

## Thesis of MICRO-COLLECTOR

- By:

Rahma Ali Bayumi

Farah Mahmoud Basuoni

Students at STEM Red Sea High School

- Supervisor:

Randh Fathy Kadees, Biology Senior Teacher at STEM Red Sea High School

### ملخص:

يهدف المشروع إلى تصميم خاص لمحطة تحلية المياه من المواد البلاستيكية الدقيقة التي تحتوي على نفايات بلاستيكية متحللة وألياف تركيبية وحببيات موجودة في منتجات النظافة الشخصية التي يبلغ طولها (5 مم) أو أقل، والتي لها العديد من الآثار الخطيرة على الحياة البحرية وحياة الإنسان والحياة الحيوانية والنظام البيئي للأرض، وقد تم إجراء تعديل في مراحل تحلية المياه بمحطات التحلية التقليدية، وذلك بالاستغناء عن مرحلتَي التبخير والتكثيف، باستخدام بكتيريا (إيديونيلا ساكايز سيس)؛ لتقليل من الطاقة المستخدمة في عملية التحلية، وكذلك استخدام (هلام السليكا) الذي يعمل على خفض درجة الحرارة، وقد أُشارت النتائج إلى إمكانية بكتيريا (إيديونيلا) على إزالة المواد البلاستيكية الدقيقة من المياه.

### Purpose:

At present, the entire world is having several problems arising from different kinds of pollution among which plastic pollution is also a notable one. And Egypt has many problems that should be solving it to make a balance in the environment and save the ecosystem from damage. one of the biggest problems in Egypt is water pollution (microplastics pollution) because the factory and human activity produce many wasted of plastic and this can make water pollution such as microplastics in seawater and freshwater. Microplastics include broken-down plastic waste, synthetic fibers and beads found in personal hygiene products and being 5 mm or less in their longest. There are two types of microplastics primary microplastics include plastics found in personal care and cosmetics products. Secondary microplastics are created when larger plastic items break down into smaller pieces, the remains of plastics effect on the marine environment and human.

The countries which produce waste plastics. They are known to harm marine life, which mistakes them for food, and can be consumed by humans too via seafood, tap water or other food. The risk to people is still not known, but there are concerns that microplastics can toxic chemicals compound. Microplastics have many dangerous effects of the marine life, human life, animal life and the ecosystem of the earth.

### Background Research:

While Japanese designers recently created a seaweed-based alternative to plastic packaging, another group of Japanese researchers were busy discovering a novel bacterium that can eat plastic. The engineering team behind the discovery published their findings in a paper Friday in the journal Science.

Pangburn, D. J. (2016, November 3). Japanese Researchers Discover Plastic-Eating Bacterium. Retrieved May 9, 2019, from <https://www.good.is/articles/plastic-eating-bacteria-pet>.

*Ideonella sakaiensis* is a bacterium from the genus *Ideonella* and family Comamonadaceae capable of breaking down and consuming the plastic poly(ethylene terephthalate) (PET) as a sole carbon and energy source. The bacterium was originally isolated from a sediment sample taken outside of a plastic bottle recycling facility in Sakai, Japan.

*Ideonella sakaiensis*. (2019, October 22). Retrieved June 16, 2019, from [https://en.wikipedia.org/wiki/Ideonella\\_sakaiensis](https://en.wikipedia.org/wiki/Ideonella_sakaiensis).

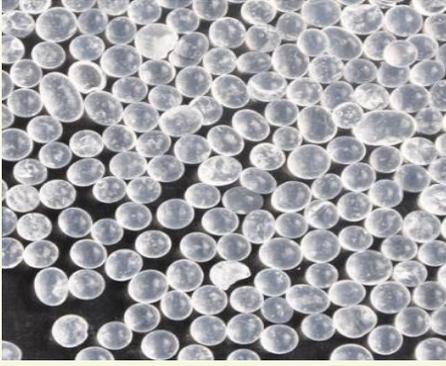
The extreme durability of polyethylene terephthalate (PET) debris has rendered it a long-term environmental burden. At the same time, current recycling efforts still lack sustainability. Two recently discovered bacterial enzymes that specifically degrade PET represent.

Structure of the plastic-degrading *Ideonella sakaiensis* MHETase bound to a substrate (2019, April 10). Retrieved August 3, 2019, from [https://www.researchgate.net/publication/332371220\\_Structure\\_of\\_the\\_plasticdegrading\\_Ideonella\\_sakaiensis\\_MHETase\\_bound\\_to\\_a\\_substrate](https://www.researchgate.net/publication/332371220_Structure_of_the_plasticdegrading_Ideonella_sakaiensis_MHETase_bound_to_a_substrate)

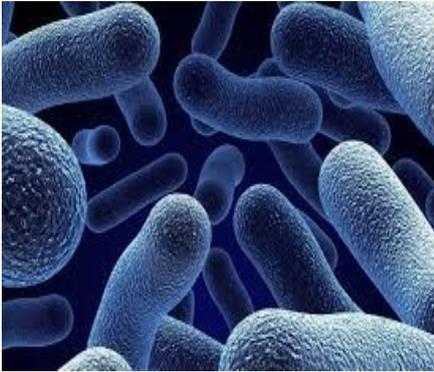
### Hypothesis:

- If the *Ideonella* effect on the (PET) Plastics only or not??
- Will *Ideonella* bacteria effect on the pH of water?
- Will *ideonella* affect total dissolved solids in water??
- Will *ideonella* affect the amount of total suspended solids in the water??

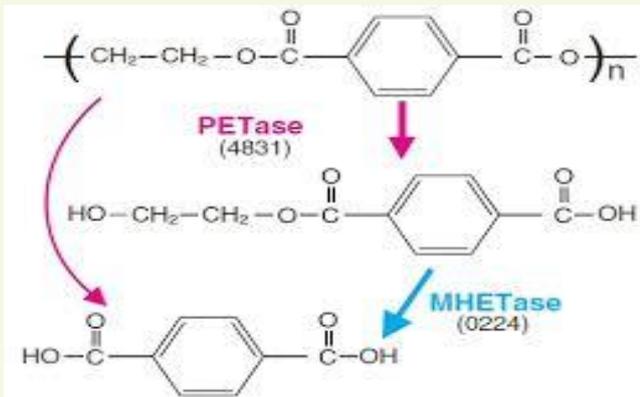
### Materials:



Silicate gel



Ideonella sakaiensis Bacteria



(MHETase and PETase)

### procedure:

To prove our hypothesis, by using our project that is made a special system of the desalination station. Water was desalinated to make it potable exclusively by using evaporation equipment that consumed a significant amount of energy. Currently, there are small desalination plants evaporating seawater or brackish well water. These plants consume small amounts of electrical energy, which can be obtained by windmills, solar panels or other renewable electrical energy sources.

This table shows the stages of conventional desalination plants:

	ENERGY	PROCESS	TECHNOLOGY
PHASE CHANGE	Thermal	Evaporation	<i>Thermal vapor compression</i>
			<i>Multiple effect distillation</i>
			<i>Multi-stage flash distillation</i>
			<i>Solar distillation</i>
		Filtration and Evaporation	Membrane distillation
		Crystallization	Freezing
	Hydrate formation		
NO PHASE CHANGE	Mechanical	Evaporation	<i>Mechanical vapor compression</i>
		Filtration	<b>Reverse Osmosis</b>
	Electrical	Selective filtration	<i>Electrodialysis</i>
	Chemical	Exchange	Ionic exchange

We modified these stages to save energy and remove the microplastic:

In our project, we will give up the evaporation and condensation stage because the bacteria work at a temperature of 30, you will need an energy amount from 0.7 to 0.9 joules, but in the old stations they used 10 joules after the bacteria work.

The second stage, we will use bacteria halophiles to remove the salts from water because they feed on salts and they are found in (seas, oceans and salt lakes) These bacteria will help us get rid of salts in the water.

The third stage, which is the water filter, we will filter the water to remove impurities and some unhelpful minerals, and we will remove the bacteria. After this stage, the water will come out pure.

▣ **Variable:-**

There are variables that affect positively or negatively on the project. These variables are either natural or unnatural:

▣ **Natural variables:**

- The natural water temperature is 2.6 ° C and Ideonella bacteria work only at temperatures up to 30 ° C, so this natural property negatively affects the efficiency of bacteria.

- The presence and availability of quantities of the main enzymes such as (MHETase - PETase) this positively affects the work of bacteria and analysis of the microplastics in an easier and faster.

#### ▣ Unnatural variables:

- The high temperature has a positive effect on the work of bacteria and its speed in the conversion of microplastics to benign monomers ecofriendly and also the addition of adsorption process to reduce water temperature by (silica gel) a positive impact on the other stages of desalination.

#### ▣ Data & Data Analysis:

*Ideonella sakaiensis* cells adhere to the PET surface and use a secreted PET hydrolase, or PETase, to degrade the PET into mono terephthalic acid (MHET). Sakaiensis PETase is the first PETase ever discovered and functions by hydrolyzing the ester bonds present in PET with high specificity (It analyzes it into mono terephthalic acid (pH 3.4: 5)). The resulting MHET is then decay into its two monomeric constituents by a lipid-anchored MHET hydrolase enzyme, or MHETase, on the cell's outer membrane. Ethylene glycol is readily taken up and used by *I. sakaiensis* and many other bacteria. (As shown in fig.1)

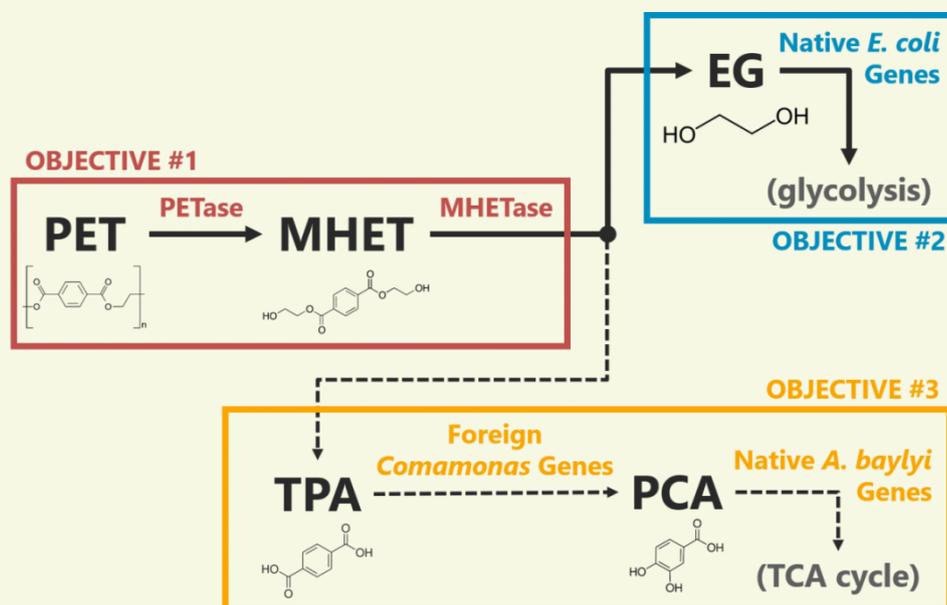


Fig.1

In 2016 sakaiensis was shown to degrade PET, a polymer widely used in bottles. Adhered to a low-grade PET film, the bacteria used two main enzymes, PETase and MHETase, (as shown in fig.2).

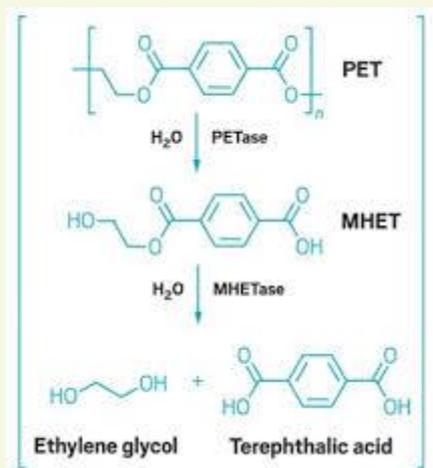


Fig.2

To decompose the plastic into two environmentally benign substances, this served as their main food source.

A colony of sakaiensis could completely degrade a low-grade plastic water bottle in six weeks. The bacteria could also be used to reduce industrial waste during plastics manufacturing.

we will use bacteria halophiles to remove the salts from water because they feed on salts and they are found in (seas, oceans and salt lakes) These bacteria will help us get rid of salts in the water.

The second stage is the adsorption process (as shown in fig.3) to reduce the water temperature to complete the rest of the dissolution stages using silica gel, because it has a specific property (as shown in table1)

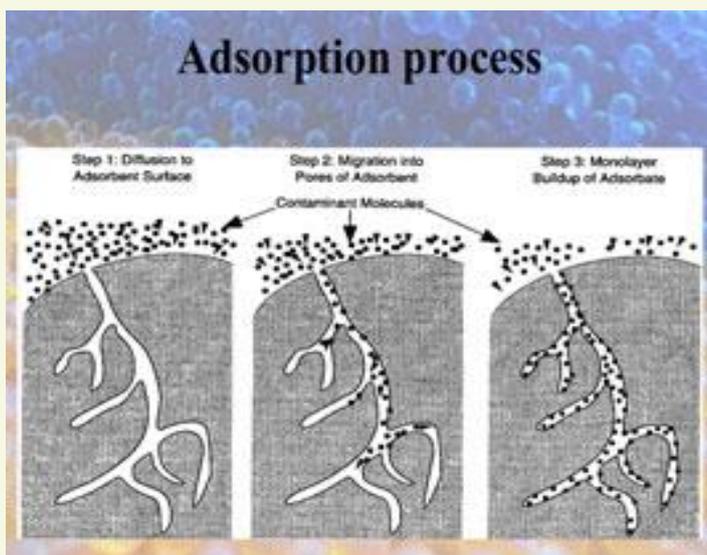


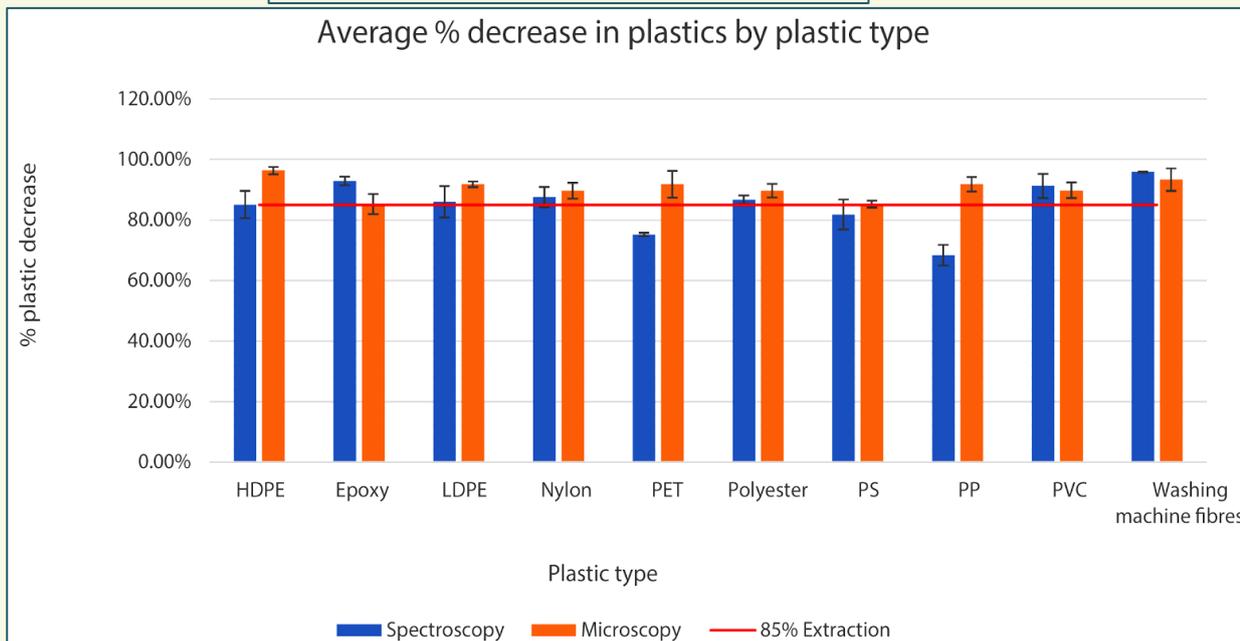
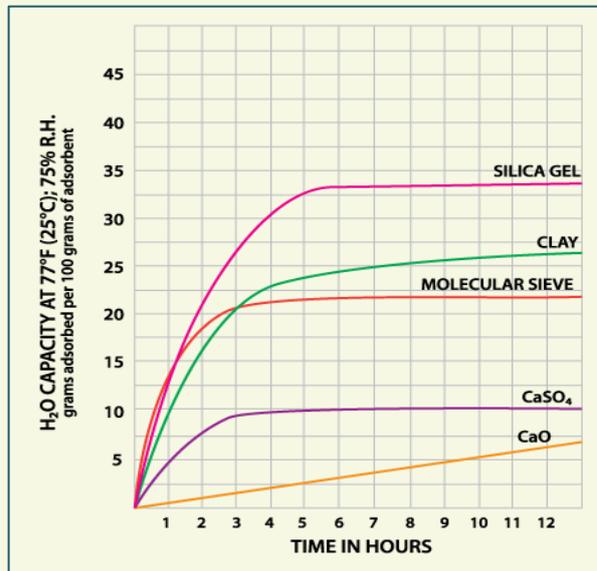
Fig.3

Property	Value	Comments
Thermal conductivity	~0.01 W/m.K	Determined by Vacuum Insulation Conductivity Tester
Bulk density	0.45 g/ml	Determined by using Helium pincometry
Porosity	80%	Determined by BET method
Specific surface area	142-357m <sup>2</sup> /g	Determined by nitrogen adsorption/desorption
Mean pore diameter	~13nm	Determined by nitrogen adsorption/desorption
Primary particle diameter	5-50nm	Determined by electron microscopy
Refractive index	1.46	Very low for a solid material
Dielectric constant	~1-2	Very low for a solid material

Table1

The use of silica gel and the non-use of other materials based on the characteristics of the candidate materials for use (graph.1 show the reason for the use of silica gel compared to available materials).

After research, the effect of ideonella on all types of microplastics was detected but with a specific percentages effect (as shown in the graph.2)



Graph.2

**Result:**

Because of the difficulty of the project in the implementation, the result was on a hypothetical basis resulting from scientific research, it is:

- At the beginning of exponential growth designated here as time  $t = 0$ , the initial concentration of bacterial cells is 1,000 CFU/mL. (Plot  $\log_{10}$  CFU/mL versus time (in hours)) At time  $t = 6$  h, the concentration of cells is 16,000 CFU/mL.
- The possibility of bacteria *Ideonella* to remove 4.6 grams of microplastics in the case of 20 grams of an enzyme MHETase, As is known that *Ideonella* bacteria contain 1.3 grams of enzyme MHETase Therefore, the required amount of *Ideonella* bacteria to remove 4.6 gram of microplastics is 15 CFU/mL, up to remove 18.53% of the amount of microplastics in 1 liter of water.
- *Ideonella* bacteria also reduce the pH of sodium acetate, which has a pH ranging from 6.8: 8.9
- After converting microplastics into terephthalic acid and ethylene glycol monomers, they can be converted into polymers by polymerization to use the ethylene gas present in the synthesis of plastics (an organic gas consisting of two molecules of carbon and four hydrogen atoms, symbolized as  $C_2H_4$ ), a derivative. Oil, because it contains carbon, which is an active ingredient in making the polymer.

#### ■ Conclusions:

- By increasing the number of bacteria in 1 L of water, increase the rate of removal microplastics, because there are 1.3 grams of MHETase enzyme/1bacteria, so the amount of microplastics that remove by 15 bacteria is 18.53% of microplastics/1L.
- When the bacteria increase, the sodium acetate is decreasing and that reduces the pH of the sodium acetate which his rate of pH is 6.8: 8.9 (one of substance in water that increase pH).
- the activity of bacteria is increasing by increase the temperature until arrived between 25:30 c, and this thing is a first disadvantage in the *Ideonella* so sodium silicate will be used which helps to reduce the temperature.

#### ■ Recommendation:

Our project can be applied in desalination stations and put our system when the water heating to remove the microplastics and we put another step to decrease the temperature to make a station complete another stages, and the filtration process will remove all microbes and bacteria. The project can be applied in any country and city, as it just depends on the amount of microplastics.

-as knowing that the 20 grams of MHETase enzyme effect on 4.6 grams from microplastics/1L in 8 hours and fully eliminate microplastics/1L in 6 weeks, so our recommendation to speed up the rate and effectiveness of Ideonella by using genetically modified organisms (GMO) by added Azotobacter SP enzyme to make Ideonella be faster and lifespan increases.

### Research sources:

- Ideonella. (2019, May 21). Retrieved August 20, 2019, from <https://en.wikipedia.org/wiki/Ideonella>.
- Austin, H. P., Allen, M. D., Donohoe, B. S., Rorrer, N. A., Kearns, F. L., Silveira, R. L., ... Beckham, G. T. (2018, May 8). Characterization and engineering of a plastic-degrading aromatic polyesterase. Retrieved August 23, 2019, from <https://www.pnas.org/content/115/19/E4350>.
- MHETase. (2019, June 9). Retrieved August 26, 2019, from <https://en.wikipedia.org/wiki/MHETase>.
- Howes, L. (2019, April 18). Structure of plastic-munching enzyme MHETase solved. Retrieved July 2, 2019, from <https://cen.acs.org/biological-chemistry/biotechnology/Structure-plastic-munching-enzyme-MHETase/97/i16>.
- Ideonella sakaiensis. (2017, October 4). Retrieved September 7, 2019, from [https://microbewiki.kenyon.edu/index.php/Ideonella\\_sakaiensis](https://microbewiki.kenyon.edu/index.php/Ideonella_sakaiensis).
- Ideonella. (2019, May 21). Retrieved August 20, 2019, from <https://en.wikipedia.org/wiki/Ideonella>.
- eval(ez\_write\_tag([[468,60],'newworldencyclopedia\_org-box-2','ezslot\_0',106,'0','0']));Silica gel. (2019, November 4). Retrieved November 13, 2019, from [https://www.newworldencyclopedia.org/entry/Silica\\_gel](https://www.newworldencyclopedia.org/entry/Silica_gel).
- Sodium acetate. (2019, October 8). Retrieved July 14, 2019, from [https://en.wikipedia.org/wiki/Sodium\\_acetate](https://en.wikipedia.org/wiki/Sodium_acetate).
- Sodium acetate. (2018, December 3). Retrieved May 23, 2019, from <https://pubchem.ncbi.nlm.nih.gov/compound/Sodium-acetate>.
- Austin, H. P., Allen, M. D., Donohoe, B. S., Rorrer, N. A., Kearns, F. L., Silveira, R. L., ... Beckham, G. T. (2018, May 8). Characterization and engineering of a plastic-degrading aromatic polyesterase. Retrieved September 17, 2019, from <https://www.pnas.org/content/115/19/E4350>.
- Azotobacter. (2019, October 26). Retrieved November 18, 2019, from <https://en.wikipedia.org/wiki/Azotobacter>.
- Quagliano, J. C., Alegre, P., & Miyazaki, S. S. (1994). Isolation and characterization of Azotobacter sp. for the production of poly-beta-

hydroxyalkanoates. Retrieved September 27, 2019, from <https://www.ncbi.nlm.nih.gov/pubmed/7938497>.

Quagliano, J. C., Alegre, P., & Miyazaki, S. S. (1994). Isolation and characterization of *Azotobacter* sp. for the production of poly-beta-hydroxyalkanoates. Retrieved September 27, 2019, from <https://www.ncbi.nlm.nih.gov/pubmed/7938497>.

Figure 2f from: Irimia R, Gottschling M (2016) Taxonomic revision of *Rochefortia* Sw. (Ehretiaceae, Boraginales). Biodiversity Data Journal 4: e7720. <https://doi.org/10.3897/BDJ.4.e7720>. (n.d.), 2–6. doi: 10.3897/bdj.4.e7720.figure2f

Tanasupawat, S., Takehana, T., Yoshida, S., Hiraga, K., & Oda, K. (2016). *Ideonella sakaiensis* sp. nov., isolated from a microbial consortium that degrades poly(ethylene terephthalate). International Journal of Systematic and Evolutionary Microbiology, 66(8), 2813–2818. doi: 10.1099/ijsem.0.001058