

Nicotine effects on the Muco ciliary motion, on the *Atlantic surf clam*

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Introduction:

Cigarette smoking includes many different effects, from the chemicals it may contain. In many cases it has resulted in pulmonary *emphysema* in lungs. (Karlinsky and Snider, 1978) It is defined as a pathological condition of the lungs marked by an abnormal increase in the size of the air spaces, resulting in labored breathing and an increased susceptibility to infection. ([Lexico Publishing Group, LLC](#), 2005). This can affect many different parts of a human body; however we study the effects of nicotine on the muco ciliary motion.

Muco cilia play a very crucial role in the lungs. It removes the foreign particles from lungs, and its efficiency depends on the interaction between the cilia and the mucous. (Karlinsky and Snider, 1978) The respiratory tract in humans is lined with cilia, which is an indication that a major part of it is ciliated. "The surface of each columnar cell carries approximately 200 cilia with an average length of 0.6micrometer and a diameter of 0.2 micrometer." (Karlinsky and Snider, 1978) The locomotion of the cilia is aided by the two microtubules in the core, and nine on the periphery, acting as a contractile factor. They form bridges namely the dynein, nexin and radial links. It causes motion by the activation of dynein and the resultant bending of the axoneme. (Karlinsky and Snider, 1978) The clam cilia, was seen to beat in one particular direction, with one power stroke and two or three recovery strokes.

Another Important is the mucous secreted by the sub mucosal glands, contributing to the ciliary motion. The mucous is comprised of 95% water and the remaining 5% of micro molecules. (Amino acids, lipids, nuclei acids, etc...) The depth of the mucous plays an important role in the ciliary motion. "In the central airway normal mucus layer is 5 – 10 micrometer." (Karlinsky and Snider, 1978)

The source of energy for the cilia beat is seen to be adenosine triphosphate, the beat usually being a 1000 beat per minute in coordinated between two adjoining cells. (Karlinsky and Snider, 1978) This coordinated beat usually forms a wave motion.

Nicotine is seen to affect (decrease) the transport of the carmine particles in the mucosa of trachea. Presence of nicotine in the respiratory tract is also seen to disrupt the ciliary motion, and also blocking the secretion of mucus from the mucosal glands. Cytotoxicity is observed on the ciliated epithelium, producing an irritant effect causing ciliostatis. (Karlinsky and Snider, 1978) Ciliostatis refers to a situation, where the cilia motion maybe in a decreased rate, but constant. ([Lexico Publishing Group, LLC](#), 2005) Nicotine stimulates the beat for a brief moment, which is followed by a constant decreased beat. The cell biology behind the effect may interpret that nicotine results in an enzyme inhibition of adenylate kinase reducing the production of ATP, which is the source of energy for ciliary motion. (Karlinsky and Snider, 1978)

Cigarette smoking encompasses the intake of various toxic chemicals, and nicotine being one of them. This experiment will allow us to understand the how much nicotine contributes towards the dys-mucociliary motion, and hence the cell damage. It will also help us confirm my hypothesis, which proved to be different from the works done by other researchers. My hypothesis stated that the nicotine would result in an increase in the ciliary motion in the short term. This hypothesis was assumed based on idea that the intake of nicotine (assumed with smoke) would cause a decrease in the oxygen intake. It was further supposed that this would mean that the cilia would have to beat faster, to get enough oxygen in the cells.

The nicotine was tested on the gills of the *Atlantic surf clam*, using sea water as the buffer. Three different sets of trials were performed on the control and the experimental each to confirm our results. This was seen as an important topic because of the increasing number of victims of nicotine, by smoking. The test results should help us understand what exactly happening, on a cell biological and a psychological level. There are many other effects of nicotine, but to begin with we try to understand its effects on the mucociliary motion.

Materials and Methods

Procedure:

The clams were put in the sea water. The clams were cut open by cutting the side muscles, and put it back in the

sea water. Gills in the control clam were identified, by comparing to a model of a dissected clam. The control clam was placed under the microscope. The clams were submerged in the sea water, to keep them alive. The control is placed under the microscope; SMZ 660. The control clam was focused on optics 1.0X. The Gills were then focused on the microscope. The BTV was opened on the desktop to be able to view the clam dissection on the computer. 'Capture' from the menu section was opened. Following the link, "capture movie" was then selected, on the "480X620" resolution.

Activated charcoal was then crushed into fine particles, in a plastic test tube using a glass rod. 1/8 of a pinch of activated charcoal was carefully on the added on the surface of the clam gills, to help us view the direction and the rate of the ciliary movement. 'Play' button was clicked to frame the gills, and to record the movies of the ciliary motion. The recorded movie clips was used to deduce data. The movie was allowed to run for 2mins approximately. Click 'stop' button after 2minutes of the movie clip. This was followed by recording the time taken for the particle of a charcoal to travel a distance 2cm. This procedure was repeated three times, so that you can confirm your data.

Similarly, the experimental clams were dissected. The gills are identified where 1/8 of a pinch of charcoal was added. After that 4 drops of nicotine are added on the gills of the experimental clam. The concentration of the nicotine used was (1.5×10^{-6}) g/L.) The videos of the experimental gills are recorded for 2mins and some frames were saved as well, to help us quantify our data later. Lastly, the time elapsed for the charcoal to move 2cm in the experimental gills was measured. This process was repeated three times.

Results:



Figure1: Experimental clam gills with charged charcoal on it, viewed on a SMZ 660 microscope with optics of 1.0 X. This figure was framed after the addition of 4drops of nicotine for exactly 2minutes.

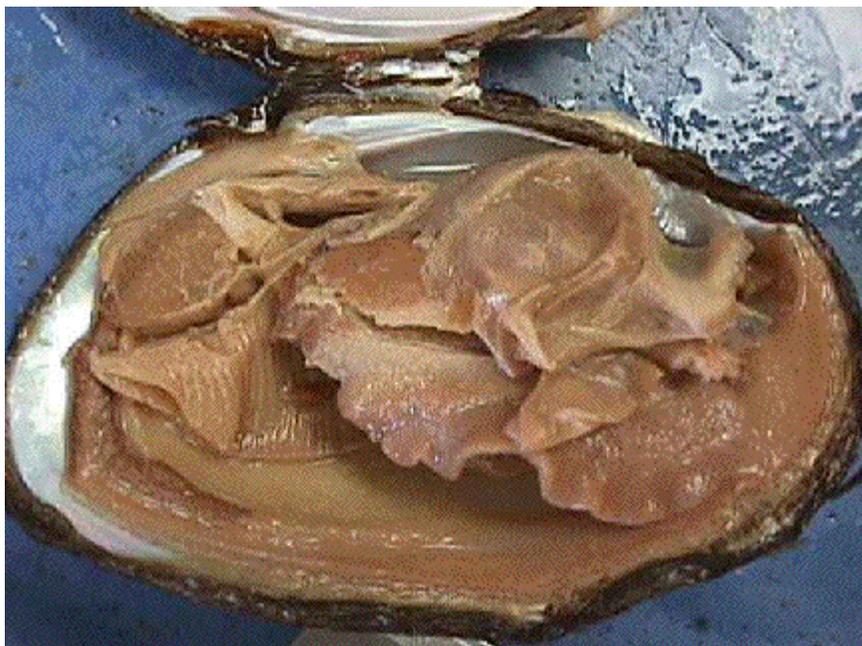


Diagram 2: picture of a dissected clam taken by a normal photo camera.
(nesc, clam dissection, 2005)

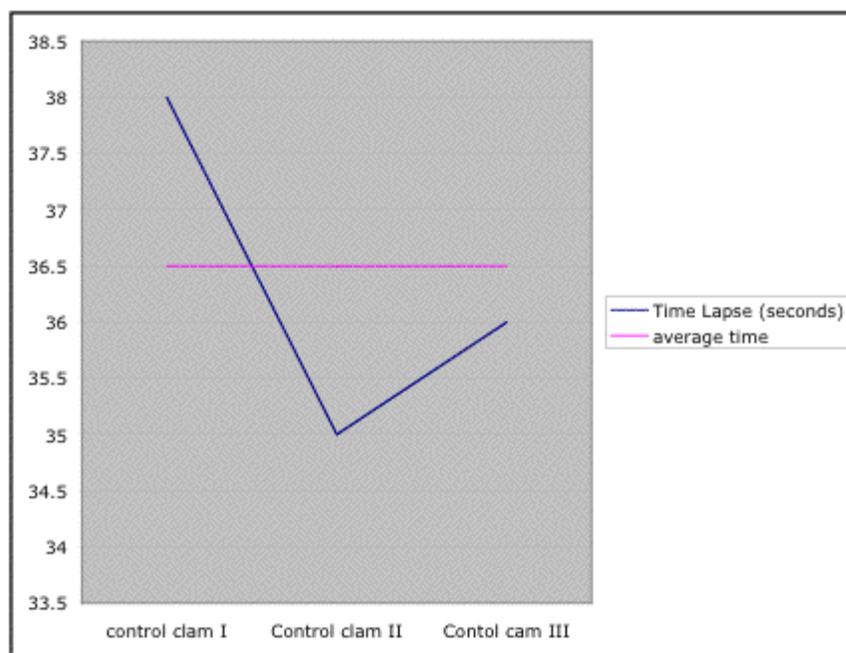


Figure 3 – Experimental clams

This y axis of the figure represents the time taken charged charcoal to move 2cm, while the x – axis represents the three different trials take on the clams. The blue curve indicates the different time taken, for each trial. The first trial took 38 seconds, second took 35 seconds and lastly the third trial took 36 seconds. The pink line indicates the average time taken in the experimental clam to travel 2cm, which was 36.5 seconds.

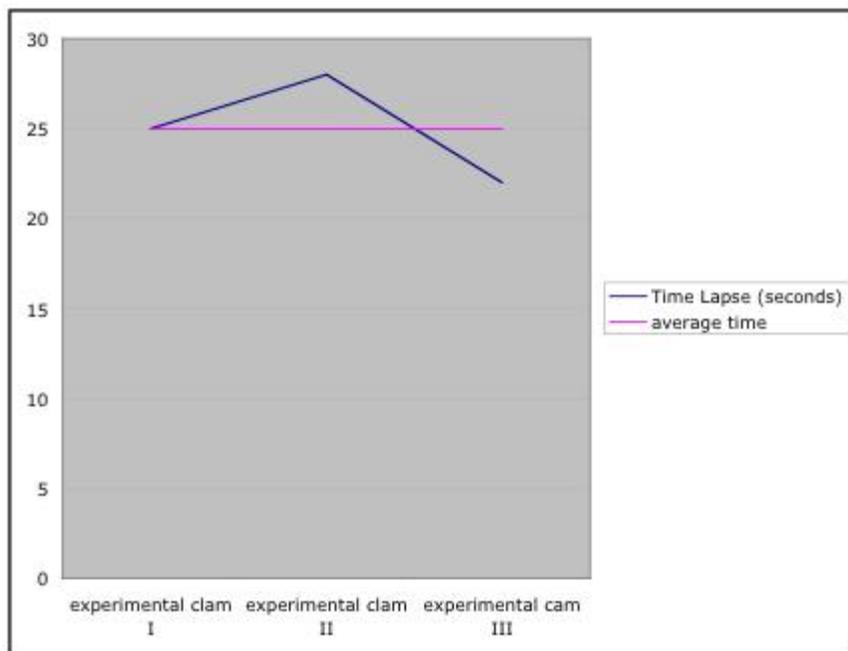


Figure 4: control clams

This y axis of the figure represents the time taken charged charcoal to move 2cm, while the x – axis represents the three different trials take on the experimental clams. The blue curve indicates the different time taken, for each trial. The first trial took 25 seconds, second took 28 seconds and lastly the third trial took 23 seconds. The pink line indicates the average time taken in the control clam to travel 2cm, which was 25seconds.

Discussion:

The results of my project disagreed to my hypothesis. My hypothesis stated that on the addition of nicotine to the surface of the clam gills, the ciliary motion would increase in the short term. This hypothesis was assumed based on idea that the intake of nicotine (assumed with smoke) would cause a decrease in the oxygen intake. It was further supposed that this would mean that the cilia would have to beat faster, to get enough oxygen to be circulated around the cells.

While on the contrary, my results indicated a decrease in the ciliary motion. This was shown by figure (3) and figure (4) in the result section. The figure (3) shows us the time take by the charcoal to travel 2minutes, in the experimental clam. There were three trails conducted on the control clam, which were quantified into 38, 35 and 36 seconds. This averaged out to be 36.5 seconds. On the other hand, figure 4 shows us the time taken by the charcoal particle to move a distance of 2cm in the control clam, in three different trials. The three different data that were collected were 23, 25 and 27 seconds, averaging out to be 25 seconds. Hence the two average data help us come to the conclusion that the cilia motion was faster in the control clam, which was why the charcoal particle in the control clam took a shorter duration to move a distance of 2cm. This shows that the cilia motion on nicotine addition, decreased.

All the sets of data recorded during the control and experimental experiments, were fairly very close to their averaged figure which accounted that minimal errors were made. However, as shown in figures (3) and (4), it lacked a smooth flow in the curves. This could have resulted due to some errors made, while performing the experiment. During the dissection, due to the possibility of some accidental incisions, tissues must be damaged resulting in adverse effects on the ciliary motion. Errors may also have resulted due to the delay while housing the clams in the sea water, which acted as a buffer.

The result supporting a decrease in mucocilia motion can be explained by the fact that nicotine affects (decrease) the transport of the carmine particles in the mucosa of trachea. Presence of nicotine in the respiratory tract is seen to disrupt the ciliary motion, and also blocking the secretion of mucus from the mucosal glands. Cytotoxicity is observed on the ciliated epithelium, producing an irritant effect, which causes ciliostatis. (Karlinsky and Snider, 1978) Ciliostatis refers to a situation, where the cilia motion maybe in a decreased rate, but constant. ([Lexico Publishing Group, LLC](#),

2005) Nicotine stimulates the beat for a brief moment, which is followed by a constant decreased beat. The cell biology behind the effect may interpret that nicotine results in an enzyme inhibition of adenylate kinase reducing the production of ATP, which is the source of energy for ciliary motion. (Karlinsky and Snider, 1978)

However, given a second chance to study cell biology, it is important that cell biologists be very delicate and yet very efficient, due to the short lived cells. Mucociliary transport declines with age, which indicates that accurate results would be achieved by studying animals of a similar range of age. (Karlinsky and Snider, 1978) It is also important that the animal not infected by any diseases, which may cause an impairment of the mucociliary function.

Although the concentration of the nicotine used, helped me deduce my results, it would be more competent if the concentration of the nicotine was higher than 1.5×10^{-6} g/L. This statement is made, assuming that higher concentrations of nicotine would create a faster and more evident difference on mucocilia motion, between the control and experimental clam. It would also be interesting to break down the tissues into individual cells by using trypsin and study an individual cell, of cilia.

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