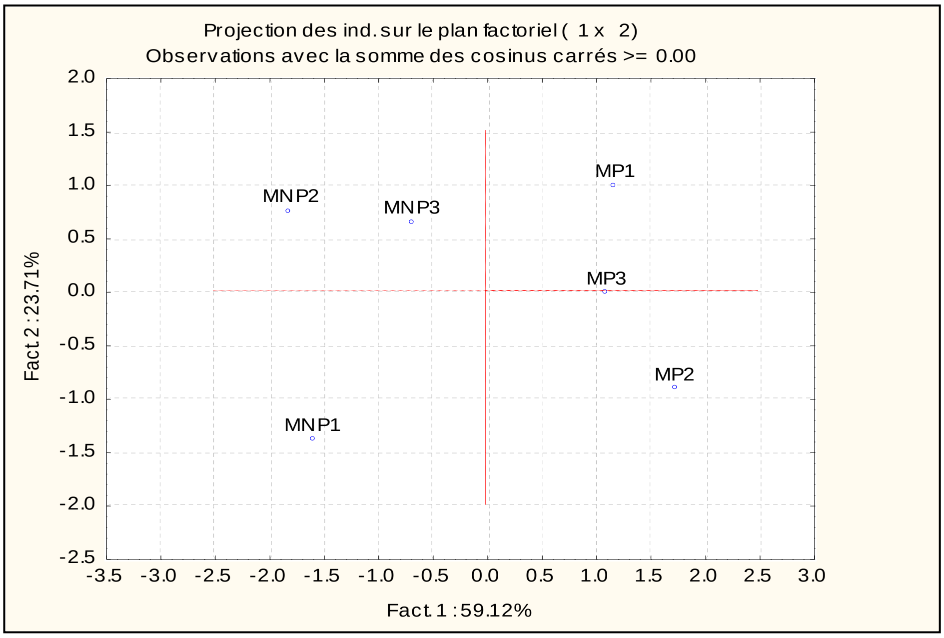
Newman-Keuls test; CE variable (Data sheet 1)
Homogeneous groups, alpha = .05000
Error: MC Inter = 12633., dl = 4.0000

	Media	CE Moyen.	1
1	MNP	856.667	****
2	MP	1015.000	****



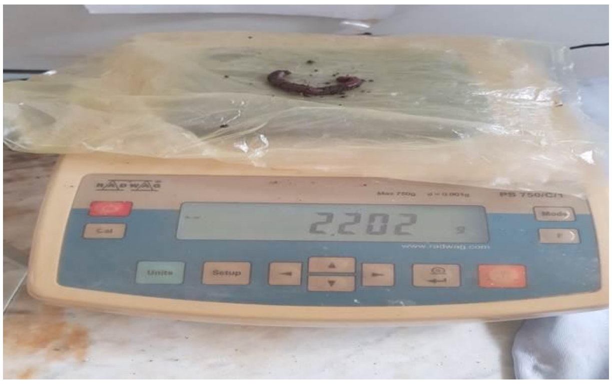
Annex 6. Analysis of main correspondences

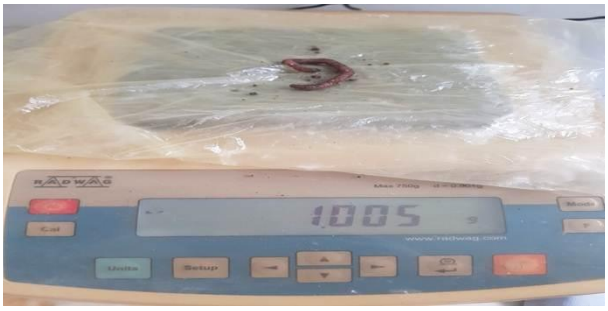


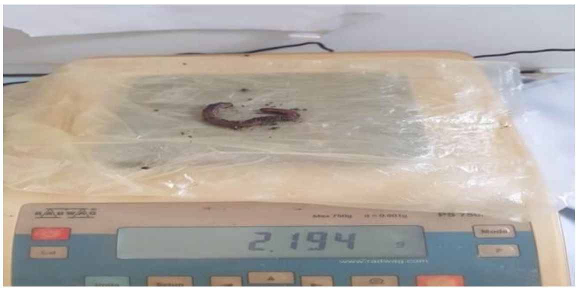


Annex 7. Sample of polluted and non–polluted

Annex 8. Biomass of earthworms from polluted sample.







Annex 9: Biomass of earthworms from non–polluted sample.

College year: 2019/2020 **Presented by : KIHONGO** RESPICIUS

EFFECT OF PLASTIC POLLUTION WASTE ON SOIL ORGANISM: THE CASE OF EARTHWORMS.

Dissertation for obtaining the Master's degree in Fundamental and Applied Ecology.

Abstract

Plastic waste and microplastic pollution have negative effect for the soil organisms. The objective of our study is to see the effect of pollution by microplastic and plastic waste on earthworms which live in soil of production. To achieve our goal we did small experiment in ecology laboratory at University Constantine 01, faculty of nature and life science. Two samples were tested; polluted sample (MP) and non-polluted (MNP). by using earthworms. Two different species were in our experiment *Aporrectodea trapezoids* and *Aperrectodea rosea*.

Results obtained revealed that earthworms which are in polluted incubation media had low biomass compared to those in non-polluted jars (BM/P = 0.69g ± 0.28 ; BM/NP = 5.61g ± 049). It seems that the pH values of plastic waste media was lower than the non-polluted ones (pH/P = 7.33 ± 0.02 ; pH/NP = 7.40 ± 0.06) . Also, the results show that the EC values are high in polluted soils compared to unpolluted ones (EC/P = 1015 µS/cm ± 30.21 ; EC/NP = 856.6 µS/cm ±138.92).

We can conclude that pollution by plastic waste can have negative effects on soil organisms, biodiversity and human.

Key words: Pollution by plastic waste, microplastic , Soil pollution , Pedofauna ,Earthworms.

Date: 06/07/2020

Evaluation Jury :

President of the jury : Dr. SAHLI Leila (University of Mentouri Brothers Constantine1).

Examiner: Dr.Kerboua Fayçal (ENS, Assia Djebbar Constantine),

Supervisor : Dr. BAZRI Kamel eddine (University of Mentouri Brothers Constantine1).