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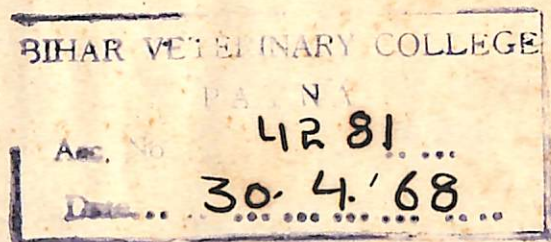
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Studies on  
REACTION TIME AND SEMEN CHARACTERISTICS OF  
THARPARKAR, CROSS-BRED AND AYRSHIRE BULLS  
AND  
THEIR CORRELATION WITH BLOOD PICTURE

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THESIS

SUBMITTED TO THE  
MAGADH UNIVERSITY

IN PARTIAL FULFILMENT OF THE REQUIREMENTS

FOR THE DEGREE OF

MASTER OF SCIENCE ( VETERINARY )

1967

BIHAR VETERINARY COLLEGE, PATNA.



Post-Graduate Department of  
Gynaecology and Sterility,  
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P A T N A.

December , 1967.

I certify that this Thesis entitled  
" Studies on Reaction Time and Semen characteristics  
of Tharparkar, Cross-bred and Ayrshire bulls and their  
correlation with Blood picture" has been prepared under  
my supervision by Shri P.M. Belorkar, a candidate for  
the M.Sc.(Vet) with Gynaecology, Obstetrics and Arti-  
ficial Insemination as major subject, and that it  
incorporates the results of his independent study.

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### ACKNOWLEDGMENTS

The author expresses his deep sense of gratitude and indebtedness to Shri B.P.S.R.Singh, G.B.V.C., P.G., F.R.V.C.S.(Sweden), Professor of Gynaecology and Sterility, Bihar Veterinary College, Patna, under whose guidance the present investigation was planned and conducted, and whose invaluable suggestions and constant encouragement have made the preparation of this Thesis possible.

I also express my feelings to Shri P.B.Kuppuswamy, Principal, Bihar Veterinary College, Patna, for kindly offering facilities for this work.

A word of gratitude is also extended to the Manager, Government Cattle Farm, Patna, for providing the facilities necessary for the present study.

My grateful thanks are also due to Shri B.K.Singh, M.V.Sc., Lecturer and Shri L.N.Purbey, M.V.Sc., Research Assistant in Gynaecology, for their cooperation and sincere advice rendered during this study.

Thanks are also due to Sri R.J.Prasad, Lecturer, Genetics, Bihar Veterinary College, Patna for his timely advice for statistical analysis.

(PRAKASH M. BELORKAR.)



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## I N T R O D U C T I O N



## I N T R O D U C T I O N

"The Sire is half the herd" is a popular saying in animal breeding. The real improvement of the animal products depends upon good sires who can transmit their inherited characters of dairy, draught and meat to their progeny who should perform better than their parents. Hence, in a herd, a sire is always valued more than the cow as she can produce hardly about half a dozen progeny in her life time, while a sire has the potentiality to produce progeny even in thousands especially with the use of the advanced technique of Artificial Insemination.

Many workers (Fraser, 1930; Scarborough, 1931-'32 and Paul, et al., 1941) have shown that males have a greater number of red blood corpuscles than females. This has been attributed to be due to the influence of sex hormones on the blood picture through their well known actions upon metabolism and general somatic growth (Paul et al., 1941). Further it is also a proven fact that testosterone initiates and maintains the production of spermatozoa and is also responsible for the male sex drive (Zarrow, 1962). From this it is clear that testosterone influences both the sperm production and the variation in the number of red blood corpuscles.

Blood and its physiological functions have been the object of great interest since ancient times. The science of



haematology in later times is, however, founded on the discovery of the blood corpuscles during the latter half of the 17th century. Swammerdam (1658) discovered the presence of the red blood cells in frogs and lice, while Malpighi (1665) found red blood cells in the mesenteric vessels of the hedgehog. Leeuwenhoek (1673) was, however, the first to prove their presence in man and also discovered the existence of lymphocytes in it. Hewson (1739-'79) after his continuous research detected the white blood corpuscles in blood.

Since then, the studies on blood picture were limited in diagnosis of the diseases only. During the first quarter of the 20th century an important advancement was made in haematological research due mainly to the discovery by Arneith (1921) and Schilling (1926) who opined that diseases in general had dissimilar effects on the different kinds of blood cells, primarily noticeable in the changes during the course of the degeneration and regeneration of the cells. Arneith's index and Schilling's haemogram still remain in force today in the orbit of haematology.

In recent years attempts have been made to correlate blood cell volume and haemoglobin contents with the quality of the bull semen (Mukherjee and Bhattacharaya, 1952). Attempts have also been made to anticipate the lactational performances of dairy animals by examination of their blood picture (Schultze, 1960). Studies have been made to estimate the breeding value of bulls by comparing their leucocyte index and



breeding performance, as evidenced by the milk yield of their daughters based on dam-daughter comparison (Grobewska, 1960). Goswami (1960) tried to establish a possible analogy of hormonal relationship with spermatogenesis as well as erythrocytogenesis.

The present study, therefore, was undertaken with a view to observe the seminal characteristics of the bulls belonging to Tharparkar, Cross-bred (Sahiwal X Brown Swiss) and Ayrshire breeds and to record correlation, if any, between the seminal picture with their blood picture.

Thus, in the present study attempts have been made;

(1) To study the seminal characteristics of bulls belonging to Tharparkar, Cross-bred and Ayrshire breeds and to compare these results;

(2) To study the normal blood picture of Tharparkar bulls at different age groups and to compare their values with the blood cellular values of Cross-bred and Ayrshire bulls; and

(3) To establish correlation, if any, between the semen picture and blood picture of the three breeds under study.

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# RESEARCH REPORT

## RESEARCH REPORT

With 1961 reported the reaction time of Tharparkar, Cross-bred and Ayrshire bulls and noted mean differences in reaction time. Valli and Choudhary (1962) reported the reaction time of Tharparkar bulls as 14.31 seconds and Ayrshire bulls as 14.31 seconds.

### P A R T - I.

#### REACTION TIME AND SEMEN CHARACTERS OF THARPARKAR, CROSS-BRED AND AYRSHIRE BULLS

The present study was conducted to determine the reaction time and semen characters of Tharparkar, Cross-bred and Ayrshire bulls. The study was conducted at the Veterinary College, Magadh University, Patna. The study was conducted during the period from 1966 to 1967. The study was conducted on 10 Tharparkar bulls, 10 Cross-bred bulls and 10 Ayrshire bulls. The reaction time was determined by the method of Valli and Choudhary (1962). The semen characters were determined by the method of Valli and Choudhary (1962). The results of the study are presented in the following tables.



## REVIEW OF LITERATURE

### Reaction time:

Smith (1951) compared the reaction time of Hereford, North Devon and Friesian bulls and noted breed differences in reaction time. Mukherjee and Bhattacharya (1952) recorded the average reaction time of Kumauni bulls as 74.34 seconds and noted highly significant difference between the bulls. Bhattacharya and Prabhu (1955) gave the average reaction time of 267.87 seconds in Sahiwal breed and noted variation among Tharparkar and among Sahiwal bulls and obtained significant difference between five Indian breeds. Bhatia (1960) reported a reaction time of  $8.63 \pm 0.69$  and  $15.3 \pm 0.53$  minutes during the year 1956-'57 and 1957-'58 in Haryana breeding bulls and noted the reaction time for Jersey bulls within a range of 30 seconds to 60 seconds for first ejaculate. The average reaction time for Sindhi bull and Gir bull was recorded as 55 seconds and 208.43 seconds respectively. Singh and Prabhu (1963) concluded that there was statistically, a significant variation among the Haryana and among Kumauni hill bulls and noted differences in reaction time between the breeds. Sinha (1964) recorded the average reaction time of Tharparkar, Haryana and Taylor bulls as 10.08, 4.71 and 11.31 minutes respectively and noted variation in reaction time among the bulls and breeds also. Kodagali (1967) recorded the average reaction time of 120 seconds in Gir bulls.



### Volume of semen:

On the basis of their research work on different breeds, Lagerlof (1934), McKenzie (1939), Herman and Regsdale (1939), Herman and Swanson (1944) and Green et al. (1941) observed the average volume of semen per ejaculate as 3.0 ml., 4.2 ml., 4.38 ml. and 4.0 ml. respectively. Anderson (1941) recorded that dairy bulls were having larger ejaculates than those of the beef bulls. In another study, he found Friesians to have larger volume than Ayrshires. Herman and Swanson (1941) found no relationship between age and Semen volume but the size of the bull had a distinct effect on the amount of ejaculate, i.e. larger bulls giving more semen than smaller bulls. Erb et al. (1942) noted highly significant variation in semen volume among bulls. Kumaran (1939, 1944, 1949) recorded average semen volume of Amritmahal, Friesian, Sahiwal and Tharparkar breeds as 4.1, 4.8, 3.9, and 3.8 ml. respectively. In a study Anderson (1945) found considerable variation in semen volume between bulls while Mercier et al. (1946) noted highly significant variation among Holstein-Friesian bulls and Guernsey bulls. Shukla and Bhattacharya (1949) reported an average volume of 2.0 ml. in Kumauni bulls, 3.16 ml. in Haryana bulls and 3.80ml. in Sahiwal bulls. Mukherjee and Bhattacharya (1952) recorded an average volume of semen in Kumauni hill bulls as 2.20 ml. and observed significant variation between bulls. Bhattacharya and Prabhu (1952) stated that the average volume of semen per



collection varied according to species and breeds. In the Bos-indicus breeds, the Sahiwal topped the list with  $3.72 \pm 0.11$  ml. and next came Tharparkar with  $3.10 \pm 0.07$  ml., followed by Nagori and Indo-European Cross-bred with  $2.84 \pm 0.11$  ml. and  $2.35 \pm 0.13$  ml. per collection respectively. They also recorded the significant variation among Tharparkar and among Sahiwal bulls and noted statistically a significant difference between the breeds. Koriath et al. (1955) found a considerable variation in quantity of ejaculate among 41 Black-Pied bulls. Bhattacharya and Prabhu (1955) recorded the average volume of semen in English bred, Sahiwal, Haryana, Nagori, Red-Sindhi and Hallikar bulls as 4.37, 5.19, 3.87, 4.05, 4.08 and 3.17 ml. respectively and noted statistically a significant difference between the breeds. Hafe et al. (1958) observed bull to bull variation in semen production but did not observe any significant difference between breeds. Zulliani and Tullio (1960) found remarkable individual variation in the amount of ejaculate in Bos-aurus species. Bhatia (1960) revealed that the average volume of semen in Haryana bull was 6.04 ml. and in Tharparkar bull it was 4.59 ml. ranging from 3.5 ml. to 8.33 ml; in Sindhi bull a range of 3.0 ml. to 6.8 ml., in Jersey bull a range of 2.63 ml. to 4.86 ml. and in Hallikar a range of 2.0 ml. to 4.8 ml. Perry (1960) stated that healthy bulls of both dairy and beef breeds had an average volume of semen as 5.0 to 6.0 ml. There seemed to be no marked breed differences in his study. Singh and Prabhu (1963) did not find significant difference in volume between Haryana



and Kumauni hill breeds. Sinha (1964) noted the average volume of semen in Tharparkar, Haryana and Taylor breed as 5.06, 5.46 and 4.09 ml. respectively and recorded highly significant variation among Tharparkar bulls and among Haryana bulls and between breeds also. Barbulescu (1965) studied on Brown, Romanian-spotted and Estonian bull semen and stated that the semen volume was affected by breed more than any other semen characteristics. Tripathi (1965) recorded an average volume of semen as 6.23 ml. in Haryana breed. Kodagali (1967) studied on semen characteristics of Gir breeds and recorded an average volume of semen in 1st year and 2nd year of study as 6.92 ml. and 6.13 ml. respectively.

#### Initial motility:

Erb et al. (1942) noted highly significant variation in initial motility between bulls. Phillips et al. (1943) analysed various characteristics of semen of three beef Shorthorn and three milking Shorthorn bulls and recorded significant variation in initial motility of spermatozoa between the bulls but did not observe significant difference between breeds. Swanson and Herman (1944) noted highly significant variation in initial motility between Missouri dairy bulls. Anderson (1945) studied seasonal variation in the reproductive capacity of bulls and noted highly significant variation between bulls and between breeds also. Mercier et al. (1945) noted significant variation in sperm motility between bulls. Mukherjee and



Bhattacharya (1952) recorded an average of 3.47 initial motility in Kumauni bulls and observed highly significant variation between bulls. Koriath et al. (1952) observed highly significant variation in initial motility of spermatozoa between bulls. Johnston et al. (1953) reported highly significant difference in motile spermatozoa among bulls and also between Guernsey, Holstein and Jersey breeds. Hafs et al. (1958) noted highly significant variation among the bulls but did not observe any variation in initial motility of spermatozoa between the breeds. Bhatia (1960) noted the average initial motility in Gir and Sahiwal breed as 4.58 and 3.82 respectively. Singh and Prabhu (1963) did not find significant difference in initial motility between bulls. Sinha (1964) recorded the average initial motility in Tharparkar, Hariana and Taylor breeds as 4.01, 3.84 and 3.79 respectively and noted highly significant variation among bulls and between breeds also. Tripathi (1965) studied on semen of Hariana bulls and noted the initial motility as 3.85, while Sexana (1965) recorded 4.40 initial motility in Hariana bulls. Kodagali (1967) studied on Gir breed and recorded 3.75 and 3.50 initial motility in 1st and 2nd year of study respectively.

#### Hydrogen-ion-concentration (pH):

Webster (1932) reported that pH of the bull semen varied from 7.0 to 7.5. Davis and Williams (1932) stated that pH of the bull semen was generally considered to be on the acid



side. Anderson (1938) reported the mean pH of the bull semen to be 6.73. Lambert and Mckenzi (1940) reported that pH of the bull semen varied from 6.5 to 7.5. Swanson and Herman (1944) on 205 samples of bull semen indicated a range in pH from 5.8 to 7.4 with an average of 6.3. Anderson (1945) noted considerable variation between bulls in pH of the semen. Mukherjee and Bhattacharya (1952) reported that the mean pH value of the Kumauni hill bulls was 6.28. Bhatia (1960) found that the initial pH varied in a very narrow range (5.96 - 6.50). According to him, Sahiwal, Gir and Sindhi breeds showed an average pH of 6.29 in each whereas, the Tharparkar and Hariana breeds were having an average of 6.31 and 6.32 respectively. Singh and Prabhu (1963) did not find significant difference in pH among the Hariana and among Kumauni hill bulls. Kodagali (1967) noted the average pH value in Gir bull as 6.25 and 6.40 in 1st year and 2nd year of the study respectively.

#### Percentage of dead spermatozoa:

Lasley (1944) recorded 16.9% dead sperm on examination of the semen immediately after collection, while Sexana (1965) reported 12.5 % dead sperm in Hariana bull and Tripathi, in the same year, noted 18.62% dead sperm in Hariana bull. Nikulenko (1965) noted that semen of Red Danish and Jersey bulls had lower percentage of dead spermatozoa than that of the Black-Pied bulls. Mukherjee and Singh (1966) stated that



percentage of dead spermatozoa did not vary significantly between the bulls. Kodagali (1967) studied on semen characteristics of Gir breed for two years and recorded 20 % dead sperm.

#### Sperm concentration:

According to Lagerlof (1934) there was a great individual variation in sperm production in the bull. Lagerlof (1934) and Herman and Regsdale (1939) listed a range of 300 to 1200 millions per ml. and 420 to 1950 millions per ml. respectively, while McKenzie (1939) and Anderson (1940) recorded a range of 300 to 2000 millions per ml. and 500 to 2000 millions per ml. respectively; but Green et al. (1941) and Lasley and Bogard (1943) noted a range of 300 to 3000 millions per ml. and 400 to 1160 millions per ml. respectively. Phillips et al. (1943) reported considerable variations in sperm concentration both within and between bulls and also observed significant differences between beef and dairy breeds. Kumaran (1944) recorded the sperm concentration within a range of 370 to 3160 millions per ml. Swanson and Herman (1944) noted considerable variation in sperm concentration both within and between the bulls. Anderson (1945) obtained individual differences in sperm concentration. Mercier et al. (1946) noted highly significant variation among Holstein-Friesian bulls and Guernsey bulls. Shukla and Bhattacharya (1949) reported a sperm concentration of 420 to 2020 millions per ml. in Sahiwal



breed, 575 to 1857 millions per ml. in Mariana breed and 250 to 1950 millions per ml. in Kumauni breed. Mukherjee and Bhattacharya (1952) noted the average sperm concentration as 634.54 millions per ml. in Kumauni hill bulls and observed highly significant variation among bulls. Johnston et al. (1953) reported highly significant difference in sperm concentration between bulls and also between Guernsey, Holsteins and Jersey breeds. Hafs et al. (1958) obtained a marked difference in sperm concentration between bulls but stated that breed did not provide a significant source of variation for sperm concentration of semen. Singh and Prabhu (1963) noted highly significant variation among Mariana and among Kumauni hill bulls. Sinha (1964) recorded the average sperm concentration of Tharparkar bulls as 1315 millions per ml. Sexana (1965) observed in Mariana bulls an average sperm concentration of  $1620 \pm 61.6$  millions per ml. and Tripathi, in the same year, recorded 1115.3 millions per ml. in Mariana bulls only. Paul et al. (1966) noted highly significant variation between five Indian breeds in sperm concentration. Kodagali (1967) recorded an average concentration, of 1310 millions per ml. and 1340 millions per ml. in 1st and 2nd year of study respectively in Gir breed. Nikulenko (1967) stated that semen of Red-Danish and Jersey bulls had higher sperm concentration than that of black-Pied bulls.

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## MATERIALS AND METHODS

In the present study, 8 Tharparkar, 4 Ayrshire and 4 Cross-bred (Sahiwal X Brown Swiss) bulls maintained at Government Cattle Farm, Patna were taken. These bulls were from two to five years of age. The bulls were maintained under existing conditions of management and feeding of the farm. The concentrates consisted of wheat bran, Ground nut cake, salt and mineral mixtures were supplied to bulls morning and evening as per feeding standards. They were given exercise regularly in early morning hours. Hay and greens were given ad libitum. The animals were allowed to drink water ad libitum.

The experiment was carried out for three months i.e. from May to July, 1967.

Tharparkar bulls which were under progeny testing scheme were already trained for semen collection in artificial vagina before the experiment was started, while Ayrshire and Cross-bred bulls were trained for the purpose of this experiment.

Always a cow in oestrus was used as a dummy for collection of semen. Semen collection was done at fortnightly intervals. Reaction time was noted at the time of collection. Every bull was allowed one false mount before taking the first ejaculate as suggested by Branton C. et al. (1952). For each bull a separate artificial vagina was used. The temperature of the water used was varying from 40°- 52°C. depending upon



the reaction of the individual bulls. Each artificial vagina was provided with an insulating bag covering the rubber cone and semen collection tube to protect the spermatozoa from heat shock caused by external atmospheric temperature during the process of collection. After collection the semen was brought immediately to the laboratory where it was kept at 37°C in a water bath and then examined for different characteristics.

The following seminal characteristics were studied:-

- I. Volume
- II. Initial motility
- III. Power of Hydrogen-ion-concentration (pH)
- IV. Percentage of dead sperm
- V. Sperm concentration

In all 32 semen samples from Tharparkar bulls, 16 from Ayrshire bulls and 16 from Cross-bred bulls were studied.

The animals were apparently healthy and free from diseases like T.B., Brucellosis and Johne's disease on the basis of their periodical tests conducted by Livestock Research Station, Patna.

The following characters along with reaction time were studied:-

Reaction time:- It was noted as the period between the approach of the bull near the cow in the crate and the ejaculation in the artificial vagina in seconds which was



recorded with the aid of a stop watch as studied by Prabhu (1956).

### Characters of Neat Semen:

I. Volume:- Volume of the ejaculate was recorded directly from the graduated collection tube up to one tenth of millilitre, Kushwaha et al. (1955).

II. Initial motility:- Immediately after collection the semen was examined under low power magnification of the microscope, putting one droplet of semen on the slide without coverslip at 30°C. The initial motility was graded in the following scale as adopted by Perry (1960).

- |   |                    |   |
|---|--------------------|---|
| 0 | ...                | No motile sperm.  |
| 1 | (Poor motility) .. | Less than 25% motile, but weak and oscillatory.                   |
| 2 | (Fair motility) .. | From 20 to 50 % progressively motile but lacking in waves.        |
| 3 | (Good motility) .. | From 50 to 70 % showing motion and waves.                         |
| 4 | (Very good " ) ..  | From 75 to 85% showing vigorous motion and recurring waves.       |
| 5 | (Excellent " ) ..  | More than 80% in vigorous motion, with waves of a billowy nature. |

III. Power of Hydrogen-ion-concentration(pH):- pH was measured immediately after the semen collection with the help of B.D.H. capillary set. Capillary tubes of similar bore



were used. They were marked with grease pencil at equal distance. A large drop of semen was taken in a watch glass. It was sucked in one of the marked capillary tubes upto the mark and was transferred to another clean watch glass. Bromothymol blue indicator, with pH range from 6.2 to 7.6 was sucked in other capillary tube upto the mark and was transferred to the watch glass containing semen. It was mixed well. The mixture was sucked in the same capillary tube and was compared with the capillary set of the same indicator and pH was noted.

IV. Percentage of dead spermatozoa:- For the estimation of percentage of dead spermatozoa eosin-nigrosin stain in the strength of 1 and 5% advocated by Swanson et al. (1951) was used. It was prepared as follows:-

|                          |     |         |
|--------------------------|-----|---------|
| Eosin (water soluble)    | ... | 1 g.    |
| Nigrosin                 | ... | 5 g.    |
| Sodium citrate dihydrate | ..  | 3 g.    |
| Distilled water to       | ... | 100 cc. |

This solution was kept on water bath for 30 minutes and filtered after cooling.

On grease free glass slides, a drop of neat semen was taken. To it two drops of the above stain was added so that the ratio between the semen and the stain was 1:2 (Swanson et al. 1951). The semen and the stain droplets were well mixed by gently blowing through a pipette and allowed to stand for one



to two minutes. Thin smears were drawn on clean slides and marked with grease pencil with bull no. and date of collection. The smears were dried in air and examined under oil immersion lense of microscope. Live spermatozoa did not take any stain and were colourless. The dead spermatozoa were seen red due to eosin stain. The back ground was violet due to the nigrosin stain. Those taking partial stain anteriorly or posteriorly were considered dead, as majority of the workers (Perry, 1960 and Maulae, 1962) were of the view that partially stained spermatozoa were on the way to death. Assessment of the dead spermatozoa were made by counting 300 spermatozoa (Rollinson, 1955) per slide on random basis.

V. Evaluation of sperm concentration:- The diluting fluid for evaluation of sperm concentration as advocated by George (1952) was used. It was prepared as follows:-

|                      |        |          |
|----------------------|--------|----------|
| Sodium citrate 3 %   | ...    | 100 c.c. |
| Commercial formaline | ...    | 1 c.c.   |
| 37 - 40 %            |        |          |
| Eosin B              | ... .. | 0.6 gr.  |

Dilution and counting of spermatozoa :- A portion of the well mixed semen sample was poured into a small vial and was drawn into a red blood cell diluting pipette upto 0.5 mark. The tip of the pipette was wiped off by hand. A small bubble of air was then sucked into the capillary tube and the tip of the pipette was inserted into a small vial containing



a portion of the diluting fluid. The fluid was then drawn up to 101 mark to give a dilution of 1:200. The dilution pipette was shaken for 3 minutes to insure thorough mixing of the semen with the diluting fluid. Three drops of the mixture were discarded to remove any high concentration of spermatozoa remaining in the capillary tube. After that, both the platforms of the haemocytometer were charged. The sperms were allowed to settle for three minutes before the counting was started. Then, the number of sperms were counted under high power objective in four corner groups and one central group-each of 16 small squares - of the improved Neubauer chamber. Both the platforms of the haemocytometer were counted for a single estimation and the result was incorporated, only when the difference of both sides, was within a range of ten percent. In case of abnormal values a fresh dilution was made and counting was repeated. The results were expressed as millions per cubic millimeter of semen.

Calculations were done on the same lines as mentioned in erythrocyte enumeration.

Statistical analysis was done according to Snedecor (1961).

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## R E S U L T S

A total number of 32 observations in Tharparkar breed, 16 in Ayrshire breed and 16 in Cross-bred were studied. The data were subjected for statistical analysis and the results on mean, standard deviation, standard error and coefficient of variation were tabulated in table No.I - 1, 2, 3 and 4. The mean values of the characters have been represented in graphs I-1 and I-2.

### Reaction time:

The mean reaction time was recorded as  $29.75 \pm 4.37$ ,  $37.81 \pm 6.2$  and  $44.68 \pm 7.27$  seconds in Tharparkar, Cross-bred and Ayrshire breeds respectively. The analysis of variance (Table No.I-5,6 & 7) showed significant difference among the bulls of all the three breeds. Significant difference was observed between the breeds also (Table No.I-8 & 9).

### Volume of the ejaculate:

The mean volume of the ejaculate was found  $5.17 \pm 0.37$ ,  $4.05 \pm 0.27$  and  $4.74 \pm 0.22$  milliliters in Tharparkar, Cross-bred and Ayrshire respectively. Analysis of variance (Table No.I-5,6 & 7) revealed that there was highly significant difference among the bulls of all the three breeds, and there was significant difference between the breeds also. But from



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TABLE I-1

Statistical analysis of different semen characteristics of Tharparkar, Cross-bred (Sahiwal X Brown Swiss) and Ayrshire bulls.

| Character                        | Tharparkar |              |          |       | Cross-bred |             |          |      | Ayrshire |              |          |       |
|----------------------------------|------------|--------------|----------|-------|------------|-------------|----------|------|----------|--------------|----------|-------|
|                                  | No.        | Mean         | Range    | S.D.  | No.        | Mean        | Range    | S.D. | No.      | Mean         | Range    | S.D.  |
| Reaction time in second.         | 32         | 29.75 ± 4.37 | 5 - 120  | 23.1  | 16         | 37.81 ± 6.2 | 10-75    | 24.9 | 16       | 44.68 ± 7.27 | 20-120   | 29.1  |
| Volume in ml.                    | 32         | 5.17 ± 0.37  | 3.0-12.5 | 2.12  | 16         | 4.05 ± 0.27 | 2.5-5.2  | 0.87 | 16       | 4.74 ± 0.22  | 2.5-6.5  | 0.91  |
| Initial motility                 | 32         | 4.26 ± 0.08  | 3.0-4.5  | 0.46  | 16         | 3.15 ± 0.07 | 3.0-4.0  | 0.3  | 16       | 3.71 ± 0.16  | 3.0-4.5  | 0.67  |
| pH                               | 32         | 6.20 ± 0.02  | 6.0-6.5  | 0.14  | 16         | 6.38 ± 0.41 | 6.1-6.5  | 1.65 | 16       | 6.26 ± 0.39  | 6.0-7.1  | 1.59  |
| % of dead sperm                  | 32         | 6.3 ± 0.18   | 1-12     | 1.0   | 16         | 22.8 ± 0.25 | 4-35     | 1.8  | 16       | 18.23 ± 1.27 | 3-25     | 5.11  |
| Sperm concentration millions/ml. | 32         | 1465 ± 125   | 310-3150 | 707.8 | 16         | 977 ± 107   | 380-1490 | 428  | 16       | 985 ± 103    | 450-1890 | 414.5 |
|                                  |            |              |          | 48.30 | 16         |             |          | 43.7 | 16       |              |          | 42.05 |



TABLE I-2

Table showing the mean, standard deviation, standard error and coefficient of variation for reaction time, volume, initial motility, pH, percentage of dead sperm and sperm concentration for Tharparkar bulls.

| Bull/No.                 | Reaction time(second) |       |       | Volume(ml.) |       |      | Initial motility |       |       |        |       |       |       |
|--------------------------|-----------------------|-------|-------|-------------|-------|------|------------------|-------|-------|--------|-------|-------|-------|
|                          | Mean                  | S.D.  | S.E.± | C.V.%       | Mean  | S.D. | S.E.±            | C.V.% | Mean  | S.D.   | S.E.± | C.V.% |       |
| 24                       | 4                     | 32.25 | 19.3  | 9.6         | 59.84 | 4.22 | 0.96             | 0.48  | 22.74 | 4.12   | 0.76  | 0.38  | 18.44 |
| 26                       | 4                     | 27.25 | 15.5  | 7.7         | 56.88 | 4.20 | 0.28             | 0.14  | 6.66  | 3.37   | 0.47  | 0.23  | 13.94 |
| 30                       | 4                     | 17.25 | 6.4   | 3.2         | 37.10 | 4.87 | 1.03             | 0.51  | 21.14 | 4.25   | 0.28  | 0.14  | 6.58  |
| 65                       | 4                     | 32.25 | 22.5  | 11.2        | 69.76 | 3.58 | 0.43             | 0.21  | 12.01 | 4.37   | 0.24  | 0.12  | 5.49  |
| 50                       | 4                     | -     | -     | -           | -     | 6.40 | 0.74             | 0.37  | 11.56 | 4.50   | 0     | 0     | 0     |
| 66                       | 4                     | 60.0  | 42.4  | 21.2        | 70.66 | 9.0  | 3.7              | 1.8   | 41.11 | 4.50   | 0     | 0     | 0     |
| 80                       | 4                     | 18.25 | 2.3   | 1.1         | 12.60 | 5.12 | 0.01             | 0.005 | 0.19  | 4.50   | 0     | 0     | 0     |
| 113                      | 4                     | 20.0  | 9.1   | 4.5         | 45.5  | 3.97 | 0.45             | 0.22  | 11.33 | 4.50   | 0     | 0     | 0     |
| pH                       |                       |       |       |             |       |      |                  |       |       |        |       |       |       |
| Percentage of dead sperm |                       |       |       |             |       |      |                  |       |       |        |       |       |       |
| 24                       | 4                     | 6.2   | 0.1   | 0.05        | 1.61  | 6.6  | 0.18             | 0.09  | 2.72  | 176.25 | 60.37 | 30.18 | 34.25 |
| 26                       | 4                     | 6.05  | 0.05  | 0.02        | 0.82  | 2.0  | 0.35             | 0.14  | 17.5  | 54.25  | 21.04 | 10.52 | 38.78 |
| 30                       | 4                     | 6.05  | 0.05  | 0.02        | 0.82  | 4.9  | 0.18             | 0.09  | 3.67  | 100.00 | 22.2  | 11.1  | 22.20 |
| 65                       | 4                     | 6.32  | 0.05  | 0.02        | 0.79  | 15.8 | 0.32             | 0.16  | 2.02  | 91.25  | 4.50  | 2.25  | 4.93  |
| 50                       | 4                     | 6.25  | 0.05  | 0.02        | 0.8   | 6.9  | 0.08             | 0.04  | 1.15  | 144.00 | 13.8  | 6.9   | 9.58  |
| 66                       | 4                     | 6.15  | 0.12  | 0.06        | 1.95  | 4.2  | 1.2              | 0.04  | 28.57 | 172.25 | 98.7  | 49.3  | 57.30 |
| 80                       | 4                     | 6.25  | 0.05  | 0.02        | 0.78  | 12.2 | 0.03             | 0.01  | 0.24  | 243.00 | 31.4  | 15.7  | 12.92 |
| 113                      | 4                     | 6.37  | 0.19  | 0.09        | 2.98  | 2.6  | 0.29             | 0.14  | 11.15 | 191.00 | 29.1  | 14.5  | 15.23 |
| Sperm concentration      |                       |       |       |             |       |      |                  |       |       |        |       |       |       |



TABLE I-3.

Table showing the mean, standard deviation, standard error and coefficient of variation for reaction time, volume, initial motility, pH, percentage of dead sperm and sperm concentration for cross-bred bulls.

| Bull<br>No. 1 | No. 1 | Reaction time (second) |       |       |        | Volume (ml.) |      |      |        | Initial motility |      |      |        |
|---------------|-------|------------------------|-------|-------|--------|--------------|------|------|--------|------------------|------|------|--------|
|               |       | Mean                   | S.D.  | S.E.  | C.V. % | Mean         | S.D. | S.E. | C.V. % | Mean             | S.D. | S.E. | C.V. % |
| 10            | 4     | 55                     | 5.7   | 2.8   | 10.3   | 5.75         | 0.1  | 0.05 | 1.77   | 3.0              | 0    | 0    | 0      |
| 17            | 4     | 67                     | 6.4   | 3.2   | 9.55   | 4.3          | 0.12 | 0.06 | 2.7    | 3.0              | 0    | 0    | 0      |
| 20            | 4     | 15                     | 0     | 0     | 0      | 2.75         | 0.28 | 0.14 | 10.5   | 3.12             | 0.25 | 0.12 | 8.0    |
| 29            | 4     | 13                     | 2.5   | 1.2   | 19.9   | 4.05         | 0.12 | 0.06 | 2.9    | 3.5              | 0.40 | 0.20 | 11.4   |
| <hr/>         |       |                        |       |       |        |              |      |      |        |                  |      |      |        |
| 10            | 4     | 6.5                    | 0.45  | 0.22  | 6.9    | 32.1         | 0.05 | 0.02 | 0.15   | 62.5             | 6.23 | 3.11 | 9.9    |
| 17            | 4     | 6.3                    | 0.42  | 0.21  | 6.6    | 8.2          | 0.43 | 0.21 | 5.23   | 132.75           | 5.5  | 2.7  | 4.1    |
| 20            | 4     | 6.3                    | 0.17  | 0.08  | 2.6    | 26.8         | 1.4  | 0.7  | 0.52   | 52.25            | 15.8 | 7.9  | 30.2   |
| 29            | 4     | 6.2                    | 0.057 | 0.028 | 0.91   | 27.7         | 0.14 | 0.07 | 0.50   | 143.5            | 4.4  | 2.2  | 2.0    |



TABLE I-4

Table showing the mean, standard deviation, standard error and coefficient of variation for reaction time, volume, initial motility, pH, percentage of dead sperm and sperm concentration for Ayrshire bulls.

| Bull No. | No. | Reaction time (second) |      |       |       | Volume (ml.)             |      |       |       | Initial motility    |      |       |       |
|----------|-----|------------------------|------|-------|-------|--------------------------|------|-------|-------|---------------------|------|-------|-------|
|          |     | Mean                   | S.D. | S.E.+ | C.V.% | Mean                     | S.D. | S.E.+ | C.V.% | Mean                | S.D. | S.E.+ | C.V.% |
| 1942     | 4   | 27.5                   | 6.4  | 3.2   | 23.27 | 4.32                     | 0.17 | 0.08  | 3.93  | 3.0                 | 0    | 0     | 0     |
| 2031     | 4   | 63.75                  | 43.0 | 21.5  | 67.45 | 4.62                     | 1.6  | 0.8   | 34.63 | 4.12                | 0.74 | 0.37  | 17.96 |
| 2281     | 4   | 60.0                   | 26.7 | 13.3  | 44.5  | 5.5                      | 0.4  | 0.2   | 7.27  | 4.12                | 0.74 | 0.37  | 17.96 |
| 2570     | 4   | 27.5                   | 6.4  | 3.2   | 23.27 | 4.52                     | 0.36 | 0.18  | 7.96  | 3.62                | 0.24 | 0.12  | 6.62  |
|          |     |                        |      |       |       |                          |      |       |       |                     |      |       |       |
|          |     | pH                     |      |       |       | Percentage of dead sperm |      |       |       | Sperm concentration |      |       |       |
| 1942     | 4   | 6.17                   | 0.1  | 0.05  | 1.62  | 14.3                     | 1.2  | 0.6   | -     | 50.25               | 4.2  | 2.1   | 8.35  |
| 2031     | 4   | 6.45                   | 0.44 | 0.22  | 6.82  | 10.5                     | 0.49 | 0.24  | -     | 135.25              | 36.9 | 18.4  | 27.28 |
| 2281     | 4   | 6.17                   | 0.14 | 0.07  | 2.26  | 7.6                      | 0.64 | 0.32  | -     | 129.25              | 20.1 | 10.06 | 15.55 |
| 2570     | 4   | 6.25                   | 0.12 | 0.06  | 1.92  | 7.4                      | 0.50 | 0.25  | -     | 79.5                | 12.5 | 6.2   | 15.72 |



the table of critical difference (Table No.I-9) it was found that the average volume of semen of Tharparkar breed was significantly superior to Cross-bred, while there was no significant difference between Tharparkar and Ayrshire, and between Ayrshire and Cross-bred.

#### Initial motility:

The average initial motility was calculated as  $4.26 \pm 0.08$  in Tharparkar breed,  $3.15 \pm 0.07$  in Cross-bred breed and  $3.71 \pm 0.16$  in Ayrshire breeds. Analysis of variance (Table No.I-5,6 & 7) showed a highly significant difference among the bulls of Tharparkar breed and among Cross-bred bulls also, while a significant difference was noted between the Ayrshire bulls. It was further noted that a highly significant difference existed between the breeds (Table No.I-8), but from the value of critical difference it was observed that the superiority of Tharparkar breed over the other two breeds in relation to average initial motility and that of Ayrshire breed over the Cross-bred breed was highly significant.

#### Hydrogen-ion-concentration (pH):

The average pH was observed as to be  $6.20 \pm 0.08$ ,  $6.38 \pm 0.41$  and  $6.26 \pm 0.39$  in Tharparkar, Cross-bred and Ayrshire breeds respectively. The data were analysed for 'F' test (Table No.I-5,6 & 7) and the result showed that there was no significant difference among the bulls of all the three



breeds. Analysis of variance (Table No.I-8) did not reveal any significant difference between the breeds.

Percentage of dead spermatozoa:

The mean percentage of dead spermatozoa recorded for Tharparkar, Cross-bred and Ayrshire breeds was  $6.3 \pm 0.18$ ,  $22.8 \pm 0.45$  and  $18.23 \pm 1.27$  respectively. From the table of analysis of variance (Table No.I-5,6 & 7) it appeared that a highly significant difference existed among the bulls of all the three breeds. The 'F' test (Table No.I-8) also showed that there was highly significant difference between the breeds but the value of critical difference (Table No.I-9) revealed that the superiority of Tharparkar and Ayrshire breed was highly significant so far this characteristic of semen was concerned.

Sperm concentration:

The mean sperm concentration was calculated as  $1465 \pm 125$  millions/ml. in Tharparkar breed,  $977 \pm 107$  millions/ml. in Cross-bred breed and  $985 \pm 103$  millions/ml. in Ayrshire breed. It was observed from the analysis of variance (Table No. I-5,6 & 7) that the difference between the bulls belonging to the Tharparkar, Ayrshire and Cross-bred breeds was highly significant. Such difference was also noted between Tharparkar, Ayrshire and Cross-bred breeds, but from the table of critical difference (Table No.I-9) it was noted that the average sperm



concentration of Tharparkar breed was significantly higher than those of Ayrshire and Cross-bred breeds, while there was no significant difference between Ayrshire and Cross-bred breeds.



TABLE 1-5

Analysis of variance on semen picture in Tharparkar bulls.

| Source of variation. | Reaction time |          |        |        | Volume |        |       |        | Initial motility |      |      |        |
|----------------------|---------------|----------|--------|--------|--------|--------|-------|--------|------------------|------|------|--------|
|                      | D.F.          | S.S.     | M.S.   | F.     | D.F.   | S.S.   | M.S.  | F.     | D.F.             | S.S. | M.S. | F.     |
| Between bull         | 6             | 5778.54  | 963.09 |        | 7      | 88.30  | 12.61 |        | 7                | 4.68 | 0.66 |        |
| Within bull          | 21            | 5302.92  | 252.52 | 4.21** | 24     | 51.91  | 2.16  | 5.83** | 24               | 2.32 | 0.09 | 7.33** |
| Total                | 27            | 11081.46 |        |        | 31     | 140.21 |       |        | 31               | 7.0  |      |        |

|              |    | pH   |       | Percentage of dead sperm |         | Sperm concentration. |           |
|--------------|----|------|-------|--------------------------|---------|----------------------|-----------|
| Between bull | 7  | 0.45 | 0.064 | 7                        | 791.97  | 113.13               | 106176.75 |
| Within bull  | 24 | 0.19 | 0.007 | 24                       | 254.04  | 10.58                | 49161.95  |
| Total        | 31 | 0.64 |       | 31                       | 1046.61 |                      | 155338    |

\*\* Significant at 1% level.



TABLE 1-6

Analysis of variance on semen picture in cross-bred bulls

| Source of variation | Reaction time |         |         |    | Volume |       |          |    | Initial motility |      |      |    |
|---------------------|---------------|---------|---------|----|--------|-------|----------|----|------------------|------|------|----|
|                     | D.F.          | S.S.    | M.S.    | F. | D.F.   | S.S.  | M.S.     | F. | D.F.             | S.S. | M.S. | F. |
| Between bull        | 3             | 9104.44 | 3034.81 |    | 3      | 11.32 | 3.77     |    | 3                | 0.67 | 0.22 |    |
|                     |               |         | 14.92** |    |        |       | 121.61** |    |                  |      | 4.4* |    |
| Within bull         | 12            | 244.0   | 20.33   |    | 12     | 0.38  | 0.031    |    | 12               | 0.69 | 0.05 |    |
| Total               | 15            | 9348.44 |         |    | 15     | 11.70 |          |    | 15               | 1.36 |      |    |

|              |    | pH    |      | Percentage of dead sperm |        | Sperm concentration |         |
|--------------|----|-------|------|--------------------------|--------|---------------------|---------|
| Between bull | 3  | 0.17  | 0.56 | 3                        | 779.67 | 259.89              | 26522.5 |
|              |    |       | 0.16 |                          |        | 28.49**             | 8841    |
| Within bull  | 12 | 40.87 | 3.4  | 12                       | 109.46 | 9.12                | 1021.5  |
|              |    |       |      |                          |        |                     | 85      |
| Total        | 15 | 41.04 |      | 15                       | 888.13 |                     | 27545   |

\* Significant at 5% level.

\*\* Significant at 1% level.



TABLE I-7

Analysis of variance on semen picture in Ayrshire bulls.

| Source of variation | Reaction time |         |         | Volume |       |      | Initial motility |      |      |
|---------------------|---------------|---------|---------|--------|-------|------|------------------|------|------|
|                     | D.F.          | S.S.    | M.S.    | D.F.   | S.S.  | M.S. | D.F.             | S.S. | M.S. |
| Between bull        | 3             | 4754.69 | 1584.89 | 3      | 9.23  | 3.00 | 3                | 3.42 | 1.14 |
| Within bull         | 12            | 4778.52 | 398.21  | 12     | 3.19  | 0.26 | 12               | 3.57 | 0.29 |
| Total               | 15            | 9533.21 |         | 15     | 12.42 |      | 15               | 6.99 |      |

|              | pH   |       |      | Percentage of dead sperm |        |       | Sperm concentration. |          |         |
|--------------|------|-------|------|--------------------------|--------|-------|----------------------|----------|---------|
|              | D.F. | S.S.  | M.S. | D.F.                     | S.S.   | M.S.  | D.F.                 | S.S.     | M.S.    |
| Between bull | 3    | 0.20  | 0.06 | 3                        | 190.98 | 63.66 | 3                    | 19940.69 | 6646.89 |
| Within bull  | 12   | 37.97 | 3.16 | 12                       | 144.96 | 12.08 | 12                   | 5839.25  | 486.60  |
| Total        | 15   | 38.17 |      | 15                       | 335.94 |       | 15                   | 25779.94 |         |

\* Significant at 5% level

\*\* Significant at 1% level.



TABLE I-8

Analysis of variance on semen characteristics between the breeds (Tharparker, Cross-bred and Ayreshire).

| Source of variation | Reaction time |          |         | Volume |         |         | Initial motility |       |      |         |
|---------------------|---------------|----------|---------|--------|---------|---------|------------------|-------|------|---------|
|                     | D.F.          | S.S.     | M.S.    | D.F.   | S.S.    | M.S.    | D.F.             | S.S.  | M.S. | P.      |
| Between Breed       | 2             | 5249.30  | 2624.65 | 2      | 20.98   | 10.49   | 2                | 13.49 | 6.74 | 26.96** |
| Within Breed        | 57            | 36489.13 | 640.16  | 61     | 164.33  | 2.69    | 61               | 15.35 | 0.25 |         |
| Total               | 59            | 41738.43 |         | 63     | 177.64  |         | 63               | 28.84 |      |         |
| pH                  |               |          |         |        |         |         |                  |       |      |         |
| Between Breed       | 2             | 0.32     | 0.16    | 2      | 2106.9  | 1053.43 | 2                | 0.44  | 0.22 | 5.5**   |
| Within Breed        | 61            | 78.85    | 1.29    | 61     | 2328.73 | 38.17   | 61               | 2.86  | 0.4  |         |
| Total               | 63            | 79.17    |         | 63     | 4435.72 |         | 63               | 3.30  |      |         |

\* Significant at 5% level

\*\* Significant at 1% level.



TABLE I-9

Table showing critical difference to test the significance of mean difference among different breeds.

| Breeds                   | Reaction time   |                     | Volume          |                     | Initial motility |                     |        |      |      |
|--------------------------|-----------------|---------------------|-----------------|---------------------|------------------|---------------------|--------|------|------|
|                          | Mean difference | Critical difference | Mean difference | Critical difference | Mean difference  | Critical difference |        |      |      |
| Tharparkar & Ayrshire    | 14.93**         | 14.23               | 20.24           | 0.43                | 1.0              | 1.33                | 0.58** | 0.30 | 0.39 |
| Tharparkar & Cross-bred  | 8.06            | 14.23               | 20.24           | 1.12*               | 1.0              | 1.33                | 1.11** | 0.30 | 0.39 |
| Ayrshire & Cross-bred    | 6.87            | 17.8                | 23.7            | 0.69                | 1.14             | 1.51                | 0.58** | 0.34 | 0.45 |
| pH                       |                 |                     |                 |                     |                  |                     |        |      |      |
| Percentage of dead sperm |                 |                     |                 |                     |                  |                     |        |      |      |
| Tharparkar & Ayrshire    | 0.06            | 0.68                | 0.90            | 3.5                 | 3.6              | 4.78                | 0.16** | 0.12 | 0.15 |
| Tharparkar & Cross-bred  | 0.18            | 0.68                | 0.90            | 16.5**              | 3.6              | 4.78                | 0.17** | 0.12 | 0.15 |
| Ayrshire & Cross bred    | 0.12            | 0.8                 | 1.06            | 13.0**              | 4.38             | 5.79                | 0.01   | 0.14 | 0.18 |

\* Significant at 5% level.

\*\* Significant at 1% level.



## DISCUSSION

### Reaction time:

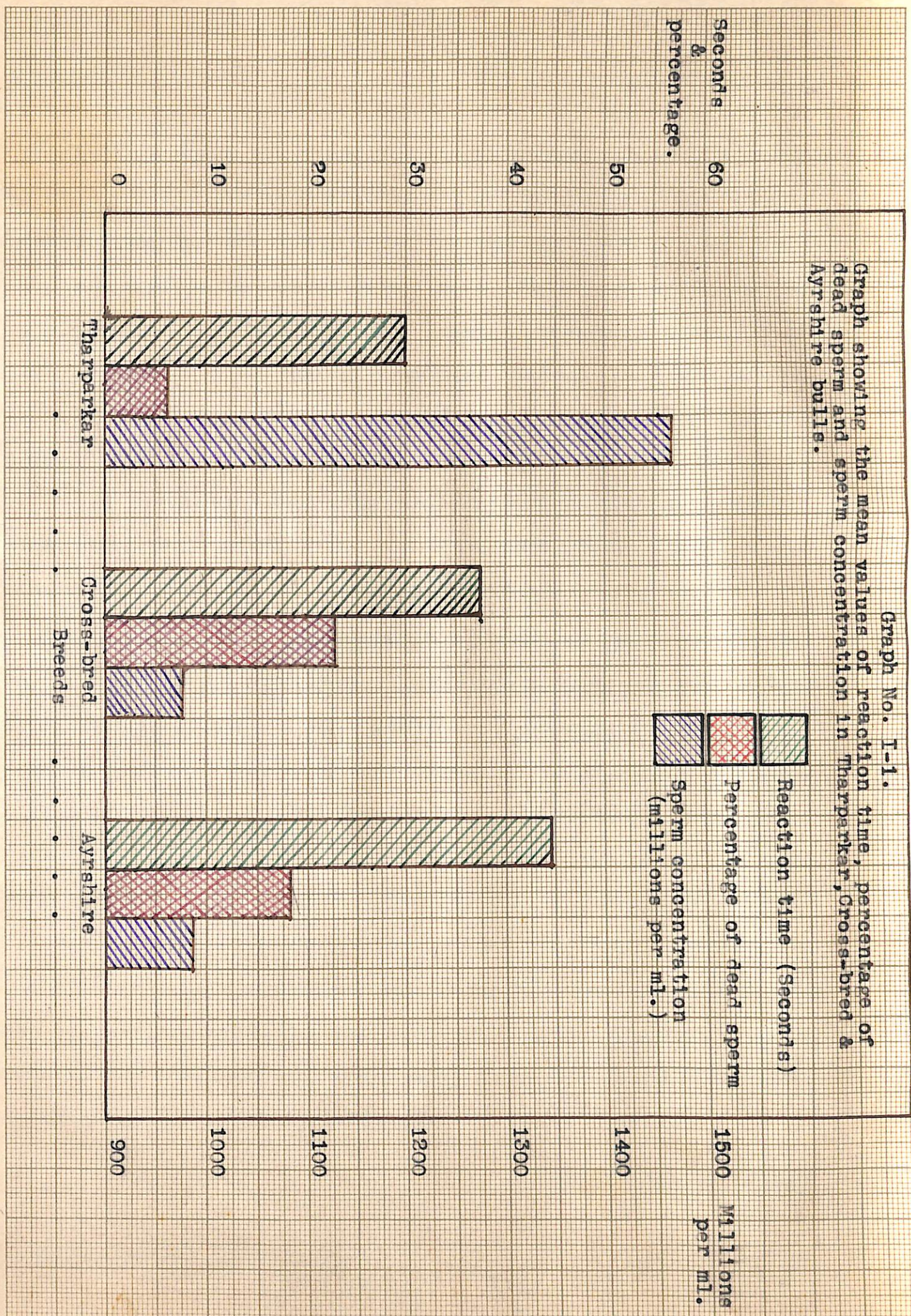
The average reaction time of Tharparkar, Cross-bred and Ayrshire bulls as recorded in the present study was  $29.75 \pm 4.37$ ,  $37.81 \pm 6.2$  and  $44.68 \pm 7.27$  seconds respectively, which was in agreement with the findings of Bhatia (1960) in Jersey and Gir bull. The observations of Mukherjee and Bhattacharya (1952) and Kodagali (1967) had shown slightly higher reaction time in Kumauni and Gir bulls but the figures were within the range of the present finding. Bhattacharya and Prabhu (1955) and Sinha (1964) recorded quite a higher reaction time in different breeds, which probably might be due to the fact that they used dummy for the collection of the semen, while in the present study the semen was collected always on cows in oestrus (Prabhu, 1956) and Russian workers as quoted by Krishnaya (1966).

A significant variation in average reaction time was observed between the bulls of all the three breeds which agrees with Mukherjee and Bhattacharya (1952), Bhattacharya and Prabhu (1955), Singh and Prabhu (1963) and Sinha (1964). Significant variation was noted between the breeds and the result was in concurrence with Smith (1951), Bhattacharya and Prabhu (1955) and Sinha (1964). Oloufa et al. (1958) suggested that this difference in reaction time may be due to difference in genetic make up of individual bull.



Graph No. I-1.

Graph showing the mean values of reaction time, percentage of dead sperm and sperm concentration in Tharparkar, Cross-bred & Ayrshire bulls.





### Volume of the ejaculate:

The mean volume of the ejaculate was  $5.17 \pm 0.37$  ml. in Tharparkar breed, which agrees with Bhattacharya and Prabhu (1955) and Sinha (1964); while in Cross-bred the average volume was  $4.05 \pm 0.27$  ml. agreeing with Kumaran (1949), Bhattacharya and Prabhu (1955) and Sinha (1964). In case of Ayrshire the average volume was  $4.74 \pm 0.22$  ml., which was in agreement with Kumaran (1939) finding. The results of Bhatia (1960), Tripathi (1965) and Kodagali (1967) were a bit higher than the present findings, while Lagerlof (1936), Bhatia (1960), Mukherjee and Bhattacharya (1952) and Bhattacharya and Prabhu (1952) recorded lesser values. Such variations in volume may be due to the breed and enviromental differences as suggested by Anderson (1941).

From the analysis of variance, it was found that there was a highly significant variation among bulls of all the three breeds. The present result agreed with those reported by Anderson (1941,45), Herman and Swanson (1941), Erb et al. (1942), Mercier et al. (1946), Mukherjee and Bhattacharya (1952), Bhattacharya and Prabhu (1952,55), Koriath et al. (1955), Bratton et al. (1956), VanDemark (1956), Haf et al. (1958), Zulliani and Tullio (1960) and Sinha (1964). Significant difference was also noted between the breeds; which finds support from Anderson (1941), Phillips et al. (1943), Bhattacharya and Prabhu (1952,55), Sinha (1964) and Barbulescu (1965). From the table of critical



difference it appeared that the average value of semen of Tharparkar breed was significantly superior to Cross-bred which elucidates that this Cross-bred although is a cross of Sahiwal and Brown-Swiss, yet so far this character is concerned Tharparkar supercedes it.

#### Initial motility:

The mean initial motility of spermatozoa in Tharparkar, Cross-bred and Ayrshire bulls were  $4.26 \pm 0.08$ ,  $3.15 \pm 0.07$  and  $3.71 \pm 0.16$  respectively and were in concurrence with the findings of Sexana (1965), Kodagali (1967) and Sinha (1964) who worked on Hariana, Gir and Taylor bulls respectively. The observations of Bhatia (1960), Mukherjee and Bhattacharya (1962) and Tripathi (1965) were more or less the same as in the present findings.

The result of the present study indicated that there was a significant variation in average initial motility between the bulls of all the three breeds. This variations agreed with those reported by Swanson and Herman (1941,44), Erb et al. (1942), Phillips et al. (1943), Anderson (1945), Mercier et al. (1946), Mukherjee and Bhattacharya (1952), Johnston et al. (1953) and Sinha (1964). A highly significant variation between the breeds concurred with Anderson (1945), Johnston et al. (1953) and Sinha (1964). Such differences thus explain that breed and the individuality have got the effect on the initial motility.



### Hydrogen-ion-concentration (pH):

The average pH recorded for Tharparkar, Cross-bred and Ayrshire bulls was  $6.20 \pm 0.08$ ,  $6.38 \pm 0.41$  and  $6.26 \pm 0.39$  respectively. Present result was quite in agreement with the findings of Mukherjee and Bhattacharya (1952), Bhatia (1960) and Kodagali (1967) who recorded in Kumauni, Sahiwal and Sindhi and Gir bull respectively.

No significant variation was noted between the bulls of all the three breeds and the result agreed with Singh and Prabhu (1963). There was no significant variation in average pH between the breeds also. Thus, it can be concluded that the average pH of the semen of the bulls is towards acidic.

### Percentage of dead spermatozoa:

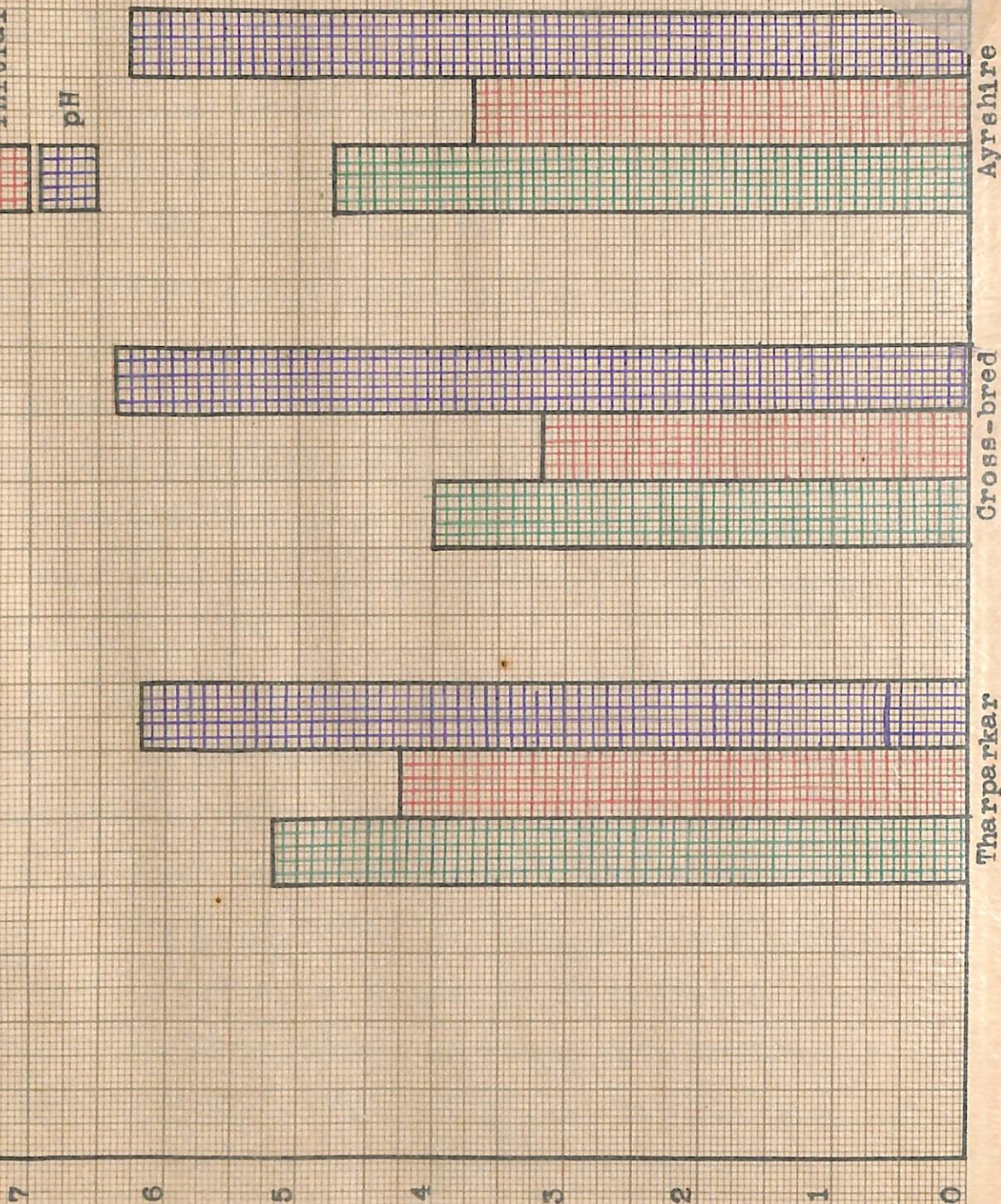
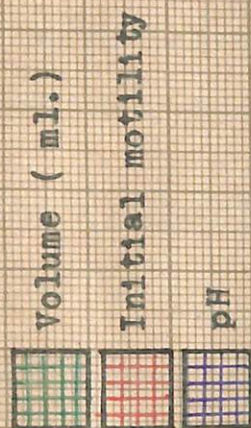
The percentage of dead sperm in fresh semen observed during the present study was  $6.3 \pm 0.18 \%$  in Tharparkar bulls,  $22.8 \pm 0.45 \%$  in Cross-bred bulls and  $18.23 \pm 1.27 \%$  in Ayrshire bulls. The findings of Lasley (1944), Tripathi (1965), Sexana (1965) and Kodagali (1967) fell in the range of present result. Highly significant variation in average percentage of dead spermatozoa was noted between the bulls of all the three breeds which agreed with the findings of Nikulenko (1965).

Highly significant variation in average percentage of dead sperm was noted between the breeds. From the table of critical difference it was observed that the Tharparkar and



Graph I-2.

Graph showing the mean values of semen volume, initial motility and pH in Tharparkar, Cross-bred & Ayreshire bulls.



.....

Breeds

.....



Ayrshire were superior to Cross-bred as they were having significantly less percentage of dead sperms. For this character also both the pure breeds viz. Tharparkar and Ayrshire supercede the Cross-bred.

#### Sperm concentration:

As recorded in the present study the average sperm concentration of Tharparkar, Cross-bred and Ayrshire bulls were  $1465.0 \pm 125.0$ ,  $977.0 \pm 107.0$  and  $985.0 \pm 103.0$  millions/ml. respectively, which was in agreement with the findings of Sinha (1964), Tripathi (1965) and Kodagali (1967) and fell in the range of other workers (Lagerlof, 1934; Anderson, 1940; Kumaran, 1944, and Shukla and Bhattacharya, 1943 etc.). But there was no agreement with the findings of Shukla and Bhattacharya (1949) and Sexana (1965), which might be due to breed or environmental differences.

Significant variation due to individuality in all the breeds under study agreed with those reported by Lagerlof (1934), Swanson and Herman (1941), Phillips *et al.* (1943), Anderson (1945), Mercier *et al.* (1946), Mukherjee and Bhattacharya (1952), Johnston *et al.* (1953), Hafs *et al.* (1958) and Singh and Prabhu (1962). A highly significant variation in average sperm concentration between the breeds also found an agreement with Phillips *et al.* (1943), Johnston *et al.* (1953), Paul *et al.* (1966) and Nikulenko (1966). Thus it proves that individuality and breed effect significantly on this character of semen.



P A R T -II.

PART -II.

BLOOD PICTURE OF

THARPARKAR, CROSS-BRED AND AYRSHIRE MALES



## REVIEW OF LITERATURE

Many workers as mentioned in the table given have worked on the blood picture of the cattle belonging to different breeds. Their results with the average value have been mentioned in the table no.II-1 for comparisons and in reference to the present study.

### Red blood corpuscles:

Storch (1901) worked on different age groups of cattle and recorded total number of Rbc. as 8.5 millions/ c.mm. in calves, 7 millions/ c.mm. in young cattle and 6.5 millions/c.mm. in bulls. According to him red blood cell count was highest in calves than the bulls.

Working on Australian cattle, Norris and Chamberlin (1929) found 9 to 17 millions/c.mm. of Rbc. in calves and heifers and 8 to 12 millions/ c.mm. in bullocks.

Fraser (1930) studied on four yearling bulls and bulls of two to five years age and observed a low red count of 5-6 millions/c.mm. in yearling bulls at grass which according to him probably due to an active and productive existence and rapid growth; and high count of 10.7 millions/c.mm. in bulls of 4 to 5 years age which he speculated due to stall feeding and production. He further attributed that this increase was greater as the age of the bulls advanced.



| Authors | Breed | R.B.C.<br>millions<br>per cmm. | W.B.C.<br>per<br>c.mm. | Lympho-<br>cyte % | Mono-<br>cyte % | Neutro-<br>phil % | Eosino-<br>phil % | Baso-<br>phil % |
|---------|-------|--------------------------------|------------------------|-------------------|-----------------|-------------------|-------------------|-----------------|
| 1       | 2     | 3                              | 4                      | 5                 | 6               | 7                 | 8                 | 9               |

|                                       |                      |       |       |      |     |      |     |   |
|---------------------------------------|----------------------|-------|-------|------|-----|------|-----|---|
| Shankar Narayanan<br>(1937)           | Sindhi bull          | 9.272 | -     | 53   | -   | 32   | 16  | - |
|                                       | Young Sindhi<br>bull | 9.476 | 12000 | 71   | -   | 20   | 8   | - |
|                                       | Gir bull             | 9.024 | 15600 | 71   | -   | 19   | 11  | - |
|                                       | Cross-bred           | 6.876 | 11200 | 61   | -   | 28   | 10  | - |
|                                       | Ayrshire bull        | 6.088 | 10200 | 56   | -   | 26   | 16  | - |
| Mullick & Pal<br>(1943)               | Bariana bullock      | 8.0   | 8400  | -    | -   | -    | -   | - |
|                                       | Dhanni "             | 7.9   | 8000  | -    | -   | -    | -   | - |
| Kehar & Murty<br>(1945)               | Bariana "            | 7.7   | 7700  | 63.1 | 6.2 | 27.4 | 3.0 | - |
| Govind Nayyar<br><u>et al.</u> (1949) | S. Indian cattle     | 6.53  | 8100  | 68   | 3   | 23   | 6   | - |
| Murty & Kehar<br>(1952)               | Kumanni bullock      | 6.5   | 8400  | -    | -   | -    | -   | - |
| Misra and Biswal<br>(1961)            | Orissa cattle        | 6.5   | 9440  | 66   | 4   | 25   | 5   | - |
| Patel <u>et al.</u> (1965)            | Gir bullock          | 6.3   | 12500 | 55   | 3   | 33   | 9   | - |
| Mithuji <u>et al.</u><br>(1966)       | Kankrej cattle       | 6.94  | 11850 | 63   | 4   | 24   | 9   | - |



TABLE II-1 (contd.)

Summary of some Haematological findings of Foreign cattle.

| 1                           | 2                        | 3     | 4     | 5    | 6    | 7    | 8    | 9   |
|-----------------------------|--------------------------|-------|-------|------|------|------|------|-----|
| Dimock & Thompson<br>(1906) | Holstein bull            | 6.670 | 4400  | -    | -    | -    | -    | -   |
|                             | Jersey bull              | 4.818 | 3555  | -    | --   | -    | -    | -   |
|                             | Guernsey bull            | 5.920 | 5350  | -    | ---  | -    | -    | -   |
| Canham<br>(1930)            | Bull calf                | 9.0   | 12000 | 68   | 4    | 30   | 2    | 1   |
|                             | Young bull               | 7.6   | 9900  | 59.8 | 4    | 29   | 7    | 1   |
| Fraser<br>(1930)            | Yearling bull            | 5.334 | 5200  | 61.2 | 5.6  | 30.3 | 0.45 | 0.7 |
|                             | Bulls                    | 8.246 | 6900  | 53.4 | 8.4  | 28.2 | 9.8  | 0.2 |
| Rusoff et al. (1954)        | Jersey bull              | 6.55  | 8580  | -    | -    | -    | -    | -   |
|                             | Guernsey bull            | 7.49  | 6444  | -    | -    | -    | -    | -   |
|                             | Holstein bull            | 7.84  | 7416  | -    | -    | -    | -    | -   |
| Holman<br>(1955)            | Ayrshire                 | 5.95  | 7030  | 51.4 | 8.32 | 29.1 | 9.87 | -   |
| Gourlay<br>(1959)           | Zebu cattle in<br>Uganda | 9.6   | 10100 | 48.5 | 12.3 | 28.1 | 9.7  | 1.4 |
| Penny et al. (1966)         | Ayrshire bull            | 6.92  | 6150  | 55.1 | 3.75 | 31.6 | 8.33 | -   |
|                             | Guernsey bull            | 6.62  | 5910  | 47.0 | 6.02 | 33.2 | 13.0 | -   |
|                             | Shorthorn bull           | 6.80  | 5890  | 50.8 | 3.13 | 32.9 | 9.78 | -   |
|                             | Hereford bull            | 7.45  | 9220  | -    | -    | -    | -    | -   |
|                             | Friesian bull            | 7.43  | 5850  | 39.9 | 4.83 | 43.2 | 10.4 | -   |



Canhom (1930) noted a decrease in red count with increasing age and he suggested that this fall was due to the fact that cattle take less and less exercise during the first two years of life; which should have shown that the bulk of the erythrocytes had not been increased. He recorded the mean number of erythrocytes as 9 millions/c.mm. from birth to 14 months; 7.6 millions/c.mm. from 14 months to 3 years and 6.66 millions/c.mm. in adult.

Shankaranarayanan (1937) reported a higher count in Indian cattle than in European cattle, but half bred cows had a value agreeing with European cattle.

Allocraft (1941) recorded a highly significant difference between breeds, which occurred mainly in Jersey cattle having low red count.

Mullick and Pal (1944) claimed that erythrocyte count of Indian Hissar cattle was highest in young animals and decreased with age.

Reid et al. (1948) worked on Holstein bulls and reported a decrease in erythrocyte count with advancing age.

Rusoff et al. (1954) studied on 5 Jersey, 5 Guernsey and 5 Holstein bulls. The Jersey bulls had the lowest mean erythrocyte values ( $6.55 \pm 0.35$ ) in millions/c.mm. as compared to other two breeds, but the difference was not found to be significant.

Greatorax (1957) studied on the Haematology of calves from birth to one year of age and recorded higher count in calves



upto 19 weeks old and then a decrease in number as the calves advanced in age. According to him the mean value of Rbc. was 8.1 millions/c.mm. in 8 to 12 weeks, 7.8 millions/c.mm. in 4 to 6 months and 7.5 millions/c.mm. in 12 months old calves.

Working on the changes associated with age in the blood picture of calves and heifers, Holman (1956) recorded high count in calves of four months age. There was a decrease in number with the age of the calves and heifers.

Again Greatedorex (1957) observed high count at birth and during the first four months after birth erythrocyte count was very irregular, but a steady decline occurred from about the fourth month to the end of the first year. This decrease continued as the age of the animal advanced. He noted a highly significant difference between the different age groups of the animals and between the breeds also.

Kupferschmied (1957) found high values at birth and observed a fall in the count till the animals reached two years. This value remained constant upto adult life.

Patel et al. (1960) worked on Kankrej calves and cows and recorded significantly higher values in calves than in cows.

Patel et al. (1965) worked on Gir cattle and observed a decrease in Rbc. count with increasing age. Significant differences were recorded between different age groups of animals.

Penny et al. (1966) studied the blood picture of 152 bulls at Artificial Insemination Centre and found a difference in the Rbc. count between the animals of different age groups



which was not statistically significant but recorded a highly significant difference between the Friesian and Guernsey breeds only.

Studying on Tharparkar and Sahiwal breeds, Khajuria and Razdan (1966) noted that erythrocyte count decreased with increasing age suggesting an increased haemopoietic activity. He did not find significant difference between the breeds.

Total leucocyte count (W.B.C.):

Schultz (1905) observed a decrease in the total leucocyte count with increasing age of animals.

Fraser (1930) also noted a decrease in the total leucocyte count with increasing age and obtained the following average values:-

| <u>Age</u>     | <u>Average</u><br>(per c.mm.) |
|----------------|-------------------------------|
| 2 to 4 months  | 8500                          |
| 4 to 6 months  | 9200                          |
| Yearling bulls | 5200                          |
| 2 to 5 years   | 6900                          |

Canham (1930) found that the young animals had a high white cell count which gradually decreased with advancing age and recorded an average value as 12000 per c.mm. in 1 day to 13 months group and 9900 per c.mm. in 14 months to young adult.

Zemljic (1936) reported a decrease in the total



leucocyte count with increasing age.

Shankaranarayanan (1937) reported higher counts in Indian cattle than European cattle, but half bred cows gave figures agreeing with the European cattle.

Braun Warner (1946) could not find significant difference in total leucocyte count in different age group of animals.

Rusoff et al. (1954) reported a considerable difference among the breeds. These difference were highly significant.

Moberg (1955) observed that total leucocyte count decreased with the age. The difference existing between the age groups was not statistically significant.

Holman (1956) recorded a total leucocyte count of 11500 per c.mm. in 4 month calves and after that there was a slight rise to 12500 per c.mm. by the 10th month followed by a gentle fall over the rest of the period. The line given by moving average was fairly definite for the leucocyte count.

Greatorrex (1957) noted slightly low values in calves than in 1 year and 2 year group followed by a gradual fall during adult life. Low values were particularly noticeable during 5th and 6th year. He recorded the following average values:-

| <u>Age</u>     | <u>Average</u><br>(per c.mm.) |
|----------------|-------------------------------|
| 2 to 4 months  | 9700                          |
| 4 to 6 months  | 9300                          |
| 1 year . . . . | 11900                         |



| <u>Age</u> | <u>Av.</u> |
|------------|------------|
| 2 year     | 11000      |
| 3 year     | 9400       |
| 4 year     | 9100       |
| 5 year     | 8600       |

He did not find any significant difference between age groups.

Patel et al. (1960) found significantly higher values in Kankrej calves than in cows.

Patel et al. (1965) recorded a decrease in total leucocyte count with increasing age in Gir cattle. Significant difference was recorded between different age groups, of animals.

Riegle et al. (1966) reported that the total leucocyte count decreased with the age in dairy bulls.

Penny et al. (1966) studied the blood picture of 152 bulls of different breeds and revealed a highly significant fall over the first year in Friesian bulls and subsequently no significant changes occurred. They further stated that Herefords had a significantly higher Wbc. than other breeds.

Khajuria and Razdan (1966) worked on 128 animals of Tharparkar and Sahiwal breeds and did not find any definite trend of Wbs. count with increasing age. Analysis of variance did not reveal significant difference between the age groups and breeds also.



Lymphocyte:

Fraser (1930) stated that the percentage and the actual number of lymphocytes rose considerably as age advanced in young stock and afterward there was a slow fall in lymphocyte value up to the adult life. He recorded average figures as follows:

| <u>Age</u>      | <u>Percentage</u> | <u>Absolute number</u><br>(per c. mm.) |
|-----------------|-------------------|--|
| 2 to 4 months.  | 61.2%             | 5084                                   |
| 6 months.       | 70.9%             | 6407                                   |
| Yearling bulls. | 61.2%             | 3178                                   |
| Bulls.          | 53.4%             | 3751                                   |

Canham (1930) reported that the average percentage of lymphocyte decreased with advancing age and observed that the percentage was 68% in 1 day to 13 month old calves, while 59.8% in 13 month to young adult bulls.

Obentraut (1934) and Storke-Baum (1951) noted that in calves during their first few days of life, there was a pronounced neutrophilia combined with eosino-lymphocyte and monocytopenia. According to them this was a rapidly passing phase and the blood picture was soon lymphocyte dominated, a condition which became less pronounced with increasing age.

Braun (1946) recorded a higher percentage of lymphocyte in young animals than in adult cattle and gave an average figure of 59.50% for young animal and 52.4% for older once ( over 30 months).



Moberg (1955) stated that the absolute number of lymphocyte decreased with age. Further he observed that the number of circulating lymphocytes was low in the new born which corresponded with the undeveloped state of lymphocytic organs. Later, as antibodies were needed, the lymphocytic tissue expanded and circulating lymphocytes increased. According to him with age, and perhaps due to atrophy of the thymus, the circulating lymphocytes decreased again.

Holman (1956) found that there was a definite fall in the absolute lymphocyte count with advancing age. He recorded 33% lymphocyte at birth which rose to 72% in four months after which there was a slow fall up to 50% to the adult level at about 2 years of life. He obtained statistically a highly significant difference between age groups.

Greatorrex (1957) observed an increase in the percentage of lymphocytes upto one year of life which, after decreasing slowly upto adult life, remained stable. He calculated an average value of 67% in young calves from first week to one year of age and 57% in adult cattle. There was a highly significant difference between age groups and breeds in his study.

Patel et al. (1960,65) found significantly higher values in Kankrej and Gir calves than cows.

Riegle et al. (1966) reported that the average percentage of lymphocyte decreased with advancing age in dairy and beef bulls.

Khajuri and Razdan (1966) worked on 128 animals of



Tharparkar and Sahiwal breeds and reported that the average percentage of lymphocyte increased with advancing age. They recorded statistically highly significant difference between age groups, but could not find significant difference between the breeds.

Penny et al. (1966) noted a decrease in the percentage of lymphocyte with advancing age (upto 7 years) after which the count remained relatively stable. Analysis of variance of absolute value failed to reveal any significant difference.

#### Neutrophil:

Fraser (1930) recorded a decrease in the percentage of neutrophil with advancing age which remained stable in adult life. The average values in his findings revealed 52.2% in calves, 27.9% in young stock and 28.4% in adult cattle.

Canham (1930) reported a decrease in the percentage of neutrophil with advancing age and observed the average values as 30% in 1 day to 13 month group and 29% in 13 months to young adult.

Moberg (1955) revealed that absolute number of neutrophil decreased with age which reflect a sluggish pituitary adrenal action in the bovine. According to him shift in neutrophil to 50 % or above with occurrence of few bands or younger forms was considered an indicative of stress reaction to bacterial infection with or without a significant increase in total count or a distinct left shift without significant neutrophilia or



leucocytosis.

Holman(1956) observed high percentage of neutrophil at birth and then a sudden fall as the lymphocyte percentage increased. Having fallen from 60% on the day of birth to 20% at 4 months, it showed some irregular increase in harmony with the proportional decrease in lymphocytes.

Greatorrex (1957) observed a high neutrophil percentage (45%) at birth with reducing values during the first week (32%). This, according to him, may be associated with stress at birth. Marked fluctuation occurred in his study throughout the remainder of the first year, with noticeable reduced values (21%) during the 5th and 8th month. By the end of the second year the neutrophil percentage had increased to 27% followed by a steady rise as age advanced. A value of 39% was recorded for animals in upper age group. He noted a significant difference between age groups and breeds also.

Patel et al. (1965) worked on Gir cattle and reported that the average percentage of neutrophils increased with advancing age and also recorded significant difference between the different age groups.

Riegle et al. (1966) found that the average percentage of neutrophil increased with the age of dairy and beef bulls.

Khajuria and Razdan (1966) observed low percentage of neutrophil in calves which increased with age. Statistically significant difference was noted by him between different age group but they could not reveal significant difference between



the breeds.

Penny et al. (1966) recorded an increase in the percentage of neutrophils with age up to seven years after which the count remained comparatively stable. Absolute count revealed very little change with age and did not show any significant breed difference also.

### Eosinophil:

Canham (1930) noted low values in calves than the young adult and recorded 2% in 1 day to 13 months old calves while 7% in 13 month to young adult.

Fraser (1930) reported that there was a gradual increase in the number of eosinophil with age of the animal and observed the average figures as follows:-

| <u>Age</u>     | <u>Percentage</u> | <u>Absolute values</u><br>(Per c.mm) |
|----------------|-------------------|--------------------------------------|
| 2 to 4 month   | 0.7 %             | 61                                   |
| 4 to 6 month   | 0.8 %             | 64                                   |
| Yearling bulls | 1.8 %             | 90                                   |
| Bulls          | 9.8 %             | 65.3                                 |

Moberg (1955) found an increase in the number of eosinophil with age in the cow and changes with the values were associated with reproductive phenomenon.

Helman (1956) reported an increase in eosinophil count which was associated with age and recorded 1.5% eosinophils in 6 month old calves and then there was a steady increase



to about 10 % by 22 months.

Greatorrex (1957) stated that during the first year of life the eosinophil count in calves varied according to the age, increased values being recorded as the age of the animal advanced. Over the whole period a range of 0 to 15 % was obtained with a marked increase in the number of eosinophils occurring during the age of 4 to 8 months. Eosinophils were more numerous in adult cattle, having an average of 11 % with a range from 2 to 30%. Significant difference was found in eosinophil percentage due to age and breeds also.

Patel et al. (1960, 1965) recorded significantly lower values in Kankrej and Gir calves than in cows.

The work of Khajuria and Razdan (1966) on Tharparkar and Sahiwal breeds could not reveal significant difference between age groups and breeds also.

Penny et al. (1966) indicated that the percentage of eosinophil increased up to 7 years of age after which no regular increase could be observed. The absolute values did not show a trend with age. No significant difference was noted between the age groups and breeds also.

#### Monocyte:

Fraser (1930) revealed that monocyte increase was transient with age, as the count in cattle from 2 months to adult stage varied only from 2 to 13% and 200 to 1500 per c.mm. with average values as follows.



| <u>Age</u>     | <u>Percentage</u> | <u>Absolute values</u><br>Per c.mm. |
|----------------|-------------------|-------------------------------------|
| 2 to 4 month   | 6.2 %             | 437                                 |
| 4 to 6 month   | 6.7 %             | 631                                 |
| Yearling bulls | 5.6 %             | 277                                 |
| Bulls          | 8.4 %             | 597                                 |

Canhom (1930) reported that the monocyte percentage had no effect on the age. The average values in his study was 4 % in 1 day to 13 month old calves as well as in 13 month to young adult. But the study of Moberg (1955) showed that the number of monocytes decreased with age.

Holman (1956) stated that the number of monocytes decreased with age and recorded 8.5 % and 4.5 % in 4 month and in 20 month calves respectively.

Greatorrex (1957) reported that the percentage of monocyte cells fluctuated considerably in both calves and adults and recorded the following findings:-

| <u>Age</u>         | <u>Av. percentage</u> | <u>Range</u> |
|--------------------|-----------------------|--------------|
| Birth to 4 month   | 2.8 %                 | 0 - 9 %      |
| 4 month to 7 month | 5.4 %                 | 0 - 12%      |
| 7 month to 13 "    | 3.0 %                 | 0 - 5 %      |
| Adult              | 2.0 %                 | 0 - 8 %      |

Analysis of variance revealed a highly significant difference between age groups but failed to show significant difference between the breeds.



Patel et al. (1965) observed that monocyte percentage was not affected with age in Gir cattle.

Khajuria and Razdan (1966) noted low count in calves which increased with advancing age. Statistical calculation revealed significant difference between age groups but failed to find significant difference between the breeds.

Penny et al. (1966) did not get any significant difference between the age group and breeds in relation to percentage as well as absolute value.

#### Basophil:

Fraser (1930) reported that basophils were very scanty in all the groups studied having an average of 0.2 % and 15 per c.mm.

Canham (1930) also recorded very scanty basophils in all age groups of animals averaging 1 %.

Holman (1956) observed an increase in the number of basophils at about 6 month after which it remained constant at the level of about 0.7 %.

Penny et al. (1966) did not find any difference in the percentage of basophils due to age or breeds.

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## MATERIALS AND METHODS

In the present study, 24 Tharparkar males of different age group, 4 Ayrshire bulls and 4 Cross-bred bulls were taken. The blood samples were collected from jugular vein at every fortnight intervals in the early hours of the morning before giving water, food and exercise. They were maintained under the existing condition of the management and feeding as mentioned earlier.

For the study of blood picture in different age groups of Tharparkar bulls, the animals were grouped as follows on the basis of their ages.

| <u>Group No.</u> | <u>Age</u>    |             | <u>No. of animal</u> |
|------------------|---------------|-------------|----------------------|
| I                | 0 to 6 month  | Bull calves | 4                    |
| II               | 6 to 12 month | Bull calves | 4                    |
| III              | 1 to 2 year   | Bulls       | 4                    |
| IV               | 2 to 3 year   | Bulls       | 4                    |
| V                | 3 to 4 year   | Bulls       | 4                    |
| VI               | 4 to 5 year   | Bulls       | 4                    |

In all, 96 samples from Tharparkar males, 16 samples from Ayrshire bulls and 16 samples from Cross-bred bulls were studied.



### Collection of blood:

For collection of blood the animal was controlled calmly and quietly without any excitement, in standing position in a service crate. The area over the jugular vein was sterilised with rectified spirit, after closely clipping the hair. The vein was made prominent with the pressure of the thumb and a sterilised stainless-steel hypodermic needle (No. 18) having uniform bore was inserted into the vein. In almost all cases the blood was drawn by the first thrust only. First few drops of blood were discarded. Blood smears were drawn on clean, dry, nongreasy slides and dried rapidly in air. About 5 ml. of blood were drawn directly through the needle into a small glass vial containing anticoagulant crystals of double oxalates. These crystals were prepared as follows, according to the recommendation of Heller and Paul, (1934).

|                   |         |
|-------------------|---------|
| Ammonium Oxalate  | 1.2 g.  |
| Potassium Oxalate | 0.8 g.  |
| Distilled water   | 100 ml. |

Of this solution, 0.5 ml. was poured into a clean glass vial and dried in hot air oven at 60°C. During and after collection the vial was gently rotated between fingers and the thumb so as to mix the anticoagulant with the blood thoroughly.

The following estimations were made and recorded.



(A) Erythrocyte enumeration:

The technique for the estimation of red blood corpuscles as recommended by Napier and Das-Gupta (1946) was adopted. The blood was shaken well to bring about a homogenous distribution of cells and was drawn in a clean, dry, red blood cell diluting pipette (Thoma type) up to the mark of 0.5. The excess blood adhering to the outside of the pipette was wiped off by hand. The Rbc. diluting fluid (Hayem's solution) was sucked in upto the mark of 101. While sucking the diluent, the pipette was rotated between the fingers and the thumb in order to mix the contents well. The composition of the diluting fluid used was as follows:-

|                             |         |
|-----------------------------|---------|
| Mercuric chloride           | 0.5 g.  |
| Sodium sulphate (Anhydrous) | 2.2 g.  |
| Sodium chloride             | 1.0 g.  |
| Distilled water             | 200 ml. |

After filling, the pipette was held in a horizontal position and shaken for about two minutes in order to ensure thorough mixing. The first few drops were discarded and the improved Neubauer haemocytometer was charged. The blood cells were allowed to settle for about three minutes before the counting was started. Then the number of red blood cells were counted under high power objective in four corner groups and one central group- each of 16 small squares - of the improved Neubauer chamber (Wintrobe - 1951). Both the platforms of the



haemocytometer were charged and counted for a single estimation and the result was incorporated, if the difference of both sides was within a range of ten percent. The results were expressed in millions per cubic millimeter of blood. In case the difference was above ten percent, a fresh dilution was made and counting was repeated. The following calculation was done to find out the total number of cells per c.mm. of blood:-

$$\text{Area of each small square} = \frac{1}{400} \text{ Sq.mm.}$$

$$\text{Depth of the chamber} = 0.1 \text{ mm.}$$

$$\text{Volume of each small square} = \frac{1}{400} \times 0.1 = \frac{1}{4000} \text{ c.mm.}$$

$$\begin{array}{l} \text{Volume of each group of} \\ 16 \text{ squares.} \end{array} = 16 \times \frac{1}{4000} = \frac{1}{250} \text{ c.mm.}$$

$$\begin{array}{l} \text{Volume of five groups of} \\ 16 \text{ small squares each} \end{array} = \frac{5}{250} = \frac{1}{50} \text{ c.mm.}$$

Suppose, N was the number of cells counted in five groups each of the 16 small squares, i.e. in  $\frac{1}{50}$  c.mm. of diluted blood, then the total number of cells in one c.mm. of diluted blood was  $N \times 50$ . Now, since 0.5 ml. of blood was diluted to 101, hence the dilution of blood was 200 times. Therefore, the number of cells present in one c.mm. of undiluted blood was  $N \times 50 \times 200$ , i.e.  $N \times 10000$ .

Thus, the number of cells counted in 80 small squares was multiplied by 10000 to arrive at the total number of cells per c.mm. of blood.



(B) Leucocyte enumeration:

The enumeration of the leucocytes was carried out in the same way as employed for the erythrocytes, with the following main differences in the method:-

(i) Instead of R.B.C. pipette, a white blood cell (W.B.C.) pipette was used.

(ii) The blood was diluted upto the mark of 11, thereby giving only 20 times dilution.

(iii) The diluting fluid (Turk's fluid) had the following composition:

Glacial acetic acid ----- 1 ml.

1% Aqueous gentian violet -- 1 ml.

Distilled water ----- 100 ml.

(iv) The count was made under low power objective.

(v) The cells were counted in four primary corner squares of the haemocytometer.

Total number of cells per c.mm. of blood was found out on the following calculations:-

Volume of fluid in each primary square of the chamber =  $1 \times 1 \times 0.1 = \frac{1}{10}$  c.mm.

Fluid in four primary squares =  $\frac{4}{10}$  c.mm.

If supposed that N was the total number of cells present in four primary squares, then  $\frac{4}{10}$  c.mm. of the diluted blood contained N number of cells. Hence, 1 c.mm. could contain  $\frac{10}{4} \times N$  number of cells. Now, since the dilution is 20 times,



therefore, number of cells present in 1 c.mm. of undiluted blood =  $10/4 \times N \times 20 = 50 \times N$ . Thus the number of cells counted in four primary corner squares was multiplied by fifty to arrive at the final number of cells per c.mm. of blood.

(C). Differential leucocyte count:

For the differential count of the white blood cells, the blood smear was properly drawn and stained well. Having drawn good smears the slides were put on the staining rack taking care that the two ends of the slides were in the same plane. From a dropper sufficient Wright's stain (B.D.H.) was poured on the slide to cover the whole film. It was allowed to stand for 1 to 2 minutes for proper fixing of the film. Without pouring off the stain, double the volume of distilled water (pH 6.9) was poured over the slide and mixed well by gently blowing an eddy in the fluid.

The mixture was allowed to settle. A bronze like scum appeared on the surface of the fluid, if the proportion of the stain and diluent was correct. The diluted stain was allowed to act for 5 to 10 minutes. The slide was then washed alternately in tap and distilled water till the film turned to pink colour and was allowed to dry in the air.

Battlement method was adopted for counting the cells as recommended by MacGregor et al. (1940). This method implies "a count of three horizontal edge fields followed by two fields towards the centre (so as to give three vertical fields),



Followed by two fields in a horizontal and then by two in a vertical direction to reach the edge again." The counting of cells was confined to an area of one mm. from the edge of the blood film. In all, 200 cells were differentiated and the percentage of lymphocytes, monocytes, neutrophils, eosinophils and basophils was calculated.

Statistical analysis was done according to

Snedecor (1961).



## RESULTS

A total number of 96 observations in Tharparkar males of different age group, 16 in Cross-bred bulls and 16 in Ayrshire bulls were studied. The data were subjected for statistical treatment and the results have been mentioned in table no. II-2 & 3. The mean values of the characters have been represented in graphs II-1, II-2 & II-3.

### Red blood corpuscles:

The mean Rbc. count in the I group i.e. from 0 to 6 month was found to be  $10.97 \pm 0.53$  millions per c.mm. which fell down after six month of life to one year coming to  $10.19 \pm 0.30$  millions per c. mm. In 1 to 2 year group and 2 to 3 year group the average values were  $7.98 \pm 0.30$  and  $6.78 \pm 0.31$  millions per c. mm. respectively. The concentration slightly rose to  $7.40 \pm 0.25$  and  $7.43 \pm 0.26$  millions per c. mm. in 3 to 4 year and 4 to 5 year group respectively.

On subjecting the whole data to treatment for analysis of variance ( Table No. II-4) it was observed that a highly significant difference was noted between the age groups. But the Bar-diagram (Table No. II-6) showed that I and II group did not differ significantly while both the groups differed significantly from all other groups. III group differed signi-



ficantly from IV group, but no significant difference existed between any other two groups.

In Tharparkar breed the over all average value for erythrocyte count was observed as  $8.46 \pm 0.25$  millions per c. mm. whereas in Cross-bred and Ayrshire breed the values were  $6.66 \pm 0.88$  and  $6.29 \pm 0.21$  millions per c. mm. respectively. Analysis of variance (Table No. II-7) of the data revealed a highly significant difference between the breeds. From the critical difference (Table No. II-8) it was found that the average erythrocyte count in Tharparkar breed was significantly higher than Ayrshire and Cross-bred breeds. There was no significant difference in average erythrocyte count between Ayrshire and Cross-bred breeds.

#### White blood corpuscles:

The overall average value of total leucocyte count in Tharparkar breed was  $12867 \pm 420$  per c. mm. The average value was highest in 0 to 6 month and 6 month to 1 year age groups being  $16034 \pm 1619$  and  $15606 \pm 719$  per c. mm. respectively. In 1 to 2 year and 2 to 3 year group, the average count was  $10709 \pm 659$  and  $12634 \pm 650$  per c. mm. respectively. While the average values were recorded as  $10419 \pm 685$  and  $11799 \pm 643$  per c. mm. in 3 to 4 year and 4 to 5 year group respectively. Analysis of variance (Table No. II-4) did not reveal any significant difference between the age groups.

The average values for Tharparkar, Cross-bred and



Ayrshire breed were  $12867 \pm 420$ ,  $10759 \pm 665$  and  $11284 \pm 750$  per c. mm. respectively. Analysis of variance ( table II-7) did not reveal any significant difference between the breeds. But from the value of critical difference ( table II-8) it was noted that Tharparkar breed had significantly higher value than Cross-bred and Ayrshire breeds.

#### Lymphocytes:

The average lymphocytes of all the groups in Tharparkar breed was  $64.77 \pm 0.16 \%$  and  $8441 \pm 357$  per c. mm. The percentage and absolute value of lymphocyte was highest in 0 to 6 month group averaging  $77.69 \pm 0.14 \%$  and  $12363 \pm 1184$  per c. mm. Then these values dropped slightly in 6 to 12 months and 1 to 2 year group having an average figures as  $67.31 \pm 0.21$  percent;  $10614 \pm 624$  per c. mm. and  $70.13 \pm 0.11$  percent;  $7478 \pm 451$  per c. mm. respectively. From 2 years of age the percentage and absolute values went on decreasing up to 5 years of age, having the average figures as  $62.5 \pm 0.3\%$  and  $7961 \pm 532$  per c. mm. for 2 to 3 year group,  $58.38 \pm 0.6\%$  and  $6146 \pm 582$  per c. mm. for 3 to 4 year groups and  $52.63 \pm 0.23\%$  and  $6084 \pm 290$  per c. mm. in 4 to 5 year group respectively. Statistically the lymphocyte percentage and absolute values were highly significant between the age groups ( table II-4 & 5). But from the Bar diagram ( table II-6) it was noted that all groups differed significantly from each other except II and III, so far values in % were concerned. In the case of absolute



values, the I and the II group did not differ significantly but both the groups differed significantly from all other groups. No significant difference existed between any other two groups.

The average value of  $64.77 \pm 0.16\%$  and  $8441 \pm 357$  per c. mm. in Tharparkar breed,  $57.4 \pm 0.21\%$  and  $6024 \pm 312$  per c. mm. in Cross-bred and  $66.4 \pm 0.20\%$  and  $7537 \pm 629$  per c. mm. in Ayrshire breed were recorded. Analysis of variance (table II- 7 & 9) of lymphocyte percentage and absolute values revealed a significant difference between the breeds, but from the value of critical difference (table II- 8 & 10) it was observed that the superiority of Tharparkar and Ayrshire breed over the Cross-bred in relation to lymphocyte % and absolute values was highly significant.

#### Neutrophil:

The overall average value in Tharparkar breed was  $24.50 \pm 0.13$  percent and  $3124 \pm 156$  per c. mm. The average value in 0 to 6 month and 6 to 12 month group was  $15.63 \pm 0.20\%$ ,  $2618 \pm 425$  per c. mm. and  $26.13 \pm 0.23\%$ ,  $3944 \pm 350$  per c. mm. respectively. After that the percentage and absolute values dropped slightly in 1 to 2 year and 2 to 3 year groups having the average values as  $22.25 \pm 0.19\%$ ,  $2413 \pm 262$  per c. mm. respectively. In 3 to 4 year and 4 to 5 year group the average value was  $23.50 \pm 0.25\%$ ,  $2477 \pm 300$  per c. mm. and  $32.44 \pm 0.23\%$ ,  $3933 \pm 437$  per c. mm. respectively. From the table of analysis of variance ( table II- 4 & 5) it appeared that a highly



TABLE II-4

Analysis of variance of blood picture in Tharparkar breed.

| Source of variation.      | R.B.C. |            | W.B.C. |              | Lymphocyte |              | absolute |       |
|---------------------------|--------|------------|--------|--------------|------------|--------------|----------|-------|
|                           | D.F.   | M.S.       | D.F.   | M.S.         | D.F.       | M.S.         | absolute | value |
| Between group             | 5      | 46.47      | 5      | 94084609.39  | 5          | 102662649.68 |          |       |
| Within group              | 90     | 1.90       | 90     | 126787806.93 | 90         | 7203389.4    | 14.25**  |       |
| Total                     | 95     |            | 95     |              | 95         |              |          |       |
| Absolute neutrophil value |        |            |        |              |            |              |          |       |
| Between group             | 5      | 8199545.2  | 5      | 2452293.4    | 5          | 249895.0     |          |       |
| Within group              | 90     | 1992901.48 | 90     | 302923.9     | 90         | 140429.8     | 1.77     |       |
| Total                     | 95     |            | 95     |              | 95         |              |          |       |

\*\* Significant at 1% level.



TABLE II-5

Analysis of variance of blood picture in Tharparkar breed.

| Source of variation | Lymphocyte % |        |        | Neutrophil % |        |        | Eosinophil % |        |        |
|---------------------|--------------|--------|--------|--------------|--------|--------|--------------|--------|--------|
|                     | D.F.         | M.S.   | F.     | D.F.         | M.S.   | F.     | D.F.         | M.S.   | F.     |
| Between group       | 5            | 489.86 |        | 5            | 239.98 |        | 5            | 335.12 |        |
| Within group        | 90           | 30.07  | 16.2** | 90           | 33.01  | 7.22** | 90           | 15.12  | 22.1** |
| Total               | 95           |        |        | 95           |        |        | 95           |        |        |

| Monocyte %    |    |      | Basophil % |      |     | Absolute basophil value |          |       |
|---------------|----|------|------------|------|-----|-------------------------|----------|-------|
|               |    |      |            |      |     |                         |          |       |
| Between group | 5  | 7.75 | 5          | 1.79 |     | 5                       | 1807.70  |       |
| Within group  | 90 | 20.5 | 90         | 2.74 | 0.6 | 90                      | 41471.22 | 0.043 |
| Total         | 95 |      | 95         |      |     | 95                      |          |       |

\*\* Significant at 1 % level.

Note- For percentage, the data were Arcsin transformed.



TABLE II-6.

Bar - Diagram

| Character                            | C.D.    | Averages of bull/arranged in Ascending order |          |          |          |          |          |    |   |    |     |    |   |
|--------------------------------------|---------|--|----------|----------|----------|----------|----------|----|---|----|-----|----|---|
|                                      |         | Group  |          |          |          |          |          |    |   |    |     |    |   |
| R.B.C. $10^6$ /c.mm.                 | 0.95    | IV   | V        | VI       | III      | II       | I        | IV | V | VI | III | II | I |
|                                      |         | 6.78   | 7.40     | 7.43     | 7.98     | 10.19    | 10.97    |    |   |    |     |    |   |
| W.B.C. per c.mm.                     | 7906.46 | - V  | III      | VI       | IV       | II       | I        |    |   |    |     |    |   |
|                                      |         | 10418 .79                                    | 10709.31 | 11798.61 | 12634.43 | 15605.86 | 16034.30 |    |   |    |     |    |   |
| Absolute lympho-<br>cyte value c.mm. | 1884.52 | VI   | V        | III      | IV       | II       | I        |    |   |    |     |    |   |
|                                      |         | 6084 .31                                     | 6146.06  | 7477.81  | 7960.62  | 10613.62 | 12363.06 |    |   |    |     |    |   |
| Lymphocyte %                         | 3.83    | VI   | V        | IV       | II       | III      | I        |    |   |    |     |    |   |
|                                      |         | 52 .63                                       | 58.38    | 62.56    | 67.31    | 70.13    | 77.69    |    |   |    |     |    |   |
| Absolute neutro-<br>phil value c.mm. | 991.23  | III  | V        | I        | IV       | VI       | II       |    |   |    |     |    |   |
|                                      |         | 2413.50                                      | 2477.37  | 2618.06  | 3360.56  | 3933.25  | 3944.12  |    |   |    |     |    |   |
| Neutrophil %                         | 4.01    | I  | III      | V        | II       | IV       | VI       |    |   |    |     |    |   |
|                                      |         | 15.63  | 22.25    | 23.50    | 26.13    | 27.06    | 32.44    |    |   |    |     |    |   |
| Absolute eosino-<br>phil value c.mm. | 386.28  | II   | I        | III      | IV       | V        | VI       |    |   |    |     |    |   |
|                                      |         | 326.25                                       | 383.81   | 438.50   | 832.75   | 1154.93  | 1186.0   |    |   |    |     |    |   |
| Eosinophil %                         | 2.72    | II   | I        | III      | IV       | VI       | V        |    |   |    |     |    |   |
|                                      |         | 2.07   | 2.62     | 3.94     | 6.44     | 9.87     | 11.87    |    |   |    |     |    |   |



TABLE 11-6 (contd.).

| Character                           | C.D.   | Averages of bull group arranged in ascending order |             |              |             |             |              |  |  |
|-------------------------------------|--------|--|-------------|--------------|-------------|-------------|--------------|--|--|
| Absolute mono-<br>cyte value c. mm. | 247.66 | III<br>372.00                                      | IV<br>452.0 | VI<br>571.18 | V<br>608.0  | I<br>637.81 | II<br>710.37 |  |  |
|                                     |        | <hr/>  |             |              |             |             |              |  |  |
|                                     |        | 6  |             |              |             |             |              |  |  |
|                                     |        | <hr/>  |             |              |             |             |              |  |  |
| Monocyte %                          | 3.17   | III<br>3.62  | IV<br>3.75  | I<br>3.88    | II<br>4.43  | VI<br>4.87  | V<br>6.0     |  |  |
|                                     |        | <hr/>  |             |              |             |             |              |  |  |
| Absolute baso-<br>phil value c.mm.  | 142.79 | III<br>7.50  | II<br>11.50 | VI<br>23.93  | IV<br>28.50 | I<br>31.56  | V<br>32.43   |  |  |
|                                     |        | <hr/>  |             |              |             |             |              |  |  |
| Basophil %                          | 0.25   | II<br>0.06   | III<br>0.06 | I<br>0.18    | VI<br>0.19  | V<br>0.25   | IV<br>0.25   |  |  |
|                                     |        | <hr/>  |             |              |             |             |              |  |  |



significant difference existed in neutrophil percentage and absolute values between the different age groups. But from the Bar-diagram ( table No. II-6) it was observed that in case of percentage I and VI group differed significantly from one another and with all other groups. II, III and V groups did not differ among themselves significantly but differed significantly with the rest. IV group differed significantly with all other groups except II and V; and in case of absolute value I, III and V group did not differ significantly each other but they differed significantly with other groups except IV. II and VI group did not differ significantly each other but both differed significantly with all other groups except IV.

The average values of  $24.50 \pm 0.13\%$ ,  $3124 \pm 156$  per c. mm.;  $30.4 \pm 0.14\%$ ,  $3417 \pm 347$  per c. mm. and  $17.1 \pm 0.15\%$ ,  $1971 \pm 205$  per c. mm. were recorded in Tharparkar, Cross-bred and Ayrshire breeds respectively. The 'F' test (table II- 7 & 9) showed a significant difference in neutrophil percentage and absolute value between the breeds. But the value of critical difference (table II- 8 & 10) revealed that the superiority of Tharparkar and Cross-bred breeds over the Ayrshire breed in neutrophil percentage and absolute values and that of Cross-bred breed over the Tharparkar breed was highly significant.

#### Eosinophil:

The average eosinophil count of all the groups of



Tharparkar breed was  $6.14 \pm 0.15 \%$  and  $720 \pm 66$  per c. mm. The percentage and absolute values were lowest in 0 to 6 month and 6 to 12 month group, averaging  $2.62 \pm 0.22 \%$ ,  $383 \pm 131$  per c. mm. and  $2.07 \pm 0.19 \%$ ,  $326 \pm 98$  per c. mm. respectively. After that the percentage and absolute values increased with age the average being  $3.94 \pm 0.27 \%$  and  $439 \pm 133$  per c. mm. in 1 to 2 year group,  $6.44 \pm 0.17 \%$  and  $833 \pm 142$  per c. mm. in 2 to 3 year group,  $11.87 \pm 0.3 \%$  and  $1153 \pm 153$  per c. mm. in 3 to 4 year group and  $9.87 \pm 0.14 \%$  and  $1186 \pm 160$  per c. mm. in 4 to 5 year group. Analysis of variance (table II- 4 & 5) showed a highly significant difference in eosinophil % and absolute values between the age groups. But from the Bar-diagram (table II-6) it appeared that, in case of percentage I and II group did not differ significantly but differed significantly from other groups except III. IV group differed significantly from all other groups except III. V and VI did not differ significantly but differed significantly from all other groups. In case of absolute values I, II and III; IV, V and VI group did not differ significantly from each other but differed significantly from all other groups.

The average value of eosinophil in Tharparkar, Cross-bred and Ayrshire breed was  $6.14 \pm 0.15 \%$ ,  $720 \pm 66$  per c. mm.,  $6.8 \pm 0.12\%$ ,  $814 \pm 154$  per c. mm. and  $10.9 \pm 0.13\%$ ,  $1249 \pm 154$  per c. mm. respectively. It was observed from the analysis of variance (table II- 7&9) that the difference between the breeds in eosinophil percentage was highly significant but



TABLE II-7

Analysis of variance of blood picture between Tharparkar,  
Cross-bred and Ayrshire breeds.

| Source of variation | Red blood corpuscles      |          |         | White blood corpuscles    |             |       | Absolute lymphocyte value |            |       |
|---------------------|---------------------------|----------|---------|---------------------------|-------------|-------|---------------------------|------------|-------|
|                     | D.F.                      | M.S.     | F.      | D.F.                      | M.S.        | F.    | D.F.                      | M.S.       | F.    |
| Between br breed    | 2                         | 47.79    | 14.35** | 2                         | 41964638.60 | 2.83  | 2                         | 42250535.3 | 4.12* |
| Within breed        | 125                       | 3.33     |         | 125                       | 14822950.6  |       | 125                       | 10251550.3 |       |
| <hr/>               |                           |          |         |                           |             |       |                           |            |       |
|                     | Absolute neutrophil value |          |         | Absolute eosinophil value |             |       | Absolute monocyte value   |            |       |
|                     | D.F.                      | M.S.     | F.      | D.F.                      | M.S.        | F.    | D.F.                      | M.S.       | F.    |
| Between breed       | 2                         | 10586545 | 5.11**  | 2                         | 1918501     | 4.71* | 2                         | 51957.5    | 0.38  |
| Within breed        | 125                       | 2069127  |         | 125                       | 407296      |       | 125                       | 135220.7   |       |
| Total               | 127                       |          |         | 127                       |             |       | 127                       |            |       |

\* Significant at 5 % level

\*\* Significant at 1% level.



10

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1

100



TABLE II-9

Analysis of variance of blood picture between Tharparkar,  
Cross-bred and Ayrshire breeds.

| Source of variation | Lymphocyte % |        |       | Neutrophil % |        |        | Eosinophil % |        |        |
|---------------------|--------------|--------|-------|--------------|--------|--------|--------------|--------|--------|
|                     | D.F.         | M.S.   | F.    | D.F.         | M.S.   | F.     | D.F.         | M.S.   | F.     |
| Between breed       | 2            | 163.73 | 3.42* | 2            | 325.82 | 8.56** | 2            | 319.51 | 7.47** |
| Within breed        | 125          | 47.76  |       | 125          | 38.04  |        | 125          | 42.75  |        |
| Total               | 127          |        |       | 127          |        |        | 127          |        |        |
| Monocyte percentage |              |        |       |              |        |        |              |        |        |
| Between breed       | 2            | 5.97   |       | 2            | 5.78   |        |              |        |        |
| Within breed        | 125          | 19.31  | 0.30  | 125          | 2.62   | 2.20   |              |        |        |
| Total               | 127          |        |       | 127          |        |        |              |        |        |
| Basophil percentage |              |        |       |              |        |        |              |        |        |
| Between breed       | 2            |        |       | 2            |        |        |              |        |        |
| Within breed        | 125          |        |       | 125          |        |        |              |        |        |
| Total               | 127          |        |       | 127          |        |        |              |        |        |

\* significant at 5 % level

\*\*= significant at 1 % level.

N.B.- For percentage the data were Aresin transformed.



TABLE II-10

Table showing the critical difference to test the significance of mean difference among different breeds.

| Breeds                  | Lymphocyte % |                     | Neutrophil % |                     | Eosinophil % |                     |
|-------------------------|--------------|---------------------|--------------|---------------------|--------------|---------------------|
|                         | Mean         | Critical difference | Mean         | Critical difference | Mean         | Critical difference |
|                         | 5            | 4                   | 5            | 4                   | 5            | 4                   |
| Tharparkar & Ayrshire   | 1.63         | 3.52                | 4.63         | 7.4**               | 3.25         | 4.27                |
| Tharparkar & Cross-bred | 7.37**       | 3.52                | 4.63         | 5.9**               | 3.25         | 4.27                |
| Ayrshire & Cross-bred   | 9.0**        | 4.7                 | 6.18         | 13.3**              | 4.25         | 5.58                |
|                         | Monocyte %   |                     | Basophil %   |                     |              |                     |
| Tharparkar & Ayrshire   | 0.07         | 2.31                | 3.03         | 0.16                | 0.84         | 1.10                |
| Tharparkar & Cross-bred | 0.23         | 2.31                | 3.03         | 0.01                | 0.84         | 1.10                |
| Ayrshire & Cross-bred   | 0.30         | 3.02                | 3.96         | 0.17                | 1.09         | 1.44                |

\*\* significant at 1 % level.



only significant difference was noted between the breeds in case of absolute value. From the value of critical difference (table II- 8 & 10) it was noted that average eosinophil percentage and absolute value of Ayrshire breed was significantly higher than Tharparkar breed. There was no significant difference between any other two breeds.

### Monocyte:

The average value of monocyte in Tharparkar breed was calculated as  $4.43 \pm 0.06 \%$  and  $559 \pm 39$  per c. mm. In 0 to 6 month and 6 to 12 month group the average values were  $3.88 \pm 0.07$ ,  $637 \pm 109$  per c. mm. and  $4.43 \pm 0.11$ ,  $710 \pm 107$  per c. mm. respectively. The values remained stable with slight variation in 1 to 2 year and 2 to 3 year group, averaging  $3.62 \pm 0.05 \%$ ,  $372 \pm 44$  per c. mm. and  $3.75 \pm 0.10 \%$ ,  $452 \pm 62$  per c. mm. respectively. A slight increase was recorded in 3 to 4 year and 4 to 5 year group having the mean values as  $6.0 \pm 0.45 \%$ ,  $608 \pm 131$  per c. mm. and  $4.87 \pm 0.14 \%$ ,  $571 \pm 78$  per c. mm. respectively. Analysis of variance ( table II- 4 & 5) of monocyte percentage and absolute values did not reveal significant difference between the age groups. From the Bar-diagram (table II-6) it was noted that there was also non significant difference in percentage between any two groups but in case of absolute value I group differed from III group and II group differed from IV group significantly. No significant difference existed between any other two groups.





The average value of  $4.43 \pm 0.06 \%$  and  $559 \pm 39$  per c. mm. in Tharparkar breed,  $4.2 \pm 0.09 \%$  and  $474 \pm 73$  per c. mm. in Cross-bred and,  $4.5 \pm 0.17 \%$  and  $527 \pm 85$  per c. mm. in Ayrshire breed were noted. Analysis of variance (table II-7 & 9 ) revealed no significant difference between the breeds.

#### Basophil:

The mean value of Tharparkar breed was found as  $0.16 \pm 0.008 \%$  and  $24 \pm 6$  per cubic millimeter. The average in 0 to 6 month and 6 to 12 month group was  $0.18 \pm 0.02 \%$ , 31 per c. mm. and  $0.06 \pm 0.01 \%$ , 12 per c. mm. respectively. The average value remained stable in 1 to 2 year group having  $0.06 \pm 0.01 \%$  and 8 per c. mm. The average values in 2 to 3 year, 3 to 4 year and 4 to 5 year groups were  $0.25 \pm 0.02 \%$  and 28 per c. mm.,  $0.25 \pm 0.03 \%$  and 32 per c. mm. and,  $0.19 \pm 0.02 \%$  and 24 per c. mm. respectively. Analysis of variance (table II- 4 & 5 ) of basophil percentage and absolute value did not reveal any significant difference between the age groups.

The average value in Tharparkar, Cross-bred and Ayrshire breeds was calculated as  $0.16 \pm 0.008\%$ ,  $0.17 \pm 0.037 \%$  and  $0\%$  respectively. Statistically (table II-9 ) there was no significant difference between the breeds.

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## D I S C U S S I O N

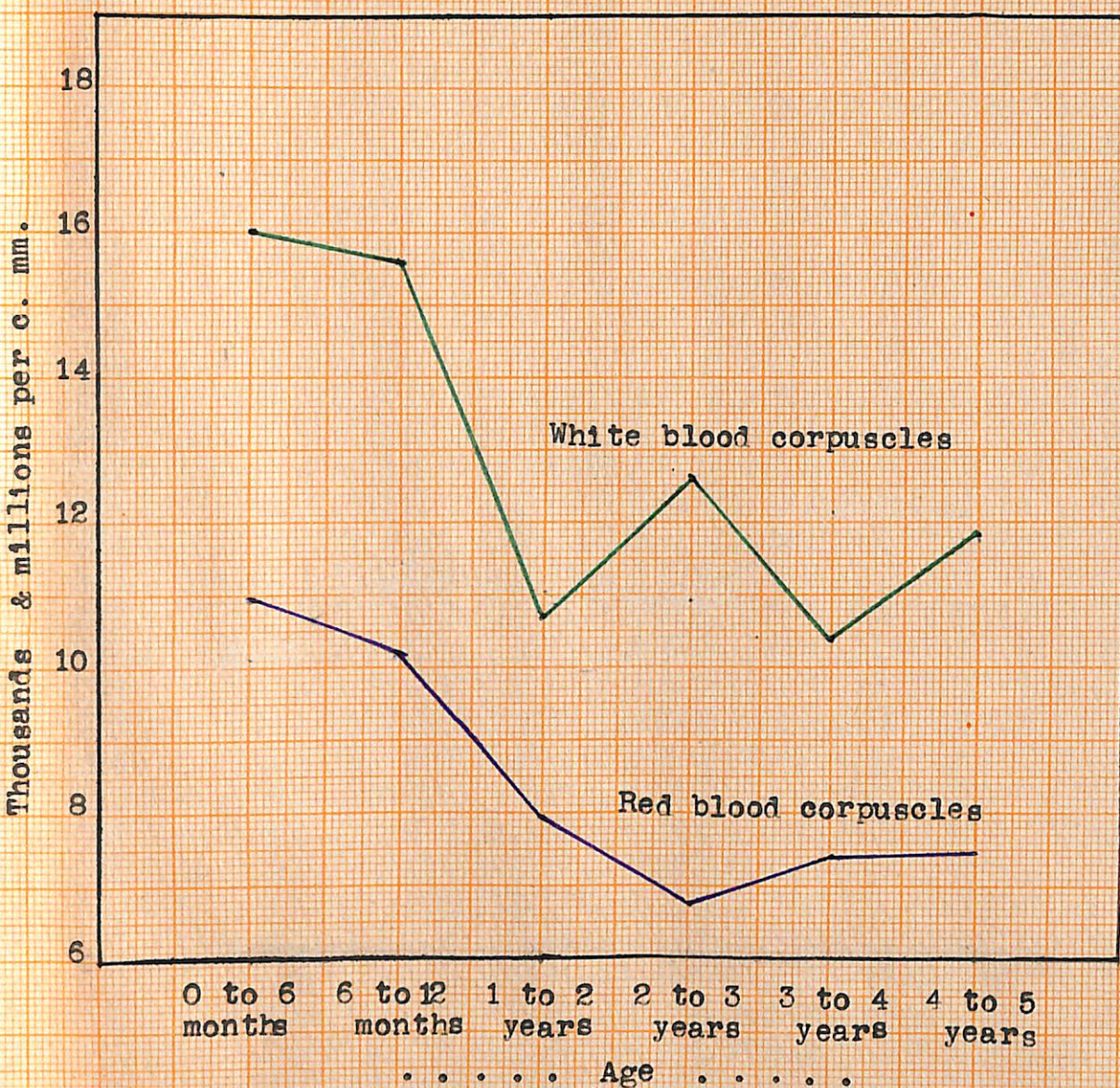
### Red blood corpuscles:

It appeared from table II-2 that the red cell count was highest in young groups; then it went on decreasing up to the age of 3 years and increased gradually up to 4 years and remained stable in 5 year group (graph II-1), which was in agreement with the works of Norris and Chamberlin (1929) and Canhom (1930). Fraser (1930) recorded increasing value with advancing age. He observed the low red count in yearling bulls at grass, which according to him probably was due to an active and productive existence and rapid growth; high count in bulls of 4 to 5 year old, which he speculated due to stall feeding, minimum activity and production, and attributed this increase to the greater age of the older bulls. Penny et al. (1966) also suspected increase in R.B.C. count with increasing age but failed to show significant age difference. Many workers (Storch, 1901; Canhom, 1930; Mullick & Pal, 1944; Greatorrex, 1957 and others) had recorded decreasing value with advancing age suggesting a increased haemopoetic activity or this decrease in number might be due to increase in size of the corpuscle as stated by Holman (1956), and the present result agreed with the findings of above mentioned workers. Analysis of variance (table II-4) revealed a highly significant difference between



Graph No. II-1.

Mean values of total Red blood corpuscles and white blood corpuscles in Tharparkar bulls in different age groups.





the age groups which was in agreement with the findings of Greatorrex (1957) and Patel et al. (1965).

The average value of Tharparkar breed was higher than that calculated for Cross-bred and Ayrshire breeds (table II-8) and it was in concurrence with the findings of Shankaranarayanan (1937). The mean R.B.C. count of Cross-bred and Ayrshire breeds were more or less same and it was in agreement with the findings of Rusoff et al. (1954), Holman (1956) and Penny et al. (1966).

Statistically significant difference was noted between the breeds (table II-7) which agreed with Shankaranarayanan (1937), Rusoff et al. (1944) and Khajuria and Razdan (1966) could not find significant difference between the breeds. The personal errors of the technicians making the counts were in partly responsible for the great disparity with regard to mean R.B.C. count. However, the differences in the red cell numbers of cattle as reported from different regions of the world were of such magnitude as to suggest that breed, climate and level of nutrition may have significant influences on red cell numbers, as revealed by Schalm (1961).

#### Total leucocyte count (W.B.C.):

It was apparent from table II-2 that leucocyte count did not show any definite trend with increasing age (graph II-1). However, high counts were recorded in young bull calves and low counts in older bulls, which might be due to



muscular activity associated with the struggling of the young animals when bled, this was explained on the basis of flushing leucocytes into the circulation that had become sequestered in the capillary bed during relative inactivity (Schalm, 1961) or it may be the sign of lessened activity of the haemopoietic organs due to increasing age (Moberg, 1955). The present result was in agreement with the findings of Schultz (1905), Holman (1956), Patel et al. (1960, 65) and Riegler et al. (1966). Analysis of variance (table II-4) did not reveal any significant difference between the age groups, which agreed with Braun Warner (1941), Moberg (1955), Grestorex (1957) and Khajuria and Razdan (1966).

The average count of Tharparkar breed was in agreement with the results of Shankaranarayanan (1937) and Patel et al. (1965). The mean leucocyte value of Ayrshire breed was higher than that reported by Rusoff et al. (1954), Penny et al. (1966) and others. While the average count of Cross-bred was in consistent with findings of Shankaranarayanan (1937) for Cross-bred animals. Average leucocyte count of Tharparkar breed (table II-8) was significantly higher to Ayrshire and Cross-bred breeds and the present result was in agreement with the findings of Shankaranarayanan (1937) as he also recorded higher values in Indian breeds than the foreign breeds and Cross-bred. Rusoff et al. (1954) and Khajuria and Razdan (1966) could not find significant difference between the breeds.



### Lymphocyte:

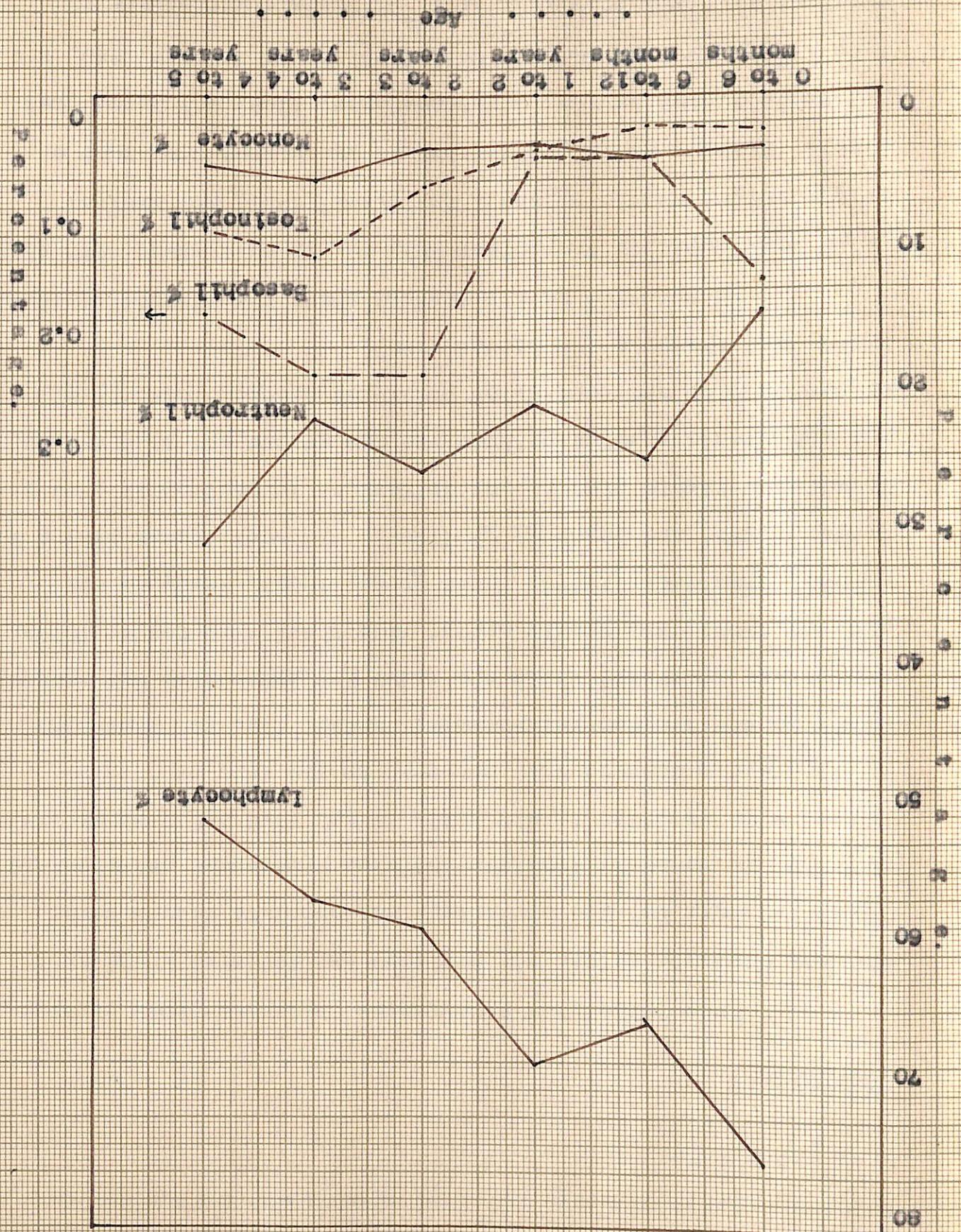
It appeared from the table II-2 that the lymphocyte percentage and absolute values were higher in young bull calves, and recorded a decrease with advancing age (graph II-2 & 3) which might be due to the atrophy of the thymus, with increase in age as stated by Moberg (1955). The present result agreed with the findings of Moberg (1955) and was in close agreement with Fraser (1930), Canham (1930), Holman (1956) and Grestorex (1957). Statistically the lymphocyte percentage and absolute values were highly significant between the age groups (table II-4 & 5), which concurred with Holman (1956), Grestorex (1957) and Khajuria and Razdan (1966).

The mean percentage of lymphocyte of Tharparkar breed was in consistent with the findings of many Indian workers (Mullick and Pal, 1943; Mishra and Biswal, 1961; Mithuji et al., 1966 & others), but it was higher than the findings of Patel et al. (1966). The average percentage for Ayrshire breed was higher than that recorded by Shankaranarayanan (1937) and many foreign workers (Holman, 1956; Penny et al., 1966 and others). The average percentage of cross-bred bull was in close agreement with the findings of Shankaranarayanan (1937) for Cross-bred bull only. Result of the analysis of variance (table II-7 & 9) of lymphocyte percentage and absolute values revealed a significant difference between the breeds, which agreed with Romano (1935) and Grestorex (1957). But Khajuria and Razdan (1966) and



Graph No. 11-2.

Mean differential count of Tharparkar  
bulls in different age groups.





Penny et al. (1966) could not find significant difference between the breeds.

### Neutrophil:

From table II-2 it appeared that neutrophil percentage and absolute values did not show any definite trend with advancing age; but low percentage and absolute values were recorded in young bull calves and high percentage and absolute values were recorded in old bulls (graph II-2 & 3), which might be due to the changes that occurred in the form of "Cross-over" in the lymphocyte:neutrophil ratio, i.e. the neutrophil percentage increased while the value for lymphocyte was reduced (Moberg, 1955). The result agreed with those reported by Holman (1956), Greateorex (1957), Patel et al. (1965), Khajuria and Razdan (1966) and Penny et al. (1966). 'F' test (table II-4 & 5) revealed a highly significant difference in neutrophil percentage and absolute value between the age groups and this was in agreement with the results of Greateorex (1957), Patel et al. (1965) and Khajuria and Razdan (1966).

As recorded in the present study the average percentage of neutrophil in Tharparkar bull was in consistant with that of the Orissa cattle (Mithuji and Biswal, 1961) and Kankrej cattle (Mithuji et al., 1966), but it was higher than that of the South Indian cattle (Govindan-Nayar, 1949) and lower than that of the Hariana and Gir bullock (Kehar and Murty, 1945, and Patel et al., 1965 respectively). The average percentage of



neutrophil in Ayrshire breed was lower than that reported by Holman (1955) and Penny et al. (1966) in Ayrshire, Hereford and Friesian breeds. The average neutrophil percentage of Cross-bred bull was in concurrence with the report of Shankaranarayanan (1937) for Cross-bred bulls. The result of the present study indicated that (table II-7 & 9) there was highly significant variation in neutrophil percentage and absolute values between the breeds. The result agreed with those obtained by Greatorrex (1957) and Penny et al. (1966).

#### Eosinophil:

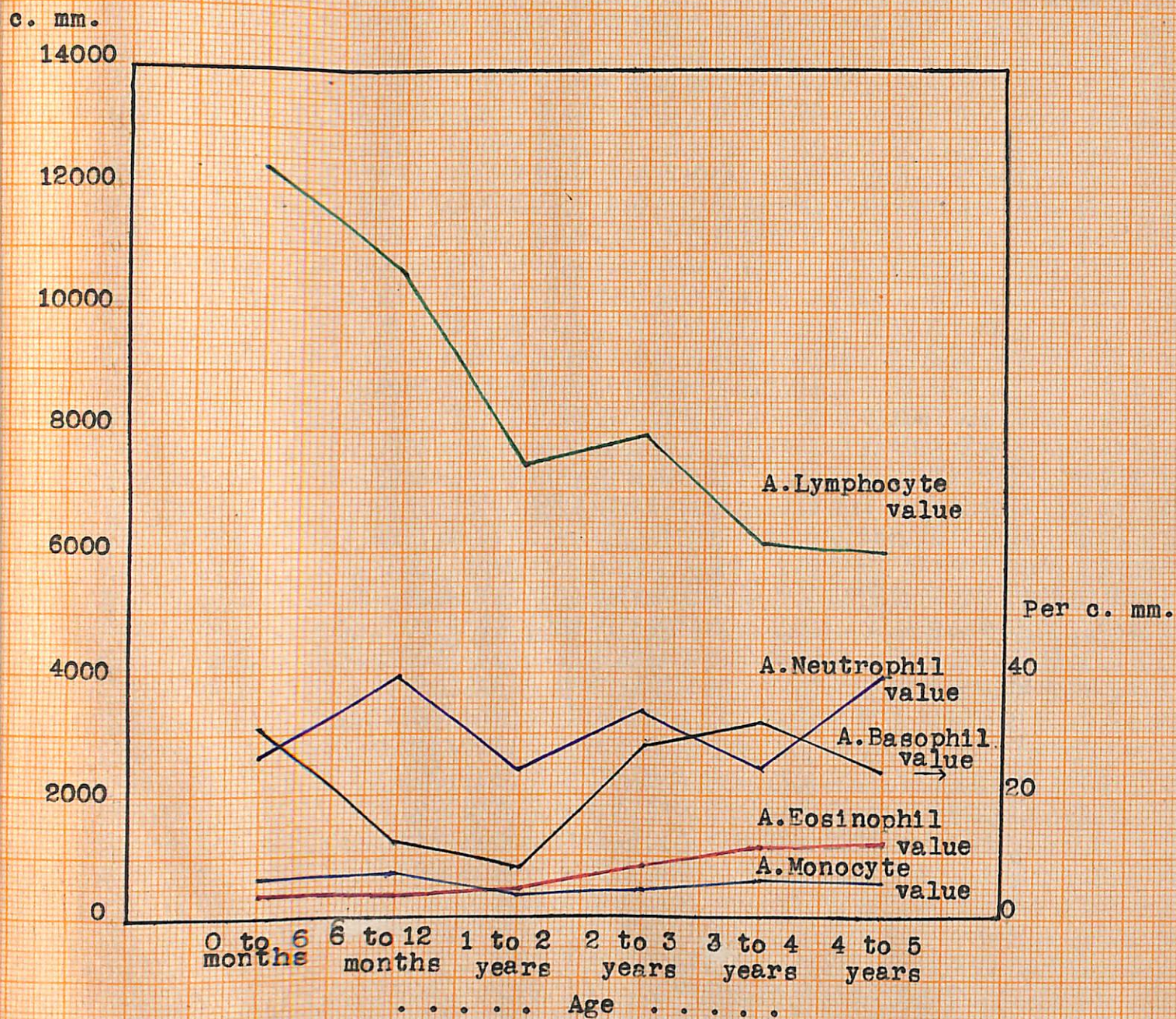
It was revealed from the table II-2 that eosinophil percentage and absolute values increased gradually with advancing age (graph II- 2 & 3), which was in consistent with the findings of Canham (1930), Fraser (1930), Moberg (1955), Patel et al. (1960, 55), Khajuria and Razdan (1966) and Penny et al. (1966) and in close agreement with the results of Holman (1956) and Greatorrex (1957). Analysis of variance (table II- 4 & 5) indicated a highly significant difference in eosinophil percentage and absolute value between age groups and this was in agreement with the results of Greatorrex (1957).

The average percentage of eosinophil in Tharparkar breed was in agreement with the reports of Govindan Nayar (1949), Patel et al. (1965) and Mithuji et al. (1965) but was lower than the findings of Shankaranarayanan (1937) in Sindhi and Gir bull and higher than the results of Kehar and Murty (1945)



Graph No. II-3.

Mean absolute values of Tharparkar bulls  
in different age groups.





in Mariana bullock. The average eosinophil percentage of Ayrshire breed was in consistent with the findings of Shankaranarayanan (1937) for Ayrshire bull, but was slightly higher than the percentage recorded by Fraser (1930), Holman (1955) and Gourley (1959). The average eosinophil percentage of Cross-bred bull was in agreement with the findings of Canham (1930). Analysis of variance (table II- 7 & 9) revealed a highly significant difference in eosinophil percentage and significant difference in absolute values between the breeds, which agreed with Greatedorex (1957). Khajuria and Razdan (1966) and Penny et al. (1966) could not find significant difference between the breeds.

#### Monocyte:

It was apparent from table II-2 that monocyte percentage and absolute values did not show any definite trend with increasing age (graph II- 2 & 3), which was in consistent with the findings of Fraser (1930), Greatedorex (1957) and Patel et al. (1965). Statistically (table II- 4 & 5) significant difference was not found in monocyte percentage and absolute values between the age groups, which agreed with the results of Penny et al. (1966).

The average percentage of monocyte in Tharparkar breed was in concurrence with Mistra and Biswal (1961), but it was slightly lower than that recorded by Kehar and Murty (1945). The average monocyte percentage of Ayrshire bull was in



agreement with Canham's (1930) finding but was lower than that reported by Fraser (1930) and Holman (1956). The average monocyte percentage of Cross-bred bull was in close agreement with the reports of Govindan Nayar et al. (1949). No significant variation (table II- 7 & 9) was found in monocyte percentage and absolute values between the breeds, which agreed with the findings of Grestorex (1957), Khajuria and Razdan (1966) and Penny et al. (1966).

#### Basophil:

It was apparent from the table II-2 that basophil percentage and absolute values did not show any definite trend with advancing age (graph II - 2 & 3). Basophils were very scanty in all the age groups (Fraser, 1930) and this was in agreement with the present findings. Analysis of variance (table II- 4 & 5) of basophil percentage and absolute values did not reveal any significant difference between the age groups and this was in concurrence with the result of Penny et al. (1966).

The average percentage of basophil for Tharparkar, Cross-bred and Ayrshire breed was very low in the present result. Hence it was difficult to compare with that of other's findings. Analysis of variance (table II- 9) failed to reveal a significant difference in basophil percentage between the breeds.

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## CORRELATION BETWEEN



## REVIEW OF LITERATURE

During the course of study it was understood that much attention had not been paid to find any direct relationship of semen picture with the blood picture of the males for drawing an influence about the breeding performance of the bulls. Of course, a few reference could reveal that attempts were made by the workers, who either tried to correlate the leucocyte picture of the bull with the performance of its dam and daughter or to correlate the other factors with the semen character, but not directly with the blood cellular count. However, the references available in relation to this are mentioned below:

In recent years attempts have been made to correlate blood cell volume and haemoglobin contents with semen quality of bulls ( Mukherjee and Bhattacharya, 1952), and also to anticipate the lactational performance of dairy heifers by examination of blood picture ( Schultz, 1960).

Groblewska (1953) attempted to asses the breeding value of bulls on the basis of the blood leucocyte picture. A brief account is given of an experiment carried out in the first half of 1953 using Red Polish bulls and closely related cows and heifers (daughters of the bulls). The results to data ( not given) indicate the possibility of using leucocyte



picture as an aid in confirming an animals constitutional type.

Groblewska (1954) again attempt to estimate the breeding value of bulls from their white blood picture. Observations on 25 Red Polish ( Polish Red), 2 Red Danish, 1 Jersey and 10 Red Polish X Red Danish bulls have indicated that there is a connection between the leucocyte index of a bull and the production level of his dam. The ratio of one granulocyte to two agranulocytes is said to be characteristic of bulls belonging to the respiratory constitutional type, descended from dams showing a high level of production. A smaller ratio is found in bulls of the digestive type, descended from dams with lower milk production.

Again in 1960, Groblewska attempts to determine the breeding value of bulls on the basis of the white blood picture. In order to test the possible usefulness of the leucocyte index ( ratio of the neutrophils to lymphocytes) in estimating the breeding value of Polish Red bulls, a comparison was made between the leucocyte indices of 6 bulls and their breeding performance, as evidenced by the milk yields of their daughters, based on 84 dam-daughter comparisons. It was found that 58.3 - 88.3% of the daughters of 5 bulls with a leucocyte index lower than 1:2 gave higher milk yielding than their dams, whereas none of the daughters of the other bull, which had a leucocyte index of 1:3, showed improved proved production, their yields being less than those of other daughters of the same dams by a different bull. A negative correlation ( $r=0.74 \pm 0.042$ ) was



found between leucocyte index and level of milk production in 166 Polish Red cows.

Goswami (1965) administered Sodium-L-Thyroxine and Equine Gonadotrophic hormones to the buffalo bulls to study on correlation of spermatozoan and blood corpuscular counts. He observed that there is no apparent effect on the white blood corpuscular counts of the animals due to the administration of the hormones and the spermatozoan counts behaved independent of that variable. It appears that the hormones administered have no direct influence on engendering of the white blood corpuscles. The spermatozoa concentration in millions per ml. and the total number of spermatozoa in millions per collection showed the same trend as that of red blood corpuscular counts of the animals. It appears that these variables are under the influence of same hormonal action. Semen volume supported the above trend of hormonal inter-relationship.

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## M A T E R I A L S   A N D   M E T H O D S

For the correlation between semen picture and blood picture eight Tharparkar, four Ayrshire and four Cross-bred (Sahiwal X Brown Swiss) bulls maintained at Government Cattle Farm, Patna were taken. These bulls were from two to five years of age.

In all thirty two semen and blood samples in Tharparkar, sixteen semen and blood samples in Cross-bred and sixteen semen and blood samples in Ayrshire breed were correlated.

Statistical analysis was done according to Snedecor ( 1961).

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## R E S U L T S

The correlation coefficient values ( $r$ ) between the semen characters and blood cellular count was calculated in all the three breeds and the results have been tabulated in table III-1.

From the table it appeared that the ' $r$ ' values between R.B.C. and sperm concentration were + 0.52 (52%) in Tharparkar, + 0.19 (19%) in Cross-bred and - 0.38 (38%) in Ayrshire breed. This means that the R.B.C. value was positively correlated with sperm concentration in Tharparkar breed only and it was significant, while although it was correlated in Ayrshire and Cross-bred but the values were nonsignificant.

In case of total leucocyte count and sperm concentration the correlation was negative having -0.28 (28%) in Tharparkar, -0.17 (17%) in Cross-bred and -0.20 (20%) in Ayrshire breeds. But there correlation coefficient values were not found significant.

The correlation coefficient between absolute lymphocyte value and sperm concentration was + 0.10(10%) in Tharparkar, + 0.25 (25%) in Cross-bred and +0.26 (26%) in Ayrshire breeds. But these correlation coefficient values were not found significant.

In case of absolute eosinophil number and sperm



TABLE III-1.

Table showing phenotypic correlation coefficient between semen picture and blood picture of Tharparkar, Cross-bred and Ayrshire bulls.

| Correlation between                                   | No. of<br>pairs | Tharparkar<br>breed | No. of<br>pairs | Cross-bred<br>breed | No. of<br>pairs | Ayrshire<br>breed |
|---|-----------------|---------------------|-----------------|---------------------|-----------------|-------------------|
| Red blood cells and<br>Sperm concentration            | 32              | +0.52 *             | 16              | +0.19               | 16              | -0.38             |
| White blood cells and<br>Sperm concentration          | 32              | -0.28               | 16              | -0.17               | 16              | -0.20             |
| Absolute lymphocyte value<br>and Sperm concentration  | 32              | +0.10               | 16              | +0.25               | 16              | +0.26             |
| Absolute eosinophil value<br>and Sperm concentration  | 32              | +0.49 *             | 16              | +0.64 *             | 16              | +0.66 *           |
| Absolute neutrophil value<br>and Sperm concentration  | 32              | +0.01               | 16              | -0.15               | 16              | +0.32             |
| Eosinophil percentage and<br>percentage of dead sperm | 32              | -0.25               | 16              | -0.10               | 16              | +0.21             |

\*Significant at 5% level.

+ Positive correlation.

- Negative correlation.



concentration, the 'r' values were + 0.49 (49%) in Tharparkar, + 0.64 (64%) in Cross-bred and + 0.66 (66%) in Ayrshire breeds. This means that the absolute eosinophil value was positively correlated with sperm concentration in all the three breeds under study which was found to be significant statistically.

It was observed to be not correlated with absolute neutrophil value and sperm concentration as indicated by 'r' value which was +0.01 (1%) in Tharparkar, -0.15 (15%) in Cross-bred and +0.32 (32%) in Ayrshire breeds because this was not significant.

The percentage of dead sperm was found to be negatively correlated with eosinophil percentage in Tharparkar and Cross-bred as the values were -0.25 (25%) and -0.10 (10%) respectively. While it was positively correlated in Ayrshire because the value was +0.21 (21%). But this correlation coefficient values were non-significant in all the three breeds.

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### DISCUSSION

A significant positive correlation was observed between sperm concentration and red cell count in Tharparkar breed (table III-1), which agreed the findings of Goswami (1966) who also reported that the sperm concentration showed the same trend as that of R.B.C. count. Logically, this result finds supports of Taber et al. (1943) who claimed male hormone was mainly responsible for the stimulation of erythrocyte forming cells in the bone marrow; and Gole and Cupps (1959), Perry (1961) and Hafez (1962) who noted that the male hormone viz., testosterone, was responsible for spermatogenesis.

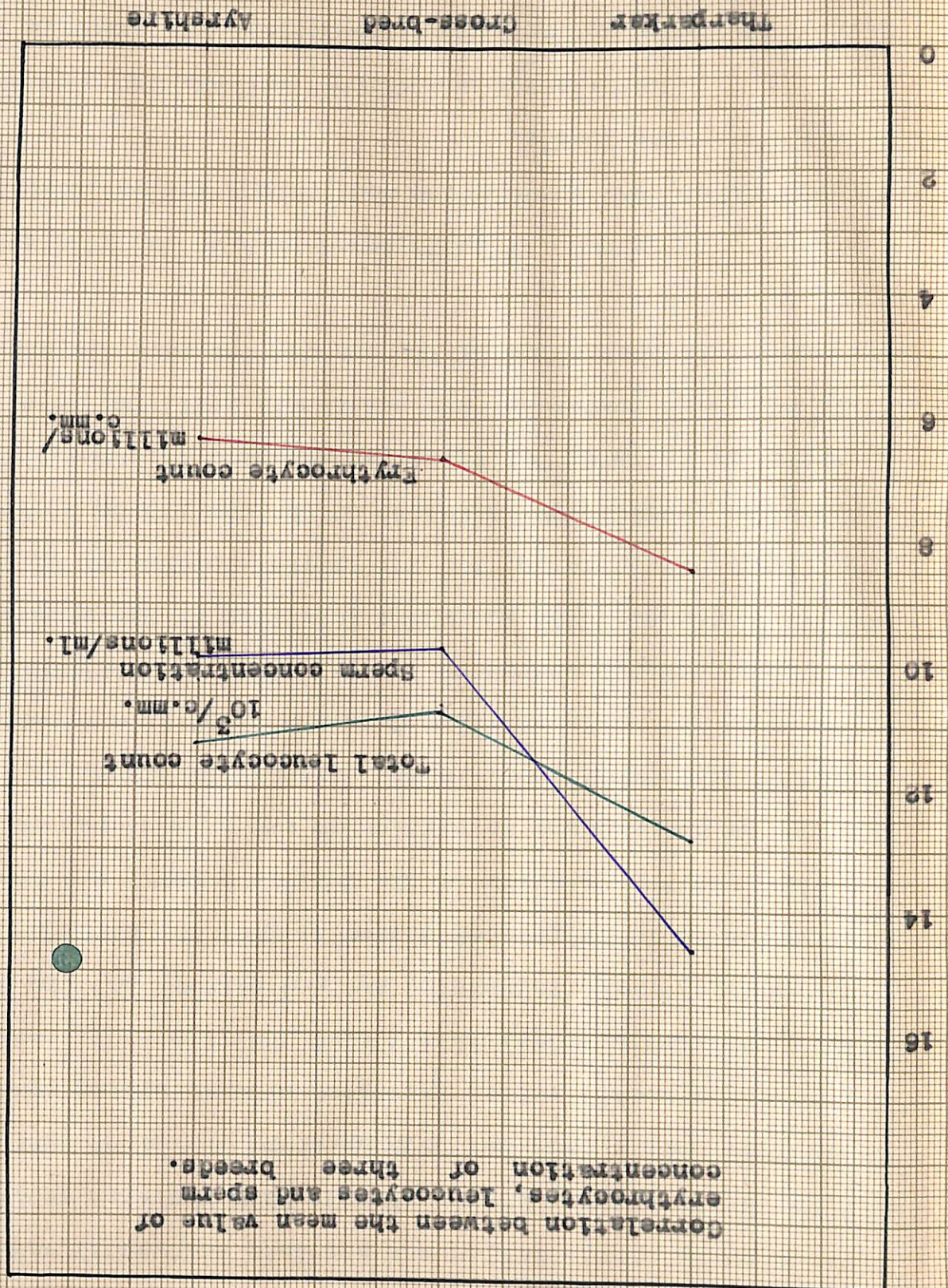
Although, there was statistically a non-significant correlation between these two characters in rest of the breeds, but the graphical trend (graph III-1) indicated a positive movement in the values of the two characters.

In case of total leucocyte count (W.B.C.), no correlation was found with sperm concentration. This finding concurred with Goswami (1966) who stated that the hormones were not having any effect on the total white blood corpuscles, thus, it also made clear that the spermatogenesis was not related with W.B.C. production.

The lymphocyte value was having a positive trend with the sperm concentration in all the three breeds, although it was statistically not significant.



Graph No. III-1.





A significant positive correlation was recorded between sperm concentration and absolute eosinophil value in all the three breeds. It has already been reviewed under Part II of this thesis, and has been observed in the present study that the eosinophil values increased with advancing age. Perry (1961) and Maulae (1962) have proved that the sperm concentration increases with increasing age and maturity of the bulls. This might be interpreted logically that the hormones responsible for the maturity of the bull in general and the production of spermatozoa in particular are in some way concerned with the eosinophil level in the circulating blood. It is well known that the eosinophil level in the blood is controlled by the adrenocortical secretion (Selye, 1950) and that the same hormone influences the masculine behaviour of the animals also viz., aggressiveness and the sex libido (Collins et al., 1951, and Prabhu, 1956). The intensity of the sex libido of the bull is taken as an index for the semen quality. It seems, therefore, that the sperm production and eosinophil level in the circulating blood is controlled by the one and the same hormonal complex.

In case of sperm concentration and absolute neutrophil value, the correlation coefficient was not significant in all the three breeds.

Correlation coefficient between the percentage of dead spermatozoa and eosinophil percentage was positive in Tharparkar and Cross-bred breeds and negative in Ayrshire



breed but the 'r' values were non-significant in all the three breeds.

Thus, it can fairly be concluded that the sperm concentration of a bull possess a positive correlation with total R.B.C. count and absolute eosinophil values. These two blood cellular characters viz., the R.B.C. and eosinophil, therefore can be taken under consideration in judging the quality of a bull in animal breeding practice. Further, research work on the aspect requires attention to draw a concrete conclusion in helping for the better livestock breeding.

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S U M M A R Y



## S U M M A R Y

A study was undertaken to observe the semen characters of Tharparkar, Cross-bred (Sahiwal X Brown Swiss) and Ayrshire bulls maintained at Government Cattle Farm, Patna and to find their correlation with the blood picture of these animals. As such, studies were made on reaction time, volume, initial motility, pH, percentage of dead sperms and concentration of spermatozoa in the ejaculate to constitute the semen characters of these bulls, while observations were made on red blood corpuscles, total leucocyte count and differential count in respect of blood picture.

An average reaction time of  $29.75 \pm 4.37$ ,  $37.81 \pm 6.2$  and  $44.68 \pm 7.27$  seconds were recorded in Tharparkar, Cross-bred and Ayrshire breeds respectively. A significant difference was found among the bulls as well as among the breeds.

The mean volume of the ejaculate was found to be  $5.17 \pm 0.37$  ml. in Tharparkar,  $4.05 \pm 0.27$  ml. in Cross-bred and  $4.74 \pm 0.22$  ml. in Ayrshire breeds respectively. Individuality and breed were noted to have significant effect in the volume of the ejaculate.

The average initial motility of spermatozoa in Tharparkar, Cross-bred and Ayrshire breed was  $4.26 \pm 0.08$ ,  $3.15 \pm 0.07$  and  $3.71 \pm 0.16$  respectively. Significant difference



due to bull and breed was also observed.

In Tharparkar the average pH was found as  $6.20 \pm 0.08$ , while in Cross-bred and Ayrshire it was  $6.38 \pm 0.41$  and  $6.26 \pm 0.39$  respectively. No significant difference among the bulls and between the breeds was recorded.

The mean percentage of dead sperm was calculated as  $6.3 \pm 0.18$ ,  $22.8 \pm 0.45$  and  $18.23 \pm 1.27$  in Tharparkar, Cross-bred and Ayrshire breeds respectively. A significant difference in this character was observed due to bull as well as due to breed.

The average concentration of sperm was noted to be  $1465.0 \pm 125.0$ ,  $977.0 \pm 107.0$  and  $985.0 \pm 103.0$  millions per milliliter in Tharparkar, Cross-bred and Ayrshire breeds respectively. Bulls and breeds both were observed to have significant effect on sperm concentration.

Highest red blood cell count was observed in young stock and then it was observed decreasing with age. Highly significant difference was noted between the age groups. The average erythrocyte count of Tharparkar, Cross-bred and Ayrshire breed was  $8.46 \pm 0.25$ ,  $6.66 \pm 0.88$  and  $6.29 \pm 0.21$  millions per c.mm. respectively. Highly significant difference due to breed was observed.

Total leucocyte count did not show any definite trend with age. However, high counts were recorded in young bull



calves and low counts in older bulls. No significant difference was found between the age groups. An average leucocyte count of  $12867 \pm 420$ ,  $10759 \pm 665$  and  $11284 \pm 750$  per c.mm. were recorded in Tharparkar, Cross-bred and Ayrshire breeds respectively. Breed was observed to have no-significant effect on the total leucocyte count.

Lymphocyte percentage and absolute values were higher in young bull calves which decreased with age. A highly significant difference was found between the age groups. The average lymphocyte percentage and absolute values in Tharparkar, Cross-bred and Ayrshire breeds were  $64.77 \pm 0.16\%$  and  $8441 \pm 357$ ,  $57.4 \pm 0.21\%$  and  $6024 \pm 312$ , and  $66.4 \pm 0.20\%$  and  $7537 \pm 629$  per c.mm. respectively. Significant difference due to breed was also observed.

Neutrophil percentage and absolute values did not show any definite trend with advancing age. However, low percentage and absolute values were recorded in young bull calves and high values in bulls. A significant difference in this character was observed due to age. The mean neutrophil percentage and absolute values were found to be  $24.50 \pm 0.13\%$  and  $3124 \pm 156$  per c.mm. in Tharparkar,  $30.4 \pm 0.14\%$  and  $3417 \pm 347$  per c.mm. in Cross-bred and  $17.1 \pm 0.15\%$  and  $1971 \pm 205$  per c.mm. in Ayrshire breed. Breed was noted to have significant effect in the neutrophil percentage and absolute values.



Eosinophil percentage and absolute values increased gradually with advancing age and a significant difference was found between the age groups. In Tharparkar the average eosinophil percentage and absolute values was found as  $6.14 \pm 0.15 \%$  and  $720 \pm 66$  per c.mm., while in Cross-bred and Ayrshire it was  $6.8 \pm 0.12 \%$  and  $814 \pm 154$  per c.mm., and  $10.9 \pm 0.13 \%$  and  $1249 \pm 154$  per c.mm. respectively. A significant difference in this character was also observed due to breed.

Monocyte percentage and absolute values did not show any definite trend with increasing age. No significant difference between the age groups was recorded. The mean monocyte percentage and absolute values were calculated as  $4.43 \pm 0.06 \%$  and  $559 \pm 39$  per c.mm.,  $4.2 \pm 0.09 \%$  and  $474 \pm 73$  per c.mm., and  $4.5 \pm 0.17 \%$  and  $527 \pm 85$  per c.mm. in Tharparkar, Cross-bred and Ayrshire breeds respectively. Analysis of variance revealed no significant difference between the breeds.

Basophil percentage and absolute values did not show any definite trend with advancing age and statistically there was no significant difference between the age groups. An average of  $0.16 \pm 0.008 \%$  in Tharparkar breed,  $0.17 \pm 0.037 \%$  in Cross-bred and  $0 \%$  in Ayrshire breed were noted. Analysis of variance did not reveal significant difference between the breeds.

Significant positive correlation was observed



between sperm concentration and red blood corpuscles count in Tharparkar only, but there was non-significant correlation in case of Cross-bred and Ayrshire breed.

Correlation coefficient between sperm concentration and total leucocyte count was non-significant in all the breeds under study.

In case of sperm concentration and absolute lymphocyte value the correlation was positive in all the three breeds but the 'r' values were non-significant.

Correlation coefficient was significantly positive between sperm concentration and absolute eosinophil value in Tharparkar, Cross-bred and Ayrshire breeds.

In case of sperm concentration and absolute neutrophil value the correlation was not significant as indicated by 'r' value.

Correlation coefficient between percentage of dead sperm and eosinophil percentage was not significant in all the three breeds under the study.

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