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A Complex Three Breakpoint Translocation in the Domestic Pig

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Mäkinen, A., M.-T. Kuokkanen, T. Niini and L. Perttola: A complex three breakpoint translocation in the domestic pig. Acta vet. scand. 1987, 28, 189–196. – A structural rearrangement involving three autosomes, numbers 2, 4 and 15, was transmitted from an A.I. boar of Finnish Landrace breed to its offspring. The boar was used extensively for breeding work; its phenotype was normal, and it had normal sexual functions and semen characteristics. However, because of the small size of its litters, blood samples were taken from the offspring for chromosome analysis.

reciprocal translocation; low fertility; sperm analysis.

Introduction

Reduced fertility, based on small litter sizes caused by structural rearrangements of chromosome segments carried by heterozygous animals, has already been noted in the domestic pig (reviewed by *Gustavsson* 1984, *Popescu & Boscher* 1986, *Mäkinen & Remes* 1986).

The fertility may be reduced by the production of genetically unbalanced gametes which are effective at fertilization but result in early embryonic loss of progeny. Sperm production in the males may be unaffected, the sperm count lying within the range for normal individuals. Reduced fertility due to early embryonic death could thus be a secondary effect of the chromosome abnormality. Hence, the number of piglets per litter or the non-return rates give practical connections to mitotic chromosome studies.

Material and methods

The pigs investigated were of the Finnish Landrace breed. Blood samples for chromosome analysis were taken originally from 4 piglets (sows numbers 1 and 2 in Table 1) in connection with halothane testing, because their sire had low fertility. We later tried to obtain whole litters from different sows inseminated by the same boar but the pig farmers had only some piglets left from the litters in question (Table 1).

The boar was slaughtered before the cytogenetic investigation was carried out, but semen had been collected once a week for 9 months by the gloved-hand method when the boar was mounting a dummy sow. The progressive motility of the spermatozoa was studied microscopically soon after semen collection and the total volume and sperm cell concentration (counted by a photometer) of each ejaculate were recorded.

The dam of the boar was alive and was studied chromosomally, but the sire, an A.I. boar, had already been slaughtered.

Karyotypes were prepared from blood lymphocyte cultures by standard techniques (*Gustavsson et al.* 1983). The chromosomes were analysed by RBA (*Dutrillaux et al.* 1983), GTG (*Seabright* 1971, modified by *Kuokkanen & Mäkinen* 1987), and CBG (*Sumner* 1972) banding techniques. They were arranged into karyotypes according to the international standard (*Proceedings of the First International Conference for the Standardization of Banded Karyotypes of Domestic Animals* 1980).

The number of piglets per litter from the A.I. boar was counted from information given to the A.I. centre. The litter results

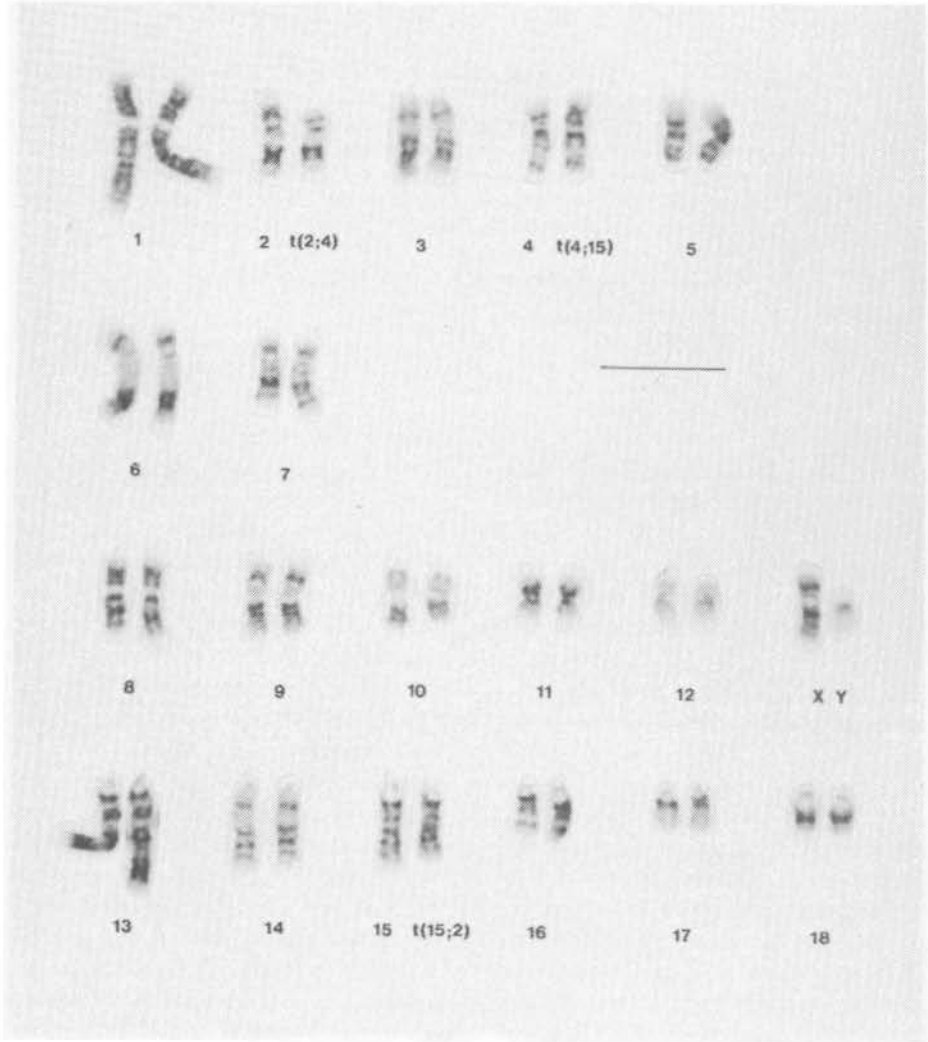

Figure 1. GTG-banded karyotype of the boar, 2n = 38, XY, t (2; 15; 4) (p13; q26; p15). Scale is 10 μm.

from the sows were collected from the farms concerned.

Results

Chromosome studies

The chromosome analysis of the offspring showed a balanced reciprocal translocation. The distal segments of autosomes 2, 4 and 15 were exchanged (Figs. 1 and 2). The exchanges, comprising 3 breakage points, occurred in the RBA-positive (GTG-negative) regions of the short arms of autosomes 2 and 4 close to the distal regions (2p13) (4p15) and of acrocentic autosome 15 in the distal region (15q26) (Fig. 3a, b). Designation of the rearrangements was made according to the recommendation of the *Paris Conference* (1971).

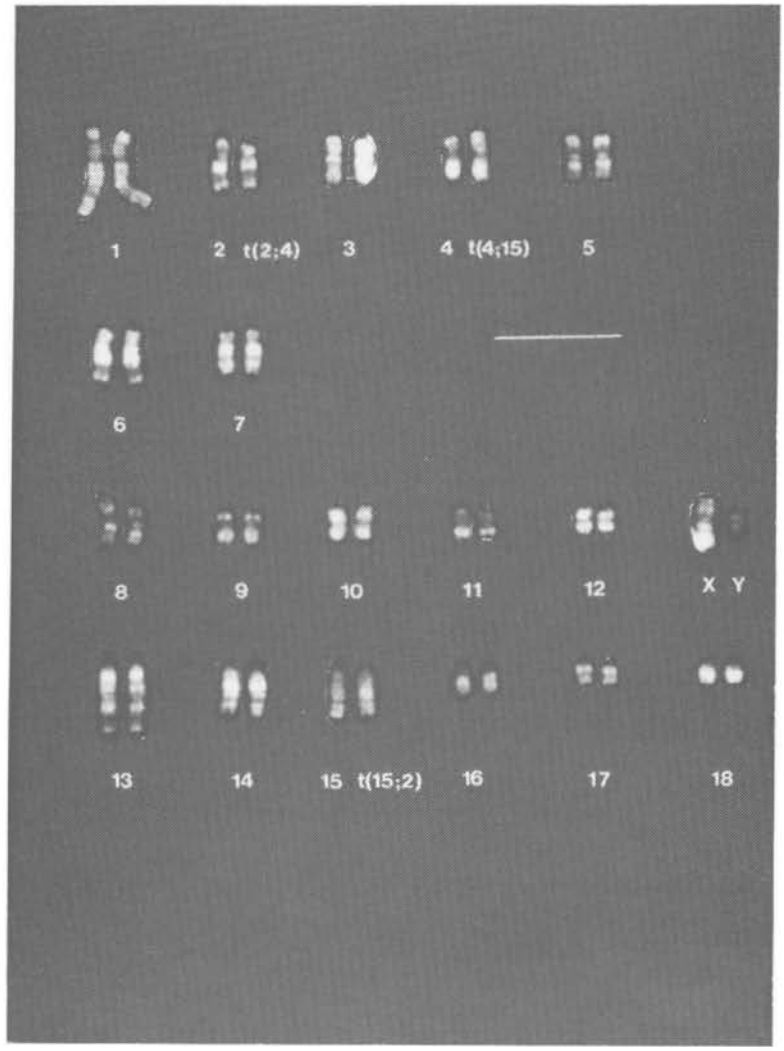

Figure 2. RBA-banded karyotype of the boar, 2n = 38, XY, t (2; 15; 4) (p13; q26; p15). Scale is 10 μm.

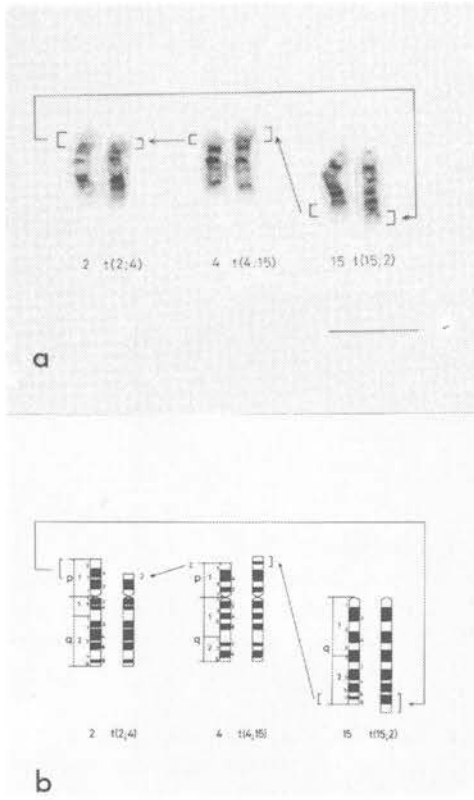

Figure 3a. Illustration of the cyclical exchange between the three GTG-banded cut-out chromosomes 2, 15 and 4 involved in the translocation. Scale is 10 μm.

Figure 3b. Their schematic presentation. According to the landmark system proposed by *Lin et al.* (1980) and *Fries* (1982) the translocation can be designated as t (2; 15; 4) (p13; q26; p15). The exchanged segments are shown by arrows.

The altered short arms in autosomes 2 and 4 could be seen easily by their length. However the GTG-negative terminal regions were devoid of characteristic bands in the exchanged segments making detection of the broken parts difficult.

The amount of constitutive heterochromatin detected by CBG-banding was constant in the rearranged autosome pairs.

Table 1. The number of piglets analysed chromosomally from different sows inseminated by the same A.I. boar.

Sow inseminated by the A.I. boar	Chromosomally-analysed piglets	
	number	female/male
1	2	2/0
2	2	1/1
3	3	2/1
4	1	1/0
5	1	1/0
6	3	3/0
7	1	1/0
Total	13	11/2

Table 2 shows the deviation of the rearranged chromosomes in the chromosomally-studied piglets and the size of the litters. Only 12 piglets distributed over 6 sows and litters were available for chromosome studies. One frozen blood sample from a piglet failed in chromosome analysis. The number of chromosomally-studied offspring within the different litters was too small for hereditary studies to be carried out.

In 3 piglets from different sows, the reciprocal translocation between autosomes 2, 15 and 4 was found in all studied lymphocytes. The other chromosomally-studied piglets had normal karyotypes. The piglets carrying the translocation were normal from a phenotypic point of view and healthy.

Fertility studies

The A.I. boar started its breeding career at the age of 9 months. Its testicles were of normal size and consistency and were symmetrical. Its sexual behaviour evaluated under A.I. station conditions where boars never see normal sows, was normal. Its reaction time was about 4 times longer than the average

Table 2. Whole litter sizes, chromosomally studied piglets and their karyotypes in offspring from 7 different sows by the boar 2n = 38, XY, t (2, 15, 4).

Sow inseminated by the A.I. boar	Number of piglets per litter	Number of chromosomally-studied piglets	Piglets with altered karyotype	Piglets with normal karyotype
1	8	2	1	1
2	10	2	1	1
3	7	3	0	3
4	6	1	*)	*)
5	7	1	1	0
6	10	3	0	3
7	8	1	0	1
Total		13	3	9

*) a culture of 1 frozen blood sample failed.

for A.I. boars at the station but this could be normal variability.

No abnormality was noted with regard to semen colour, odour and liquification. Its semen-producing capacity was good; an approximate gel-free semen volume was 242.5 ± 45.26 ml. Semen concentration was 665.64 ± 117.65 (x 10^6) per ml and progressive motility 74.15 ± 1.46 %. Exact morphological evaluation of the sperm cell of this boar was not made because at least 70 % of the cells appeared normal.

The boar was slaughtered before the cytogenetic investigation. It had been used for insemination of 290 sows with an average of 8.2 piglets per litter. The deviation of litter size was from 2 to 17 piglets.

In the Finnish Landrace breed artificially inseminated sows have an approximate average of 11.6 piglets per litter. Hence, the complex rearrangement of the chromosomes 2, 4 and 15 gave an average reduction in litter size of 30 %.

The non-return rate of sows inseminated by this boar was 84.9 % compared with non-return rates of 83.2 % for A.I. Finnish Landrace breed sows counted 60 days after insemination. Hence, the rate was not higher after insemination by this chromosomally-rearranged boar.

The dam of the boar was alive. It was cytogenetically investigated and found to be normal. It had farrowed 6 litters with an average of 12.7 piglets per litter. The deviation was from 10 to 14 piglets per litter.

The sire of the boar was no longer alive but had been used as an A.I. boar and had produced 690 litters with an average of 11.8 piglets per litter.

Discussion

A structural rearrangement involving an exchange between the broken segments of 3 chromosomes has previously been found in healthy and phenotypically normal pigs, referred for cytogenetic study because of fertility impairment reflected by reduced litter size of their sire, a Finnish Landrace breed A.I. boar. This boar showed normal sexual behaviour and a semen picture within the normal range of variability (*Gibson & Johanson* 1980).

In domestic pigs, reciprocal translocations where the broken ends of chromosomes are joined in new combinations, are typical chromosome abnormalities. Each transloca-

tion is likely to be unique because breakpoints are widely, if not randomly, distributed over the whole chromosome complement (*Gustavsson et al.* 1983). These structural variants of the chromosomes in balanced form do not appear to have any effect on the phenotype.

Since homologous chromosome segments tend to pair in meiosis, it is expected that each segment of the rearranged chromosomes will pair at pachytene with its homologous segment in the unchanged chromosomes and a cross-shaped hexavalent configuration will be formed in the 3 breakagepoints translocation (*Meer et al.* 1981, *Saadallah & Hulten* 1985).

The complex structural rearrangement in the genome may impair the zygotene and later stages of gametogenesis because the derivative chromosomes fail to pair characteristically, which affects the unbalanced gametes. The gametes mature and are capable of fertilization, but the chromosomally unbalanced zygotes tend to die in early embryogenesis (*King et al.* 1981, *Popescu & Boscher* 1982, *Gustavsson et al.* 1983).

Because only the embryos with balanced and normal karyotypes survive, a reduction in litter size is clearly seen. In our present study there was no indication of an increased incidence of stillborn or malformed pigs. It should be noted, however, that repeated matings with other boars at the farms may possibly explain for the very high litter sizes of some sows inseminated by the chromosomally-rearranged boar. Accurate figures can only be obtained by extensive and controlled matings.

Unfortunately, information is not available from heredity studies, because the number of chromosomally-studied piglets per litter was small, the slaughter of the offspring being fast. Only 12 piglets from 6 different sows could be chromosomally investigated; 3 of them had an altered karyotype, the others a normal one.

A meiotic study can be a useful adjunct to the lymphocyte chromosome investigation. In fact, basic information concerning the behaviour of structural and other chromosome abnormalities throughout gametogenesis can only be gained by a thorough knowledge of the meiotic process. Unfortunately in this study, one male offspring, a carrier of the rearranged karyotype failed.

Rearranged chromosomes can have a de novo origin or they can be inherited from the parents. In this A.I. boar the complex chromosome rearrangements seem to have had a de novo origin, because its dam had a normal karyotype and the litter sizes of its sire were within the normal range for this breed.

It is evident that selection of breeding animals can eliminate most chromosome mutations from the population, but it is also clear that there are many aberrations which can only be eradicated after cytogenetic evaluation.

The practical problems involved in analysing the somatic chromosome constitution of every A.I. boar are considerable. Although the lymphocyte culture technique involved is straightforward and reliable, facilities are needed as well as trained staff with considerable experience in analysing pig chromosome preparations. Moreover, such analysis is time-consuming. Therefore, selection of boars for chromosome examination could be made by fast-screening their litter size results at the beginning of their breeding use. Only boars with reduced litters or complete sterility would be referred for chromosome analysis.

Speed in detecting chromosome abnormalities in breeding animals specially used for A.I. work, is of great economical value for the pig farmers.

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Sammanfattning

En komplex trepunkts translokation hos tamsvin.

Strukturella kromosomförändringar mellan tre olika autosomer hos en A.I. galt minskade grisantalet i kullar med ca. 30 %. Spermanalyser samt galtens fenotyp var normala.

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