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Transgenic animals

A transgenic animal is the one that contains a cloned gene in all of its cells. Knock-out mice used to study the function of human and other mammalian genes are the similar to transgenic animals but in such animals a particular natural gene is deleted. There are several different approaches of creating transgenic animals as depicted in the diagram below. On the other hand the term chimera is used for that animal in which all the cells do not carry the cloned gene.

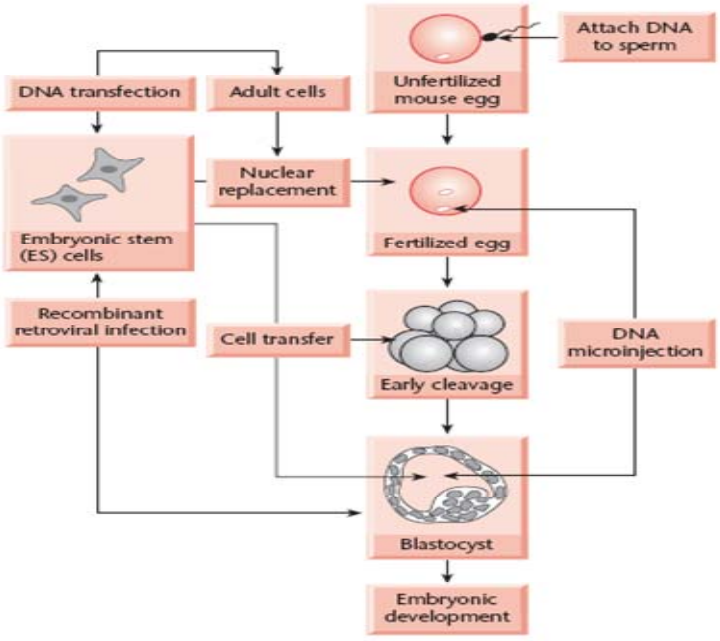

Fig: Different approaches of making transgenic animal.

1. Attachment of DNA to sperm: The gene to be inserted into the animal is inserted into the genome of sperm that is then allowed to fertilize the ova. The ova will now develop to a transgenic animal.
2. Embryonic stem cells (ES cells): The embryonic stem cells are the cells that have the capability to differentiate into any type of cell. These cells are cultured in vitro and DNA is transfected into the cell culture of ES cells. The nucleus of these transfected cells are taken out and replaced with the nucleus of fertilized egg. The egg now contains the 2n nucleus of ES cells. The ova will be allowed to go for further embryonic development that will develop to a transgenic animal.
   Alternatively whole of the ES cell can be transferred to blastocoel of blastocyst stage of embryo. In this case as all the cells of the embryo will not be having the cloned gene. Such embryo will develop to a chimera instead of transgenic animals.
3. DNA microinjection: The external gene can be transferred to cell directly via process called DNA microinjection. As shown in the figure, the DNA microinjection can be

ensued in either of two stages. Stage I: at the stage of zygote. Stage II: at the stage of blastocyst (at this stage, the animals will be chimera)

4. Recombinant Retroviral infection: The recombinant retrovirus where the gene of interest is inserted into its genome is infected to the ES cells or at the stage of blastocyst as shown in the figure. When the retroviral infection is performed in the stage of blastocyst, the animal will develop to chimera.

Biopharming:

Biopharming refers to production of recombinant protein in transgenic animals. For instance, production of human insulin protein in milk of transgenic cattle. This is done by incorporating the human insulin gene in the genome of an animal (for instance cattle). For this purpose, the appropriate promoter is to be selected that has to be placed before the gene of interested that is to be incorporated into the genome of the animal.

Say for instance, if the human insulin gene is tagged with the casein promoter (promoter of the casein gene) than the insulin gene will be expressed in the milk of cattle. The reason behind this is that the casein gene is expressed only in the mammary gland owing to the casein promoter. So, if the foreign gene is tagged with the casein promoter, it would also express in mammary gland only.

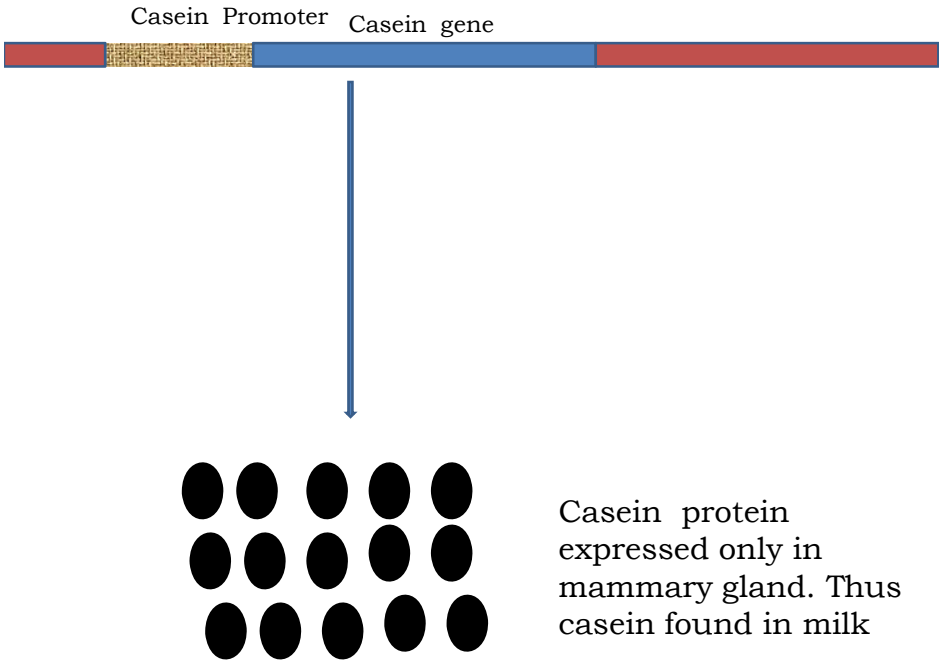

Fig 1. The casein gene is expressed only in mammary glands.

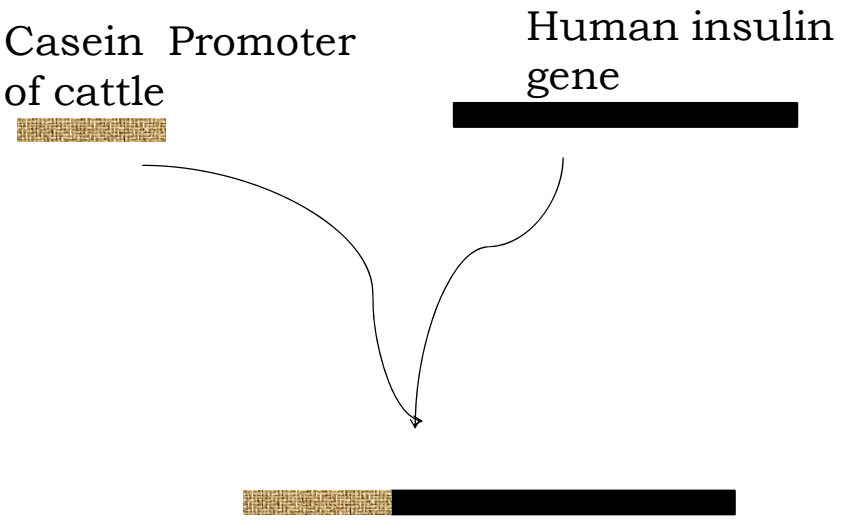

Fig 2. The human insulin gene is purified and the casein promoter of cattle casein gene is tagged before the human insulin gene.

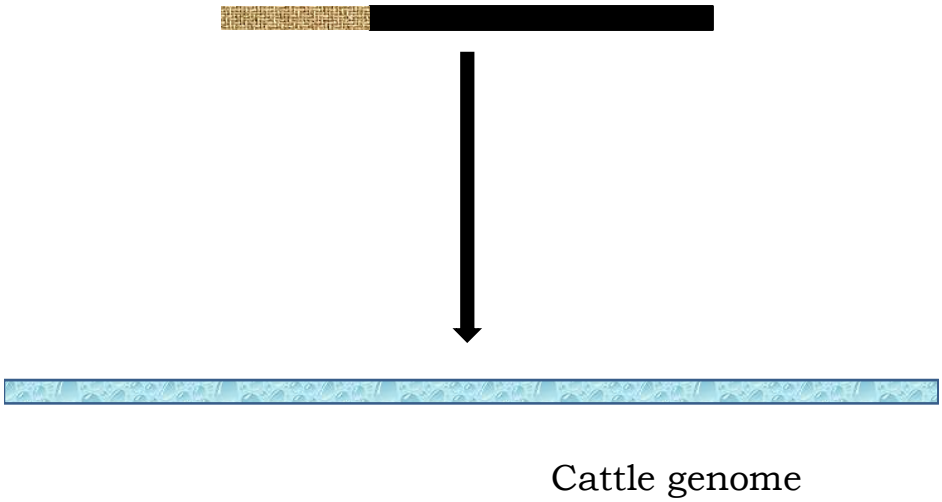

Fig 3. The human insulin gene tagged with casein promoter of cattle is inserted into the genome of cattle.

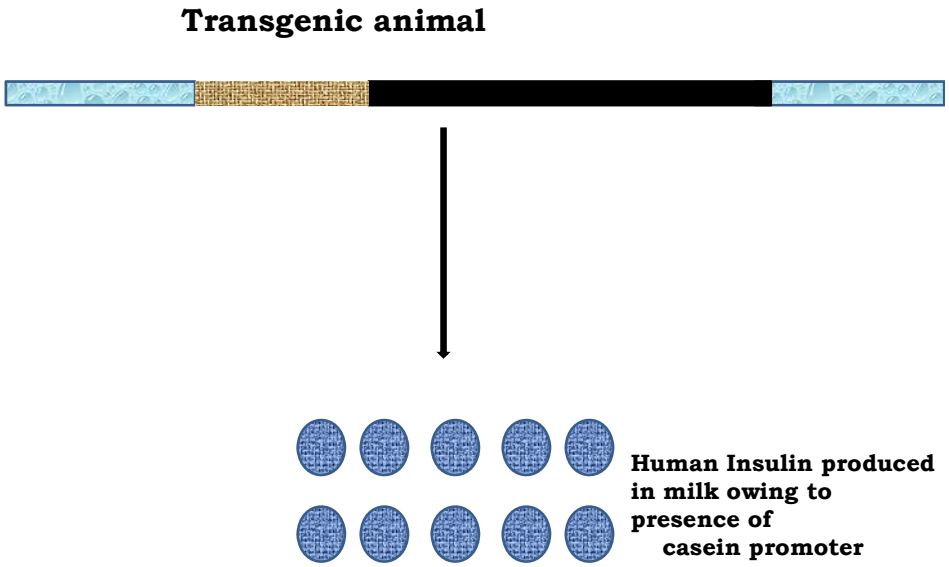

Fig 4. The human insulin gene after being inserted into the cattle genome is expressed only in milk of cattle owing to presence of cattle casein promoter. After the human insulin gene is inserted into the cattle genome, the cattle is said to be **transgenic cattle**.