The Effect of Spray Paint on Athletic Fields

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#### Background: The Effect of Spray Paint on Athletic Fields

On athletic fields, spray paint is used to mark sidelines and other important lines during a sporting event. Even though it seems necessary for a game, certain chemicals in the paint are probably affecting and damaging the soil underneath it. Not only can the chemicals in the spray paint be dangerous to humans, but they can also be just as harmful to the soil. There are many bacteria inside of soil that affect the growth of things planted in the soil. When certain chemicals are added to the soil, it decreases the number of bacteria. This is an important issue because the bacteria in the soil are very helpful and the chemical Aliphatic Hydrocarbon may be destroying them. In our experiment, we set out to find the effect athletic spray paint has on the bacteria in the soil. To do this, we have decided to spray athletic field paint on a strip of grass and compare the amount of bacteria after 2 days with the number of bacteria without paint. We think the number of the bacteria will decrease in the soil with the spray paint.

Bacteria, which are usually only one micron in length, can be very beneficial to soil. Even though they are very small in size, they make up for it in their amount. These small but abundant organisms cause many chemical transformations. The bacteria decompose the dead matter on the soil and bring the carbon dioxide back up, either being absorbed or brought up through the roots of the plants. The carbon dioxide allows the plants to photosynthesize and to produce the glyceraldehyde phosphate. The glyceraldehyde phosphate is the foundation of molecules. It has all the molecules in it, carbon, oxygen, hydrogen, nitrogen, and phosphate. Animals get all of the molecules from the eating the plants which have the glyceraldehyde phosphate in them (Brock, 2005).The basic carbon cycle is quite simple. Carbohydrates and oxygen are combined to create carbon dioxide, water and energy. Then during photosynthesis, carbon dioxide and water are combined to create carbohydrates and oxygen. Photosynthesis takes energy from the sun and keeps it in the carbon-carbon bonds within carbohydrates. (Marietta College. 2004) The chemicals within the spray paint could affect this cycle if they decrease the number of bacteria in the soil. If the paint kills the bacteria, then the bacteria cannot give the plants the glyceraldehyde phosphate, meaning the plant, or animal that eats the plant, would not have the five molecules.

Another important cycle is the nitrogen cycle, which has a great impact on every living thing. The bacteria in the ground help to transform the nitrogen into ammonia, which is used by some plants, and for other plants, the ammonia is transformed (through bacteria) into nitrate. This happens through one of two main ways: one being the nitrogen from the air, and the other is the nitrogen from the dead plant and animal material. Bacteria decompose the dead material that is on top of the soil. By decomposing the dead material, it is breaking down the nitrogen and "chewing" it up. It is chewing it up into ammonia so that the plants are able to use it. It is also "chewing" up the carbon, making it into carbon dioxide, leading to the creation of the glyceraldehyde phosphate. With the nitrogen it creates new molecules of nitrogen, carbon, water, and phosphate. Once the nitrogen is able to be used again, it is brought up through roots of plants so that they are able to grow and have energy. The plants are also used as food for the animals to receive energy (Coastal Carolina University, Biochemical cycles, 1998).

The bacteria allow the plants and animals to use carbon dioxide and nitrogen. Without carbon dioxide, the plants and animals would not have the glyceraldehyde phosphate in their systems, meaning they would not have the major molecules. Without nitrogen, proteins would not be formed, which, in turn, means no enzymes. Without enzymes, there is nothing to control the start and stop of chemical reactions. If there aren't any chemical reactions, then the four tasks cannot be completed, which ultimately leads to death. In addition, plants and bacteria would not be able to survive. Bacteria eat the dead plants and animals to gain nutrients, but if they do not have nitrogen in their systems, it will not travel to the bacteria. Without the nitrogen, there would be a decrease in the amount of nucleic acids, and DNA. If there isn't DNA, then RNA could not be produced. With the lack of DNA and RNA, the chemicals cannot react, meaning they cannot do their four tasks. The four tasks include reproduction, respiration, synthesizing of proteins, and regulation of the environment.

The four tasks are performed by the presence of the five major biochemicals; lipids, proteins, nucleic acids, carbohydrates, and water. The biochemicals are made up of carbon, oxygen, hydrogen, phosphate, and nitrogen. One of the molecules, phosphate, plays a very important role in the cell. In the cell, the phosphate makes ATP which is used to create energy for the cell. Without energy, there is no work, causing the cell to die. Therefore, without the bacteria in the soil to transform nitrogen to make it usable, nothing would survive (Brock, 2005).

Since bacteria can be actually helpful in the soil, we think that the Aliphatic Hydrocarbon will decrease the number of bacteria, therefore hurting the soil. The Aliphatic Hydrocarbon is toxic; therefore it could kill the bacteria. It is made up of carbon and hydrogen atoms. It can either be saturated or unsaturated. When it is saturated, all bonds in the hydrocarbon are single bonds, meaning that they share one electron per atom. When it is unsaturated it becomes and double, or even triple bond. Aliphatic hydrocarbon is the chemical that one smells in the paint. It can eventually affect ones central nervous system, and it is also dangerous because it has additional oils added to it ensure that it stays together. Aliphatic Hydrocarbon is important because it is the actual paint in the spray paint. Instead of evaporating, this chemical seeps into and affects the bacteria in the soil. Aliphatic Hydrocarbon is made up of three types of hydrodcarbons that will all affect the soil in different ways (MSDS HyperGlossary: Aliphatic, 2004). These hydrocarbons are present in the spray paint affecting the bacteria in the soil.

We think that the soil most in contact with the paint will be much less healthy than the soil without any paint. All of the chemicals inside the Pioneer Quik Stripe Athletic Field Marking Paint can be very dangerous and toxic. Though the paint is only sprayed onto the grass, it still seeps into the soil eventually. The area with the most paint sprayed on, will consequently be the least healthy area.

To prove this, we are going to test a patch of regular soil and a patch of soil that has Pioneer Quick Stripe Athletic Field Marking to see if the patch with the athletic field marking is not as healthy or has been affected in a negative way. This idea appealed to us because it is a very common use on the Roland Park Country School campus as well as many campus's in the world. The spray paint is mainly used for athletic field lines. These lines are in the same general area of the fields year round and it is not a natural aspect of an ecosystem. This could affect the soil and overall affect the humans. If the soil is affected, then the entire ecosystem is affected. If we found that the chemicals in the spray paint affect the soil, we could ask the school to buy a more nature-friendly spray or even write to Pioneer Quick Stripe Athletic Field Marking's producer and ask them to make their spray paint more environmentally safe. If we prove that the Pioneer Quick Stripe Athletic Field Marking is harmful to bacteria, it could be possible that some bacteria are immune to the toxic substances in the spray. These bacteria would survive and be the successful species in natural selection. This could lead to the eventual evolution of bacteria that would be resistant to many toxic substances or chemicals (Groffman, 2005).

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## Problem:

How does the presence of aliphatic hydrocarbons change the density of bacteria in the soil?

# **Hypothesis:**

Aliphatic Hydrocarbons will decrease the density of bacteria in the soil.

# **Experiment:**

- A) independent variable: amount of Aliphatic Hydrocarbon
- B) Dependent variable: number of bacteria colonies
- C) Negative control: grass without spray paint and samples of the sites before the experiment
- D) Controlled variables:
  - amount of grass
  - location of grass
  - distance of trees/plants away from the testing plots
  - elapsed time for paint to seep into soil
  - height of spray paint can (when being sprayed)
  - the length of time sprayed
  - size of plots
  - amount of soil taken out to test
  - distance from one plot to another
  - type of water (sterile)
  - amount of water for dilution
  - type of pipettes
  - types of sheets- petrifilm
  - amount of dilution placed on petrifilm- 100 μl
- E) Procedure: We did the follow steps at N39.35536, W076.63267

1. Plot a soil area that is 2.5 meters long and 25 cm wide. (as seen in the picture below) 2. Mark flags on the ground, starting at 0 meters and mark after every .5 meter, there should be 6 flags. (as seen in the picture below)

3. Mark these flags(left, to right) A, B, C, D, E, F. (as seen in the picture below)

4. Also mark flags on the ground at the two 25 cm marks(so you know where the boundaries are, as seen in the picture below)

5. 10 cm away from the each flag (A, B, C, D, E, F), punch a 2 cm diameter hole that is 5 cm deep into the soil and put each soil sample into a seperate labeled plastic bag. As shown in the picture below:

6. Steps 6-17 need to be done in the same day. Start by labeling 5 cultured tubes:  $A10^{0}$ ,  $A10^{-1}$ ,  $A10^{-2}$ ,  $A10^{-3}$ ,  $A10^{-4}$ 

7. Using a serological pipette, put 9 ml of sterile water into tubes  $A10^{-1}$ ,  $A10^{-2}$ ,  $A10^{-3}$ ,  $A10^{-4}$ .

8. Put 1 cc of your soil sample into the culture tube labeled  $A10^{\circ}$ .

9. Add 10 ml of sterile water to tube  $A10^{\circ}$ .

10. Cap tube  $A10^0$  and shake vigorously.

11. Using the serological pipette, take out 1ml of the soil/water mixture in tube  $A10^{0}$  and put it into tube  $A10^{-1}$ .

12. Shake vigorously and take out 1ml of mixture in A10<sup>-1</sup> and putting it in A10<sup>-2</sup>

13. Shake vigorously and take out 1ml of mixture in  $A10^{-2}$  and putting it in  $A10^{-3}$ 

14. Shake vigorously and take out 1ml of mixture in  $A10^{-3}$  and putting it in  $A10^{-4}$ .

15. Using tube A10<sup>-3</sup>, with a micro pipette, put the pipette tip on it and take out 100  $\mu$ l of the mixture and put it onto a petrifilm sheet.

16. Repeat step 13 with tube  $A10^{-4}$ .

17. Repeat steps 6-16 using samples B, C, D, E, and F (label the tubes accordingly to the letter)

18. After 48 hours, examine the sheets and count the number of bacteria colonies.

19. Use the equation below for samples  $10^{-3}$  and  $10^{-4}$ , that have at least 5 bacteria colonies:

# colonies on plate •  $10^2$  = # of bacteria in dilution tube;# of bacteria dilution tube• $10^{|# of}$ dilutions| = # of bacteria in original sample

20. Record data

21. With a piece of cardboard, create a 10 cm by 5 cm rectangular template.

22. 10 cm above flags A, C, E, using the template, spray the paint in the 10 cm by 5 cm rectangle, hold the spray can 40 cm above the ground and spray the paint for 5 seconds into the rectangle. As shown in the picture below:

- 22. Do not mark above flags B, D, or F.
- 23. Wait 5 minutes after you spray the paint on the grass.
- 24. pour 500mL liter of water, for each patch of soil, directly on top of the soil above all
- the flags; A, B, C, D, E, F.
- 25. Wait 48 hours
- 26. Repeat steps 5-20

### Data:

Density of bacteria	a in soil(bacteria p	er cubic cm)				
	soil A	soil B	soil C	soil D	soil E	soil F
Before spray						
paint	11,000,000	7,000,000	6,000,000	700,000	5,000,000	400,000
After spray paint	6,000,000	12,000,000	6,000,000	900,000	3,900,000	700,000
Averages:	treated with spray paint	untreated without spray paint				
before spray	•	•				
paint	7,333,333	2,700,000				
after spray paint	7,100,000	4,533,333				

### Analysis:



Average number of bacteria(per cubic cm) in soil

One has to look at the corrected differences in the negative control. By looking at the negative control, one can see that the bacteria in the soil are increasing in a high amount. The negative control is showing that something in the environment, as a whole, is making the bacteria increase. This means that for the soil with spray paint, the bacteria has actually decreased more than it looks. The bacteria in the soil with spray paint should have increased the same amount as the negative control, but it did not only not increase, but it also decreased in the number of bacteria.

#### **Conclusion**

Our hypothesis was correct by saying that the aliphatic hydrocarbons would decrease the density of bacteria in the soil. By looking at the graphs one can see that there is a decrease in the amount of bacteria that was in the treated soil; with spray paint. One can also see that there is a high increase in the amount of bacteria in the untreated soil; without spray paint. Bacteria is reproducing and growing rapidly each day. This is a reason why there is such a high increase in the bacteria that was in the soil without spray paint. By looking at the graph for the treated soil, there is no increase. Seeing the results, it seems that the spray paint is not only stopping the bacteria from growing/increasing, but it is also killing it. This could be hazardous not only to the soil, but also to the surrounding environment. Without the bacteria in the soil one of the major ways to get energy to plants would not be able to happen. Without the bacteria, the nitrogen cycle process could not be done successfully. Meaning the nitrogen in the air would not be able to transform into ammonia (used for plant energy). Therefore it would not provide energy for the plants and furthermore it would not provide energy for the animals that eat those plants. If the dead plants and animals do not have nitrogen in their systems, then when the bacteria decompose them, they will not survive without nitrogen.

Along with the nitrogen cycle, the decrease in bacteria is also affecting the carbon cycle. The carbon cycle is done to give carbon dioxide to plants so they can perform photosynthesis. The purpose of photosynthesis is to create sugar and energy for the plant, and also for the animal that eats the plant. The bacteria plays a part in this by taking the carbon dioxide from decomposing matter and bringing it back up through the plants. Without the bacteria to bring the carbon dioxide back up, then the plants would not be

able to create the glyceraldehyde phosphate. The glyceraldehyde phosphate can not only be made into sugar, but it provides the five molecules; hydrogen, nitrogen, oxygen, carbon, and phosphate. Without the glyceraldehydes phosphate the plant would not have these molecules.

In conclusion, the spray paint is included with a toxin that is stopping the growth of the bacteria and killing the bacteria in the soil.