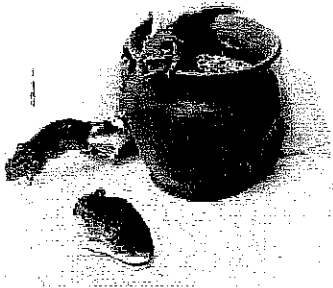
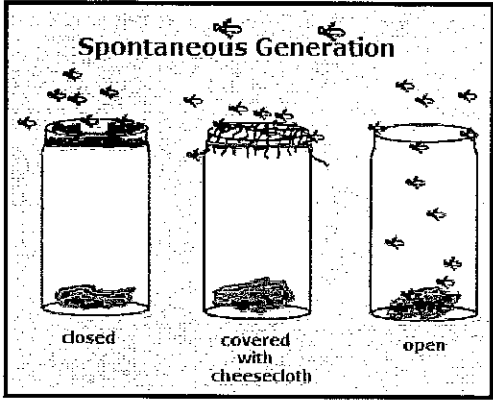


Spontaneous Generation

What is it?

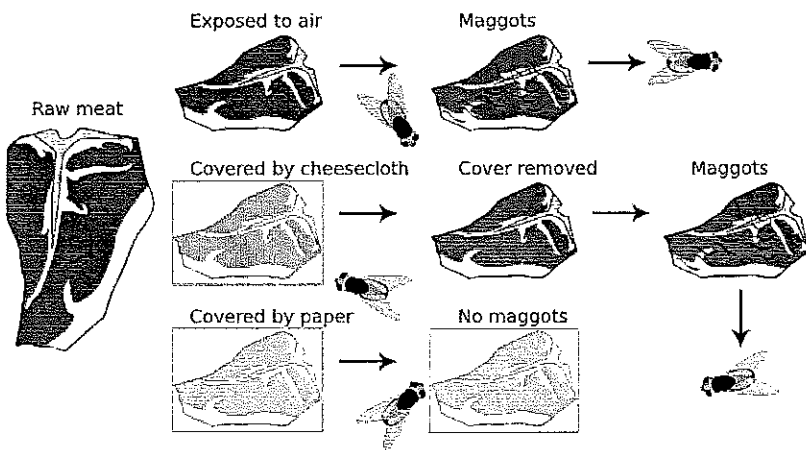
What do these pictures have to do with spontaneous generation?



What was really going on?

Other Naturalists

- Francesco Redi



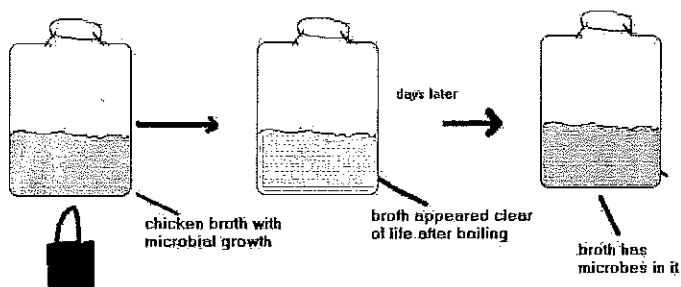
- Describe his experiment.

- Did his experiments support or falsify the hypothesis of spontaneous generation?

- John Needham

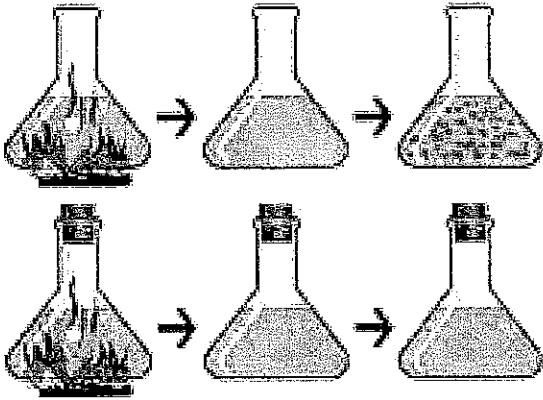
- Describe his experiment.

John Needham Experiment (1748)



- Did his experiment support or falsify the hypothesis of spontaneous generation?

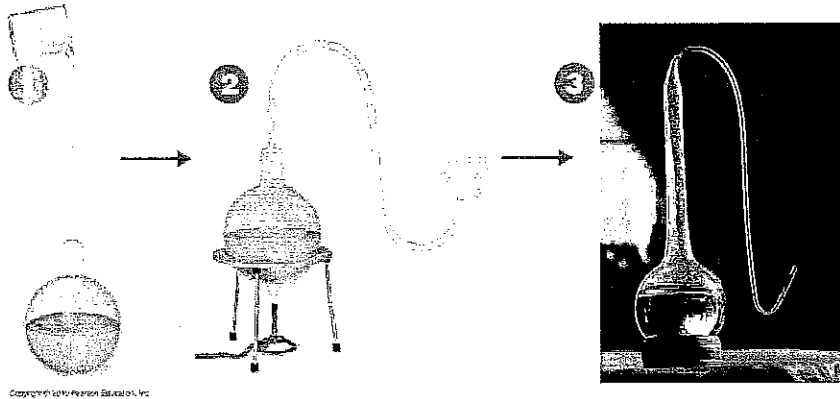
- Lazzaro Spallanzani



- Describe his experiment.

- Did his experiment support or falsify the hypothesis of spontaneous generation?

- Louis Pasteur



- Describe his experiment.

- Did his experiment support or falsify the hypothesis of spontaneous generation?





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Origin of Life

Spontaneous Generation

It was once believed that life could come from nonliving things, such as mice from corn, flies from bovine manure, maggots from rotting meat, and fish from the mud of previously dry lakes. Spontaneous generation is the incorrect hypothesis that nonliving things are capable of producing life. Several experiments have been conducted to disprove spontaneous generation; a few of them are covered in the sections that follow.

Origin of Life

- Introduction
- Spontaneous Generation
- Early Earth Environment

Redi's Experiment and Needham's Rebuttal

In 1668, Francesco Redi, an Italian scientist, designed a scientific experiment to test the spontaneous creation of maggots by placing fresh meat in each of two different jars. One jar was left open; the other was covered with a cloth. Days later, the open jar contained maggots, whereas the covered jar contained no maggots. He did note that maggots were found on the exterior surface of the cloth that covered the jar. Redi successfully demonstrated that the maggots came from fly eggs and thereby helped to disprove spontaneous generation. Or so he thought.

In England, John Needham challenged Redi's findings by conducting an experiment in which he placed a broth, or "gravy," into a bottle, heated the bottle to kill anything inside, then sealed it. Days later, he reported the presence of life in the broth and announced that life had been created from nonlife. In actuality, he did not heat it long enough to kill all the microbes.

Spallanzani's Experiment

Lazzaro Spallanzani, also an Italian scientist, reviewed both Redi's and Needham's data and experimental design and concluded that perhaps Needham's heating of the bottle did not kill everything inside. He constructed his own experiment by placing broth in each of two separate bottles, boiling the broth in both bottles, then sealing one bottle and leaving the other open. Days later, the unsealed bottle was teeming with small living things that he could observe more clearly with the newly invented microscope. The sealed bottle showed no signs of life. This certainly excluded spontaneous generation as a viable theory. Except it was noted by scientists of the day that Spallanzani had deprived the closed bottle of air, and it was thought that air was necessary for spontaneous generation. So although his experiment was successful, a strong rebuttal blunted his claims.

Pasteur's Experiment

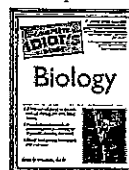
Louis Pasteur, the notable French scientist, accepted the challenge to re-create the experiment and leave the system open to air. He subsequently designed several bottles with S-curved necks that were oriented downward so gravity would prevent access by airborne foreign materials. He placed a nutrient-enriched broth in one of the goose-neck bottles, boiled the broth inside the bottle, and observed no life in the jar for one year. He then broke off the top of the bottle, exposing it more directly to the air, and noted life-forms in the broth within days. He noted that as long as dust and other airborne particles were trapped in the S-shaped neck of the bottle, no life was created until this obstacle was removed. He reasoned that the contamination came from life-forms in the air. Pasteur finally convinced the learned world that even if exposed to air, life did not arise from nonlife.

Bloterns

Pasteurization originally was the process of heating foodstuffs to kill harmful microorganisms before human consumption; now ultraviolet light, steam, pressure, and other methods are available to purify foods—in the name of Pasteur.

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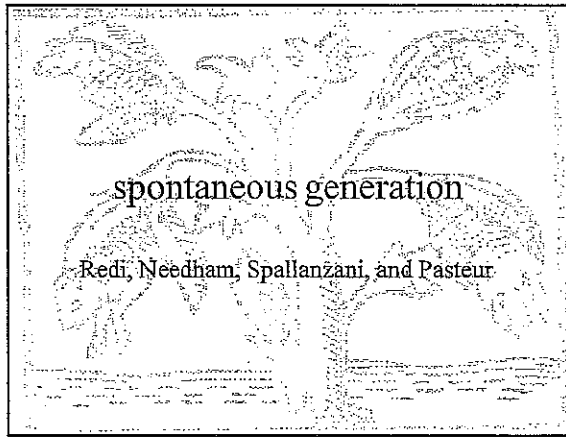
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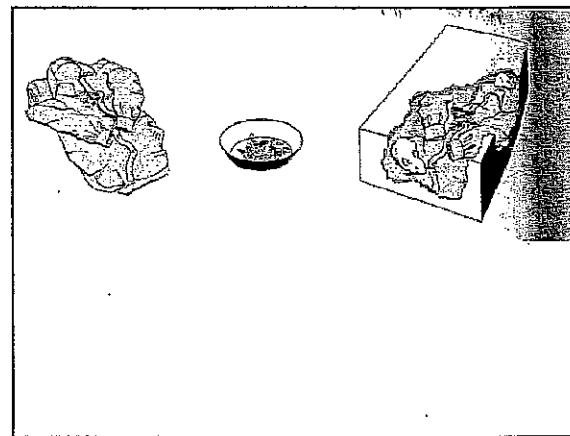
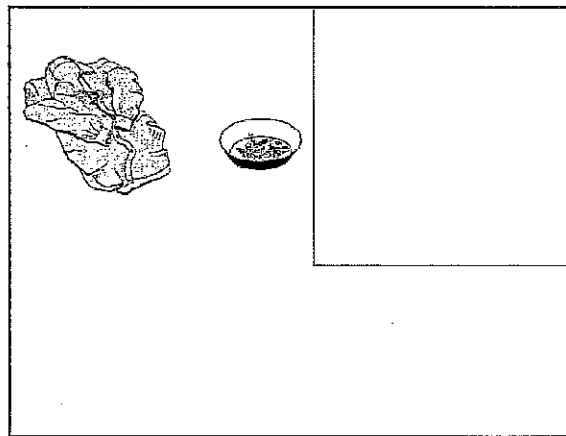
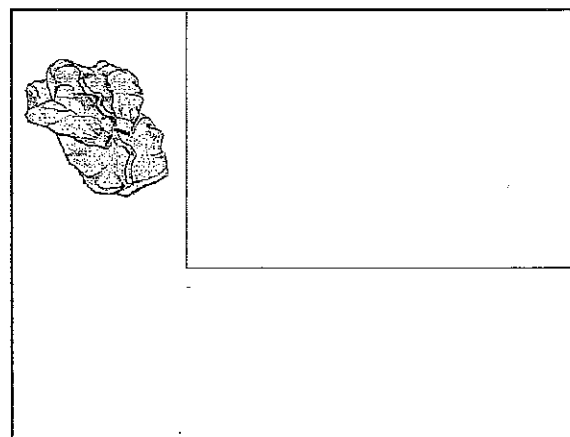

Spontaneous Generation

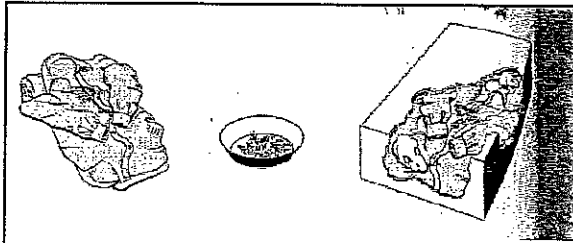
- For much of history, people believed that animals could come from non-living sources. They thought:
 - Frogs developed from falling drops of rain
 - mice arose from sweaty underwear
 - and flies arose from decaying meat.
- This is called **abiogenesis**
- Also known as **spontaneous generation**



The Power of Authority

- In the past, people believed what they were told by "authorities" such as the Church, or the ancient Greek philosopher Aristotle
- Questioning Aristotle was like questioning the Church....





- One "scientist" put forward the belief that mice could be generated spontaneously from wheat and a sweaty shirt.
- The wheat provided the "nutritive power" and the shirt provided the "active principle."
- "active principle" = a mysterious "life-force" that allowed spontaneous generation to occur.

1668 -- Francisco Redi (Italian physician & poet)-- attempted to disprove the theory of Spontaneous Generation.

Experiment 1

beginning of experiment open jars end of experiment

closed jars end of experiment

"The flesh of dead animals cannot engender worms unless the eggs of the living being deposited therein"

- Put dead snakes, eels, and veal in large wide mouthed vessels. Sealed one set with wax and left the other set open to air.
- Decaying meat was teeming with maggots, sealed meat had no maggots
- Wax sealed vessels failed to produce maggots because flies were unable to reach the meat

Redi's critics said:

- You have too many variables
- There is a lack of access and a lack of air.
- We ALL know that everything needs air
- Of course no flies grew!
- **You haven't proven anything.**

Redi part 2 -- answer to critics

Experiment 2

fine mesh allows in air, but not flies

flies laid eggs on top of mesh, no maggots in meat

fine netting

Redi's Conclusions:

- "All living beings come from seeds of the plants or animals themselves"
- However, if someone were to demonstrate even one exception to this hypothesis, then Redi's hypothesis would be rejected.

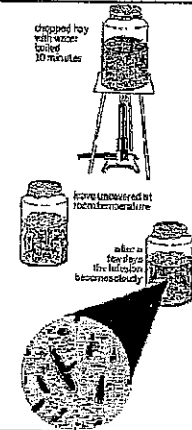
John Needham (English Clergyman) wondered if this would work with microorganisms in 1745

- Everyone knew that boiling killed organism
- Needham prepared various broths and show that they contained microbes.
- Then he boiled them, and showed that there were no longer any microbes.
- He ensured the stoppers were loose, so that would not be excluded
- Then, after a few days, microbes had reappeared!
- This was "proof" that the microbes had spontaneously generated from the non-living broth.



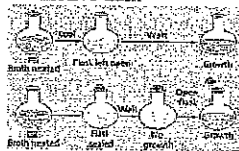
Needham's error

- **BUT: how was this evidence of a faulty experiment?**
 - what ERROR in experimental method is shown here?
- **Hypothesis:** microbes **MUST HAVE** arisen spontaneously *from the broth*.
- **Assumption:** there is no other place the microbes could come from (other than the broth).
- **error:** microbes could have come from the air!



Spallanzani's (Italian Naturalist) -- 1745

- Disagreed with Needham
- Claimed he didn't seal jars well enough
- He said microbes could have come from the air
- He repeated Needham's experiment, but changed two things:
 - boiled flasks longer, and
 - **SEALED THEM** after boiling by fusing the glass tops shut
 - (hermetically sealed – absolutely airtight)
- Result: **NO growth in ANY flask**



Needham criticizes Spallanzani's first experiment

- **BUT** Needham said: you boiled it **TOO LONG**, and:
- You spoiled the *vegetative power* by boiling.
- You killed the ability of the broth to give life.
- Life *can still* come from broth -- but the broth must not be "damaged" by boiling.



Spallanzani's second experiment

- he did **TIMED BOILINGS**
- then left them partially sealed
- some partially sealed, some hermetically sealed as in his previous experiment
- hypothesized that more boiling should lead to less life
- he left some jars as Needham had (leaky seals), to ensure "active principle" was not damaged

	tight seal	loose seal
30 mins		
60 mins		
90 mins		
120 mins		

Spallanzani's second experiment -- results

- this showed **TWO** main things:
- boiling did **NOT** damage broth's ability to support life
- growth depended on the **SEAL** only

	tight seal	loose seal
30 mins		
60 mins		
90 mins		
120 mins		

Louis Pasteur 1859— (French chemist) entered a contest sponsored by French Academy of Sciences to prove or disprove Spontaneous generation.

- used swan-necked flask
- flask allowed in air, but trapped dust (and microbes)
- boiled infusion
- showed that **NO growth occurred**, even after many days
- BUT -- what about damaging the "active principle"?

- Pasteur showed that the active principle was **NOT** damaged
- at any later time, he could tip the flask
- this allowed nutrient broth to contact the dust
- this carried microbes into the broth
- result: growth!

Pasteur squashes the idea of abiogenesis completely!

- Since then, no one has been able to refute Pasteur's experiment
- scientists everywhere soon came to accept that abiogenesis did **NOT EXIST**.
- but: then how did life on this planet start in the first place?

Spontaneous Generation

Origin of Life

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Bioterms

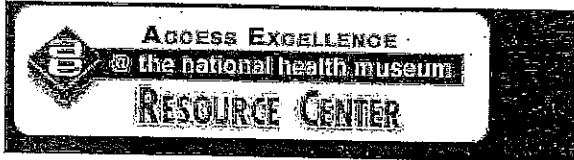
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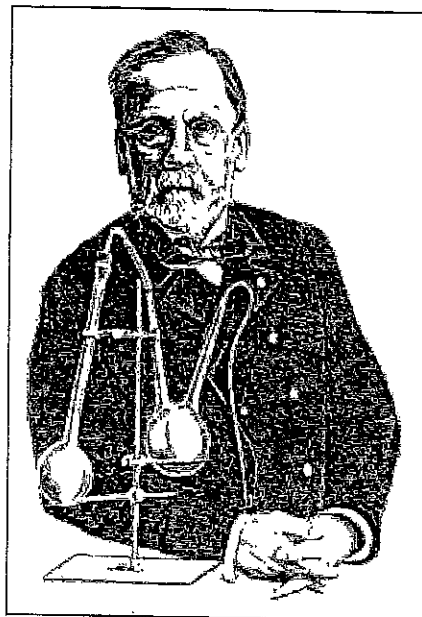


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Louis Pasteur (1822-1895)

Seung Yon Rhee



Line drawing of Louis Pasteur drawn by David Wood from Genentech, Inc. Graphics Department.

If one were to choose among the greatest benefactors of humanity, Louis Pasteur would certainly rank at the top. He solved the mysteries of rabies, anthrax, chicken cholera, and silkworm diseases, and contributed to the development of the first vaccines. He debunked the widely accepted myth of spontaneous generation, thereby setting the stage for modern biology and biochemistry. He described the scientific basis for fermentation, wine-making, and the brewing of beer. Pasteur's work gave birth to many branches of science, and he was singlehandedly responsible for some of the most important theoretical concepts and practical applications of modern science.

Pasteur's achievements seem wildly diverse at first glance, but a more in-depth look at the evolution of his career indicates that there is a logical order to his discoveries. He is revered for possessing the most important qualities of a scientist: the ability to survey all the known data and link the data for all possible hypotheses, the patience and drive to conduct experiments under strictly controlled conditions, and the brilliance to uncover the road to the solution from the results.

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On the discipline of rigid and strict experimental tests he commented, "Imagination should give wings to our thoughts but we always need decisive experimental proof, and when the moment comes to draw conclusions and to interpret the gathered observations, imagination must be checked and documented by the factual results of the experiment."

The famous philosopher Ernest Renan said of Pasteur's method of research, "This marvelous experimental method eliminates certain facts, brings forth others, interrogates nature, compels it to reply and stops only when the mind is fully satisfied. The charm of our studies, the enchantment of science, is that, everywhere and always, we can give the justification of our principles and the proof of our discoveries."

The pattern of logic in Pasteur's scientific career and the brilliance of his experimental method are well documented. It all started from studying crystal structure. As a student at the Ecole Normale, Pasteur observed that the organic compound tartrate, when synthesized in a laboratory, was optically inactive (unable to rotate the plane of polarized light), unlike the tartrate from grapes, because the synthetic tartrate is composed of two optically asymmetric crystals. With careful experimentation, he succeeded in separating the asymmetric crystals from each other and showed that each recovered optical activity. He then hypothesized that this molecular asymmetry is one of the mechanisms of life. In other words, living organisms only produce molecules that are of one specific orientation, and these molecules are always optically active.

This hypothesis was tested again by utilizing a synthetic tartrate solution that had been contaminated with mold. He found that this solution became more optically active with time and concluded that the mold was only utilizing one of the two crystals. Later in his career, he was approached with a contamination problem in alcoholic fermentation, which was thought to be an entirely chemical process at the time. After careful examination, he found that the fermenting solution contained optically active compounds and concluded that fermentation was a biological process carried out by microorganisms. This hypothesis, called the germ theory, was followed by many elegant experiments that showed unequivocally the existence of microorganisms and their effect on fermentation.

The germ theory was the foundation of numerous applications, such as the large scale brewing of beer, wine-making, pasteurization, and antiseptic operations. Another significant discovery facilitated by the germ theory was the nature of contagious diseases. Pasteur intuited that if germs were the cause of fermentation, they could just as well be the cause of contagious diseases. This proved to be true for many diseases such as potato blight, silkworm diseases, and anthrax. After studying the characteristics of germs and viruses that caused diseases, he and others found that laboratory manipulations of the infectious agents can be used to immunize people and animals. The discovery that the rabies virus had a lag-time before inducing disease prompted the studies of post-infection treatment with weakened viruses. This treatment proved to work and has saved countless lives.

All of these achievements point to singular brilliance and perseverance in Pasteur's nature. His work served as the springboard for branches of science and medicine such as stereochemistry, microbiology, bacteriology, virology, immunology, and molecular biology. Moreover, his work has protected millions of people from disease through vaccination and pasteurization.

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Lazzaro Spallanzani

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In 1770, Lazzaro Spallanzani an Italian priest and a Ph. D in philosophy set out to disprove the long held theory of spontaneous generation, that life could derive from non-living matter. Lazzaro heard about John Needham's experiment with chick broth that 'proved' spontaneous generation, but Lazzaro believed he had discovered a flaw in the experiment making it invalid. When Needham conducted the experiment he heated the chicken broth and then sealed the container, but the broth was exposed to the air before being fully sealed, the introduction of air to the chicken broth is what Lazzaro Spallanzani believed was the flaw in Needham's Experiment. Even though in his experiment John Needham had boiled away the bacteria initially, having exposed the broth once again to air, and bacteria being in the air (although this was unknown at the time) his experiment was flawed. To disprove the Needham experiment, and that spontaneous generation did not in fact exist Lazzaro Spallanzani conducted his own experiment.



Lazzaro Spallanzani



John Needham

To properly and scientifically prove that John Needham's experiment to prove that spontaneous generation existed, Lazzaro Spallanzani would do a near identical experiment, albeit not allowing the chicken broth to come in contact with the outside air. Lazzaro had two different flasks both of which were heated so that all bacteria would be eliminated from them, Lazzaro had one chicken broth sample come in contact with the air and the other did not. The results of his experiment was that the chicken broth sample that had come in contact with air had life and things growing, the same results Needham had gathered. However Lazzaro's sample that had not come in contact with air did not have life growing in it at all, proving not only that Needham's experiment was invalid but that spontaneous generation did not exist. But even though Lazzaro Spallanzani had proven that spontaneous generation did not exist it was still debated and considered possible

in some cases.

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The Microbial World And You

1. Introduction	2. History	3. The Debate Over Spontaneous Generation	4. The Golden Age Of Microbiology	5. Modern Developments Microbes and Disease
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The Debate Over Spontaneous Generation

Until the mid-1880s, many people believed in spontaneous generation, the idea that living organisms could arise from nonliving matter.

Francesco Redi demonstrated that maggots appear on decaying meat only when flies are able to lay eggs on the meat (1668). This was the first real example of modern experimentation with both experimental and control groups.

Even though Redi thought he had disproved spontaneous generation, for maggots anyway, he still believed it occurred in some cases. Everybody was aware that you could put hay in water and in a few days you'd have a bunch of those animalcules that van Leeuwenhoek kept talking about, so for years people continued to believe that microorganisms at least arose via spontaneous generation.

In 1745 John Needham claimed to show that microorganisms could arise spontaneously from heated nutrient broth. Everyone was aware that boiling animalcules would kill them, so Needham boiled broth, sealed the flasks, and got growth. He claimed that these results supported the idea of spontaneous generation.

In 1765 Lazzaro Spallanzani repeated Needham's experiments and suggested that Needham's results were due to microorganisms in the air entering his broth before he sealed the flasks. Spallanzani sealed the flasks, evacuated the air, and then boiled. When no growth occurred the conventional wisdom was that the "mysterious life force", which was required for spontaneous generation, was excluded.

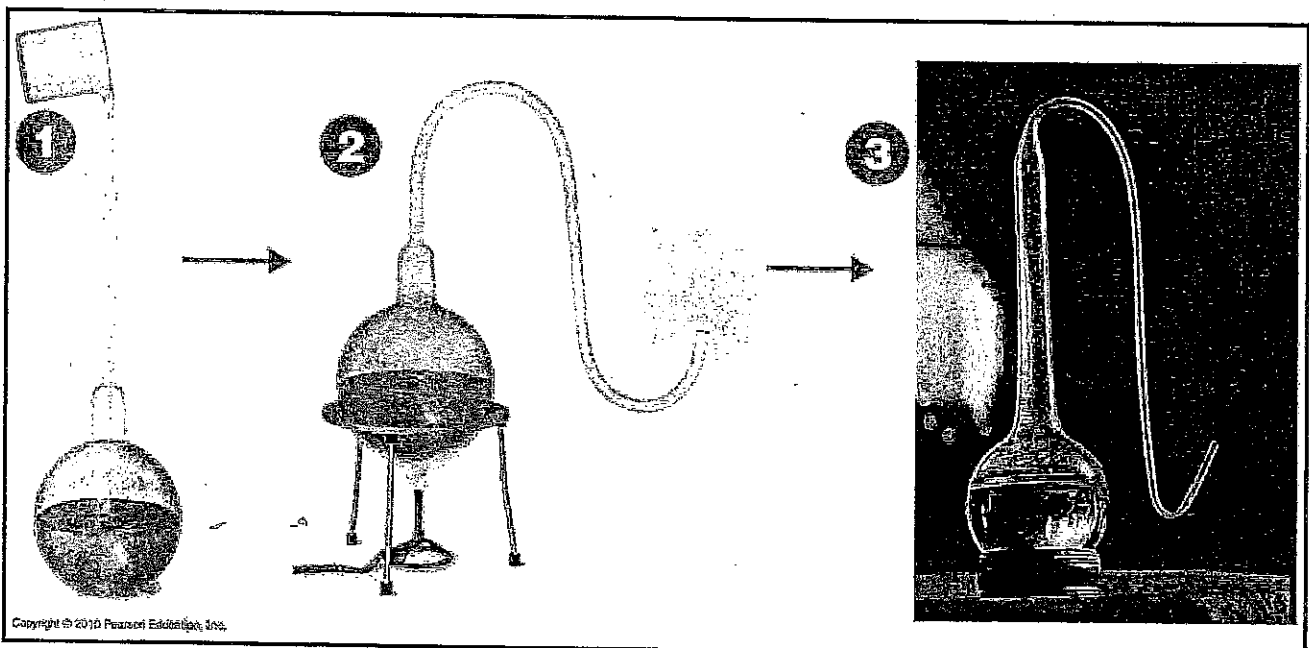
While that sounds like a bunch of hokum it was not far from the truth, at least in terms of the requirements for a number of living organisms. It was around this same time Laurent Lavoisier demonstrated the oxygen requirement of living organisms, and Spallanzani was back to square one.

In 1858 Rudolf Virchow introduced the concept of biogenesis: living cells can arise only from preexisting cells ("Life from life").

Louis Pasteur demonstrated that microorganisms are in the air everywhere and offered proof of biogenesis with a set of elegant experiments in 1861.

To allow air to enter the flasks and at the same time prevent air-borne bacteria from gaining entry, Pasteur bent the necks of his flasks after he added broth.

He then boiled the broth, killing any microorganisms that were present. If the theory of biogenesis was valid there should be no growth in the sterilized broth.



And sure enough, that's exactly what happened. As a matter of fact, some of the original flasks are still on display at the Pasteur Institute today. (The personnel in charge of the flasks did eventually seal them to prevent jokesters from trying to

blow bubbles, plug the ends with their gum, etc.)

Pasteur's discoveries led to the development of aseptic techniques used in laboratory and medical procedures to prevent contamination by microorganisms that are in the air.

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Francesco Redi and Controlled Experiments

Most people can name one 17th century Italian scientist based in Florence who challenged the widely accepted theories of Aristotle and was to influence the way Science was done in the centuries to come? There were actually two! The other besides Galileo was Francesco Redi. Francesco Redi is most famous for his excellent demonstration of the use of controlled experiments and his challenge to the theory of spontaneous generation.

When a scientist designs an experiment it is important to eliminate as many unknowns as possible. For instance, if one were trying to assess the health effects of a drug on humans, there are many factors which may affect health..simply counting how many of the patients get better or worse when given the drug is not good enough. We want to know how many got better or worse specifically from the drug. One solution might be to introduce a control to compare the drug-based tests against some standard case. In these drug-tests one group is commonly given the drug and another group, the control group, is given a placebo (commonly a sugar-pill with no known health effects). The subjects do not know which type of pill they have been given. The drug results from the test group can then be compared against those of the control group and we can get a better idea of which effects result from the drug. It is interesting that this important (and often ignored) advance in scientific methods was introduced only 25 years after the death of Galileo and only a few kilometres away from where he lived.

The Francesco Redi Experiment

Contemporaries

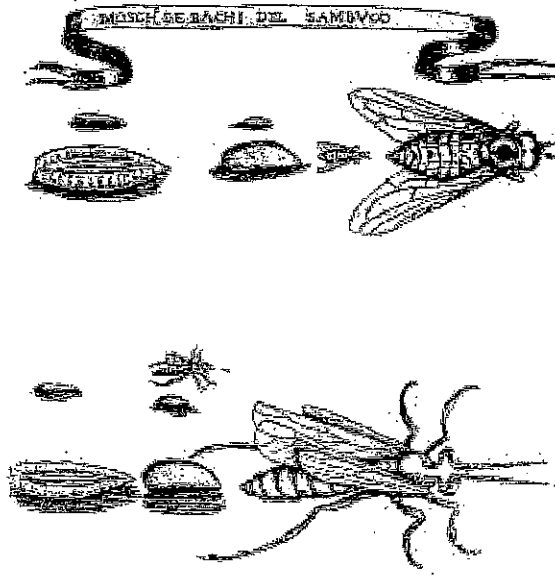
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Francesco Redi was able to disprove the theory that maggots could be spontaneously generated from meat using a controlled experiment. Spontaneous generation, the theory that life forms can be generated from inanimate objects, had been around since at least the time of Aristotle. Francesco took eight jars, placed meat in all the jars, but covered four of the jars with muslin. Maggots developed in the open jars but did not develop in the muslin-covered jars. Today controlled experiments are commonly demanded by scientific journals and are sometimes legally required by regulatory bodies (especially for pharmaceuticals). The image below is taken from *Esperienze intorno alla generazione degl' Insetti* (p. 187) where Francesco Redi published a description of the experiment in 1668 (see sidebar for digital copies of book).



Francesco Redi is often credited with developing the controlled experiment just as Galileo is often credited with introducing the modern experimental method. Both beliefs may be simplistic, however. Francesco Redi and Galileo Galileo demonstrated the effectiveness of their methods using very simple experiments, then explained their procedures and results in clear and compelling ways. These experiments were watershed events in the history of science. That is not to say that experimental scientists before Francesco Redi didn't realize the need to control variables or that the sequence of steps involved in Galileo's method hadn't been described by earlier natural philosophers (e.g. professors from the School of Padua). When Galileo was still a young boy, Giuseppe Moletti, the professor who he would eventually succeed at the University of Padua, decided to conduct some experiments on free fall by dropping weights in different media (see Timeline of

Classical Mechanics). His test with free fall in water and air specified that the balls must be of the same substance, weight and figure in order to remove doubt. In the same book, when Moletti described dropping balls of wood and lead from a tower to demonstrate that free fall doesn't depend on weight (as Aristotle had said) he was careful to eliminate size as a nuisance variable by conducting the experiment with wooden balls of different sizes. Moletti's experiments certainly weren't as elegant as Francesco Redi's, but they indicate that he understood the need for controls [1].

Being careful to control for the known variables doesn't guarantee that you will get the correct results. That is because "you don't know what you don't know". What if there are variables that need to be controlled that you don't even know exist. This could explain why the famous Tower of Pisa experiment actually came up with incorrect results. Many consider the legend of the Tower of Pisa experiment to be a myth. The experiment did occur. It was conducted by Vincenzo Renieri, a Catholic monk (see Galileo's *Battle for the Heaven's*) and not Galileo as is commonly thought. Vincenzo was a friend of Galileo's. Like Moletti before him, Renieri, controlled for size when he dropped two balls of the same size (one of wood and one of lead.) He came up with the wrong results. There was almost 2 metres difference between the heavier and lighter balls when they hit the ground. Galileo described similar results in some of his works. These scientists could not have known that they needed to control for human physiology as well. Modern experiments with humans dropping balls of markedly different weights show that there is a tendency to grip the heavier ball more tightly and release it more slowly [2].

Francesco Redi and the Galileo Affair

Francesco Redi is very important to any discussion of events relating to the Galileo and the Church. These events are most often portrayed as dramatic evidence for the "the recurring clash between religion and science" (see Galileo's *Battle for the Heaven's*). But so many of the facts relating to Francesco Redi seem to conflict with this interpretation. Francesco Redi lived a comfortable life in Florence, walking the same streets and working for the same people that Galileo did (the Medicis). He died without encountering any problems with the Church. Depending on the author, Galileo's

use of Italian instead of Latin was supposed to be a problem with the Church. But with Francesco Redi, it wasn't. Any challenge to Aristotle was also supposed to be a problem for the Church. But it was Aristotle who proposed life-forms such as maggots spontaneously generated, and Redi proved that to be false (although he continued to believe other life-forms might spontaneously generate). The Galileo Affair was supposed to have caused the decline of science in Italy, yet we we have one of the most important advances in the scientific method happening a short time later in the very same city.

The life and work of Francesco Redi provides cause to rethink the most common interpretation of the Galileo Soundbite; that it was, in essence, a conflict between science and the church. If the Galileo Affair demonstrated the inherent conflict between the church and science why wasn't this conflict repeated 25 years later when very similar circumstances arose. Could there have been other factors involved in what happened. Could Galileo's personality and his personal and professional disagreements with the other scientists of the day have played a more central role. Certainly, some of the most important historians and philosophers think so. And leaving personality aside, it could also be that Francesco Redi may have demonstrated better methodology in disproving Aristotle than did Galileo.

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Francesco Redi is often credited as being to first scientist to conduct a controlled experiment.