A Cytogenetic Study of XX/XY Chimerism and Other Anomalies of Bos taurus in a Sampled Dairy Herd

Ferne M. Rogers

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A CYTOGENETIC STUDY OF XX/XY CHIMERISM AND OTHER ANOMALIES OF Bos taurus IN A SampLED DAIRY HERD

(TITLE)

BY

FERNE M. ROGERS

THESIS

SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF SCIENCE

IN THE GRADUATE SCHOOL, EASTERN ILLINOIS UNIVERSITY
CHARLESTON, ILLINOIS

1977
YEAR

I HEREBY RECOMMEND THIS THESIS BE ACCEPTED AS FULFILLING THIS PART OF THE GRADUATE DEGREE CITED ABOVE

ADVISER

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A CYTOGENETIC STUDY OF XX/XY CHIMERISM AND OTHER
ANOMALIES OF Bos taurus IN A SAMPLED DAIRY HERD

By

FERNE M. ROGERS
B.S., Eastern Illinois University, 1974

ABSTRACT OF A THESIS

Submitted in partial fulfillment of the requirements
for the degree of Master of Science at the Graduate School of
Eastern Illinois University

CHARLESTON, ILLINOIS
1977
ABSTRACT

The establishment of normal chromosomal patterns for a wide range of plants and animals has led to the recognition of abnormalities as well as their relationship to phenotypic irregularities. Research dealing with the expression of human karyotype is of prime interest; but, closely rivaling that is the genetic study of domestic animals which are of economic importance to man.

One observed anomaly which has come under scrutiny by cytogeneticiests is that of the female member of a heterogeneous bovine twin pair, the freemartin. This is a sterile animal with internal morphology showing varying degrees of intersex. Cytogenetic studies reveal XX/XY chimerism in blood and various other tissues due to interuterine vascular anastomosis.

This study resulted in collection of the information regarding this anomaly into a relevant body of reference. Observing stained metaphase chromosomes from leukocyte cultures, experimental data were established for 109 bovine individuals, sampling the University of Illinois Dairy Research herd, the sires, and offspring. Differences were demonstrated by photomicrographs. Eight animals (7.3%) were found to carry XX/XY chimerism, only one of which was a member of a known multiple birth. The early death rate among these individuals is twice that of an average herd in Illinois. No relationship between a specific breed and appearance of this anomaly was seen.

As karyotype determinations were made, they were surveyed for other aberrations and counted to detect any variance from the normal
diploid number, $2N = 60$. Tetraploid-diploid mosaicism was seen in 5.5% of the animals in this study. No pathology was seen in connection with this condition. Three female animals (2.8%) carried trisomy XXX in mosaic condition with normal XX cells. Two cases (1.8%) of autosomal aneuploidy were seen, both dying of pneumonia at an early age. A Robertsonian translocation involving a centric fusion of chromosomes 1 and 29 was seen in one individual. These anomalies were demonstrated by photomicrographs.
# TABLE OF CONTENTS

<table>
<thead>
<tr>
<th>LIST OF ILLUSTRATIONS</th>
<th>iv</th>
</tr>
</thead>
<tbody>
<tr>
<td>LIST OF TABLES</td>
<td>v</td>
</tr>
<tr>
<td>ACKNOWLEDGMENTS</td>
<td>vi</td>
</tr>
</tbody>
</table>

## Chapter

### I. INTRODUCTION

1. Introduction 

### II. LITERATURE REVIEW

- **A. Historical perspective of cytogenetics**
  1. Descriptive period 
  2. Experimental period 
  3. Cytology plus embryology 
  4. Cytology plus genetics 
  5. Role of chromosomes 
- **B. Gonad development**
- **C. Gamete formation**
- **D. Role of sex chromosomes**
- **E. Role of hormones**
- **F. Chromosomal abnormalities**
  1. Freemartin chimerism
     - a. In leukocytes 
     - b. In erythrocytic antigens 
     - c. In hemopoietic tissue 
     - d. In other somatic tissue 
     - e. In gonads 
     - f. Morphology of freemartin chimeras 
     - g. Mechanism of chimera origin 
  2. Tetraploidy
  3. Trisomy XXX
  4. Autosomal aneuploidy
  5. Robertsonian translocation 

### III. MATERIALS AND METHODS

- **A. Chromosome preparations**
- **B. Cytogenetic survey**
- **C. Photomicrograph preparations**
- **D. Individual histories**
LIST OF ILLUSTRATIONS

Figure
1. An XX-bearing Cell of Animal #356, a Phenotypic Male .................. 36
2. An XY-bearing Cell of Animal #356, a Phenotypic Male .................. 36
3. An XX-bearing Cell of Animal #367, a Phenotypic Male .................. 37
4. An XY-bearing Cell of Animal #367, a Phenotypic Male .................. 37
5. An XX-bearing Cell of Animal #372, a Phenotypic Male .................. 38
6. An XY-bearing Cell of Animal #372, a Phenotypic Male .................. 38
7. An XX-bearing Cell of Animal #2456, a Phenotypic Male .................. 39
8. An XY-bearing Cell of Animal #2456, a Phenotypic Male .................. 39
9. An XX-bearing Cell of Animal #3292, a Phenotypic Female ................. 40
10. XY-bearing Cells of Animal #3292, a Phenotypic Female ................. 40
11. An XX-bearing Cell of Animal #3204-s, a Phenotypic Male ................. 41
12. An XY-bearing Cell of Animal #3204-s, a Phenotypic Male ................. 41
13. Tetraploid Cell of Animal #2834-s1 ........................................ 42
14. Tetraploid Cell of Animal #3308 ............................................. 42
15. Tetraploid Cell of Animal #3208 ............................................. 43
16. Diploid Cell of Animal #3208 ................................................. 43
17. Cell Demonstrating Trisomy XXX in Animal #3204, an XXX/XX Mosaic ........ 44
18. Cell Demonstrating Trisomy XXX in Animal #3289, an XXX/XX Mosaic ........ 44
19. Cell Demonstrating Trisomy XXX in Animal #3297, an XXX/XX Mosaic ........ 45
20. Cell Demonstrating Autosomal Aneuploidy of Animal #2961 ................. 45
21. Cells Demonstrating Robertsonian Translocation of Chromosomes 1 and 29 in Animal #360 ................. 46
LIST OF TABLES

1. Incidence of XX/XY Chimerism in a Sampled Dairy Herd, *Bos taurus* .......... 47
2. Incidence of Tetraploidy in a Sampled Dairy Herd, *Bos taurus* ................. 48
3. Other Observed Anomalies in a Sampled Dairy Herd, *Bos taurus* ............... 49
ACKNOWLEDGMENTS

I would like to express my sincere gratitude to Dr. Eugene Krehbiel for his guidance, patience, and encouragement in this research effort. His scholarly competence and his willingness to share it have enriched my experience in graduate study.

I also wish to express appreciation to Dr. John Speer for time spent reading this manuscript and for invaluable suggestions given, to Dr. J. R. Lodge for use of staff and facilities of the Dairy Science Department at the University of Illinois, to A. L. Smith and P. S. Weathersbee for technical advice and assistance on several occasions.

Special thanks go to my parents, Mr. and Mrs. Leal Isaacs, for their initial encouragement and their faith that I could attain this goal.

I would like to dedicate this thesis to my husband, Don, for understanding why I wanted to do it, and to my daughters: Amy, Jill, Michelle, and Debi for supporting my decision. Without the cooperation of my family, this could not have been completed.
I. INTRODUCTION

Knowledge and understanding of the role of chromosomes have developed over the last three centuries, starting with the first observations of cells and gradually culminating with the sophisticated techniques of cytogenetics presently available (Mittwoch, 1967). The development of this knowledge is an integral part of a cytogenetic study.

Significance of the sex chromosomes has been learned during the past 65 years. First efforts were toward understanding normal sex determination. That paved the way for consideration of abnormalities exhibited in phenotypes, either behavior or anatomical. It also spurred efforts to relate them to genetic anomalies (Mittwoch, 1967).

Of prime interest is the study of human chromosomes, but closely rivaling it is the genetic study of domestic animals which have economic importance. Because of the practical applications in this field, once the significance was recognized, a considerable amount of study has been done in this area (Hutt, 1964).

As with human genetics, the study of normal karyotypes led to the discovery of abnormal arrangements. One abnormality seen in bovine and, to a lesser degree, ovine species is the freemartin, a female born twin to a male. This is a sterile animal with internal morphology showing varying degrees of intersex. While the problem has long been recognized by breeders (Hutt, 1964), discovery of cell chimerism and implications of it have depended on the body of knowledge gathered in cytogenetics.
It is the intent of this study to review the information yielded by the literature, to trace its progress, and to unify it into a relevant body of reference.

Most of the study regarding this abnormality has been done with animals of known multiple births or with animals exhibiting phenotypic abnormalities. It was the experimental goal of this study to determine if the phenomenon of chimerism might exist undetected in a large dairy here and, if so, to what extent it might occur among members of the herd, their offspring, and the sires. Karyotypes were scanned for chromosomal aberrations and the diploid number (2N) established for each animal. This was accomplished by observation of stained, metaphase chromosomes from leukocyte cultures. Differences were demonstrated by photomicrographs.
II. LITERATURE REVIEW

Historical perspective of cytogenetics

The earliest basis for cytogenetic study dates to Robert Hooke's first observation of cork cells in 1665. During the next century and a half, optics were improved and eventually achromatic lenses were introduced (Nordenskiold, 1927). This allowed the observation of much greater detail than previously possible and paved the way for a descriptive period of cytogenetic study (Swanson, 1957).

It was in this environment that M. J. Schleiden in 1838 and T. Schwann in 1839 independently proposed what has come to be widely known and accepted as the cell theory (Mittwoch, 1967). Two more decades passed before Rudolph Virchow was to postulate that every cell is the offspring of a pre-existing cell. This laid the foundation for the development of the science of evolutionary genetics, since acceptance of this theory meant an unbroken line of descent from the first cell to all presently existing cells. It also stimulated more questions regarding the manner in which this was done. This eventually meant the birth of cytogenetics.

Gregor Mendel's conclusive work on inheritance patterns in garden peas in the 1860's should have provided to Charles Darwin an explanation of individual variation. Yet, typical of the thinking of that time, Darwin wrote in 1872:

The laws governing inheritance are for the most part unknown. No one can say why the same peculiarity in different individuals of the same species, or in different species, is sometimes inherited and sometimes not so; why the child often reverts in
certain characteristics to its grandfather or grandmother or even more remote ancestor.  
(as quoted by Borek, 1965)

Indeed, Mendel's work was to lie buried in the literature until the end of the 19th century when it was rediscovered and widely acclaimed in the scientific community (Borek, 1965).

Another area that contributed heavily to the development of cytogenetic study was the analytical investigations of the mechanism of fertilization in the late 1800's. Roux, Weismann, Van Beneden, and others investigated the maturation and fertilization of the egg and development of the embryo. Flemming observed mitosis and conferred the name of chromatin on the stainable portions of the nucleus, but most importantly, he observed that these stained threads divide in a lengthwise fashion. Oscar Hertwig showed that fertilization involved the union of egg and sperm nuclei and began to experiment with portions of eggs. This marked the beginning of the experimental period of cytology (Swanson, 1957).

By the turn of the century, E. B. Wilson had clearly outlined the chromosome theory of inheritance (Swanson et al., 1967); the mechanisms of meiosis were being uncovered (Mittwoch, 1967); and the Mendelian laws were revived and applied (Borek, 1965). These events initiated the new hybrid science, cytogenetics—the study of genetics at the cellular level. Through this century it has concentrated on three general areas: the nature of the gene, its manner of reproduction, and its mode of action in phenotype determination (Swanson, 1957). It has used chemistry, physics, physiology, statistics, biochemistry, virology, and enzymology as its tools. In turn, cytogenetics has become the tool of evolutionists, population geneticists, embryologists, endocrinologists, as well as
medical practitioners. Improved techniques of fixation and staining, in vitro culture methods, electron microscopy, spectrophotometry, phase-contrast microscopy, autoradiography are some of the developments which have elevated it to its present level of sophistication (Hutt, 1964).

One area of cytogenetic study which has intrigued investigators is the behavior, influence, and mechanisms of the sex chromosomes. Several early workers observed the "accessory chromosome," as it was termed, with its more intense staining and late replicating properties. C. E. McClung (Mittwoch, 1973) observed spermatogenesis in insects, discovering two different types of spermatozoa in equal numbers. He was the first to suggest that this might influence sex determination. E. B. Wilson verified the role with his studies on large numbers of hemipterans (Wilson, 1905).

Gonad development

Since there is such diversity in the reproductive mechanisms of living things, the general material herein included shall apply to mammalian species only. In mammalian embryos the gonadal primordia, or genital ridges, arise from the mesonephros, presumably under the influence of the genetic sex (McFeely et al., 1967). The primordial germ cells which will occupy that tissue originate in the endodermal epithelium of the yolk sac and migrate to the genital ridges. These cells and the ridges in which they proliferate appear to be identical in both sexes, with all embryos possessing structures for development of either sex. In this undifferentiated state, the chromosomal constitution determines which set of organs will develop and which will be suppressed.
Normally, if a Y chromosome is present, the gonad becomes a testis; if it is absent, ovarian development occurs (Mittwoch, 1973).

The developing testes seem to grow at a much faster rate than the ovary in studies of the rat (Mittwoch et al., 1969). It seems reasonable to assume that the presence of the Y chromosome is the triggering mechanism to accelerate that rate (Jost, 1970). Dain (1974) suggests this possibility as an explanation for a preponderance of XY cells in three of four pairs of twins studied among chimeric sheep.

Gamete formation

Understanding the operational mode of the sex chromosomes requires some explanation of early syngamy. In normal ovulation, one or a few ova are produced by mammalian females, all bearing the haploid number (N) of chromosomes including one X chromosome. These ova are fertilized by mature sperm also carrying the haploid complement of chromosomes, but the sperm are not all identical. Half of them carry a single X chromosome and half bear the Y chromosome. In this respect, females are said to be homogametic and males are heterogametic. It is the moment of fertilization that determines the genetic sex of most mammals (McFeely et al., 1967). Once fertilization takes place, cleavage proceeds through blastulation, gastrulation, and finally the differentiation process.

Role of sex chromosomes

O. H. Mohr suggested a morphological difference between the X chromosome of the egg and that contributed by the X-bearing spermatozoa. (Mohr as reported in Mittwoch, 1967). This idea was not seriously suggested again until Lyon (1961) published his single active X hypothesis
and not really substantiated until cytological evidence was presented (Murherjee et al., 1964) and enzymatic studies were completed (Deys et al., 1972). Mittwoch (1973) segmented the Lyon hypothesis in four parts:

1. The genes on the sex chromatin forming one X chromosome are inactive.

2. Inactivation of the X chromosome occurs at random, so that paternal and maternal X chromosomes have an equal chance of being inactivated.

3. The process of inactivation occurs early in embryonic life.

4. Once inactivation has begun, the same X chromosome will always be inactivated in the descendents of each cell.

More recent work seems to indicate that in one marsupial species (kangaroo) the paternal X is the one whose genes are inactivated (Sharman, 1971). One X chromosome is found to be late replicating (an indication of inactivity) in the female of the primate marmoset (McLaren, 1974). It is late replicating in both sexes of chimeric marmosets as shown by autoradiography.

Some workers do not accept the Lyon theory completely. The abnormal situation of more than two X chromosomes in XXX or XXXX females or of one excess X chromosome in XXY males apparently does not show inactivation of the additional X chromosome (s) since the presence of it is related to pathology of the bearers in a number of species. However, they may still have no active genetic effect, but rather play a role in the rate of cell proliferation (Barlow, 1972). In view of the volume of supportive literature, it seems that the Lyon theory is valid, at least for some species.
Role of hormones

Hormones secreted by the developing testis appear to play a role in development of the Wolffian duct derivatives: vas deferens, seminal vesicles, a penis or copulatory organ, and also the male sex accessory glands (Herschler and Fechheimer, 1967). A similar pattern is seen in females regarding the development of the Mullerian duct and its derivatives: the Fallopian tubes, uterus and vagina. However, these develop, in the absence of virilizing hormone, whether or not estrogens are produced (Jost, as reported in Dunn et al., 1968). It is generally accepted that these hormones are necessary for the production of secondary sex characteristics such as mammary glands (Banerjee and Rogers, 1971).

Chromosomal abnormalities

Definitions of abnormalities as used in this discussion:

chimerism--condition of individuals with two or more cell lines, each of which has a different genetic origin. These cell lines are capable of production of different tissue lying adjacent or overlapping (Stern, 1968).

freemartin--this is a condition of intersex resulting in sterility of a female born twin to a male in bovine and occasionally ovine species.

intersex--an animal with congenital anatomical variations that confuse the diagnosis of sex; may be chimeric, hermaphroditic, or mosaic (McFeely et al., 1967).

mosaicism--condition of individuals with two or more cell lines differing in chromosomal constitution, but originating from a single zygote. This condition arises from errors in mitosis after fertilization (Williams, 1968).

pseudohermaphrodite--an animal having gonads of one sex and the reproductive organs characteristic of the other sex (McFeely et al., 1967).

true hermaphrodite--an animal having the gonads of both sexes (Goodfellow et al., 1965).
Understanding of normal development has been both increased by
and a factor in the understanding of abnormalities. Receiving the
most attention are abnormalities that affect man, but certainly second
to that category are those abnormalities affecting domestic animals of
economic importance. Because of this interest, as well as the visible
nature of certain abnormalities, there is written mention of such dating
to ancient times (Hutt, 1964).

**Freemartin chimerism**

One such anomaly is the occurrence of bovine twins of opposite
sexes. It has long been observed that the female twin shows varying
degrees of masculinity in morphology and behavior and is, in nearly all
cases, sterile (Moore and Rowson, 1958). Such females are known as
freemartins.

Lillie (1916) showed that in twin calves placental blood vessels
anastomose in the connecting part of the chorion during early stages of
uterine life. This allows mixing of the primordial blood lines so that
both lines are established in each animal. In like-sexed twins this
presents no problem, although it can be established by blood antigen
studies that it does occur (Owen, 1945). However, in mixed twins,
according to Lillie,

> ... the reproductive system of the female is largely suppressed
and certain male organs even develop in the female. This is un-
questionably to be interpreted as a case of hormone action.

(Lillie, 1916)

So plausible was Lillie's explanation that it wasn't questioned
to any degree for nearly thirty years. Then R. D. Owen (1945) showed
that most dizygotic twin calves share identical blood antigens. Some
forty antigens were known at that time, so the possibility that the
same combinations could occur by chance is nil. Since full siblings are not found to have this erythrocytic similarity and since monozygotic twins are extremely rare in cattle, it appeared that there was an actual blood exchange before the development of immunological competence (Owen, 1945; Moore and Rowson, 1958). In further support, Owen cited a case of superfecundation in cattle where twins of opposite sex and by different sires had identical blood types, each possessing antigens that could not possibly have come from one sire and the dam. In each case, however, they could have come from the co-twin. These cell lines persisted to adult life, indicating not only an exchange of blood, but of precursor cells capable of being established in the hemopoietic tissues of their co-twin hosts (Owen, 1945; Laster et al., 1971).

It naturally follows that if the co-twins are immunologically compatible, then they should be receptive to mutual skin grafts as are monozygotic twins. This was found to be successful in cases of dizygotic twins of unlike sex where the female is a freemartin in cattle (Anderson et al., 1951) and in sheep (Moore and Rowson, 1958). Although more recent work indicates it is successful at a lower level than vascular anastomosis is found to occur (Slee, 1963; Stone, as quoted by Basrur and Kanagawa, 1969), it still has a success level much higher than might be expected for siblings.

Chimerism is not a quality seen only in leukocytes and erythrocytic antigens. With the production of the mixed cell population maintained throughout life and the hemopoietic system intimately involved, it was not surprising that a chimeric relationship was found in the bone marrow of both twins in unlike-sexed pairs (Ohno et al., 1962). The
The ratios of XX:XY cells were weighted in favor of the apparent genetic sex of the animals in the case of one pair, but showed a similarity between twins, regardless of sex, in the other pair studied. This latter result is in agreement with the findings of others in the instance of bovine quadruplets (Basrur et al., 1971).

Chimerism is readily demonstrated in a number of additional somatic tissues. It was observed in liver of a set of bovine triplets at the 30th day of gestation (Ohno and Gropp, 1965), in spleen of several bulls of unlike-sexed pairs (Teplitz et al., 1967), in lung of cattle (McFeely et al., 1967), and in epithelial cells of a man with normal phenotype (Sperling, 1974).

The one area that has most interested and intrigued investigators since Ohno (1962) suggested it, is the chimerism of gonads. He found XX cells present in the testes of bulls of both twin sets he studied. In one case there were more than twice as many XX cells as XY, although the testis was normal in every respect and histological examination showed vigorous mitotic activity of germ cells. (A similar ratio was seen in the bone marrow of this animal.) None of the XX germ cells was seen beyond metaphase, so it is not known if they can actually differentiate into viable sperm. Ohno speculated that they might have arrived in the genital ridges via an anastomosed blood vessel. At an early stage mammalian germ cells migrating from the yolk endoderm are undifferentiated, as also is the gonad. Hence, they might be carried there through the blood exchange, settle in the indifferent gonad without rejection, and be expressed as a spermatocyte, despite the genetic expression of the XX complement.

In the case of XX/XY chimerism of a human male the somatic cells
were found to contain about equal amounts of XX and XY cells while the
testes showed only XY (Sperling, 1974). Benirschke and Brownhill (1963)
found germ cell chimerism in the gonads of both species of the marmoset.
Some XX cells in the testis of the male had progressed to diakinesis.
Teplitz et al. (1967) found the germ cell chimeric ratios in bulls which
were born twin to females to be much lower than for somatic tissue of
the same animal. Possible explanations for this are "... early re-
jection of the donor germ cells by the host or selective elimination of
donor cells during gonadal reconstitution at sexual maturity or the
brevity of the period during which germ cells migrate, limiting the
number of donor germ cells ... completing the trip." (Teplitz et al.,
1967). Weiss and Hoffman (1969) had similar results with samples from
bulls less than three months of age. However, while the leukocyte
chimerism persisted, the testicular sample taken at 18 months of age
showed no XX cells remaining there. This is in agreement with the
fact that no deviation from the one to one (1:1) normal sex ratio is seen
in the progeny of chimeric bulls (Dunn, as reported in Teplitz et al.,
1967).

One puzzling aspect of Ohno's work (1962) was that not one XY
germ cell was found in the female gonads. The number of XX metaphase
figures was also very low, perhaps indicating that most germ cells (of
either type that may be present) are eliminated in the freemartin
gonad by androgens from the male co-twin (Ohno, 1962). Goodfellow et al.
(1965) also found very low levels of XX germ cells in the gonads of the
two freemartins they studied.
Morphology of the freemartin and in utero siblings

The morphological features of the freemartin include female external genitalia, but with the Mullerian duct derivatives missing or greatly reduced. Ovaries are also reduced and non-functional, often with some portions containing seminiferous tubules (Mittwoch, 1973). Many freemartins show an enlarged or segmented clitoris. Vaginal length is reduced and often ends as a blind passageway. Laster et al. (1971) found the mean vaginal length to be 11.2 cm ± 1.9 cm. The total length in normal females is 37.0 cm ± 4.1 cm. Freemartins show varying extents of internal masculinization. Some show vas deferens, seminal vesicles, and rudimentary epididymis as well as gonads with testicular tissue (Ohno et al., 1962; Goodfellow et al., 1965; Herschler and Fechheimer, 1967). The same developmental errors are seen in sheep (Dain, 1974) and in two pigs suspected of being freemartins (Somlev, 1970). Anatomical abnormality does not occur, however, in female twins of marmosets showing blood cell chimerism (Benirschke and Brownhill, 1963) or in humans (Woodruff, 1962). This may be due to enzymes present in the placenta of these primates, but not in lower forms. Herschler and Fechheimer (1967) suggest this may be due to poor acceptance of donor tissue (such as germ cells) and a delay in the time at which chorionic anastomosis takes place in these primate species.

While bulls born twin to a freemartin seem to be phenotypically normal (Goodfellow et al., 1965; Ohno et al., 1962) and functionally effective (Lillie, 1916; Mittwoch, 1967), this is not always the case. There is a higher mortality rate among sheep chimeras, perhaps indicating a general condition of poor health (Dain, 1974). Dain suggests there may be some immunological disadvantage in chimerism of the
reticulo-endothelial system. This may be correct, but it is not in agreement with the selective advantage of hemoglobin heterozygotes of sheep in the mountains of Pamir (Raushenbakh, 1976). There is a higher rate of chimerism among inbred flocks which might tend to have some loss of vitality for reasons unrelated to the chimeric condition. Makinen (1974) followed the case of an XX/XY chimera bull for five years and reported consistently poor quality semen with a high percentage of dead sperm and low motility of the remainder. Similar findings were reported by Stafford (1972) leading to Makinen's conclusion that such animals are best excluded from artificial insemination programs.

Laster et al. (1971) have found a correlation between the degree of masculinization and the proportion of male:female calves in multiple births. Those having a greater proportion of females were more likely to show production of gonadal tissue, less likely to have seminal vesicles, and reproductive tracts were generally less masculinized. Herschler and Fechheimer (1967) report similar findings. Such females were also found to have lower percentages of littermate donor cells circulating in peripheral blood (Basrur et al., 1971). Dain (1974) however, did not find a relationship between percentages of XY cells in cultured leukocytes and the degree of masculinization of freemartin sheep. For the present, that appears to be a matter of species difference, but more work is indicated in this area.

Mechanism of chimera origin

With the passage of more than 30 years since Owen's work (1945) elucidating the basic mechanism of chimerism, there is still considerable mystery surrounding the details. There are several reasons
for this. The generation time of cattle is quite long: 278-284 days for gestation (Salisbury and VanDemark, 1961), 15-18 months to breeding levels of sexual maturity (Lerner and Donald, 1966), and perhaps several years before reproductive patterns are seen. Breeders are anxious to rid themselves of unproductive stock, so these animals very often become beef. Test herds are costly to acquire and maintain, so it is tempting to generalize on the basis of smaller laboratory animals. Because of these time and cost factors, conclusions are often drawn from a relatively small number of cases. This results in some ambiguity and impedes the search for definitive information.

In spite of these handicaps, a considerable body of information has become available in recent years regarding the mechanism of freemartin origin. Some workers still embrace the humeral theory of Lillie which accepts the contribution of androgens from the male twin to the freemartin as the causative agent in freemartin development (Dain, 1974). Others modify it with the hypothesis of Jost (as reported in Dunn et al., 1968) suggesting the production of at least two substances by the male co-twin testicular tissue. One would stimulate the male portion of the indifferent gonad while the other would inhibit the expression of the female portion. Dunn et al. (1968) believes the freemartin could arise without the presence of XY cells as long as the hormonal contribution is made to her circulatory system.

There are several problems with the humeral theory. No embryonic substance, produced by male gonad, has been identified which will alter gonadal tissue (Herschler and Fechheimer, 1967). Also, laboratory attempts to simulate such a condition with both young and adult animals were not successful in altering gonadal tissue (Jost as reported in Dunn
et al., 1968). Some intersexes were produced with modification of external genitalia and production of male sex accessory structures, but these are known to be under the influence of androgens (Mittwoch, 1967) and therefore did not add credibility to the above-mentioned hypotheses. The most remarkable problem, however, to the acceptance of hormones as the cause of the freemartin syndrome is the observation by Goodfellow et al. (1965) of asymmetry within the freemartin gonad. Since both gonads reside in the same hormonal environment, such a difference would not be expected if, indeed, hormones were the sole causative agent.

McFeely et al. (1967) suggest that the X chromosome may contain homologous, male-determining genes that can be either repressed or de-repressed. In the presence of a Y chromosome, the genes are de-repressed and maleness prevails. In the absence of a Y chromosome, the genes are repressed and femaleness is expressed. Therefore, an intersex will result with testicular tissue produced in an area occupied by XY cells and ovarian tissue located where cells are XX-bearing. They concur with other workers in the method of transfer of primordial germ cells via vascular anastomoses (Ohno and Gropp, 1965; Goodfellow et al., 1965).

The cell admixture theory of the origin of the freemartin as first suggested by Fechheimer et al. (1963) is perhaps the most widely accepted. This theory is based on the observation of cells donated by the male co-twin found migrating throughout the body of the prospective freemartin (Ohno et al., 1962; Goodfellow et al., 1965). It is known that, prior to the 42-54th day of gestation, the bovine gonad is undifferentiated. It is also known that vascular anastomoses occur prior to the 30th day of development (Ohno and Gropp, 1965). During
this period of time primordial germ cells are migrating from the endoderm of the yolk sac to the forming genital ridges. Since germ cells of one twin, as identified by alkaline phosphatase reaction, pass to the other (Ohno and Gropp, 1965) as also do hemopoietic and other somatic cells, then it is logical to assume that some may find their way to the forming gonad (Fechheimer et al., 1963). Cells bearing the XX chromosome complement have been demonstrated in the testes of bulls, co-twin to a freemartin, but so far there have been no discoveries of XY cells in the female gonad (Goodfellow et al., 1965). Since there are very few mitotic cells seen there, it is still quite possible that they have not been detected. The work of Goodfellow et al. (1965) supports this theory and further suggests that the condition is similar in result to double fertilization in man.

The primary etiological factor of gonad modification, according to Herschler and Fechheimer (1967), is probably the Y chromosome-bearing cells that interfere with the development of the female gonad. The Y chromosome is more foreign (and thus more pathological) in the body of a female than an X chromosome is in the tissues of a male. Thus, the exchange of cells is more devastating to the development of the female than it is to the male. This view is supported by the finding of Laster et al. (1971) of the presence of a higher proportion of sex nodules (indicating more X chromosomes and fewer Y chromosomes) in the less masculinized fremartins.

**Tetraploidy**

It has been known for some time that intense inbreeding of cattle leads to the expression of some undesirable phenotypes due to the increased probability of recessive genes occurring in homozygous conditions
Such an example is that of dwarf calves occurring in several beef cattle breeds (Gardner, 1972). With the widespread use of artificial insemination, a few sires are responsible for a large share of the present Hereford beef population. In propagating the desirable characteristics of these bulls, certain recessive alleles have been expressed phenotypically.

The effects of inbreeding are quite evident in these cases, but a more subtle loss of productivity is seen in other studies, such as depressed birth weights, impaired growth rate, and reduced reproductive success (Woodward and Clark, 1959; Carter et al., 1963). Falconer (1960) found a linear relationship between this fitness decline and the intensity of inbreeding, but did not identify the specific mechanism involved. Zartman and Fechheimer (1967), theorizing that this could be related to abnormal chromosome numbers in somatic cells, compared cytogenetic data on 17 inbred and 15 linecross bulls. They found no significant differences in the totals of polyploid cells when data were pooled within each group. However, when only the data from inbred sons were pooled, differences among sire lines approached significance.

**Trisomy XXX**

The occurrence of three X chromosomes is a type of primary trisomy in which one additional X chromosome is added to the normal diploid complement. This occurs with relatively high frequency as a result of non-disjunction of the female gamete. The primary oocyte then produces gametes which are either N+1,XX or N−1,0. When the XX egg cell is fertilized by an X-bearing spermatozoa, the resulting zygote is 2N+1,XXX (Hamerton, 1969). In humans this condition does not produce significant
abnormalities although it is associated with a slightly higher rate of mental retardation and/or lowered fertility (Sybenga, 1972) than in the normal population. Khush (1973) states that the tolerance for this form of aneuploidy in animals is much greater than the tolerance for aneuploidy of autosomes. This is presumably because of the inactivity of a large portion of chromatin contained in the sex chromosomes (Lyon, 1961).

**Autosomal aneuploidy**

The deletion or addition of one or more autosomal chromosomes in higher animals is very often accompanied by some form of pathology: lethality at early developmental stages or serious malformation of the affected organism which reduces vitality and longevity (Puck, 1972). However, the mosaic condition resulting when some somatic cells carry the aberrant chromosomal number and others the normal diploid complement can often be tolerated successfully. Zartman and Fechheimer (1967) found a population of apparently normal yearling bulls carrying 16.6% peridiploid cells among 1,200 counted for 40 animals.

Spontaneous, incidental non-disjunction of one chromosome is the most common source of aneuploidy when only one chromosome is deleted or added (Sybenga, 1972). Exposure to ionizing radiation can cause damage to the spindle apparatus and produce a population of aneuploid cells in this way also (Fechheimer, 1968). The condition can be artificially induced by treatment with certain physical and chemical agents (Khush, 1973).

**Robertsonian translocation**

Numerous types of translocations can occur in chromosomes of plants and animals with variable effects on the organism. Animals are better
able to accommodate this type of change than one in which genetic material is added or deleted. It is thought likely that certain kinds of translocations have played an important role in the development of recent evolutionary variation (Sybenga, 1972). One such process resulting in a reduced chromosome number is centric fusion. Centric fusion occurs when two acrocentric chromosomes become unified by merging the centromeres. This type of translocation reduces the diploid (2N) number by one, but maintains the same number of chromosome arms—that is, the entire chromatin complement. For the early work of W. R. B. Robertson (1916) with this aberration, it has been given the name, Robertsonian centric fusion (Sybenga, 1972).

This type of fusion can result from several different combinations, most of which form trivalents at meiosis with the remaining, minute chromatin particles being lost eventually. However, it is necessary that the gametes be genetically balanced to survive, so only those fused chromosomes which are able to achieve this would be expected to succeed in a population.

A translocation of this type is seen in one breed of Swedish and Norwegian cattle at levels in excess of 14%. It involves the fusion of acrocentric chromosomes, numbers 1 and 29, which join to form a new sub-metacentric chromosome (Gustavsson, 1969). A translocation involving the same two chromosomes was seen by Herschler and Fechheimer (1967) in the leukocytes of female origin in a set of bovine triplets. All animals, two males and a female, exhibited XX/XY chimerism with the XX cells also carrying the 1,29 Robertsonian translocation and a diploid number of 59 (sixty being the normal diploid number in Bos taurus, Hsu and Benirschke,
1967). The female was a freemartin with a grossly maldeveloped reproductive tract, while the males appeared to be normal. The abnormalities of the female were thought to be due to the XX/XY chimerism rather than to the translocation.
III. MATERIALS AND METHODS

Animals used in this study are members of the University of Illinois dairy research herd, their offspring, and sires. The herd numbers approximately 300 members and is composed of Holstein, Brown Swiss, Jersey, Guernsey, Ayrshire and mixed breeds.

Chromosome preparations

Venipuncture blood samples were collected aseptically from 109 bovine individuals and placed in culture within a few hours of collection. Whole, heparinized blood was cultured in TC Medium 199 (Difco, Detroit) containing pokeweed mitogen (secured from Grand Island Biological Company, New York). Colcemide was used as the mitotic spindle inhibitor. A vortex mixer was utilized for mixing during harvesting of the leukocytes. Smears of the cell suspension were prepared, fixed, and stained with buffered Geimsa solution. The techniques used in this procedure are described by Smith et al. (1976).

Cytogenetic survey

Examinations were made by scanning the slides for metaphase spreads with the Ph 2 Neofluar objective of a Zeiss phase-contrast microscope at a magnification of 400X. In cattle (Bos taurus, 2N = 60), the sex chromosomes are quite easily identified (see figs. 1 and 2) as they are submetacentric while all the autosomes are acrocentric or telocentric (Hsu and Benirschke, 1967). In well-prepared spreads it was possible to determine sex and the diploid number at this magnification. When it was necessary, examinations were made under oil immersion at a magnification
of 1000 X. A minimum of five spreads was routinely examined under oil for each individual while a total of at least 25 metaphase spreads was counted for each animal. Karyotypes were surveyed for irregularities in addition to assessment of sex and 2N number. If abnormalities were found in less than 100% of the mitotic spreads of an individual animal, the percentages in which they occur were computed.

Photomicrograph preparations

Examples of any karyotypes varying from the normal (2N = 60, XX or 2N = 60, XY) were recorded by photomicrographs. They were taken on a Nikon AFM camera, M-35s, mounted on an AO-Spenser research scope. Kodak Panatomic X film was used at ASA 32 and printed on standard Kodabromide paper.

Individual histories

Informational data were secured and recorded for animals exhibiting any karyotype abnormality. This included birth date, age, breed, phenotypic sex, reproductive history, significant medical history, final disposition. However, this was not done until all counts were completed to prevent biasing the data.
IV. RESULTS

XX/XY chimeras

There were eight chimeric individuals discovered in this mixed population of dairy animals (see figs. 1-12). Only one was a known freemartin, Q 3292 (see figs. 9 and 10). Unfortunately, none are available for observation or histologic examination. Data in table 1 show the relative percentages of male and female leukocytes in these animals, their breeds and final disposition.

The freemartin female, 3292, is the only animal with more than 50% XX cells although three of the steers, numbers 2456, 372, and 367, approach that amount with 40%, 47%, and 48% respectively. The reproductive history on this animal is incomplete. She was diagnosed in estrum at 15 months of age, but not bred. A month later she was sold and no further records are available.

Of the seven steers, four were raised to the age of 16-20 months and then slaughtered for beef. All were noted in good health at the time of slaughter. The other three died soon after birth. Pneumonia was the probable cause in one, but cause was unknown in the other two. It is of interest to note that these three animals had the lowest rates of XX cells, 13%, 4%, and 4%. Of the eight chimeras, four were shown to be Holstein, two were cross-breeds and one each Guernsey and Brown Swiss. This is a similar ratio to the composition of the total herd. With eight total chimeric individuals, only one of which was known to be a twin, the rate of chimerism in the sampling of this dairy herd is found to be 7.3%.
Tetraploidy

A mosaic condition for tetraploidy (see figs. 13-16) was seen in six individuals as shown in table 2. At least 8% of the mitotic spreads observed in these individuals were tetraploid (4N) with others ranging as high as 28% 4N. One steer, 355, was plagued with poor nutrition for several months, eventually requiring a rumen plug to prevent bloating. He died at 12 months of age. One cow, 3308, in three pregnancies produced one normal calf, one third-month abortion and was the dam of one of the XX/XY chimeras described above, 3308-s. That calf died four days after birth of unknown causes.

The remaining four animals include three steers slaughtered at ages of 14 months, 15 months, and 18 months all in good health at time of slaughter, and one cow successfully reproducing at the present time. The case histories of these animals were not remarkable. All animals showing this condition were Holstein.

Trisomy XXX

Three female individuals were found carrying an extra X chromosome (see figs. 17-19) in a small percentage of cells (see table 3). The XXX condition was seen to exist in percentages of 4%, 8%, and 12% in these three cows. This condition did not seem to produce any reproductive deficiency in these small amounts as all three animals are productive members of this dairy herd. All have produced calves and are in good health. Breeds include one each of Holstein, Jersey, and a cross-breed.

Autosomal aneuploidy

Two male individuals were seen to have some cells with a chromosome number that is not an exact multiple of the monoploid (N) or basic number
In the steer, 2729-s, 32% of the metaphase spreads were seen to contain 58 or 59 chromosomes. The other individual, 2961-s, showed 6% of the metaphase spreads with 2N = 61 (see fig. 20). Both these animals died of pneumonia at ages of three months to five months. One was Holstein and the other Brown Swiss.

Robertsonian translocation

One Brown Swiss steer, 360, was found to carry an anomalous submetacentric chromosome in addition to the X and Y sex chromosomes (see figs. 21 and 22). This submetacentric chromosome is distinguished from the normal X chromosome by the length of the long arms. Those on the translocated chromosome are 50% longer than those of the X chromosome. This results in a total chromosome length increase of 33%. There is some variability in the size of chromosomes from cell to cell depending on the stage of the mitotic process, but within a cell the relative sizes are highly uniform. It is therefore concluded that this is a Robertsonian translocation (Herschler and Fechheimer, 1967).

This pattern was seen in 81% of the cells counted and was apparent in all cases where the chromosomes were particularly well spread (see table 3). Most of these cells were seen to have 59 chromosomes rather than a normal 60. In some cases, however, a small bit of chromatin was seen as a possible chromosome number 60.

This animal exhibited good health throughout his life. He was shown at the World Dairy Exposition in Wisconsin, where he brought a top beef price at the age of 19 months.
V. DISCUSSION

XX/XY chimeras

It is interesting to note that seven of the eight leukocyte chimeras were steers. This is possibly because the role of females in a dairy herd—to produce young and hence to lactate—is impaired by the reproductive limitations known to result from chimerism (Lillie, 1916). Since the major interest in a dairy research herd is that of quality and quantity of milk produced, any female chimera occurring from either a heterogeneous multiple birth or an apparent single birth might be selectively removed.

There is still disagreement in the literature regarding the effectiveness of XX/XY chimera bulls. They are phenotypically normal and have been thought to be functionally effective (Mittwoch, 1967). However, recent findings (Stafford, 1972; Mäkinen, 1973) indicate production of low quality semen in these animals. Therefore, male calves exhibiting XX/XY blood cell chimerism would, in all probability, be castrated or eliminated.

Since breed is an artificial distinction within a species, one would not expect to see major chromosomal differences between breeds. However, the inbred nature of domestic animal herds has been shown to contribute to a loss of vitality in some cases (Gardner, 1972). In this study there did appear to be a higher rate of Holstein animals showing this anomaly than any other breed, but the composition of the herd is predominately Holstein at any given time. Hence, there is no significant
relationship between breed and the occurrence of chimerism in this dairy herd.

The mortality rate in the first three months of life was high for this group of animals, 37.5%. The normal expected loss rate for cattle in the state of Illinois is 16.4% (Salisbury and VanDemark, 1961). Since the young of the research herd were subjected to optimum environmental, nutritional, and medical conditions and the dams to good husbandry practices, the loss percentage would be expected to be considerably less than state averages.

Dain (1974) has found a similar result among a small population of chimeric sheep and speculates it may be due to some immunological disadvantage of the reticulo-endothelial system. Since Dain's population and ours are both quite small, and since some do exhibit good health, it is not possible to determine if the early deaths are significant.

It is of interest to note that the three young males that died, 2593-s2, 3204-s, and 3308-s, had the lowest percentages of XX cells in the leukocyte cultures: 4%, 13%, and 4%, respectively. In Dain's study (1974) of sheep, two of the three which died at an early age showed low percentages of XX cells although all three were phenotypic females. It is tempting to speculate regarding a survival advantage conferred by the higher rate of XX cells, but again with such small numbers, this may be a coincidental finding.

The data show that XX/XY chimerism, as demonstrated in figs. 1-12, exists in a given population of dairy cattle at a total rate of 7.3%. The known heterogeneous twin birth, 3292, in this statistic would place the expected rate at 1%. According to Johansson and Venge (as quoted in Salisbury and VanDemark, 1961) the rate of twining is 1.82%. 
Approximately 50% of these would be expected to be heterogeneous. The experimental finding, then, represents a seven-fold increase in the predicted level and a similar increase over the expected rate. The question, then, is how XX/XY chimerism can occur in seven individuals not recorded as members of multiple births. In the cases of the very young male animals, a small amount of maternal blood might have seeped across the placental barrier prior to or during birth (Baker, 1967) and be found circulating in the blood stream of the young male calf. Leukocytes are thought to survive in the human circulatory system from 24 hours to a few days (Leeson and Leeson, 1976). Assuming this is common to other mammals, it is theoretically possible to see an occasional XX cell from this source.

In animals past the first week or two of life, and in the cases with higher percentages (356, 367, 372, and 2456) this could not be the explanation. In these cases and probably in all, it is our view that multiple ovulation produced a pair of heterogeneous twins (or other multiple embryos). Both fetuses survived at least to the 30th day when vascular anastomosis occurred. Prior to delivery, the female co-twin was aborted, so evidence of her presence was not obvious at the time of birth of the male. This theory is supported by the fact that multiple ovulation in cattle is known to occur much more often than the frequency of twining (Hafez, 1974). The demise of one member would explain the variation in these statistics as well as the phenomenon of apparent single birth chimeras.

Tetraploidy

Six animals were seen to be 2N/4N mosiacs with a minimum ratio of
11.5:1 or 8% tetraploid. If only one cell per 25 counted was found to be tetraploid, it was recorded, but not included in these statistics. Centerwall (1973) suggests that polyploid cells are more likely to occur as a culture ages and as subcultures are produced from it. It is our view that a random 4N cell might be an artifact of culture technique.

While not strongly supported by their grouped data, Zartman and Fechheimer (1967) did see mosaic polyploidy in bovine inbred sons more frequently than in control groups or in linecross bulls. The six animals identified as tetraploid mosaics in this study, however, were each sired by different bulls. The sire of 3187-s1 has produced an inordinate number of polyploid offspring (personal communication, Albert Smith). Additional investigation of this lineage will be made in a future report.

The six animals showing more than 8% tetraploid cells were all Holstein. At cursory glance, this might suggest a relationship between some of the features for which Holsteins are bred and the production of polyploidy. However, that conclusion cannot be drawn on the basis of our findings since, at any one time, this particular herd is predominately Holstein. The finding of six animals of the total 109 is not significant at the P<.05 level when considering that 67 of that total are Holstein. It is, however, an area that requires further investigation.

Trisomy XXX

The three animals showing trisomy X (see figs. 17-19) in at least 4% of the cells counted are all successful producers in this dairy herd. There are no apparent phenotypic or behavioral abnormalities. It appears that the additional X chromosome carried in a small percentage of the total cell population has no harmful effect on these animals. There is
no relationship between the occurrence of this anomaly and the breeds of animals.

**Autosomal aneuploidy**

The two animals identified as being hypo- or hyper-diploid (see fig. 20) died of pneumonia within a few months of birth. One is tempted to speculate that the abnormal chromosomal condition contributed to the poor health of those individuals, since any variance from the normal diploid number is so devastating in man (Hamerton, 1969) and reduces hatching success in chickens (Zartman and Smith, 1975). However, with such a small sample, the deaths of these two individuals could be unrelated.

**Robertsonian translocation**

The translocation seen in 81% of the cells of steer 360 appears to be a centric fusion (see figs. 21 and 22) as described in the literature (Gustavsson, 1969; Herschler and Fechheimer, 1967) and involving autosomes 1, 29. A 2N number of 59 is the result of this fusion. With such a high percentage of the translocated cells, it is our view that this animal produced 100% of this cell type. The fusion between the two separate chromosomes, joined by centromeres, is quite loosely held since minute amounts of chromatin are being shared. It is entirely logical that such an adhesion would be subject to a high rate of disturbance during the culture procedures. Hence, a certain portion of the cells (in this case, 19%) are seen as the normal diploid complement.

Of further interest is the fact that animal 360 was a high-quality steer, shown at the World Dairy Exposition and brought a maximum price there. Herschler and Fechheimer (1967) suggest that the Robertsonian
centric fusion of chromosomes may provide a selective advantage for cattle as has been reported for some other species. Our findings of this champion specimen would lend support to that theory.

The origin of this aberration is still in doubt. It may occur during meiosis, mitosis, or be transmitted genetically through one of the parents. Present data do not allow us to speculate, but suggest an area for further study.
VI. CONCLUSIONS

1. Of the University of Illinois dairy research cattle, their offspring, and sires sampled in this study, 7.3% are found to be XX/XY chimeras although only one was a product of a known heterogeneous multiple birth. The remainder are presumed survivors of multiple, mixed-sex incubations in which the female sibling(s) were subject to spontaneous abortions after the time of vascular anastomosis. There is found to be a higher rate of early death among these animals than the population in general. No relationship is shown between a specific breed and the occurrence of this anomaly.

2. The occurrence of tetraploid-diploid mosaicism is seen in 5.5% of the animals in this study. While one steer died after experiencing nutritional difficulties and one female had a poor reproductive record, there is no evidence of a correlation between these physiological problems and the tetraploid-diploid condition. While all individuals showing this condition are Holstein, the predominance of this breed in the herd makes this finding of questionable significance.

There was no relation between this condition and sire lineage among these six individuals. The sire of 3187-s1 has been reported producing other tetraploid animals, however.

3. Mosaics showing trisomy of the X chromosome, 2N + 1, XXX/2N, XX, are found in 2.8% of the animals sampled in this study. There is
no pathology evidenced from this anomaly. No relationship is seen between breed and the occurrence of this condition.

4. Autosomal aneuploidy is observed in 1.8% (two cases) of the individuals in this sample, both of whom died of pneumonia at an early age.

5. A Robertsonian centric fusion of chromosomes 1 and 29 was seen in 81% of the cells of one individual in this sample. This steer displayed superior form. This type of centric fusion may provide a selective advantage in *Bos taurus*.
VII. APPENDIX
Fig. 1. An XX-bearing cell of animal #356, a phenotypic male. (1000 X)

Fig. 2. An XY-bearing cell of animal #356, a phenotypic male. (1000 X)
Fig. 3. An XX-bearing cell of animal #367, a phenotypic male. (1000 X)

Fig. 4. An XY-bearing cell of animal #367, a phenotypic male. (1000 X)
Fig. 5. An XX-bearing cell of animal #372, a phenotypic male. (1000 X)

Fig. 6. An XY-bearing cell of animal #372, a phenotypic male. (1000 X)
Fig. 7. An XX-bearing cell of animal #2456, a phenotypic male. (1000 X)

Fig. 8. An XY-bearing cell of animal #2456, a phenotypic male. (1000 X)
Fig. 9. An XX-bearing cell of animal #3292, a phenotypic female. (400 X)

Fig. 10. XY-bearing cells of animal #3292, a phenotypic female. (1000 X, 400 X)
Fig. 11. An XX-bearing cell of animal #3204-s, a phenotypic male. (1000 X)

Fig. 12. An XY-bearing cell of animal #3204-s, a phenotypic male. (1000 X)
Fig. 13. Tetraploid cell of animal #2834-sl. (1000 X)

Fig. 14. Tetraploid cell of animal #3308. (1000 X)
Fig. 15. Tetraploid cell of animal #3208. (1000 X)

Fig. 16. Diploid cell of animal #3208. (1000 X)
Fig. 17. Cell demonstrating trisomy XXX in animal 
#3204, an XXX/XX mosaic. (1000 X)

Fig. 18. Cell demonstrating trisomy XXX in animal 
#3289, an XXX/XX mosaic. (1000 X)
Fig. 19. Cell demonstrating trisomy XXX in animal #3297, an XXX/XX mosaic. (1000 X)

Fig. 20. Cell demonstrating autosomal aneuploidy of animal #2961. (1000 X)
Figs. 21, 22. Cells demonstrating Robertsonian translocation of chromosomes 1 and 29 in animal #360. Double line indicates translocation. (1000 X)
TABLE 1.—Incidence of XX/XY chimerism in a sampled dairy herd, *Bos taurus*.

<table>
<thead>
<tr>
<th>Animal number</th>
<th>Sex</th>
<th>Breed</th>
<th>% XX cells</th>
<th>% XY cells</th>
<th>Fate</th>
</tr>
</thead>
<tbody>
<tr>
<td>356</td>
<td>M</td>
<td>Cross</td>
<td>27%</td>
<td>73%</td>
<td>Slaughtered in good health at 19 months</td>
</tr>
<tr>
<td>367</td>
<td>M</td>
<td>Hol.</td>
<td>48%</td>
<td>52%</td>
<td>Slaughtered in good health at 18 months</td>
</tr>
<tr>
<td>372</td>
<td>M</td>
<td>Cross</td>
<td>47%</td>
<td>53%</td>
<td>Slaughtered in good health at 20 months</td>
</tr>
<tr>
<td>2456</td>
<td>M</td>
<td>Hol.</td>
<td>40%</td>
<td>60%</td>
<td>Slaughtered in good health at 16 months</td>
</tr>
<tr>
<td>2593-s2</td>
<td>M</td>
<td>Br. Sw.</td>
<td>4%</td>
<td>96%</td>
<td>Died, 3 weeks, probable pneumonia</td>
</tr>
<tr>
<td>3204-s</td>
<td>M</td>
<td>Hol.</td>
<td>13%</td>
<td>87%</td>
<td>Died, 9 days, cause unknown</td>
</tr>
<tr>
<td>3292</td>
<td>F</td>
<td>Guernsey</td>
<td>60%</td>
<td>40%</td>
<td>Sold without successful breeding</td>
</tr>
<tr>
<td>3308-s</td>
<td>M</td>
<td>Hol.</td>
<td>4%</td>
<td>96%</td>
<td>Died, 4 days, cause unknown</td>
</tr>
</tbody>
</table>
TABLE 2.—Incidence of tetraploidy in a sampled dairy herd, *Bos taurus*.

<table>
<thead>
<tr>
<th>Animal number</th>
<th>Sex</th>
<th>Breed</th>
<th>% 4N cells</th>
<th>% 2N cells</th>
<th>Fate</th>
</tr>
</thead>
<tbody>
<tr>
<td>355</td>
<td>M</td>
<td>Hol.</td>
<td>24%</td>
<td>76%</td>
<td>Died at 1 year, poor nutritional state</td>
</tr>
<tr>
<td>405</td>
<td>M</td>
<td>Hol.</td>
<td>8%</td>
<td>92%</td>
<td>Slaughtered in good health at 18 months</td>
</tr>
<tr>
<td>2834-s</td>
<td>M</td>
<td>Hol.</td>
<td>28%</td>
<td>68%</td>
<td>Slaughtered in good health at 15 months</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>(also 4% 3N)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3187-s</td>
<td>M</td>
<td>Hol.</td>
<td>8%</td>
<td>92%</td>
<td>Slaughtered in good health at 14 months</td>
</tr>
<tr>
<td>3208</td>
<td>F</td>
<td>Hol.</td>
<td>12%</td>
<td>88%</td>
<td>Successfully reproducing</td>
</tr>
<tr>
<td>3308</td>
<td>F</td>
<td>Hol.</td>
<td>8%</td>
<td>92%</td>
<td>Had one abortion, 1 calf died at 4 days, 1 healthy calf</td>
</tr>
</tbody>
</table>
TABLE 3.—Other observed anomalies in sampled dairy herd, *Bos taurus*.

**Trisomy XXX**

<table>
<thead>
<tr>
<th>Animal number</th>
<th>Breed</th>
<th>% XXX cells</th>
<th>% XX cells</th>
<th>Fate</th>
</tr>
</thead>
<tbody>
<tr>
<td>3204</td>
<td>Hol.</td>
<td>8%</td>
<td>92%</td>
<td>Successfully reproducing</td>
</tr>
<tr>
<td>3289</td>
<td>Jersey</td>
<td>4%</td>
<td>96%</td>
<td>Successfully reproducing</td>
</tr>
<tr>
<td>3297</td>
<td>Cross</td>
<td>12%</td>
<td>88%</td>
<td>Successfully reproducing, sold</td>
</tr>
</tbody>
</table>

**Autosomal Aneuploidy**

<table>
<thead>
<tr>
<th>Animal number</th>
<th>Sex</th>
<th>Breed</th>
<th>% aneuploid</th>
<th>% 2N</th>
<th>Fate</th>
</tr>
</thead>
<tbody>
<tr>
<td>2729-s</td>
<td>M</td>
<td>Hol.</td>
<td>32%</td>
<td>68%</td>
<td>Died, 5 months, pneumonia</td>
</tr>
<tr>
<td>2961-s</td>
<td>M</td>
<td>Br. Sw.</td>
<td>6%</td>
<td>94%</td>
<td>Died, 3 months, pneumonia</td>
</tr>
</tbody>
</table>

**Robertsonian Translocation**

<table>
<thead>
<tr>
<th>Animal number</th>
<th>Sex</th>
<th>Breed</th>
<th>% translocated</th>
<th>% normal</th>
<th>Fate</th>
</tr>
</thead>
<tbody>
<tr>
<td>360</td>
<td>M</td>
<td>Br. Sw.</td>
<td>81%</td>
<td>19%</td>
<td>Slaughtered in good health at 19 months, shown World Dairy Exposition</td>
</tr>
</tbody>
</table>
VIII. LITERATURE CITED


