J. Appl. Genet. 37(3), 1996, pp. 293-298

Evaluation of the effect of centromeric heterochromatin polymorphism on pig fertility

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Abstract. C-banding identified centromeric heterochromatin polymorphism, most often located in pair no. 16, was a basis for selecting animals for the experiment. The aim of the experiment was to assess the impact of centromeric heterochromatin polymorphism on pig fertility, expressed by litter size. The research included the first two litters obtained from 30 pairs of parents with different genotypes as regards the parameter under study. A statistical analysis of the number of offspring from different mating types showed no significant differences between the experimental groups of animals. The results obtained did not confirm suggested correlation between polymorphism of the centromeric heterochromatin regions and pig fertility, estimated by mean litter size. Thus the polymorphism cannot be regarded as a selection criterion.

Key words: C-banding, constitutive heterochromatin, fertility, karyotype, litter size, pigs, polymorphism.

Introduction

Distribution of genetically inactive constitutive heterochromatin is a stable characteristic of every genome. In pigs, as in other mammals, constitutive heterochromatin is located mainly in the centromeric regions.

Cytogenetic classification of swine constitutive heterochromatin, with the use of staining with fluorochromes specific of DNA regions in combination with C-banding (barium hydroxide treatment), differentiates four types of con-

Received: January 1996.
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stitutive heterochromatin in pig chromosomes (SCHENDL et al. 1981, LIN et al. 1982). Specific base-pairs contents for these types were found.

Constitutive heterochromatin regions are characterized by great variability, as evidenced by C-band variants, which according to of polymorphism definition, are stable, occur in random proportions in a population, and are hereditary in nature.

In pigs, most recent studies showed that C-band heteromorphism can have a negative influence on fertility. CHRISTENSEN and PEDERSEN (1991) showed a statistically significant, negative effect of pair 16 or 17 chromosome C-band polymorphism on mean litter size. They also found mean litter weight to decrease and ascertained correct semen parameters of polymorphism-carrying boars. Thus, the above mentioned authors excluded spermatozoa defects as a cause of lower fertility. They suggested that differences in the size of C-bands might cause non-disjunction of chromosomes in meiosis and a higher embryonic mortality. In order to define the effects of C-band polymorphism on the fertility of Polish Landrace pigs (using litter size as a criterion) five experimental groups, genotypically different with regard to pair-16 chromosome C-band polymorphism, were compared.

Material and methods

In order to select animals with the required genotypes for the experiment, 300 Polish Landrace pigs were examined cytogenetically. C-banding was carried out (SUMMER 1972) and 40 animals demonstrating polymorphism in pair 16 were selected for the experiment. The RBA technique (DUTRILLAUX et al. 1973) was applied to confirm the existence of polymorphism in pair 16. Two forms were distinguished: small heterochromatic block and large heterochromatic block. Polymorphism of heterochromatic block in pair 16 chromosome was marked p^{+} and the lack of polymorphism (both centromeric regions small, equivalent size) was marked p^{-}. Carriers of different polymorphic variants of C-bands were classified into genotypic groups, accounting for all possible mating types (Table 1).

The control group comprised pigs without C-band polymorphism in any of acrocentric chromosomes (small centromeric region). The data on the number of piglets (live and still-born), obtained from the first two litters from each pair of parents (60 litters) were compared using one-way analysis of variance. All the animals involved in the experiment were kept under the same conditions on the same farm.

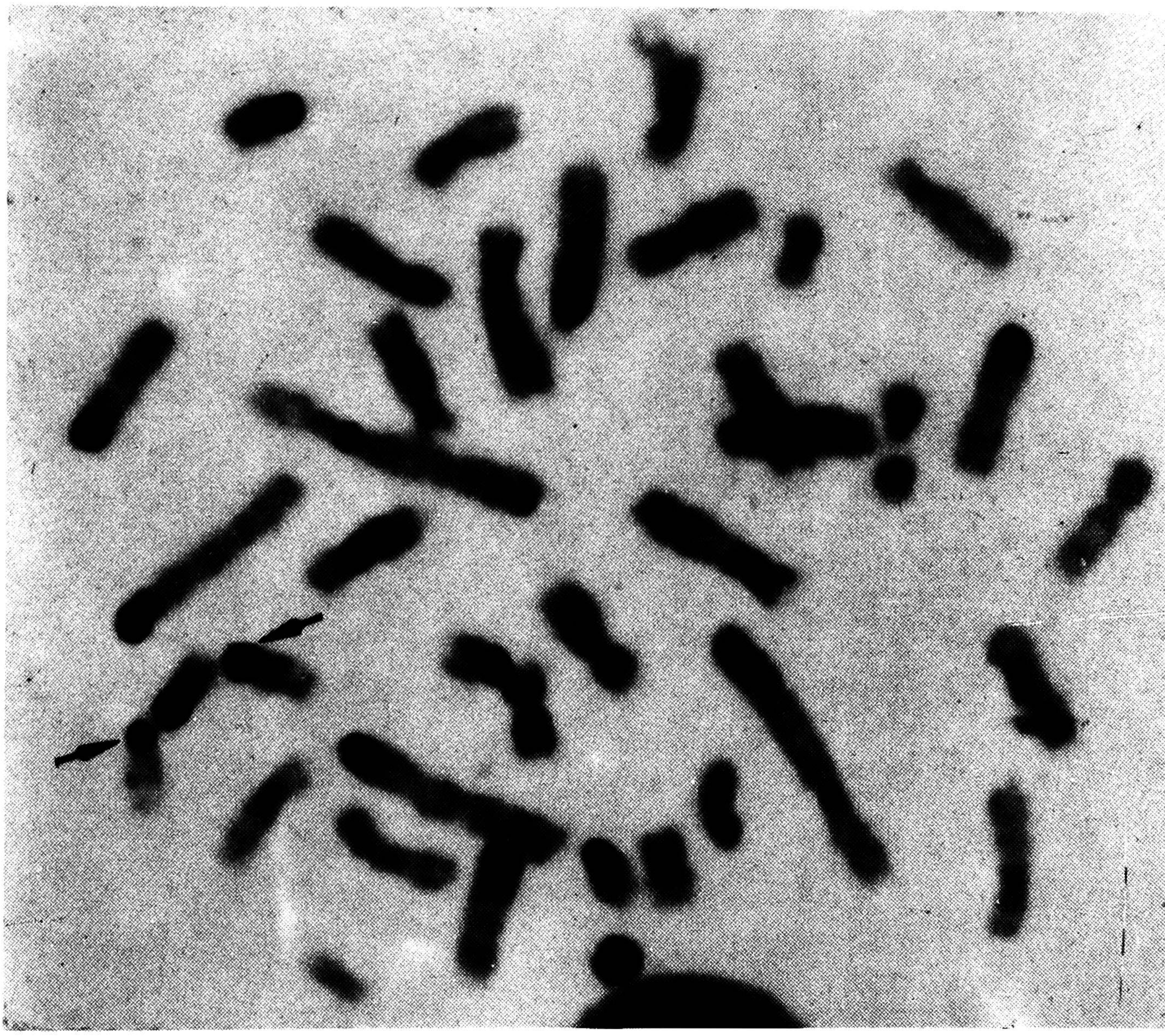

Fig. 1. Centromeric heterochromatin polymorphism of pair 16 chromosomes in the homozygotic form in pigs

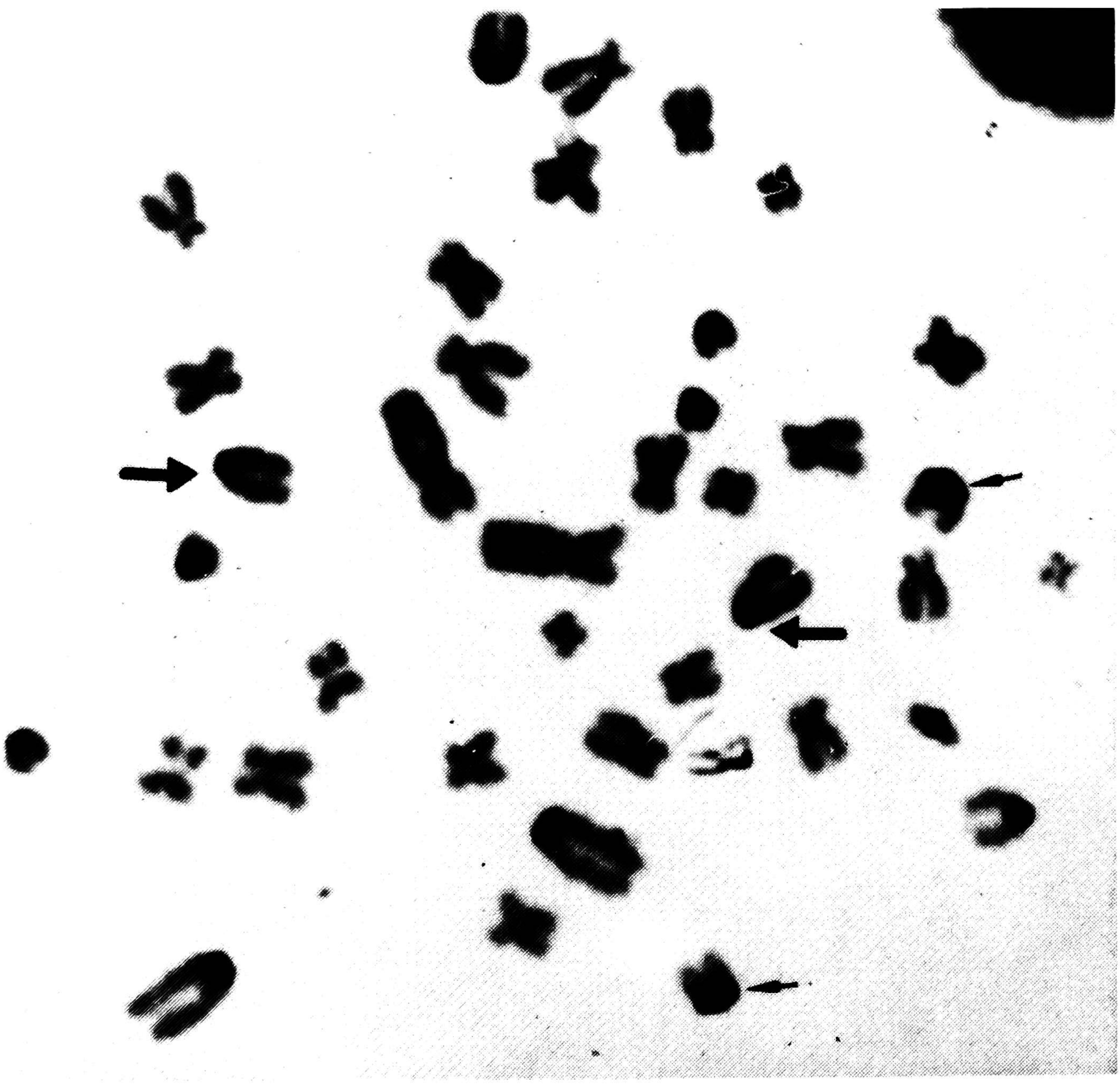

Fig. 2. Centromeric heterochromatin polymorphism of pair 16 chromosomes in the homozygotic form and of pair 14 chromosomes in the heterozygotic form in pigs

Results

On the basis of the C-band analysis conducted on 300 pigs, 157 carriers of polymorphism of centromeric heterochromatin blocks of 13-18 chromosome pairs were singled out, which constituted 52% of the population under study. The polymorphism occurred in all acrocentric chromosome pairs: in one pair – in 104 pigs (Fig. 1), simultaneously in two pairs – in 45 pigs (Fig. 2), and in three chromosome pairs – in 8 pigs.

The polymorphism was most frequently recognized in pairs: 16 (123 animals), 17 (36 animals) and 13 (33 animals). In the other pairs this phenomenon was not so often: pair 18 (10 animals), pair 15 (9 animals) and pair 14 (7 animals).

Table 1. Mating programme and results obtained for experimental groups of pigs

Experimental group of carriers	Mating type ♂ × ♀	Number of matings	Piglets No. (stillborn included) in litters		Mean litter size (live and still-born)
			first	second	
1	$p^+/p^+ \times p^-/p^-$	6	60 (2)	64 (2)	10.33
2	$p^+/p^- \times p^-/p^-$	6	57 (7)	65 (5)	10.17
3	$p^-/p^- \times p^+/p^+$	6	63 (3)	70 (3)	11.09
4	$p^-/p^- \times p^+/p^-$	6	61 (4)	67 (2)	10.67
Control	$p^-/p^- \times p^-/p^-$	6	67 (8)	68 (2)	11.25

The data on litter size (Table 1) were analysed twice by the analysis of variance. In the first comparison, the mean number of piglets from two litters, for each pair of the parents was considered (F_{calc} = 0.50, $F_{0.05}$ = 2.76, $F_{0.01}$ = 4.18). In the second comparison, each litter corresponded to one trial to increase the number of experimental factors (F_{calc} = 0.85, $F_{0.05}$ = 2.54, $F_{0.01}$ = 3.68). The analysis of variance showed no statistically significant differences between the groups of animals under study.

Discussion

Size variability of centromeric heterochromatin of acrocentric chromosomes in pigs, was frequently reported to concern 30-56% of populations under study (HIDAS, SZALAY 1990, ŚWITOŃSKI, PIETRZAK 1992, SŁOTA et al. 1994).

According to the literature the C-band polymorphism carriers have so far been identified in the following breeds: Yorkshire, Large White, Pietrain and Duroc, and in many Landrace breeds: Danish, German, Swiss, Hungarian, Polish (HANSEN-MELANDER, MELANDER 1974, CHRISTENSEN, SMEDEGARD 1979, SYSA 1980, GLAHN-LUFT et al. 1982, ŚWITOŃSKI et al. 1983, HIDAS, SZALAY 1990, ŚWITOŃSKI, PIETRZAK 1992).

Only HIDAS and SZALAY (1990) attempted to estimate the frequency of polymorphism by comparing the frequency of its carriers in four breed groups, but they found no statistically significant differences probably due to a small size of the tested animal groups. However, a comparison they made showed that there was C-band polymorphic variants had a clear tendency to occur more frequently in Duroc than in other breeds, and that most cases of pair 16 polymorphism were found in Hungarian Landrace boars.

In the population of Polish Landrace pigs (300 head) used in this experiment, the number of pigs carrying polymorphism of acrocentric chromosome C-bands constituted 52%.

As it was found in other herds, pair 16 was the most polymorphic and constituted 56% of the cases of heterochromatic block polymorphism. Similar results were obtained by SŁOTA et al. (1994), who found pair 16 polymorphism in 58% of the animals studied.

The first attempts to define correlation between C-band polymorphism and fertility have been made by CHRISTENSEN and PEDERSEN (1991) in an experiment carried out on two groups of pigs selected from a large population of Danish Durocs. Two factors were taken into consideration: the co-occurrence of polymorphism and smaller litter size, and the lack of polymorphism and greater litter size. It must be emphasized that the differences, though statistically significant, when expressed in absolute values, showed that the litter size had been decreased by one piglet only.

Such a correlation was not assumed to occur in our experiment carried out on the Polish Landrace pigs. In this connection only one factor – centromeric heterochromatin polymorphism – was taken into account. On the basis of the chromosome pair 16 C-band variants, four experimental groups were formed and the control group of polymorphism-free pigs was a reference point. Such experiment gives a greater probability to obtain objective results.

The analysis of variance of the litter size was done twice. In the first comparison, one trial corresponded to a mean number of piglets from two litters, calculated for each pair of the parents, while in the second comparison, each litter corresponded to one trial to increase the number of the components of the experiment. The use of two calculation variants aimed to double the number of degrees of freedom, which could increase the sensitivity of the analysis. Irrespective of the calculating method, however, the analysis of variance in both cases showed no statistically significant differences between the groups under study. Attention should be paid to the exceptionally great variability of the parameter studied (litter size), registered both within the group of polymorphism carriers and in the control group. The fact that no significant differences between groups were shown, with a clear variability within each of the groups, may indicate the lack of correlation between the litter size and centromeric heterochromatin polymorphism. These findings do not confirm the results of the experiment conducted by CHRISTENSEN and PEDERSEN (1991). They suggest that differences in the size of centromeric heterochromatin blocks may be a cause of non-disjunction of chromosomes at meiosis, so it can lead to an early death of embryos. However, in humans this phenomenom has not been observed, probably due to a stable character of C-band units and due to almost the lack of unequal crossover within the repetitive C-band DNA.

Conclusions

The results obtained did not confirm the presence of correlation between polymorphism of centromeric heterochromatin regions and pig fertility. The results permit to infer that centromeric heterochromatin polymorphism in combination with litter size cannot be referred to as a selection criterion.

Acknowledgements. This work was supported by State Committee for Scientific Research, Project No. 55 304 015 05.

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