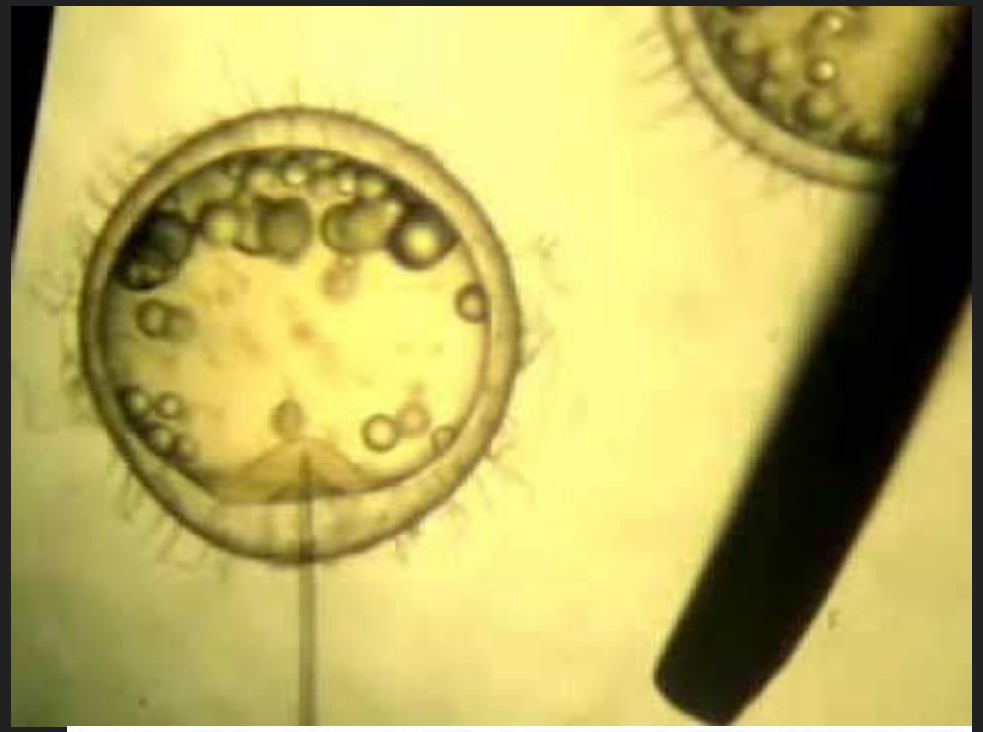
Microinjection

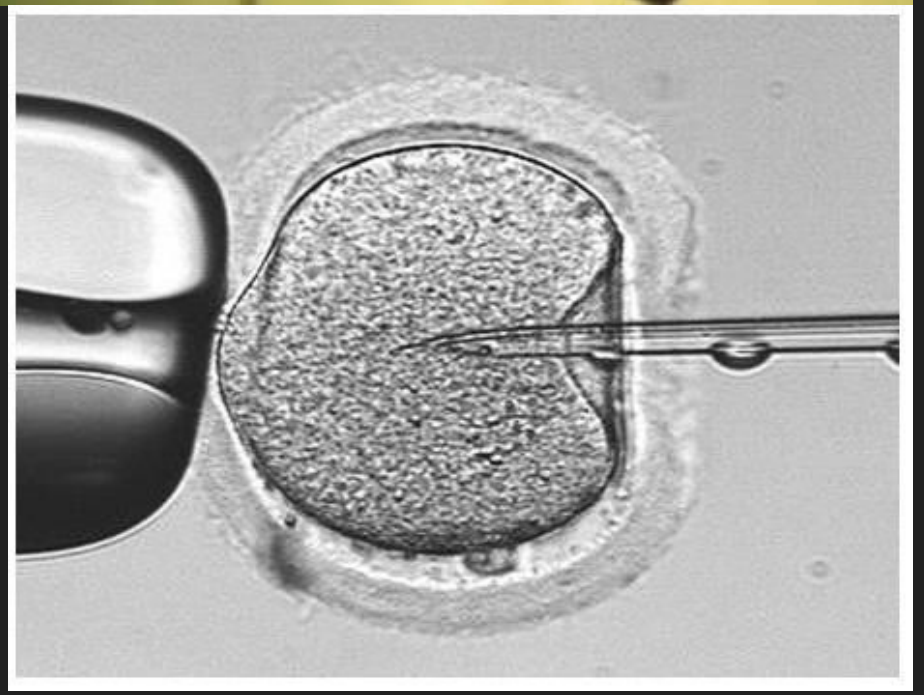
B.Sc. 4th Semester

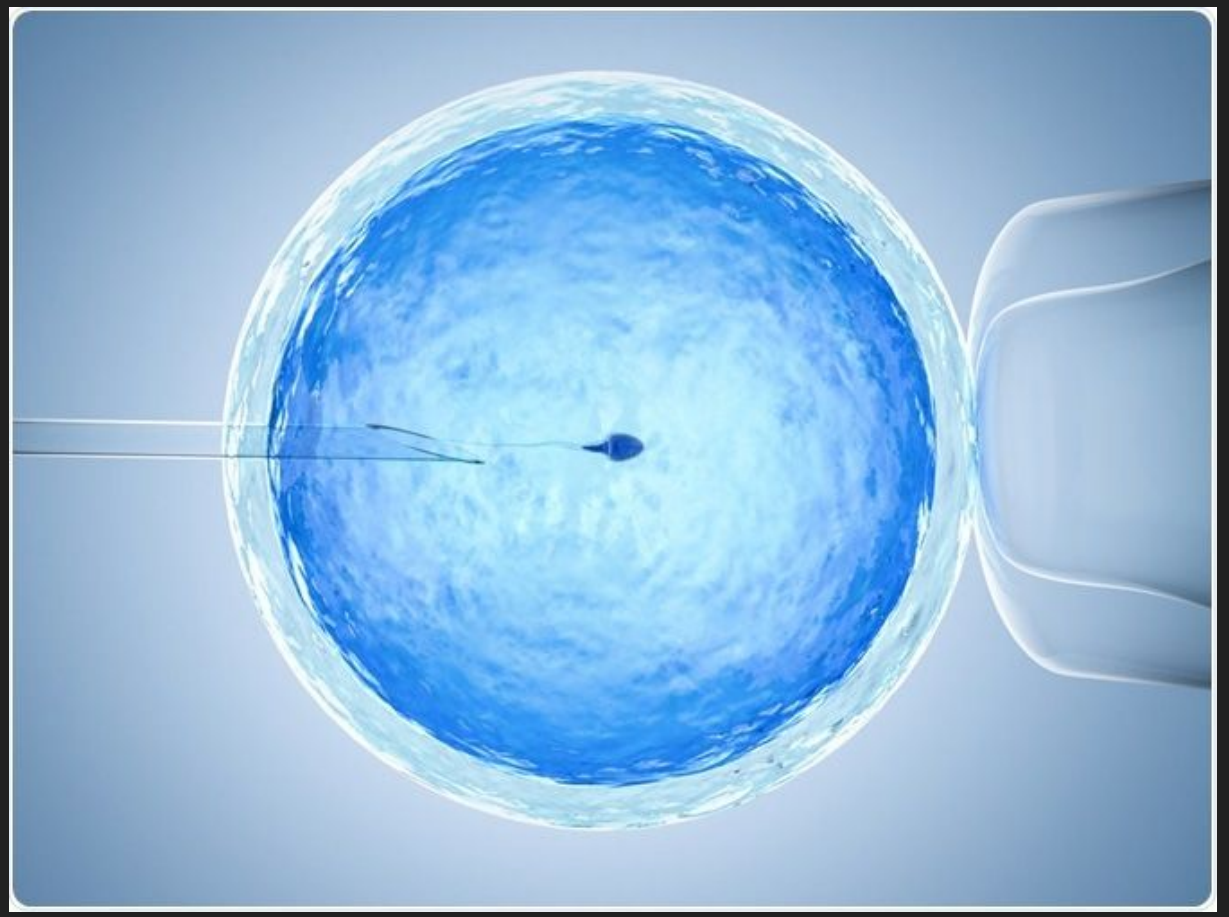
A hundred years ago, Dr. Marshall A. Barber proposed a new technique - the **microinjection** technique. He developed this method initially to clone bacteria and to confirm the germ theory of Koch and Pasteur. Microinjection can also be used in the cloning of organisms, in the study of cell biology and viruses, and for treating male subfertility through intracytoplasmic sperm injection.
There are two basic types of microinjection systems. The first is called a *constant flow system* and the second is called a *pulsed flow system*.

Microinjection is a direct method to introduce DNA into either cytoplasm or nucleus. It is a microsurgical procedure conducted on a single cell, using a glass needle (i.e., a fine, glass microcapillary pipette), a precision positioning device (a micromanipulator) to control the movement of the micropipette, and a microinjector. Extrusion of fluid containing the genetic material through the micropipette uses hydrostatic pressure. Injections are typically carried out under direct visual control, using a microscope. The small tip diameters of these micropipettes, combined with the high precision of the micromanipulator, allow accurate and precise DNA delivery. Conceptually, microinjection is the simplest gene delivery method. However, it is difficult to apply. Although pronuclear injection of DNA is very efficient, it is a laborious procedure; only one cell at a time can be injected, typically allowing for only a few hundred cells to be transfected per experiment. The cytoplasmic injection of DNA has been observed to be less effective probably because of cytoplasmic degradation of DNA by cytoplasmic nuclease enzymes.

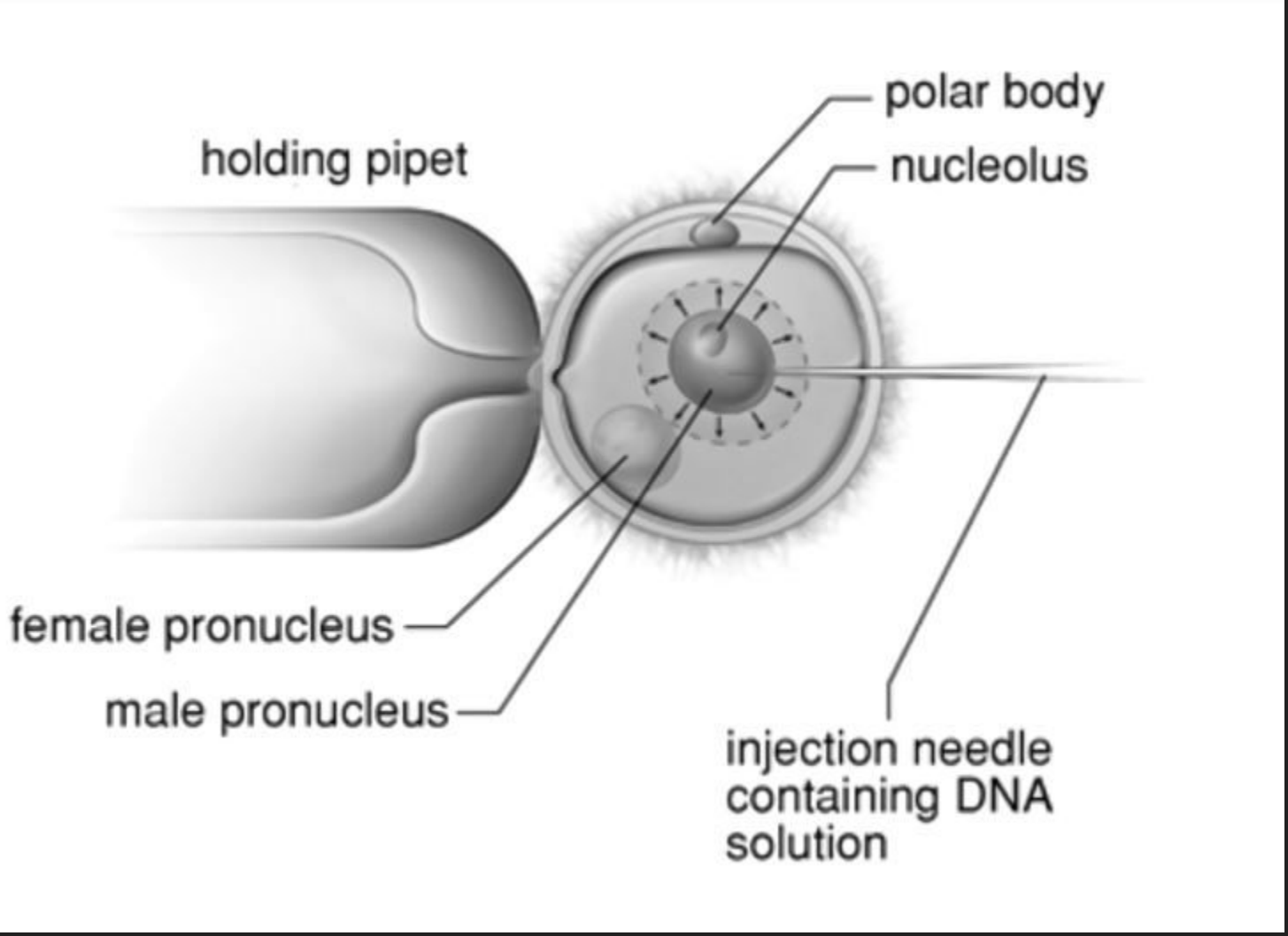
Microinjection is a simple mechanical process usually involving an inverted microscope with a magnification power of around 200x (though sometimes it is performed using a dissecting stereo microscope at 40–50x or a traditional compound upright microscope at similar power to an inverted model).








Plan Apo
1x


polar body
holding pipet
nucleolus
female pronucleus
male pronucleus
injection needle
containing DNA
solution