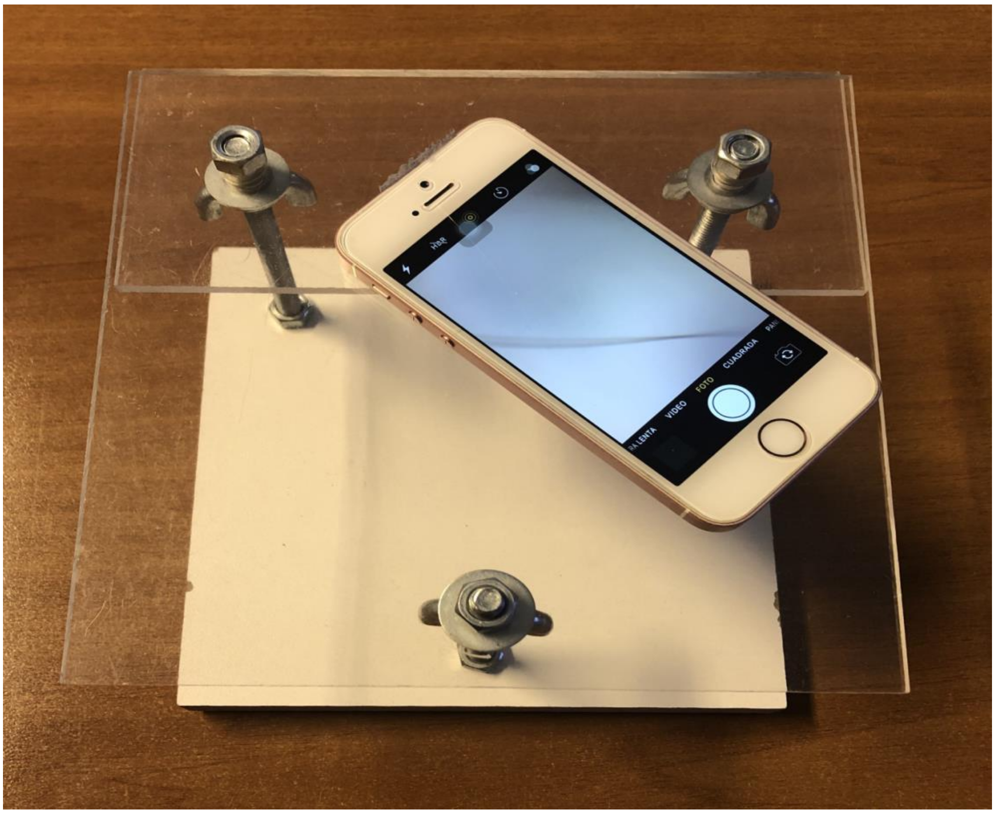
I can make my own microscope!

Miss: can we also show our families microbes?



Homemade microscope. Image by Maria José González & Paola Scavone.

María José González[1], Nicolás Navarro[1,2] and Paola Scavone[1]

[1]Departamento de Microbiología, Instituto de Investigaciones Biológicas Clemente Estable, Montevideo, Uruguay, and [2]Advanced Center for ChronicDiseases (ACCDiS), Santiago, Chile.

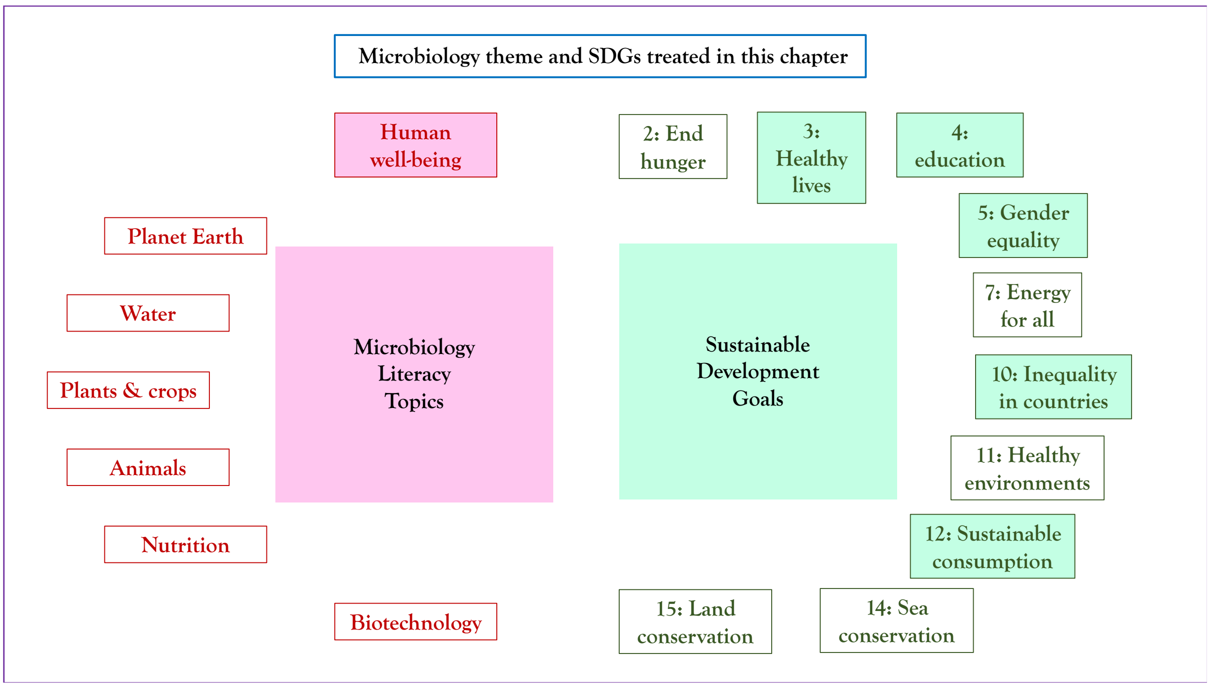
I can make my own microscope!

Storyline

Bacteria, fungi, and viruses are tiny organisms living among us. Most of them help us, a few, less so. Microbiology is the study of these teeny-tiny organisms, but how can we see them if they are so small? The size of microorganisms is around 1 micrometer (1 million part of a meter), and we cannot see them by eye. One instrument that allows the necessary magnification for observation of these small guys is the microscope. The first microscope was made in 1590 by Zaccharias Janssen. But it was Antoine van Leewhenhoek who was the first to use this instrument to observe microbes. The relevance of microscopy in microbiology is enormous as it allows us to see and understand different microbial processes and to discover which of them are the good, the bad, or the ugly. Nowadays, the evolution of microscopy permits the observation of bacteria in real time, swimming, and forming biofilms. These instruments are expensive, but we can construct a low-cost one with everyday materials. How to do it is revealed below.

The Microbiology and Societal Context

The microbiology: history of microscopy; construction of simple microscopes; accessible lenses. *Sustainability issues:* health; equitable education; gender equality; reduce inequality among countries; sustainable consumption.



I can make my own microscope!

1. ***History of Microscopy.*** The first question we might ask is, what is microbiology? And where does it come from? The etymology of the word microbiology comes from the Greek *mikros* that mean small, *bios* life and *logia* the study of bacteria, viruses, protoctists, fungi, and parasites, which are the subject of study of microbiology. All microbes have a small size, in the range of micrometers, and are not possible to see by eye. Thus, microbiology is the science that studies small forms of life. The first problem encountered by teachers is that they have to explain something that it is not possible to see by eye. Even more, children have to comprehend the subject without seeing it.

Microscopy, since its development, allowed us to discover the microbial world. Antoine van Leeuwenhoek, a Dutch businessman who we know as the Father of Microbiology, constructed his first microscope in 1670.

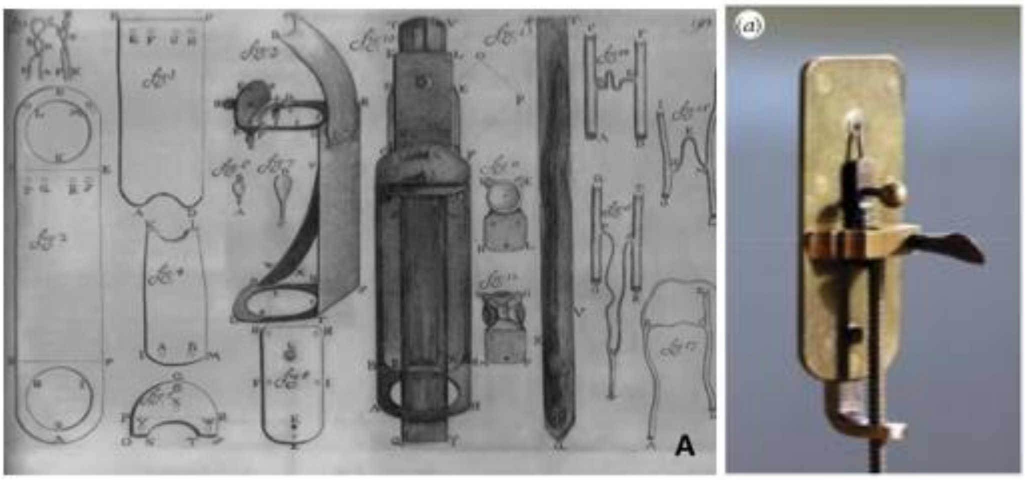


Figure 1: van Leeuwenhoek original drawings (1689) showing a microscope with its constituent parts, from Robertson, 2015. B) Replica of a single-lens microscope by Leeuwenhoek (Image by Jeroen Rouwkema. Licensed under CC BY-SA 3.0 via Wikimedia Commons).

He built more than 200 microscopes to see the fibers in the fabric he was trading (Figure 1). However, his curiosity led him to observe the small ¨animalcules¨ that inhabit his mouth; this was the first-ever observation of bacteria in history (Figure 2).

Figure 2. First drawings and observation of bacteria from Leeuwenhoek's mouth. From Leeuwenhoek, An Abstract of a Letter from Mr. Anthony Leewenhoeck at Delft dated September 17. 1683. containing some microscopical observations.

After van Leeuwenhoek, several advances were made, leading to the construction of different types of microscopes. It was in 1931 when Ernst Ruska and Max Knoll created the first electron microscope (Figure 3). In the late 1930s, electron microscopes with a theoretical resolution of 10 nm were developed (20 times higher than the resolution of optical microscopes which have a theoretical resolution of 200 nm). In the next decades, several technological improvements lead to an increase in the resolution of these devices. In 1986 a significant invention was made by Gerd Binning, Calvin Quate, and Christoph Gerber who invented the first Atomic Force Microscope (AFM) with a resolution on the nanometric scale. Nowadays, new techniques and technologies have been developed that allow correcting objective aberration, leading to increasing resolution of electron microscopes, and increased AFM resolution surpassing the nanoscale; we can even see individual molecules!

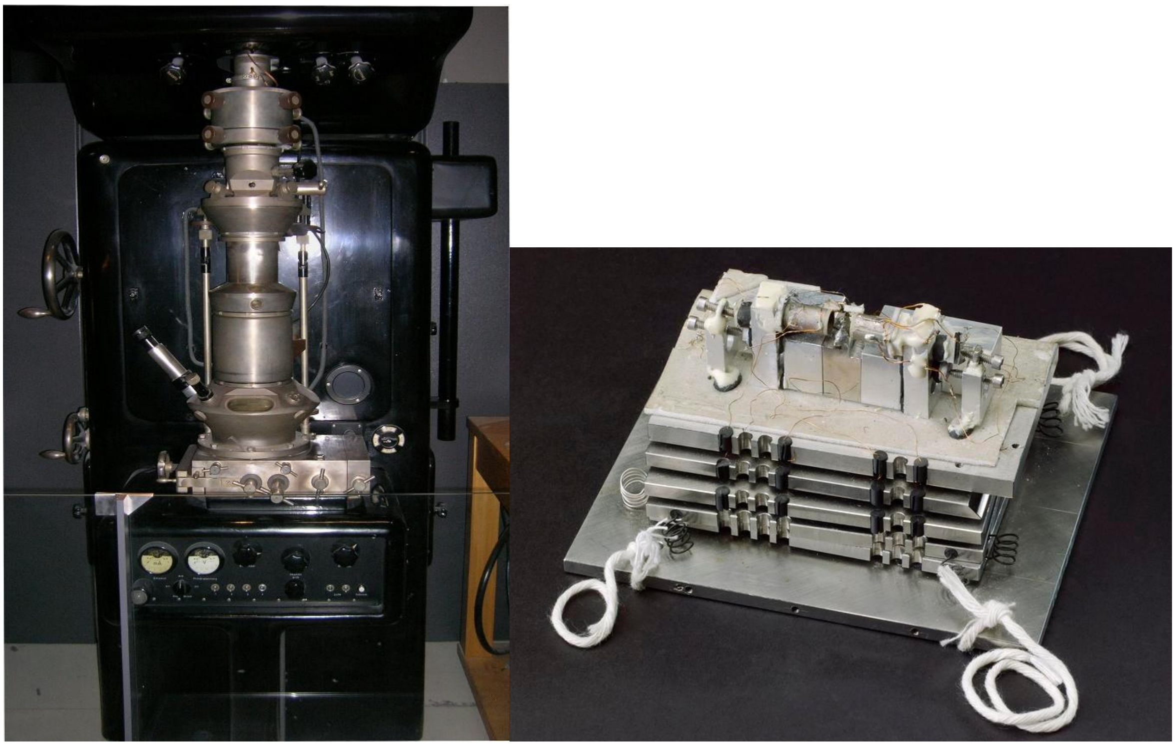


Figure 3. Replica of the first electron microscope created by Ernst Ruska and Max Knoll (Shared under Creative Commons License) and the first Atomic Force Microscope, built by Quate, Binnig, and Gerber in 1985 (© The Board of Trustees of the Science Museum).

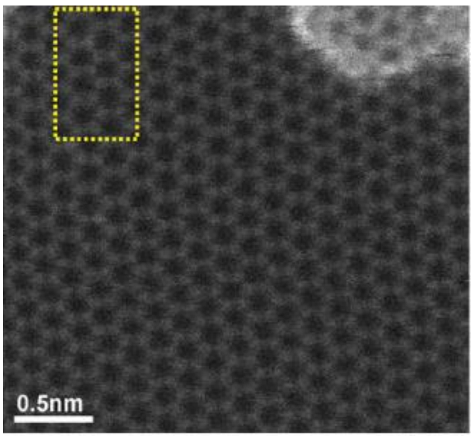


Figure 4. A) Annular dark-field imaging of a scanning transmission electron microscope (HAADF STEM) of graphene samples.

2. ***Why can we not see bacteria by eye?*** The size of a common bacterium is around 1 µm, which is one millionth of a meter, or one 10.000th part of a centimeter. Imagine that you have a ruler, so in 1 cm, you can put 10,000 bacteria in line. Now that we know the size of a bacterium, we realize that we cannot see it by eye. But why is this? Well, our eyes can see objects that are larger than 100 µm (Figure 5). This is the limit of the resolution of our eyes. So, we cannot see an object with a smaller size than that. Although there are some bacteria we can see by eye, like *Thiomargarita namibiensis*, the largest bacterium in the world, with a size of 0.2 mm, it is still really hard to see it by eye.

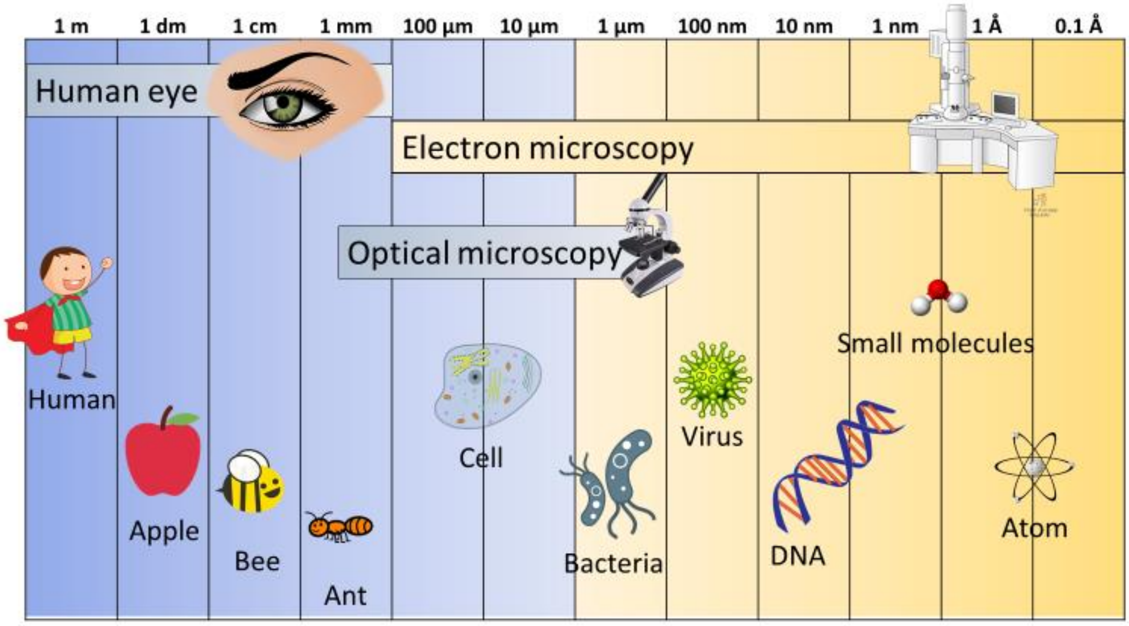


Figure 5: Scale resolution of the human eye, an optical, and an electron microscope.

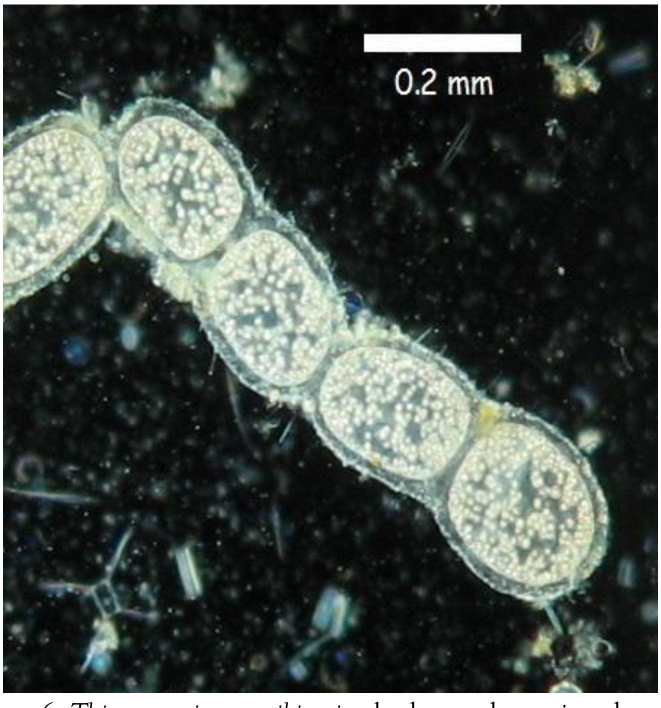


Figure 6. *Thiomargarita namibiensis*, the largest bacterium known.

3. Construction of a simple microscope

a. Materials (Figure 7)

- Three 4½ × 5/16 (11.43 x 0.8 cm) carriage bolts
- Six 5/16 (0.8 cm) nuts
- Three 5/16 (0.8 cm) wingnuts
- Three 5/16(0.8 cm) washers
- Two rubber separators
- One-piece ¾ × 7 × 7 (1.85 x 17.6 x 17.6 cm) plywood for the base
- One-piece ⅛ × 7 × 7 (0.4 x 17.6 x 17.6 cm) Plexiglas for the camera stage
- One-piece ⅛ × 3 × 7 (0.4 x 17.6 x 17.6 cm) Plexiglas for the specimen stage
- One lens (use two for increased magnification)
- One LED click light (necessary only for viewing backlit specimens)

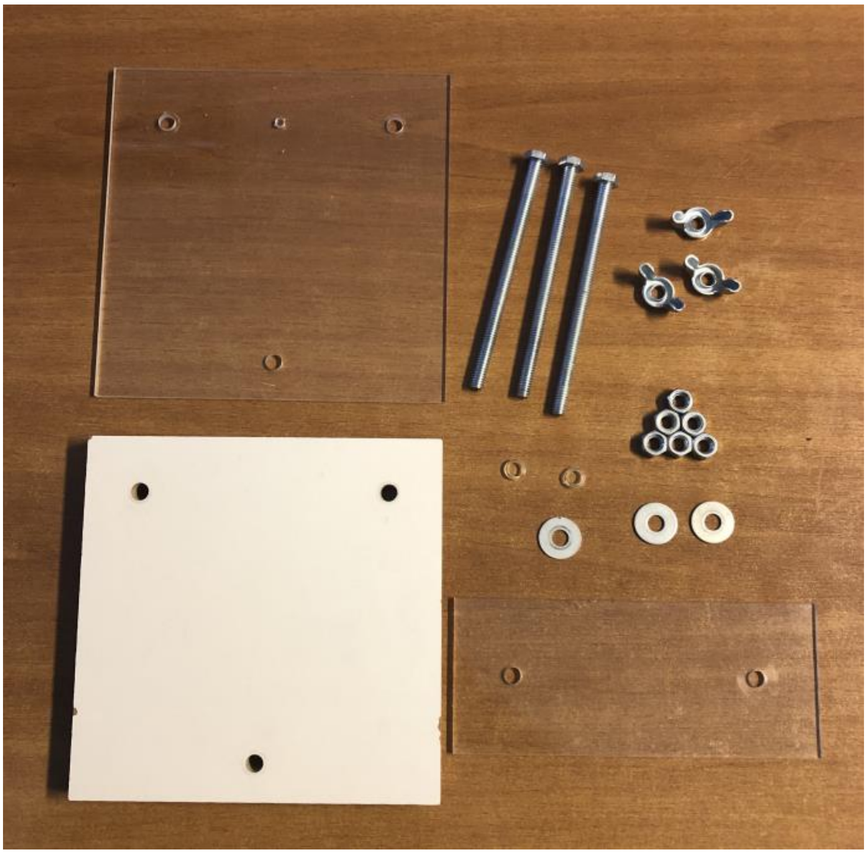


Figure 7: Materials needed for the construction of a home-made microscope.

b. Microscope assembly

First of all, you will need help to use a drill: ask an adult to do this part.

i. Make holes in the plywood base and the plexiglass plates with a drill, using a 0.8 cm drill bit. Before that, make the marks in the wood with a pencil to know where to make the holes (Figure 8). The distance between the two upper holes is 4.8 inches (12.2 cm); the distance between the upper and the lower holes is 5.7 inches (14,5 cm), making an isosceles triangle. For the small and large plexiglass plates, use the same distances as in the base so all the holes match (Figure 8). You also need to make a small hole (0.4 cm) in the large plexiglass plate where you will place the lens, precisely between the two upper holes.

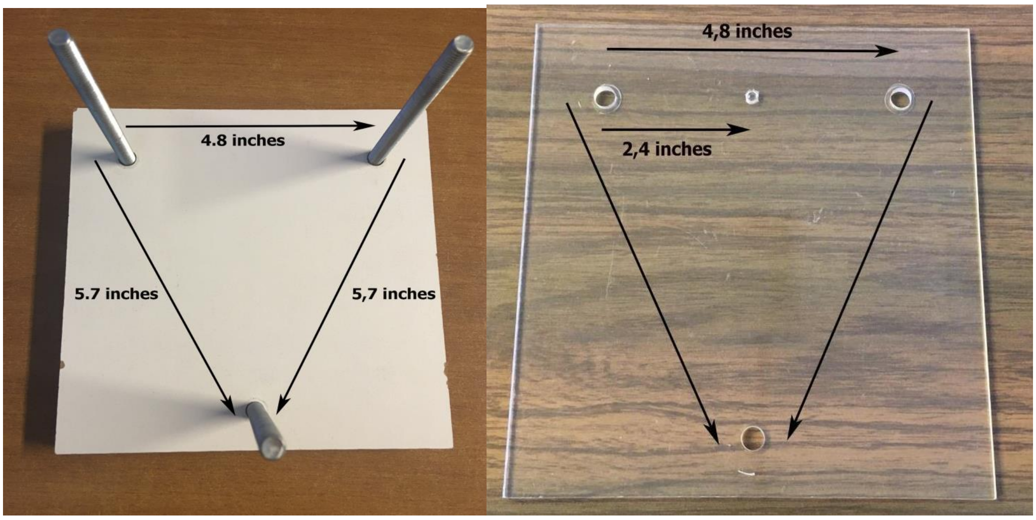


Figure 8: Distance between the holes needed in the wood base and the plexiglass. Place where the lens goes.

ii. Put carriage bolts through the holes in the plywood base, as shown in Figs. 8 & 9.

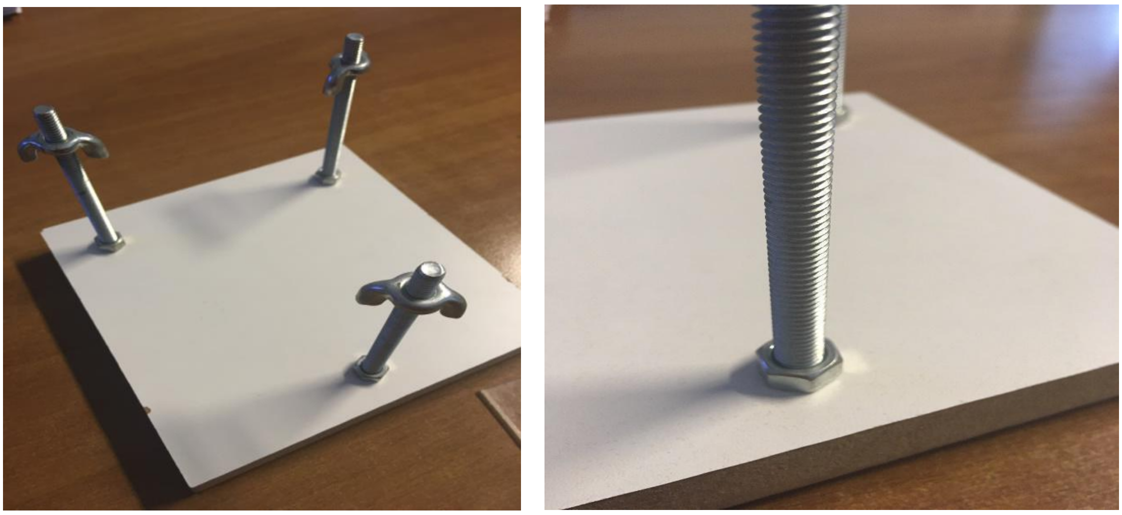


Figure 9: Bolts & wingnuts

iii. Fix the carriage bolts with the nuts, then add the wingnuts, as shown in Fig. 9.

iv. Put the metal washers on top of the wingnuts, then the small plexiglass plate, followed by the rubber separators to separate it from the large plate (Figure 10).

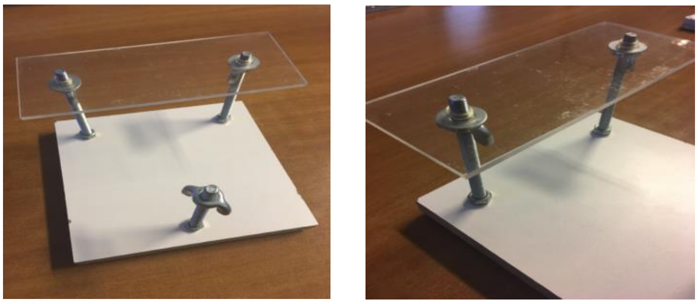


Figure 10: The small plexiglass that works as the microscope stage.

v. Finally, place the large plexiglass plate on the bolts, fix it with the remaining nuts, and insert the lens. Your home-made microscope is ready to use (Fig. 11)!

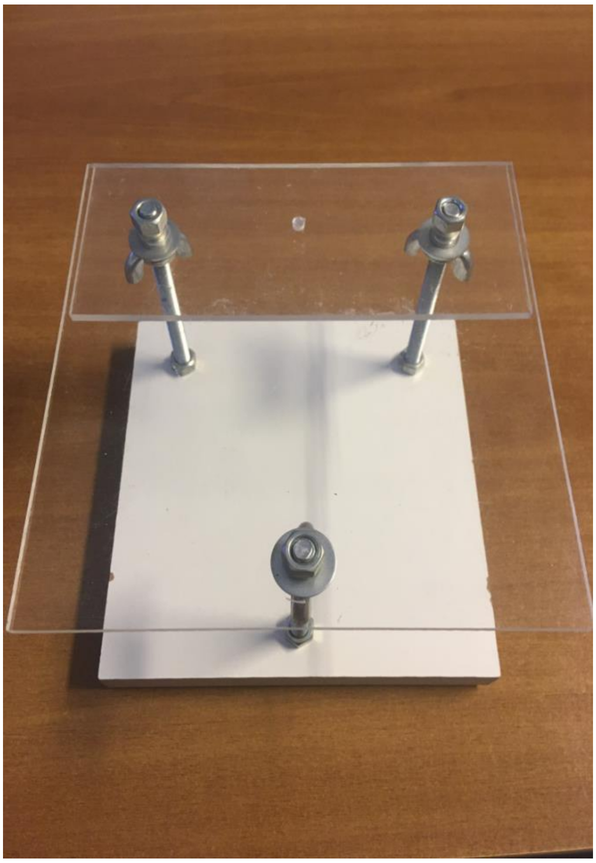


Figure 11: The home-made microscope is ready

4. ***Microscope parts and usage.*** The microscope is composed of several parts, the wooden base, the adjustable stage, and the cell phone platform where the lens is inserted (Figure 12).

a. The stage.

The stage is where the samples are placed. If glass slides are available, put the sample between the slide and the coverslip (Figure 12). Another option for samples is to put them between tape (see next chapter for recommendations for visualization and sample preparation). Place the camera of the cell phone exactly over the lens, turn on the camera, and start looking!

Probably the first image will be out of focus. How to focus on the sample? It's simple: you have to move the adjustable stage, turning both wingnuts at the same time until the image appears in focus. Then you can take a photo or make a video.

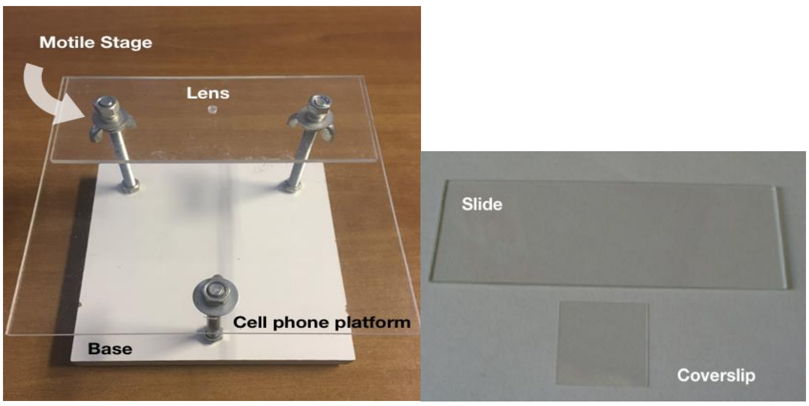


Figure 12: Home-made microscope parts, glass slide, and coverslip.

b. The lens

The other important part of the microscope is the lens, which provides magnification. You will notice that the lens is not symmetrical. You will see a thin translucent strip (about 1mm) on one side of the lens (Figure 13). This side should NOT be the side facing the camera. The correct orientation can be determined by observing the lens between the teeth of a fork or putting the lens with a hairclip in the smartphone camera and observing what happens if you use it on one side or the other. The correct orientation will give you a greater field of view. It is also possible to use two lenses to have a better magnification.

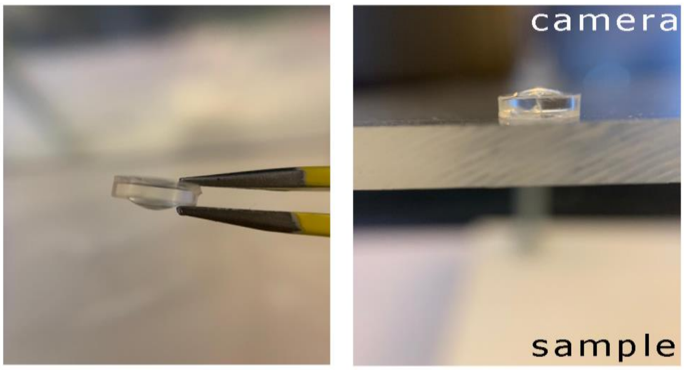


Figure 13: The lens and how to check the correct orientation.

c. Source of light (&UV led).

Sometimes supplementary lighting is needed or improves the observation. This illumination can be from a lantern, an led light, or even from another cell phone. The lighting goes below the stage, just in front of the lens, where we see the object (Figure 14). This way, when we need it, we can see the object in full view.

Figure 14: Light source

There are some materials in nature that can emit phosphorescence or fluorescence when they are illuminated with ultraviolet (UV) light. For this reason, we can use a UV led light instead of white light. This can be made very easy at home if we want to explore the world with another source of light. You only need clear adhesive tape, one blue marker, and one violet marker (see Figure 15). Put the adhesive tape over the led light of the cell phone, or other light source. Then, you paint over the light with the blue marker. Add a new tape and paint again with the blue marker. Finally, add a new tape but paint it with the violet marker. When you turn on the light of the cell-phone, the light will be violet.

Figure 15: Materials and step by step of making a UV filter.

Do you know that your fluorescent markers are really fluorescent? To test the home-made UV light, you can draw some bacteria with the fluorescent marker in a white piece of paper and illuminate with the UV filter in your cell phone (Figure 16).

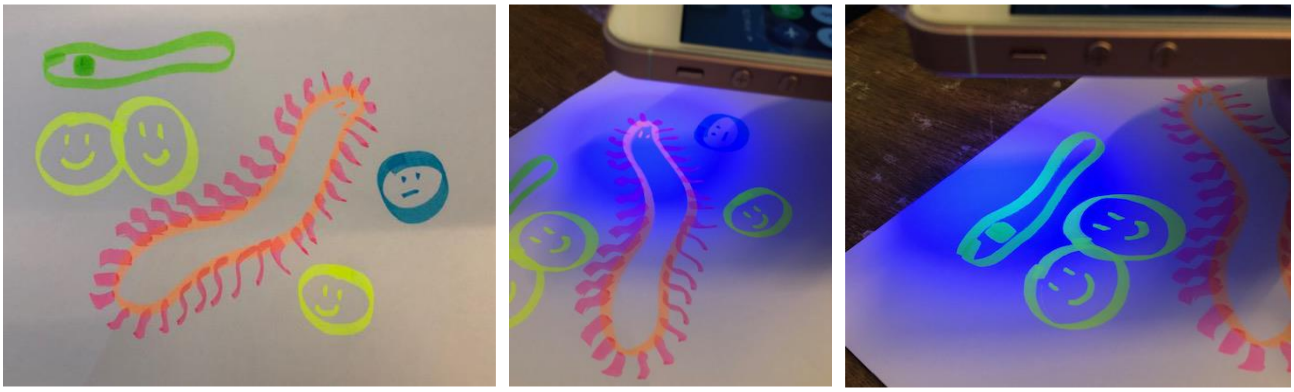


Figure 16: Some fluorescent bacteria and illumination with the UV-mobile filter.

5. ***Foldscope.*** Another affordable way to discover the microbial world is through Foldscope® (www.foldscope.com). This origami microscope was developed some years ago by **Manu Prakash** and **Jim Cybulski** to give accessibility to powerful low-cost tools instruments with 140x magnification and 2 µm resolution (Figure 17). The idea came to them on a field trip to Thailand where investigators were afraid of using an optical microscope and damaging it. Assembly of the foldscope is not complicated, but it is essential to follow the instructions carefully, as some tricky parts could lead to incorrect join up.

Foldscope microscopes are very versatile and some researchers have developed new tools for enhancing the images obtained with this type of "cell phone microscopy." Jawale et al. developed an Open Source 3D-printed focusing mechanism to control the focus of a ball lensbased microscope for cellular microscopy (Figure 18). These devices could help to improve the reproducibility of focal positioning in clinical and classroom contexts.

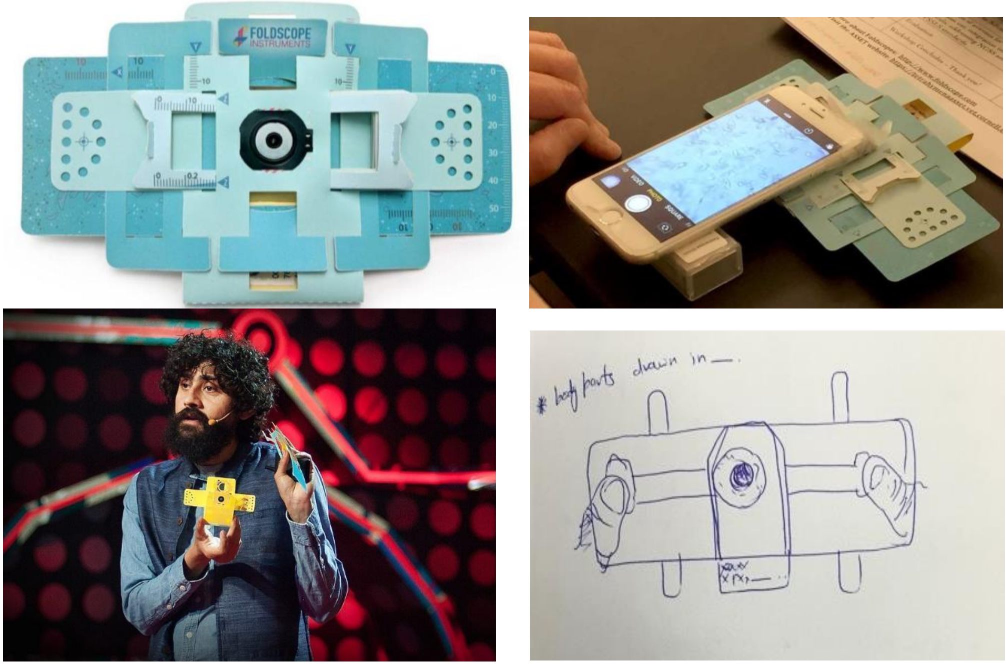


Figure 17. Foldscope® images and creator Manu Prakash (Shared under Creative Commons License).

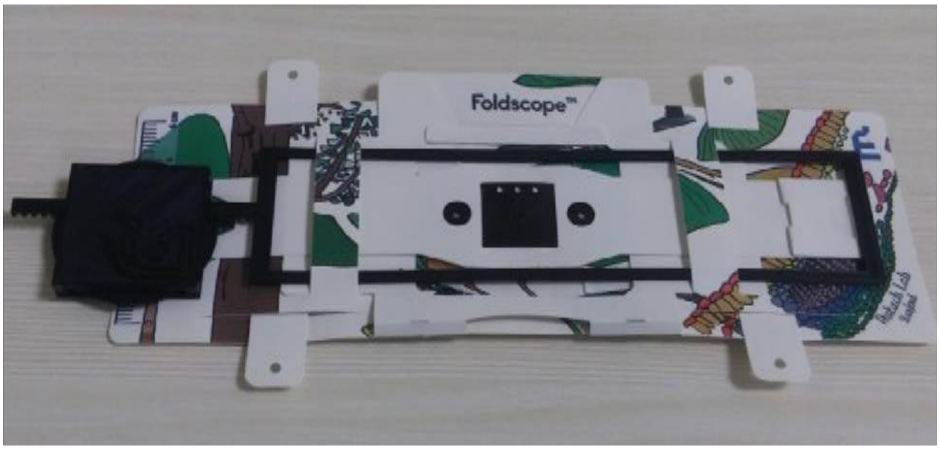


Figure 18. 3D-printed focussing mechanism for Foldscope microscope.

Relevance for Sustainable Development Goals and Grand Challenges

Since the UN articulated the Sustainable Development Goals agenda, major efforts are being made to achieve a better and more sustainable future. Educational resources contribute to these goals, and some examples of these in relation to simple microscopes are given here:

- **Goal 3. Ensure healthy lives and promote well-being at all ages.** The study of the microbial world that the microscopy enables us to access helps to understand the importance of washing our hands, drinking potable water, among other practices that directly affect our well-being.

- **Goal 4. Ensure inclusive and equitable quality education and promote lifelong learning opportunities for all.** One of the major concerns in the educational field is the quality and access to education at every level. For many reasons, there are millions of out-of-school children in the world. But it is essential that even those children can access tools to learn how our world works. These activities can be done not only at school, but also at home with the family, thereby extending education into the home and promoting lifelong learning. The development of 21-st Century skills contributes to SDG 4. The scientific method enables us to discover the world around us, and the use of a home-made microscope will induce children to pose hypotheses and objectives, design an experiment, observe, and to have conclusions. For this reason, STEM (Science, technology, engineering, and mathematics) skills must be included if we want to empower the next generation to address the future and global challenges.
- **SDG 5. Achieve gender equality and empower all women and girls.** The search for knowledge must be part of the equitable education of children and young people, and a key element of strategies to empower girls. All materials and resources that bring children closer to science must be accessible for both girls and boys.
- **SDG 10. Reduce inequality within and among countries.** To advance towards equal opportunity and promote the social, economic, and political inclusion of all, irrespective of age, sex, disability, race, ethnicity, origin, religion, or economic or another status, the means have to be free and easy to access. STEM education requires expensive laboratory equipment for teaching through experimentation, so the possibility of creating a personal microscope at home allows people from different social strata to have access to scientific knowledge. This enables and promotes the democratization of science at all levels.
- **SDG 12. Ensure sustainable consumption and production patterns.** Learning to make home-microscopes leads to learning to use materials we can find in every home, materials we will recycle and give new value, before discarding. The children will learn how sustainable consumption and production is about doing more and better things with less. It is also about decoupling economic growth from environmental degradation, increasing resource efficiency, and promoting sustainable lifestyles. It also helps us to change the way we think to be more conscientious about the importance of our planet. We need the next generation to be prepared for future challenges.

Potential Implications for Decisions

1. ***Individual***
    a. Encourage children to learn about the micro world.
    b. 21st Century skills development.
    c. Stimulate the capacity for wonder that children already possess.

2. ***Community policies***
    a. Educate the population about the importance of understanding the world that surrounds us.
    b. Generate equitable spaces for learning.
    c. Generate affordable tools for children so they can learn with their family, not only in the educational centers.

Pupil Participation

1. ***Class Discussion***
   - ***a.*** What do we want to see with our home-made microscope?
   - ***b.*** Do you think it is important to know the microbial world? Why?

The Evidence Base, Further Reading and Teaching Aids

The first microscopes: Zaccharias Janssen, Hans Lippershey, and Antony van Leeuwenhoek: https://www.aps.org/publications/apsnews/200403/history.cfm

Dobell C. Antony Van Leeuwenhoek and His "Little Animals". (1960) Dover Publications INC, NY, USA 239–255

Lesley A. Robertson (2015), van Leeuwenhoek microscopes—where are they now? *FEMS Microbiology Letters*, Volume 362, Issue 9, May 2015, fnv056, https://doi.org/10.1093/femsle/fnv056

Leewenhoeck A. (1684) An Abstract of a Letter from Mr. Anthony Leewenhoeck at Delft, dated September 17. 1683. containing some microscopical observations. Phil. Trans. 14, 568 – 574.

Science Museum Group. First atomic force microscope, United States, 1985. Science Museum Group Collection Online. Accessed September 13, 2020. https://collection.sciencemuseumgroup.org.uk/objects/co8685/first-atomic-force-microscope-united-states-1985-atomic-force-microscope

Kepaptsoglou, D., Hardcastle, T. P., Seabourne, C. R., Bangert, U., Zan, R., Amani, *et al.* (2015). Electronic structure modification of ion implanted graphene: the spectroscopic signatures of p- and n-type doping. *ACS nano*, *9*(11), 11398-11407.

Institute for Chemistry and Biology of the Marine Environment, University of Oldenburg, accessed on September 2020 http://www.pmbio.icbm.de/mikrobiologischer-garten/eng/enanswer038.htm

Cybulski, J. S., Clements, J., & Prakash, M. (2014). Foldscope: origami-based paper microscope. PloS one, 9(6), e98781.

Jawale, Y. K., Rapol, U., & Athale, C. A. (2019). Open Source 3D-printed focussing mechanism for cellphone-based cellular microscopy. Journal of microscopy, 273(2), 105-114.

Foldscope: https://www.foldscope.com/our-story

Glossary

Bacteria are single-celled microscopic organisms, not visible with the naked eye, that can be found everywhere.

Lens is a curved glass that is used to magnify an object.

Objectives are the optical element that gathers light from the object being observed and focuses the light rays on producing a real image.

Resolution the minimum distance at which two distinct points of a specimen can still be seen - either by the observer or the microscope camera - as separate entities.