

- Gene transfer research with fish began in the mid 1980's utilizing microinjection
- Zhu *et al.* (1985) published the first report of transgenes microinjected into the fertilized eggs of goldfish. In almost all fish gene transfer research, the foreign gene was microinjected into the cytoplasm of one-to- four cell embryos as pronuclei are extremely difficult to visualize in live one-cell fish embryos.
- An average of about 5% of the surviving microinjected embryos integrate the foreign DNA.
- Microinjection is a tedious and slow procedure and can result in high egg mortality.

• After the initial development microinjection, new techniques such electroporation, retroviral integration, liposomal-reverse-phase-evaporation, spermmediated transfer and high velocity microprojectile bombardment were developed that sometimes can more efficiently produce large quantities of transgenic individuals in a shorter time period.

#### Electroporation

• Electroporation involves placing the eggs in a buffer solution containing DNA and applying short electrical pulses to theoretically create a transient openings of the cell membrane, allowing the transfer of genetic material from solution into the cell. The efficiency of the electroporation is affected by a variety of factors including voltage, number of pulses and frequency of pulses.

- The first successful gene transfer utilizing electroporation produced integration rates and survival similar to that for microinjection, then demonstrated that electroporation can be more efficient than microinjection with integration rates sometimes as high as 30-100%.
- Hatching rates were higher for electroporated embryos than for microinjected fish embryos, and post-fertilization electroporation treatments had higher hatching rates than electroporation of sperm and then eggs prior to fertilization.

• The introduction of exogenus DNA into the host genome resulting in its stable maintenance, transmission and expression.

• The first Indian transgenic fish was produced in 1991 by Dr. T. J. Pandian in carps.