CHIMERISM IN A TWIN PREGNANCY

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INTRODUCTION

Chimerism is an exceptional event, produced by the fusion of two different zygotes. The result is the presence of two genetically distinct cell lines in the same organism. They are usually discovered because the two cell lines have different sexual chromosomes, i.e. 46,XX/46,XY.
Less than 40 cases have been described in literature. As a matter of fact, only a small number have been studied using microsatellite markers.
We report on a new tetragametic chimera case with the particularity of having been found in a twin pregnancy.

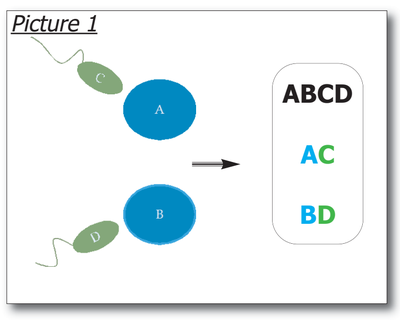
DATA ON 46,XX/46,XY

Phenotype

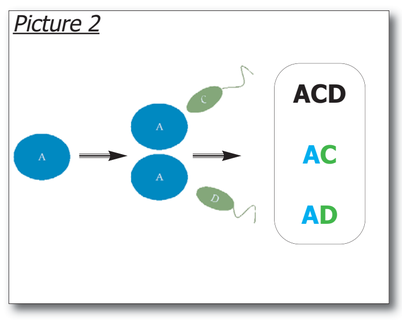
Sexual determination: The phenotype may be a **normal female or a normal male, but ambiguous genitalia** are often observed. 13% of true hermaphroditism (ovotestis) have a 46,XX/46,XY karyotype, with a 3% risk of gonadoblastoma. **Sterility** is usual. There is **only one case reported with an abnormal phenotype** including mental retardation. The abnormal phenotype is linked to the mechanism of formation of the chimera (see below mechanism of chimera).

4 mechanisms of formation of 46,XX/46,XY

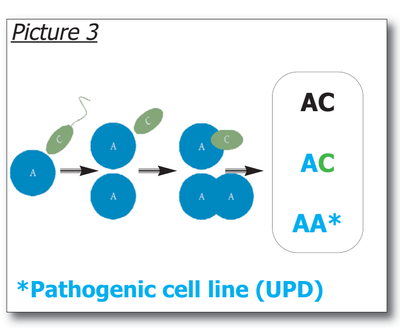
Picture 1



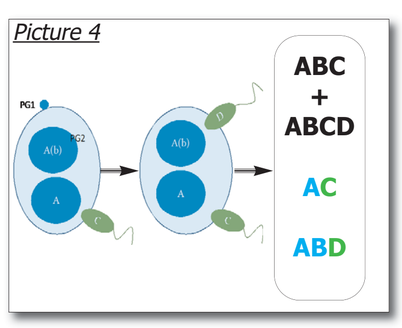
Picture 2



Picture 3



Picture 4



1) Tetragametic chimera *(Picture 1) is the most common mechanism: two distinct oocytes (A and B haploid sets) are respectively fertilized by two distinct spermatozoa (C and D haploid sets). So the zygote has two cell lines: AC and BD and its haplotype is* ***ABCD****.*
2) A parthenogenetic activation *(Picture 2) of the oocyte leads to the formation of two identical haploid cells (A and A). Each haploid maternal cell is fertilized by two distinct spermatozoa (C and D haploid sets). The zygote has two cell lines: AC and AD and its haplotype is* ***ACD****.*
A variant of a parthenogenetic activation *(Picture 3) has been described by Strain et al (1995). As above, a parthenogenetic activation of the oocyte led to the formation of two identical haploid cells (A and A). Then, the oocyte was fertilized by a spermatozoon (C haploid set). One of the resulting haploid cells with the male pronucleus produced a diploid cell AC. The other haploid cell A became diploid through endoreplication. This cell line was AA, with complete uniparental disomy (UPD), and was responsible for an abnormal phenotype: small stature, left hemifacial microsomy, bifid uvula with a submucous cleft palate and mild mental retardation. The zygote had two cell lines: AC and AA and its haplotype was* ***AC****.*
3) *chimera resulting from* ***fertilization of the second polar body*** *(Picture 4) has never been proved. The mechanism could be the following: after having been fertilized by a spermatozoon (C haploid set), the oocyte (A haploid set) does not expel its second polar body (A haploid set for proximal markers, B haploid set for distal markers because of crossing over). The second polar body is fertilized by another spermatozoon (D haploid set). The zygote has two cell lines: AC and AbD and its haplotype is* ***ACD*** *for proximal markers and* ***ABCD*** *for distal markers.*
4) *Different mechanisms of* ***mosaicism*** *(mitotic corrections) can occur. One of them is the double correction of a zygote 47,XXY leading to a cell line 46,XX and a cell line 46,XY. The haplotype is* ***AC*** *(A is the maternal haploid set, C is the paternal haploid set) for autosomic chromosome and we can find a maternal X, a paternal X and a paternal Y. The correction of a triploidy is another possible mechanism.*

It is important to determine the mechanism to exclude the risk of a complete UPD cell line responsible for an abnormal phenotype. This is possible through the study of the haplotype.

CASE REPORT

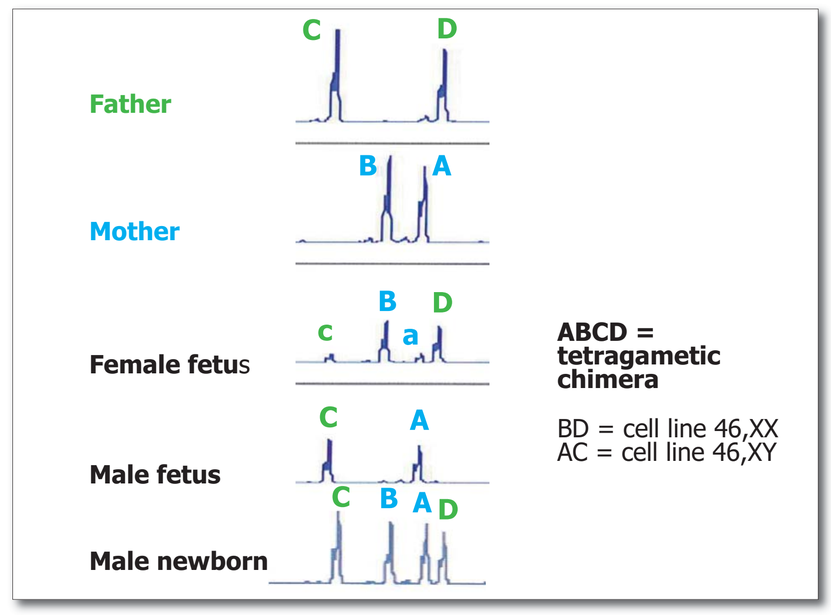
We report a first pregnancy conceived by **IVF**, with the **implantation of 2 zygotes**.
The first trimester ultrasound examination showed a twin monochorionic, biamniotic i.e. **MONOZYGOTIC** pregnancy. But, the ultrasound examination of the second trimester revealed a **female twin** (F1) and a **male twin** (F2).
An amniocentesis was performed. Results are set out in *table 1*:
- We can conclude that in F1 there are two cell lines 46,XX/46,XY seen by two different and independent techniques. But we cannot distinguish if the XX cell line in F2 is maternal or fœtal (no microsatellite result) so it is not possible to conclude between a simple maternal cell contamination or a true fœtal 46,XX/46,XY.
- The microsatellite markers on uncultured amniotic fluid study (*picture 5*) show that the **F1 haplotype is ABCD and signs the tetragametic chimera**.

Genetic counseling was reassuring because ultrasound examination did not find any sexual ambiguity and Tetragametic chimera mechanism excluded a complete UPD cell line. So the pregnancy was continued.
After birth, the microsatellite markers on lymphocytes confirmed the **chimera on the female** newborn in which ovotestis has not yet examined. On the **male newborn**, who finally has a **micropenis**, the microsatellite markers on lymphocyte found a tetragametic **chimera**.

Table 1

Prenatal study	**F1**	**F2**
Ultrasound examination	Female	Male
FISH on uncultured amniotic fluid	75% XX 25% XY	90% XY 10% XX
Microsatellite markers on uncultured amniotic fluid	Two cell lines: haplotype ABCD (*picture 5*)	Only one cell line (*picture 5*)
Caryotype and FISH on cultured amniotic fluid	117 mitoses and 500 nuclei XX	93 mitoses and 500 nuclei XY
Conclusion of prenatal study	Chimera or mosaicism XX/XY	Maternal contamination or Chimera or mosaicism
Postnatal study	**Female newborn**	**Male newborn**
Microsatellite markers on blood lymphocyte after birth	Two cell lines: haplotype ABCD	Two cell lines: haplotype ABCD (*picture 5*)

Picture 5



CONCLUSION

Chimera is a rare event. The mechanism must be studied by microsatellite markers to exclude the risk of a complete UPD cell line and phenotypic abnormalities.
In our case, the general mechanism is this :
- Implantation of two zygotes,
- Subsequent fusion of those two zygotes with the formation of a tetragametic chimera,
- Followed by split between day 3 and day 7 with the formation of a monochorial, biamniotic twin.

Long-term follow-up will be necessary for the risk of ovotestis (risk of gonadoblastoma) and pubertal and reproductive difficulties.

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