# Chasing chimeras: The challenge to identify rare individuals containing cells with distinct genomes.

**Durgadas P Kasbekar, FASc,**
**Centre for DNA Fingerprinting and Diagnostics, Hyderabad**
**Email : kas@cdfd.org.in**

In 2018, the *Journal of Assisted Reproduction and Genetics* reported the case of a couple in America who had undergone a successful IVF (*in vitro* fertilization) procedure at a fertility clinic. To everyone's consternation, a DNA test turned up a negative paternity result for the man. It excluded him as the baby's father. This raised the unsettling possibility of a sample mix-up. However, no evidence could be found of wrongdoing by the clinic. A team of researchers, including scientists from the Massachusetts General Hospital and Stanford University School of Medicine, then set out on their "quest for truth". The team analysed additional tissue samples from the man. The analysis revealed him to be a chimera. That is, he harboured cells bearing two distinct genomes. The sperm cell responsible for conceiving his child had a different genome than his buccal or peripheral blood cells used for the paternity tests, and hence the tests reported non-paternity (https://doi.org/10.1007/s10815-017-1064-6). The fertility clinic was exonerated of charge of sample mix-up, and, to the family's relief, his paternity was affirmed.

The case showed that some allegations of fertility clinic missteps might be accounted for by undiagnosed chimerism. It also highlighted the need to create guidelines to help identify and rectify cases of false exclusions of paternity.

**An unknown unknown.**

Most of us began life as a single cell called the zygote. The zygote was produced when the father's sperm fused with the mother's egg. Beginning with the zygote, a sequence of cell divisions then generated all the other cells of our body, including those of the adult. Consequently, all of our cells, whether from blood, teeth, hair, spit, sweat, or any other tissue, have the same DNA. It is the same as the DNA in the zygote.

Occasionally, however, the mother simultaneously releases two eggs, and their fertilization by two sperms creates two zygotes. Each sets out on its pathway of baby development. Twins produced in this way are called dizygotic twins, and they differ in their DNA as would any other pair of siblings.

Two embryos developing in parallel, however, have the possibility of exchanging cells. Thus, one or both of them can contain cells derived from the other's zygote. In extreme cases the two embryos can even amalgamate to become one. Chimeras are individuals with cells from more than one zygote. Embarrassingly, we do not know how frequently this can happen among humans.

The original Chimera was a fantastic monster in Greek mythology. It possessed a lion's head, a goat's body, and a serpent's tail. Human chimeras reported in the medical

literature are not quite as fantastic. In fact, most appear to be indistinguishable from non-chimeric individuals (https://doi.org/10.1016/j.ejmg.2020.103971 ).

Ordinarily, zygotes bearing the XX and XY sex-chromosome constitution develop, respectively, into females and males. When cells from XX and XY twin zygotes mix, a fraction of the chimeras produced show abnormalities of sex development, and some are even true hermaphrodites with both ovarian and testicular tissue. Hence such chimeras are more likely to be discovered and reported. Chimeras from same-sex zygotes, in contrast, are detected only accidentally, for example during routine blood-group testing. Therefore, the medical consequences of chimerism are still not fully known.

IVF procedures involve the physical manipulation of multiple zygotes. It has been suggested that this can increase the risk of cells from two embryos coming together to form chimeras. Given that more and more couples now resort to IVF to raise families, chimera frequency might show an uptick in the coming years.

**A monkey's aunt.**

Marmosets nearly always give birth to twins with extensive exchange of cells between them. Chimerism in marmosets was found to even extend to their germ cells. That is, the cells which produce the sperm and eggs. One female marmoset, whose uterine twin was male, even produced offspring that had inherited her sibling's genes. That is, her egg cell was derived from her male sibling's zygote. This effectively made the baby marmoset's mother also its biological aunt ( 10.1073/pnas.0607426104 ). These findings showed that an XY germ cell was capable of maturing and producing viable eggs in a female primate, and raised the possibility that a female could pass on a Y chromosome to her offspring.

However, so far there have been no reports of germ cell chimerism in human sex-discordant twins.

**So close, yet so far.**

The Centre for DNA Fingerprinting and Diagnostics (CDFD) in Hyderabad offers DNA-based investigative services to hospitals that conduct organ transplant procedures. It handled the case of a father who offered to donate an organ to his ailing son. DNA profiles were generated of the donor, the patient, and the patient's mother. While the mother and son profiles were consistent with their claimed mother-son relationship, those of the father and son were not. Not all of the patient's paternal genes were found in the DNA of the woman's husband. This implied he was not the actual father.

However, the DNA profiles also showed the actual father was closely related to the woman's husband. Possibly, even an amalgamated twin brother. Since both donor and recipient belonged to the same family, the family could go ahead with the transplant.

The scientists were excited by the prospect that the woman's husband could be a chimera. However, an alternative possibility was that the patient was the product of a levirate marriage. Levirate marriage is a custom practiced in some communities in India and abroad in which a woman who is widowed, or one whose husband is mentally or physically incapacitated, has children fathered by her husband's brother.

Had the scientists interviewed the family or their doctor they could have asked whether the woman's husband's had a brother who putatively was the patient's father. But the imperative to respect a family's privacy ruled out such an interview. Since the could not rule out levirate marriage, the scientists had to forgo the chance of discovering another father who was also his child's uncle.