Variation in patterns of recombination result in genetic variation in intrachromosomal shuffling in the domestic pig

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Abstract
Meiotic recombination leads to shuffling of loci located on the same chromosome. The amount of intrachromosomal shuffling from one generation to the next is affected by the number of crossovers, location of crossovers and crossover interference. In the domestic pig, *Sus Scrofa*, genome wide recombination rates are higher in females than in males. However, in this study we find that the genome wide intrachromosomal shuffling between pairs of loci is higher in males than in females due to difference in distribution of crossovers along the chromosome in the sexes. We show that this pattern is consistent in four of five different pig breeds and that there is a genetic component to the variation in genetic shuffling.

Introduction
An important part of meiosis is the exchange of genetic material between homologues chromosomes through meiotic recombination, which results in gametes with novel haplotypes. This event breaks down linkage disequilibrium and lead to haplotypic diversity that can be exploited in selection. Recombination also has a vital role in the proper alignment and segregation of homologues chromosomes. (Sherman et al., 1991; Koehler et al., 1996; Hassold et al., 1995; Fledel-Alon et al., 2009). Recombination can however also break up beneficial linkage previously built up by selection (Charlesworth and Barton, 1996), and there is evidence for increased mutation rates in recombination hotspots (Halldorsson et al., 2019; Arbeithuber et al., 2015). Recombination rates vary between taxa and species, and even within and between closely related populations (reviewed in Ritz et al., 2017; and Stapley et al., 2017). Most species show some level of heterochiasmy (Burt et al., 1991) and in some species there is also a substantial sexual dimorphism in the distribution of crossovers along the genome (Sakamoto et al., 2000; Lien et al., 2011; Tortereau et al., 2012; Johnston et al., 2016). In breeding this variation is of interest because it affects the production of novel allelic combinations. Veller et al (2019) suggests shuffling of maternal and paternal alleles from one generation to the next as an alternative measure that pics up both the number and distribution of crossovers. The aim of this study was to compare our previous results of variation in recombination rates within and between five domestic pig breeds with measures of intrachromosomal genetic shuffling and investigate whether there is variation between sex and breeds.

Materials & Methods
Data. This study focused on five purebred commercial breeding populations with pedigree and genotype data: two sow breeds, Landrace (LR) and Large White (LW); and three boar breeds, Duroc (DU), Synthetic (SY) and Pietrain (PI). The genotype data and filtering are described in detail in Brekke et al. (2022).
Linkage mapping and crossover detection. Detailed description of linkage mapping, gamete phasing, crossover detection and fine scale recombination mapping can be found in Brekke et al. (2022).

Genetic shuffling within individuals. Genetic shuffling was calculated as the probability that a randomly chosen pair of loci was shuffled during gamete production following the method suggested by Veller et al. (2019). We defined the parameter $\bar{r}$ as the probability of two alleles on a chromosome being shuffled due to recombination, i.e excluding the part due to independent assortment in equation (4) in Veller et al. (2019):

$$\mathbb{E}[\bar{r}] = \sum_{k=1}^{n} 2p_k(1-p_k)\,L_k^2$$

where k is the chromosome number (1-18), p is the proportion of grandpaternal alleles, $1\text{-}p$ is the proportion of grandmaternal alleles and L is the length of the chromosome as a fraction of the total length of the genome. For each phased gamete transmitted from a mother or father to an offspring, shuffling was calculated following equation 1 and assigned as an observation to the parent, resulting in multiple observations for the phenotype "genetic shuffling", hereafter referred to as $\bar{r}$, for each parent (hereafter focal individual, or FID).

Genetic variation. We estimated variance components for individual $\bar{r}$ with a repeatability model using the restricted maximum likelihood (REML) method and average information (AI) algorithm in DMU v 6 (Madsen et al., 2014). The following model was used:

$\bar{r}$=*id1*+*id2*+**e**

where **id1** is the random additive genetic effect of the FID, **id2** is the random effect of the FID permanent environment (i.e. individual and/or environmental effects affecting all gametes from an FID) and **e** is the residual effect. The narrow-sense heritability (h^2) was defined as the proportion of phenotypic variance explained by the additive genetic effect and was estimated separately for each breed and sex.

Results

Mean $\bar{r}$ is significantly higher in males than in females in the LR ($p=3.12e^{-105}$), DU ($p=2.51e^{-85}$), PI ($p=3.70e^{-72}$) and SY ($p=2.38e^{-53}$) breed. In the LW line $\bar{r}$ is only slightly, but significantly ($p = 1.40e^{-21}$) higher in females. Means and distribution are plotted in Figure 1. Intrachromosomal shuffling is a heritable trait in females in all breeds, but only in the LR breed in males. These results are presented in Table 1.

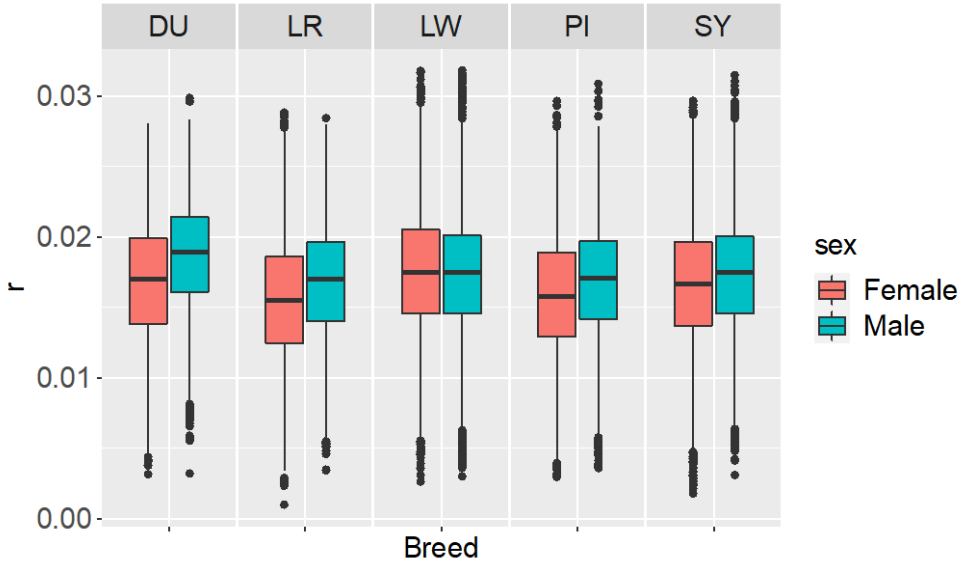

Figure 1. Sex difference in mean and distribution of shuffling.

Table 1. Results from variance component estimation of $\bar{r}$.

Breed	Sex	Mean(SD)	h^2(SE)	No obs[1]	No ind[2]
LR	M	0.0167(0.0039)	0.03(0.01)	11805	155
LR	F	0.0155(0.0043)	0.13(0.02)	11805	1960
DU	M	0.0186(0.0038)	0.03(0.02)	4090	89
DU	F	0.0168(0.0043)	0.09(0.02)	4090	661
LW	M	0.0172(0.0039)	0.04(0.01)	41237	273
LW	F	0.0175(0.0042)	0.15(0.01)	41237	4704
PI	M	0.0168(0.0040)	0.03(0.01)	12159	196
PI	F	0.0158(0.0042)	0.09(0.01)	12159	1355
SY	M	0.0172(0.0039)	0.03(0.01)	25705	224
SY	F	0.0166(0.0043)	0.05(0.01)	25705	2635

[1]Total number of observations (gametes).
[2]Total number of unique males or females with repeated observations.

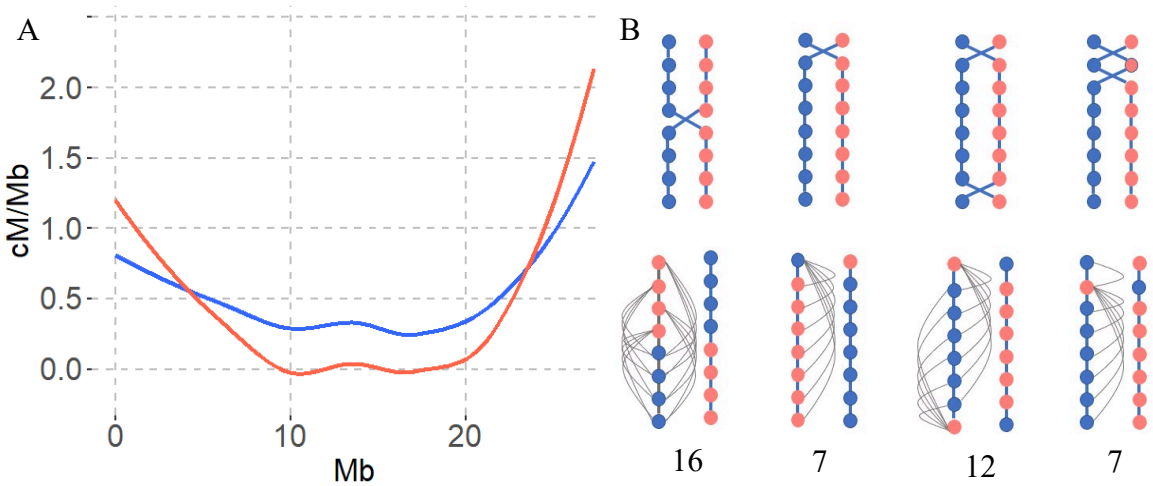

Figure 2. A) Variation in recombination rates along chromosome 1 in the PI breed for males in blue and females in red plotted with the loess method in the geom_smooth function in ggplot2 (Wickham, 2007). B) Example of crossover positions and the resulting number of pairs of loci shuffled (modified from Veller et al. (2019)).

Discussion

Our results show that the probability of shuffling between two loci on the same chromosomes is low relative to the shuffling of alleles at different chromosomes (average of 0.5 due to Mendelian segregation), but also that the intra-chromosomal shuffling differs between breeds and sexes and is a heritable trait in the domestic pig. Genome wide recombination rates in these pig breeds are higher in females than in males, and rates tend to be elevated in the telomeric regions in both sexes (Brekke et al. 2022). In some chromosomes this pattern is more extreme in females, i.e. recombination rates are higher than males in the telomeric regions, but lower than in males closer to the centromere (e.g. as in Figure 2a). This could explain why $\bar{r}$ is lower in females despite higher genome wide recombination rates. Figure 2b illustrates why a central crossover leads to more shuffling and why the position of the crossover may have a higher impact on the probability of shuffling between two loci than the number of crossovers. It is however puzzling that one of the breeds show the opposite sex difference in $\bar{r}$ (Figure 1). This breed (LW) is the breed with the highest genome-wide recombination rate (Brekke et al. 2022). More evenly spread crossovers lead to more

shuffling (Figure 2b). Differences in genetic shuffling can thus also be caused by differences in crossover interference between the breeds, explaining part of the difference in $\bar{r}$. Our results show that even if overall levels of sex-differences in recombination is the same in closely related populations, the shuffling might be different, potentially because of rapidly evolving hotspot usage (Paigen and Petkov, 2010; Weng et al., 2014, 2019). It is not clear, however, how the difference between the sex is maintained in a population from generation to generation as each offspring receives a paternal and maternal gamete. Even if a different number of unique sires and dams mated in each generation, the number of maternal and paternal gametes in each generation is always exactly the same. A next step could be to look at differences in recombination and genetic reshuffling between X and Y paternal gametes. The population level shuffling might be more influenced by variation in $\bar{r}$ within the sex, and in males in particular in pigs as the selection pressure is higher. In conclusion this study shows that variation in crossover distribution affects the production of novel haplotypes from one generation to the next and that there is variation in the shuffling caused by recombination between breeds, sex and individuals in the domestic pig.

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