Changed sex ratio in BB rat offspring

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INTRODUCTION

The BB rat is an inbred laboratory rat strain characterized by spontaneous development of diabetes mellitus and various autoimmune phenomena. It is widely used in scientific research, mostly because of the rapid onset of clinical diabetes (*Mordes et al.* 1987) with hyperglycemia, glucosuria, ketonuria, hyperinsulinemia and weight loss as well as pathological findings such as insulitis (*Seemayer et al.* 1983) and decreased β-cell volume (*Lucke et al.* 1988). Not only as a model of human type-1 diabetes (*Marliss et al.* 1982) is the BB rat a usefull model. It is used in studies of such disorders as thyroiditis (*Sternthal et al.* 1981) and lymphopenia (*Jackson et al.* 1983). Strong evidence suggests that the immune system (*Like & Rossini* 1984) plays a principal role in most of the abnormalities in the BB rat.

The BB rat has been used in laboratory experiments for more than a decade, and has been thoroughly investigated and described. Yet studies on the sex distribution in the offspring have remained unpublished.

In view of the role of this rat strain in animal research, we find it important to publish our findings of changed sex ratio in this rat compared to other rat strains and to other mammalian species.

MATERIALS AND METHODS

We have compared the sex ratio of three rat strains: 624 BB rats (BB/Wor/Mol-BB), 505 Wistar rats (Mol/WIST) and 587 Sprague-Dawley rats (Mol:SPRD). The Wistar rat was chosen for this comparison because the BB/Wor/Mol strain is inbred from the 1974 Wistar stock at the Bio Breeding Laboratories, Ottawa, Canada.

The animals in our study originated from Møllegaard Breeding Centre, Lille Skensved, Denmark. One third of the BB rats and all the Wistar and SPRD rats were bred under barrier conditions at the Møllegaard Breeding Centre. The rest of the BB rats were bred under semibarrier conditions at the Bartholin Institute. The stables were illuminated from 6:00 until 18:00. For most of the animals sex was determined at weaning (age 21 days). For a part of the BB stock, however, the sex was determined on day 7. This was done to minimize the influence of a conceivable difference in the late infantile mortality rate. An even earlier determination of sex is associated with much uncertainty.

No parents in any of the BB cages were treated with insulin before delivery of during nursing. All parents were tested for glucosuria every five days and checked for clinical diabetes twice a day. On sign of diabetes the whole litter was excluded from the study, since maternal diabetes in BB rats is known to impair viability in the offspring (*Brownscheidle et al.* 1983). All the newborns were kept in cages with both parents for the entire three week period until weaning. They were breastfed for the complete period with free access to fresh water and parental show (Altromin 1314).

Sex ratio was defined as number of males in the offspring divided by the total number of animals. Litter size was determined at the time of weaning.

Statistical analyses

Data are presented as mean ± SEM. The significance of differences was evaluated by application of the *t*-test or by means of the X^2 test. The level of type 1 error (2α) was set at 0.05.

Table 1. Scheme showing the number of animals, the sex ratio, X^2 result and p values in a comparison of the population concerned and the entire group of BB rats.

	Male	Female	Total	Sex ratio	X^2	p-value
BB rats total	274	350	624	43.9		
Wistar rats	258	247	505	51.1	5.77	0.016
SPRD rats	294	293	587	50.1	4.63	0.031
Non-BB rats total	552	540	1092	50.5	7.01	0.0081

RESULTS

During the study period from autumn 1988 to spring 1990 we found a total of 274 males and 350 females in our BB colony, which means a sex ratio of 43.9 % at age 21 days. A subpopulation of 91 males and 105 females was counted and sex determined at day seven, corresponding to a sex ratio of 46.4 %. The BB litter size was on average 6.5 (± 0.6).

In the Wistar colony a total of 505 newborns were weaned – 258 males and 247 females. The sex ratio was 51.1 %. The average litter size was 10.6 (± 0.4).

Among the SPRD rats, a total of 587 rats were weaned – 294 males and 293 females. The sex ratio was 50.1 %. The average litter size was 10.9 (± 0.5).

Comparison of the sex ratio of the BB stock with all the other stocks gives the statistical results shown in table 1. There is a significant difference between the BB rats and the non-BB rats ($p = 0.008$).

The litter size is significantly lower ($p < 0.001$) in the BB rats than in the Wistar or SPRD rats.

DISCUSSION

This study elucidates the difference in sex ratio of an inbred strain compared to two outbred strains. We have found a significantly changed sex ratio in the BB/Wor/Mol strain, whereas the outbred strains in our study show sex ratios slightly above 50 %.

A reduced litter sixe is seen in connection with the low sex ratio. Since we consider the parents to be non-diabetic the low litter size is probably more an effect of inbreeding than of metabolic impairment.

Among most mammals there is a slight numerical male dominance at birth and a shorter length of male life. The result is a higher percentage of females in the elder but not in the very young part of the population. An example of such a sex distribution is seen among humans. The sex ratio at birth in Denmark is 51 % steadily declining to 42 % among people above the age of 65 (*Danish Statistical Yearbook* 1985). Also the sex ratio of the progeny of a $(B6 \times A)F_1$ mouse strain was thoroughly described by *Bennett & Boyse* (1973). They found the sex ratio to be 53.3 %. Finally in a classical study on 35.102 newborn calves, *Foote* (1977) also found a sex ratio above 50 % (50.8 %).

The sex ratio in this study is 43.9 % in the BB colony and 50.5 % in the Non-BB colony. Since most of the rats were sex determined at weaning, it could be disputed whether the changed sex ratio was a result of different mortality rates during the first three weeks of life. This seems, however, not to be the case since we have seen a similarly changed sex ratio among BB rats at age 7 days.

In principle there could be four explanations for our findings of a lowered sex ratio. First, a lower rate of Y sperm to X sperm in the ejaculate would explain the lowered sex ratio. Second, the function of the Y sperm could be altered in the BB rat, either as a result of changed pattern of conception or as a result of immune reactions in the mother directed against the sperm. Third, the observed changes could be a consequence of intrauterine death based on mechanisms such as autoimmunity or metabolic or endocrine impairment. The process might be directed against

any stage of the development from conceptional product to neonate individual. Fourth, it can not be excluded that our findings result from neonatal death. This seems, however, unlikely since the same tendency is seen in the BB litters sex determined at day 7 as in the total number of BB rats, as explained above.

Our study has shown that the offspring of the BB rat has a significantly lowered male ratio (43.9 %).

Summary

We have found that the offspring of the spontaneously diabetic BB rat shows a lower (p = 0.008) male ratio (43.9 %) than do other rat strains (50.5 %). The lower number of males is seen both at day seven after birth and at weaning. Further we have seen a reduced litter size in the inbred BB rat compared to other rat strains. The reason for the unequal sex ratio in the BB rat is unknown but is discussed in view of the versatile autoimmune capacity of the BB rat.

Resumé

I dette studium har vi fundet, at den spontant diabetiske BB-rotte føder færre hanner end hunner (p = 0.008). Sexratioen blandt 21 dage gamle BB-rotter er 43.9 % hvorimod den hos kontrolstammerne er 50.5 %. Hos de kuld, der er kønsbestemt 7 dage gamle, ses en lignende ændring. Endvidere har vi fundet en betydeligt mindre kuldstørrelse hos BB-rotten end hos kontrolstammerne. Årsagen til den uens sexratioen hos BB-rotten kendes ikke, men den diskuteres i lyset af BB-rottens kendte autoimmune egenskaber.

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Key words: sex ratio, litter size, BB rat.

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