SOME FACTORS INFLUENCING THE PERCENTAGE OF NON-RETURNS OF DAIRY BULLS IN ARTIFICIAL BREEDING

By

Bhalchandra Tanaji Sangle

A THESIS

Submitted to the School of Graduate Studies of Michigan State College of Agriculture and Applied Science in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

Department of Dairy

1953

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Approved M. P. Ralston

Bhalchandra Tanaji Sangle

THESIS ABSTRACT

The breeding and semen collection records of bulls belonging to Michigan Artificial Breeders Association were analyzed for a period of about a year in order to study some of the factors influencing the percentage of non-returns. The factors studied were age of semen, bull, breed, month and season of breeding, registered and non-registered females.

The data were compiled by IBM machines and analyzed yielding following results:

In general, the yearly percent of non-returns for bulls and for breeds declined as age of semen advanced from 1 to 5 days. Hence semen should be used as fresh as possible.

Bulls as well as breeds varied widely in the decline of percent of non-returns for day-to-day storage of semen from 1 to 5 days.

The over-all yearly percent of non-returns was the highest for Angus and was the lowest for Red Dane breed. Holstein ranked 2nd whereas Jersey ranked 3rd to Angus in percent of non-returns.

The over-all yearly percentages of non-returns for dairy bulls for 1, 2, 3, 4, 5-day-old semen were 69.59, 63.05, 56.20, 51.54 and 50.84 percent, respectively.

In Red Dane, the percent of total services for 1-day-old semen was markedly low compared to other breeds.

The percent of non-returns was lowest in May for Holstein. It was lowest in August for Guernsey, Jersey and Red Dane, whereas for Angus, it was lowest in September.

For all dairy breeds, the percent of non-returns was the highest in fall whereas it was highest in spring for Angus. For Holstein and Red Dane, the percent of non-returns was the lowest in winter whereas for Guernsey and Jersey, it was lowest in summer.

The weighted average of percent of non-returns with 1-day-old semen was 10.9% greater in 1st service registered females than 1st service non-registered females.

The number of females inseminated with a day's collection of semen for a bull was the highest in Holstein breed.

Concentration of semen was the highest in Jersey breed. Semen volume, sperm concentration, percentage of sperms alive in non-stored semen and progressive motility of sperms in non-stored semen were higher in dairy bulls than the Angus bulls. However, the raw semen movement was lower than Angus bulls.

A highly significant positive correlation was found between raw semen movement and progressive motility of sperms in non-stored semen ($r=+0.1465$).

A highly significant (1% level) positive correlation was noted between each of the following semen characteristics and percent of non-returns:

Raw semen movement ($r = +0.195$)

Percentage of sperms alive in 4-day-old re-examined semen ($r = +0.1262$)

Concentration of sperm per 1/1000 cubic mm. of diluted semen ($r = +0.1213$)

Progressive motility of sperms in non-stored semen ($r = +0.0884$)

Observations on progressive motility of sperms either in 3-day or in 4-day-old re-examined semen seem to be of very little use in predicting the percent of non-returns.

VITA

Bhalchandra Tanaji Sangle

Candidate for the degree of

Doctor of Philosophy

Final examination, July 24, 1953, 10:00 A. M., Room 213, Dairy Building

Dissertation: Some Factors Influencing the Percentage of Non-returns of Dairy Bulls in Artificial Breeding

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INTRODUCTION

Dairy cattle breeding is one of the main tools for the improvement in the performance of a dairy herd. It can either "make or mar" the improvement depending on the ability in the selection of the sires. Judicious selection of the bulls, therefore, is very important. However, it is rather a difficult task as it requires a trained personnel having a good knowledge of genetics and animal breeding. A sound breeding program, therefore, is less likely to be within the reach of an average dairymen unless he resorts to artificial insemination. This is because experts in cattle breeding are concerned with the selection of superior sires for the artificial breeding association.

A constantly growing, increased interest for the use of artificial insemination has been demonstrated by Michigan dairymen over the past number of years. This can be easily seen from the reports of the Michigan Agricultural Statistics (1951), Michigan Cooperative Crop Reporting Service (1953), and from Annual Report of Mr. A. C. Baltzer (1951-1952). According to these reports, 6 percent of the total number of cows (2 years old and over) on Michigan farms were bred artificially in the year 1946; 12.7 percent in 1948; 14.8 percent in 1949; 17.1 percent in 1950; 20.5 percent in 1951 and 22.3 percent in the year 1952.

About 90 percent of the dairymen in Michigan could have a fairly rapid and substantial genetic improvement in their dairy herds if only they would use the superior inheritance that is provided for them by

Michigan Artificial Breeders Cooperative, Incorporated. Dairymen are becoming more conscious of the fact that artificial insemination does possess great potentialities for great improvement through the use of outstanding sires which few dairymen could afford to own. Artificial insemination is now considered to be the cheapest and easiest way of introducing the desired inheritance in the average quality herd in Michigan.

In order that the artificial breeding program be a success not only the proper selection of bulls for high production and good type is essential but also bulls that produce high quality semen which will stand up during processing, shipping and insemination, thus resulting in a high conception rate. Equally important is the quality of the inseminator-technicians and the use of the best techniques of insemination. The herd owner also has a major role to play in the success of artificial breeders cooperatives. Besides good breeding, he must be conscious of many factors such as proper feeding (nutrition), efficient management and disease control which are responsible for getting a high conception rate in his herd.

Educational programs beamed at the men working in the stud, the inseminators working in the field, the individual herd owners, and the agricultural public service personnel are important and must be maintained with a great deal of enthusiasm and finesse. The most efficient teaching is through the use of current local personal information to demonstrate the proper and improper techniques, methods and procedures. By constantly developing a program through different approaches by compiling,

analyzing and interpreting the work of the organization, the morale and ehthusiasm of the personnel can be maintained. This also favors the development of new ideas for more efficient methods and ways of bringing the latest information about the details of operation that are so important if high conception rates are to be obtained through artificial insemination.

GENERAL OBJECTIVES

The objectives of this investigation are:

I. To study some of the factors which may influence percent of non-returns:

(A) Age of semen

(B) Bull

(C) Breed

(D) Month and Season of year

(E) Registered and non-registered females

II. To study the inter-relationship of certain semen characteristics and relationship between various semen characteristics and percent of non-returns.

MATERIALS AND METHODS USED

The data used in this investigation were taken from the records kept at the Michigan Artificial Breeders Cooperative, Incorporated, East Lansing. In 1951-52 the Michigan Artificial Breeders Association consisted of 144 local organizations located throughout the State of Michigan. The bull stud is located at the central headquarters in East Lansing. The semen was collected from bulls representing 5 breeds. Of the 5 breeds, 4 were of dairy type (Holstein, Guernsey, Jersey, Red Dane) and one was of beef type (Angus).

(A) Collection of Semen

In the majority of cases, semen was collected from each bull at an interval of 6 or 8 days. The ejaculates (semen) collected at less than 5 days apart from a bull is not included in the present investigation. This is because of the fact that the current services resulting from each of these ejaculates cannot be completely separated by the procedure used in this study for determining the percent of non-returns. The collection of semen with very few exceptions, was made at night every day, except Saturday, starting from about 9 p.m. to about 12 midnight. Semen was collected by means of an artificial vagina.

(B) Examination of Semen (techniques, ratings, etc.)

Immediately after collection, the semen samples were examined in the laboratory by trained, experienced personnel, for the following semen characteristics:

Volume of semen: It was measured to the nearest 0.1 ml. by means of a graduated measuring cylinder.

Raw semen movement: It was determined by examining a drop of undiluted fresh semen under a microscope by using 100 magnification. An appropriate rating for movement of raw semen depending on the vigor of movement of the sperm mass was estimated as follows:

Rating	Explanation of the Rating	Estimated Value for the Rating
+	Maximum raw semen movement	90 percent
/ or M	Intermediate raw semen movement	70 percent
-	Very little or no raw semen movement	0 percent

No semen sample was discarded on the basis of raw semen movement alone even though the sample had - rating for raw semen movement.

Concentration of semen: It was determined by a photelometer as described by Willett and Buckner. One-tenth ml. of undiluted fresh semen was mixed thoroughly with 4 ml. of 3.2 percent sodium citrate ($Na_3C_6H_5O_7.2H_2O$) in the glass cylindrical absorption cell (vial) and the photelometer reading was taken. The reading was then converted into concentration (number of spermatozoa per cubic mm. of undiluted semen) after referring to a standard table. Semen samples showing less than 500,000 spermatozoa per cubic mm. of undiluted semen were not sent for insemination and hence could not be included in the present investigation.

Progressive motility of sperms: A drop of fresh semen diluted with egg-yolk citrate buffer was examined under a microscope using a magnification of 400. An appropriate rating for progressive motility of sperms depending on the nature of forward movement (fast or sluggish) of live-sperms in the semen sample was estimated as follows:

Rating	Explanation of the Rating	Estimated Value for the Rating
E	Excellent progressive motility of sperms	90
VG	Very good progressive motility of sperms	80
G+	Good plus progressive motility of sperms	70
G	Good progressive motility of sperms	60

In very rare cases, semen samples having less than G+ rating for progressive motility of sperms, were obtained. Such samples were discarded on the basis of other semen characteristics such as percentage of sperms alive, raw semen movement and concentration of semen.

Percentage of sperms alive: A drop of fresh semen diluted with egg-yolk citrate buffer was observed under the microscope using a magnification of 400 and the percentage of sperms alive in the sample was estimated as 80, 75, 70 etc. Semen samples with a percentage of live-sperms less than 70 were usually discarded and hence could not be included in the present investigation.

When the semen obtained during the first ejaculation from a bull was small in volume, a second ejaculate was collected subsequently from the same bull. The first and second ejaculates (non-stored semen) were examined separately for volume of semen, raw semen movement, concentration of sperm, progressive motility of spermatozoa and percentage of sperms alive. After examination, the two ejaculates were mixed together for insemination. However, the number of mixed ejaculates was small. In the present study, whenever two ejaculates from a bull were mixed, weighted averages for raw semen movement, concentration of semen, progressive motility of sperms and percentage of live-sperms were calculated on the basis of semen volume used for dilution. The weighted averages thus obtained represent the ratings for various semen characteristics of the mixed semen. In the case of volume of semen, the arithmetic average of the volume of two ejaculates was taken to represent the volume of mixed ejaculate.

In some cases, the first ejaculate from bulls was lost and hence the second ejaculate from the same bull was collected subsequently.

(C) Dilution and Storage of Semen

While a portion of the semen sample was being examined microscopically, the remaining portion contained in a glass tube was placed in warm water at about 90° F. If the semen sample was found to meet the minimum requirements of semen characteristics or combination of seme characteristics such as percentage of sperms alive, progressive motility of sperm, raw semen movement and concentration of semen (characteristics listed in order

of importance for judgment), it was poured into 55 ml. of egg-yolk citrate diluter previously warmed to 90° F. The diluted semen was then placed in a refrigerator and cooled on the average not faster than 1° F. per minute to 40° F. More diluter at 40° F. was then added to the original 55 ml. dilution making up a total volume of diluted semen on each bull according to the semen needs for a particular day's collection. About 3 to 4 ml. of diluted semen was stored in the refrigerator at 40° F. in order to re-examine on the 3rd and/or 4th day of storage for progressive motility of sperms and percentage of sperms alive. The remaining, fresh, diluted semen was poured into 8 ml. glass vials. The vials were plugged with a cork stopper and the top of the vial along with the stopper was paraffined.

(D) Shipment of Semen

1. Parcel post shipment: Glass vials containing diluted semen were placed in a hollow tin can with a jacket containing ice. The can was wrapped in an insulated bag which was then packed for shipment by parcel post. The semen was in transit from 16 to 24 hours at about 40° F. and was received by inseminators after about 12 to 35 hours from the time of collection. However, the majority of the inseminators received the semen about 34 hours from the time of collection.
2. Airplane shipment of semen: Glass vials containing diluted semen were wrapped in several thicknesses of paper and were placed in the insulated bag in contact with a tin can enclosing ice. The bag was then packed in a cardboard box for shipment

by airplane. The semen was in transit from 0 to 5 hours at about 40° F. and was received by the inseminators after 11 to 16 hours (on an average 13½ hours) from the time of semen collection.

Semen received by the inseminators either by parcel post or by airplane delivery, was kept in a refrigerator at about 40° F. until used. The semen was carried to the farms for insemination in a thermos bottle or ice chest at temperature of about 40° F.

(E) Re-examination of the diluted stored semen.

Semen samples stored in a refrigerator at 40° F. were re-examined under a microscope (400 magnification) for progressive motility of sperms and percentage of live sperms on the 3rd and/or 4th day of storage. An appropriate rating for progressive motility of sperms depending on the nature of forward movement (fast or sluggish) of live-sperms was estimated as follows:

Progressive Motility of Sperms

Rating	Explanation of the Rating	Estimated Value for the Rating
G+	Good plus progressive motility of sperms	70
G	Good progressive motility of sperms	60
G-	Good minus progressive motility of sperms	50
F+	Fair plus progressive motility of sperms	40
F	Fair progressive motility of sperms	30
F-	Fair minus progressive motility of sperms	20
P	Poor progressive motility of sperms	10
Dead	No progressive motility of sperms	0

Percentage of sperms alive: It was estimated as 60, 55, 50, 45, etc.

(F) Preparation of Egg-Yolk citrate diluter:

The egg-yolk citrate diluter was made up of 1 part of egg-yolk to 1 part of citrate buffer. To this egg-yolk citrate mixture, 500 mgms. of crystalline dihydro-streptomycin (Merck) and 500,000 units of crystalline penicillin (Merck) were added for every 1000 ml. of the mixture. The egg-yolk citrate was prepared every day except Saturday, about 8 hours before use. The diluter was kept in a refrigerator at 40° F. until it was needed for dilution.

Citrate buffer that was used for preparing the diluter was made every week by adding 60 grams of sodium citrate (Merck) ($Na_3C_6H_5O_7.2H_2O$) and 2000 ml. of distilled water. The citrate solution was heated in a pressure cooker at 212° F. for 20 minutes at 15 pounds pressure. It was then cooled and stored in the refrigerator at 40° F. and used whenever needed during the week.

GENERAL INVESTIGATIONAL PROCEDURE

(A) Source of Data

The data on semen characteristics was taken from the semen collection data sheets (records) of the Michigan Artificial Breeders Association and covers the period from the semen collection date of June 1, 1951 to May 31, 1952. The breeding record corresponding to these ejaculates was obtained from IBM cards (Standard Card No. 1) punched according to the outline mentioned below and covers the period from June 1, 1951 to September 4, 1952. Breeding records were analyzed on the basis of 90 day non-returns for 1, 2, 3, 4, 5 day old semen. A female (cow or heifer) was assumed to be pregnant if she was not bred 90 days following the last service. The percent of non-returns was calculated on the total of first, second, third, fourth, fifth and over services to a female.

Outline of Standard Card No. 1 (Breeding Record)

Columns punched	Description
1	First service females registered or non-registered
2-4	Code number for local organization of Michigan Artificial Breeders Association
5-6	Month of current service to the female
7-8	Day of current service to the female
9	Year of current service to the female
10-12	Code number of bull used on current service to the female
13-14	Service number of the female

15-16	Month of previous service to the female
17-18	Day of previous service to the female
19	Year of previous service to the female
20-22	Code number of bull used on previous service to the female
23	Automatic punch out.

(B) Placing Data on IBM Cards

IBM cards (Standard Card No. 2) were punched showing information from Michigan Artificial Breeders Cooperative, Inc. (MABCI) semen collection data sheets starting from the semen collection date of June 1, 1951 to May 31, 1952. There were some calculations made on this card. The data were punched in the columns indicated according to the following instructions:

Columns Punched	Description
1-3	Code number of bull used for semen collection; also punch high "X" in column 1 for bulls which are used for semen collection at less than 5 days apart, (encircled by blue pencil in MABCI semen collection data sheets)
4-5	The month of semen collection
6-7	The day of semen collection
8	The year of semen collection, that is punch the year 1951 as 1 and the year 1952 as 2
9	Name of the day coded as: 1 for Monday, 2 for Tuesday, 3 for Wednesday, 4 for Thursday, 5 for Friday, 6 for Saturday, 7 for Sunday
10	Season* of semen collection coded as: 1 for fall season which includes October, November and December

Columns Punched	Description
	2 for winter season which includes January, February and March
	3 for spring season which includes April, May and June
	4 for summer season which includes July, August and September
11-13	Volume of semen in milliliters
14-17	The concentration of semen per 1/1000 cubic millimeter
18-20	The dilution rate of semen, for example, 1 to 100, punch out the latter figure of the ratio only, that is 100
21-23	The total volume of semen after it has been diluted
24-25	The percentage of sperms alive in non-stored semen
26-27	The progressive motility of sperms of non-stored semen. Motility ratings are given in the semen collection data sheets as E, VG, G+, G Punch the figure 90 for E (Excellent) Punch the figure 80 for VG (Very Good) Punch the figure 70 for G+ (Good plus) Punch the figure 60 for G (Good)
28-29	The raw semen movement If there is a plus sign following the figure of percentage of sperms alive in the non-stored semen it means that there is maximum raw semen movement which is estimated to be 90%. If there is a diagonal line or letter M after the figure of the percentage of sperms alive in the non-stored semen it means that the raw semen movement is only intermediate and estimated to be 70%. If there is a dash mark after the figure of percentage of sperms alive in the non-stored semen, it means that the raw semen movement is very little or none and therefore it is estimated to be 0% In brief, for raw semen movement punch the figure: 90 for + 70 for/or for letter M 0 for -

Columns Punched	Description
30	One day old re-examined semen. Punch out the figure 1
31-32	The progressive motility of sperms of one day old re-examined semen. Progressive motility ratings are given in the semen collection data sheets as: G+, G, G-, F+, F, F-, P, Dead Punch the figure 70 for G+ (Good plus) Punch the figure 60 for G (Good) Punch the figure 50 for G- (Good minus) Punch the figure 40 for F+ (Fair plus) Punch the figure 30 for F (Fair) Punch the figure 20 for F- (Fair minus) Punch the figure 10 for P (Poor) Punch the figure 0 for Dead
33-34	The percent of sperms alive in one day old re-examined semen.
35	It is the three day old re-examined semen. Punch figure 3
36-37	The progressive motility rating of sperms of the three day old re-examined semen. Estimation (values) of motility ratings is similar to that in the columns 31-32
38-39	The percent of sperms alive in three day old re-examined semen
40	It is the four day old re-examined semen. Punch out figure 4
41-42	Progressive motility ratings of sperms of the four day old re-examined semen. Estimation of progressive motility ratings is similar to that in the columns 31-32
43-44	The percent of sperms alive in four day old re-examined semen
45-47	The concentration of sperms per 1/1000 cubic millimeter of diluted semen. This can be obtained by dividing the figures in columns 14-17 by the figures in columns 18-20 of this same card (Standard Card #2)

Columns Punched	Description
48-52	The volume of semen in milliliters squared. This can be obtained by squaring the figures in columns 11-13 of this card.
53-59	The concentration of raw semen per 1/1000 cubic millimeter squared. This can be obtained by squaring the figures in columns 14-17 of this card.
60-63	The percentage of sperms alive in non-stored semen squared. This figure can be obtained by squaring the figures in columns 24-25 of this card.
64-67	The progressive motility of sperms of non-stored semen squared. This can be obtained by squaring the figures in columns 26-27 of this card.
68-71	The movement of raw semen squared. This can be obtained by squaring the figures in columns 28-29 of this card.
72-77	The concentration of sperms per 1/1000 cubic millimeter of the diluted semen squared. This can be obtained by squaring columns 45-47 of this card.

(Columns 24-44: These data are given (written) under the heading of "Motility rating" in MABCI semen collection data sheets.)

Note: Forget the sign (+ or -) before the figure of percentage of sperms alive. For example, +70 means that the percentage of sperms alive is 70. Similarly, -70 means percentage of sperms alive is 70.

* In the present investigation, regrouping of months into seasons was done as follows:

Fall season includes September, October and November
Winter season includes December, January and February
Spring season includes March, April and May
Summer season includes June, July and August

(C) Compilation of Data from IBM Cards (Standard Card Nos. 1 and 2) for:

(a) Calculation of percent of non-returns

Preparation of "Current service" summary card: -

Steps:-

(1) For each date of semen collection (columns 4-8 of Standard Card No. 2) per bull, 5 master cards were made to represent five consecutive dates starting from the date of semen collection. On these master cards were punched the bull code number and the age of semen in days and the serial number. The date of semen collection is 1-day-old semen, whereas the next four consecutive dates represent 2, 3, 4, 5-day old semen, respectively. All the master cards were sorted by bull code number and by date of semen collection in order to arrange them by bull and by date of semen collection

(2) All IBM cards (Standard Card No. 1) having the date of current service from June 1, 1951 to June 4, 1952, were sorted by date of current service (columns 5-9 of Standard Card No. 1) and by bull used on current service (columns 10-12 of Standard Card No. 1) so as to arrange them by bull and by date of current service.

(3) IBM cards sorted in step 2 were collated with master cards arranged as mentioned in step 1, for two objectives: (i) to eliminate the current services corresponding to semen of a bull collected at less than 5 days apart, (ii) to eliminate the current services corresponding to two ejaculates from a bull which were not mixed but sent to inseminators separately.

(4) Only the IBM cards that matched in step 3 were used to cut "Current service" summary cards showing the bull used on current service and the date of current service. Age of semen

in days and the serial number were then transferred from master cards of step i, into "Current service" summary cards after collating by date of semen collection and bull code number.

Preparation of "Return service" summary card:-

(5) All IBM cards (Standard Card No. 1) having the date of previous service from June 1, 1951 to September 4, 1952, were sorted by date of previous service (columns 15-19 of Standard Card No. 1) and by bull used on previous service (columns 20-22 of Standard Card No. 1) so as to arrange them by bull used on previous service and by date of previous service. Later, they were collated with "Current service" summary cards mentioned in step 4 for two purposes: (i) to eliminate the breeding record corresponding to semen of a bull collected at less than 5 days apart, (ii) to eliminate the breeding record corresponding to two ejaculates from a bull which were not mixed but sent to inseminators separately.

(6) Ninety-day master cards were prepared as follows:-

A number from 1 through 462 was assigned for the date (current service and previous service) from June 1, 1951 to September 4, 1952. IBM cards were punched showing the date of current service, the date of previous service and the numbers assigned for the date of current service and for the date of previous service.

(7) IBM cards arranged by date of current service as mentioned in step 2 were matched with 90-day master cards and the number assigned corresponding to the date of current service was gang-punched on each card.

(8) IBM cards matched in step 5 were further collated with 90-day master cards and the number assigned corresponding to date of previous service was gang-punched on each card.

(9) The number corresponding to date of current service was subtracted from the number corresponding to date of previous service so as to get return period in days. The return period thus obtained was punched on each IBM card (Standard Card No. 1). These IBM cards were then sorted by return period in days in order to obtain 1 to 90-day returns for each bull for each date of semen collection. Then they were resorted by date of previous service (columns 15-19 of Standard Card No. 1) and by bull used on previous service (columns 20-22 of Standard Card No. 1). They were collated with the "Current service" summary cards and the matching cards were used to cut the "Return service" summary cards showing the bull used on previous service and the date of previous service.

Preparation of "Final" summary card:-

(10) The "Return service" summary cards were collated with the "Current service" summary cards and the returns were transferred to the corresponding "Current service" summary cards so as to get "Final" summary cards. The percent of non-returns

was calculated according to the following formula:-

$$\text{Percent of non-returns} = \frac{\text{Current services - Returns}}{\text{Current services}} \times 100$$

(11) The "Final" summary cards were used to arrange the breeding data conveniently in order to study the various phases of this investigation.

"Final" Summary Card

Columns punched	Description
1 - 3	Bull code number
4 - 5	Month of semen collection
6 - 7	Day of semen collection
8	Year of semen collection
9 - 10	Current first services to registered females
11 - 12	Returns of first services to registered females
13 - 15	Current first services to all females
16 - 18	Returns of first services to all females
19 - 21	Percent of non-returns for first services to all females
22 - 24	Current second services to all females
25 - 27	Returns of second services to all females
28 - 30	Percent of non-returns for second services to all females
31 - 32	Current third services to all females
33 - 34	Returns of third services to all females
35 - 37	Percent of non-returns for third services to all females
38 - 39	Current fourth services to all females
40 - 41	Returns of fourth services to all females
42 - 44	Percent of non-returns for fourth services to all females
45 - 46	Current fifth and over services to all females
47 - 48	Returns of fifth and over services to all females
49 - 51	Percent of non-returns for fifth and over services to all females
52 - 54	Total current services (1, 2, 3, 4, 5 and over) to all females
55 - 57	Total returns for 1, 2, 3, 4, 5 and over services to all females
58 - 60	Percent of non-returns for total current services (1, 2, 3, 4, 5 and over) to all females
61	Age of semen in days

62 - 64	Percent of non-returns for first services to registered females
76	Season of year
77	Day of week
78 - 80	Serial number

(b) Correlation between semen characteristics

IBM cards (Standard Card No. 2) were used to obtain ΣX, ΣX^2, ΣY, ΣY^2, ΣXY and N for various semen characteristics of Holstein, Guernsey, Jersey and Red Dane bulls.

(c) Correlation of semen characteristics and percent of non-returns

In view of the results reported by Ludwick et al. (1948) and Erb et al. (1950) and also because of the fact that about 83 percent of total services in the present investigation were to first and second service females, it was decided to use the semen samples each of which bred to forty or more females, in order to determine the correlation of semen characteristics with percent of non-returns. Accordingly, the "Final" summary cards were sorted on the number of total current services (1, 2, 3, 4, 5 and over service females) so as to eliminate cards (bulls) showing less than forty total current services.

IBM cards (Standard Card No. 2) and the "Final" summary cards having 40 or more total current services were collated so as to obtain ΣX, ΣX^2, ΣY, ΣY^2, ΣXY and N for each of the semen characteristics under study.

SOME OF THE FACTORS INFLUENCING PERCENT OF NON-RETURNS OR RATE OF CONCEPTION

(A) REVIEW OF LITERATURE

(a) Age of Semen in Relation to Percent of Non-returns

Milovanov (1932) reported that diluted semen could be stored for 6 hours with good results. According to Komarao and Gladcinova (1937) semen may be stored successfully at temperature of 15-25°C. for 12 hours, or at 8-12°C. for 24 hours. Hatziolos (1937) reported conception with semen stored up to 48 hours. Edwards and Walton (1938) also obtained conception with bull semen stored for as much as 57 hours. Altara and Adriano (1938) reported that the vitality of semen was unaffected after storing for 100 hours. Kust (1939) using 48-hour-old semen found that 60% of cows became pregnant. He also noted conception in some of the cows inseminated with 72-hour and 120-hour-old semen stored at low temperatures. Success with stored semen was also reported by Bonadonna (1939) who concluded that semen of cattle can be stored from 50 to 120 hours.

Phillips (1939) using egg-yolk phosphate diluter reported conception with 100-hour-old semen. Successful conception was also reported by Phillips and Lardy (1940) by using egg-yolk phosphate buffered bull semen stored for 150 to 180 hours. Herman et al. (1943) obtained successful results with semen having 70-90% motility before shipment and which was in transit from 48 to 130 hours. Out of 12 cows that were inseminated, 8 conceived. Vaidya and Bhattacharya (1945) reported conception in cows with semen stored for 6 days.

Attempts have been made by some of the research workers to determine the effect of length of semen-storage on the rate of conception or percent of non-returns. Herman and Ragsdale (1939) found that approximately one-third more services were required per conception with 4 to 5-hour-old dairy bull semen as compared with semen less than 1/2 to 2-hour-old.

Henderson (1939) using SGC-2 as diluter (Milovanov formula) observed no decrease in rate of conception (apparent pregnancy) with diluted semen stored up to 30 hours.

Burch (1939) reported that the use of good samples of stored semen for insemination of cows, 24 or more hours after collection, resulted in a conception rate which compared favorably with that obtained by the use of average fresh semen samples.

Margolin et al. (1943) observed a slight decrease in the percentage of conception (conception was ascertained by palpation or by calving) with the use of 1-day-old semen (61.9%) as compared to that obtained with fresh semen (63.7%).

Lasley and Bogart (1943) in a study on range cattle failed to store non-diluted semen successfully. However, with the use of egg-yolk-buffer, they were able to maintain a satisfactory level of fertility of diluted semen stored for 2 days. These investigators obtained 60% conception rate for non-stored semen and 57.1% conception rate for diluted semen stored for 24 hours and for 48 hours.

Underbjerg et al. (1942) in a study involving 931 cows reported that regardless of the kind of diluter used, the conception percentage (conception ascertained by palpation or by calving) obtained with the use of

semen stored from 24-177 hours was much less than that with the use of fresh semen samples (1-hour-old). The difference in conception rate between the stored and fresh semen samples was found to be highly significant.

Schultze et al. (1948) in a study with 25,146 first and second services from over 2,500 semen samples used in 13 artificial breeding locals at Nebraska found that the percent of non-returns decreased an average of 4.61 percent per day up to 4 days of storage of semen. The average decline in percent of non-returns from the 1st day to the 2nd day of storage of semen; from the 2nd to 3rd day; from 3rd to 4th day were 5.32, 4.63 and 4.54 percent, respectively. The percent of non-returns for the 2-year period of study for 1, 2, 3 and 4-day-old semen was found to be 63.21, 57.89, 53.26 and 48.72 percent, respectively. The Chi-square test showed a highly significant difference in the non-return rates resulting from the use of 1 to 4-day-old semen.

Decrease in conception rate with increase in the age of semen was also noted by Schaefer (1948) in a study of breeding records of NEPA Artificial Breeding Co-op, Tunkhannock, Pennsylvania. Schaefer reported that with the use of fresh, 1-day, 2-day, and 3-day-old semen, the percent non-returns obtained was 70, 66, 61 and 56%, respectively.

Erb et al. (1950) in a study involving 23,938 first and second services from bulls at Northwest Co-op Breeders (Mt. Vernon, Washington) and Evergreen Breeders (Chehalis, Washington) found that the average of percent of non-returns with 1-day-old semen was equal to that of 2-day-old semen (64%). However, a decrease in non-return rate was noted with 3-day-old semen (59%). These investigators reported an over-all percent of non-returns to be 63%.

Swanson (1951) analyzed the records of the East Tennessee Artificial Breeders' Association, Inc. during two management periods. In the first period when semen was collected in the early morning and no bactericidal substances added to semen, he obtained non-return rates of 63.3, 62 and 55.7% with 1-day-old (5 to 13-hour-old), 2 day-old (29 to 37-hour-old), and 3-day-old (53 to 61-hour-old) semen, respectively. Statistically, the difference in non-return rate between 1-day and 2-day-old semen was not significant. However, a highly significant difference was noted in non-return rate between 2-day and 3-day-old semen. During the second period of management, semen was collected in late afternoons and penicillin and streptomycin were added to the semen. The non-return rates obtained during this period with 1-day-old (18 to 26-hour-old) semen and with 2-day-old (42 to 50-hour-old) semen, were 61.5 and 58.2 percent, respectively. Statistically, the difference in non-return rate between 1 and 2-day-old semen was found to be highly significant.

Sweetman (1953) under Alaska conditions reported that the percentages of non-returns on 1st and 2nd services for 1 to 4-day-old semen were 58.7, 53.3, 48.1 and 45.3%, respectively.

Controversial results have been reported by various investigators regarding the effect of storage of semen on the conception rate. Willett et al. (1940) after a study involving more than 1,500 inseminations came to the conclusion that with proper precautions in handling the semen and diluent, the percent of non-returns resulting from diluted semen stored up to 4 days is as satisfactory as that obtained with semen used on the day of semen collection. Further investigations confirmed the same general

conclusion (Salisbury 1941). Similar results have also been reported by various investigators. Henderson (1946) stated that with good quality semen that is properly cooled and diluted, no significant decrease in fertilizing capacity should result up to 4 days of storage. Anderson (1945) reported that conception rate from the use of semen stored up to 72 hours was equal to that of fresh semen.

The literature reviewed above shows minor disagreement in the results obtained with the use of stored semen. The controversial results reported may be due to one or more factors affecting conception rate or percent of non-returns. Variation in potential fertility of semen, or variation in method of handling of semen might be responsible for the disagreement. Various factors have been reported to affect conception rate. Some of the important factors are:

(b) Relative Fertility of Bull

Bulls vary in fertility. White et al. (1925) in a study on 40 bulls noted a great variation in the fertility of bulls. The number of services required for conception varied from 1 to 3.18. A still larger variation was reported by Miller and Graves (1932) who observed that the number of services for a conception from the bulls varied from 1 to 5.85. Schultze et al. (1948) in an investigation to determine the effect of 1 to 4-day-old semen on percent of non-returns, found that bulls varied in the rate of decline of conception rate. However, a daily decline in fertility of semen from a group of bulls having more than 60% of non-returns did not differ significantly from that of semen obtained from group of bulls having less than 55% of non-returns. Swanson (1951) also noted a great variation

in the decline of breeding efficiency between bulls with age of semen. However, no significant difference in the decline of breeding efficiency with age of semen was found between the group of bulls having low breeding efficiency and the group of bulls having high breeding efficiency.

(c) Relative Fertility of Cow and Cow Families

According to Herman and Ragsdale (1946) cows are more to be blamed for lowered conception rate rather than the impotency of the bull. Erb et al. (1940) and Trimberger et al. (1945) noted a variation in the fertility (breeding efficiency) between cow families. Seath et al. (1943) reported a large variation in the breeding efficiency between cow families, ranging from a low of 40.6% to a high of 92.3% conception rate. These investigators, therefore, believed that breeding efficiency may be inherited.

(d) Breed of Animal

Schultze et al. (1948) found that the over-all percent of non-returns for Holstein, Guernsey and Jersey were 57.66, 56.14 and 50.72 percent, respectively. Lewis (1948b) reported that the percent of non-returns for the Holstein breed was 6.3% greater than that for the Guernsey breed. On the contrary, Erb et al. (1950) found no difference in percent of non-returns for Holstein (64%) and Guernsey (64%). However, the Jersey was found to have the lowest percent of non-returns (59%).

Mercier and Salisbury (1946) reported that the conception rate based on two months' non-returns was greater by 12.2 percent in the Holstein breed than the Guernsey breed. In an attempt to attribute this breed difference

to semen quality, these investigators found no statistically significant difference between breeds in semen volume, percent motile sperm and concentration. However, the breed difference in percentage of morphologically abnormal spermatozoa was highly significant. Contrary results were obtained in a later study by Mercier and Salisbury (1947) who observed that the Guernsey bulls had a slightly higher fertility (percent of non-returns) than the Holstein-Friesian bulls.

(e) Health of Animal

White et al. (1925) studied the influence of B. abortus infection and reported that 1.82 services were required per conception in the case of non-reactors whereas 2.09 services per conception were required in the case of the reactor cows. Fitch et al. (1929) reported 1.7 services per conception for a clean herd as against 2.4 services per conception in the herd infected with Bang's disease. According to Perry et al. (1952) severe abscesses and swelling, trauma (inflamed scrotum), epididymitis and hypoplasia adversely affect the breeding powers in bulls.

(f) Age of Animal

Gowen and Dove (1931) in a study involving 7,679 cows observed some decline in conception rate as age of the cows advanced. Conception rate was found to be 70% for the females under one year of age whereas it was 7% for cows of 17 years of age. A decline in the fertility of the bulls as age of bull advanced was also noted.

Miller et al. (1932) found that more services were required per conception to the heifers than to the cows. These investigators also noted

that more services were required by females for conception with mature bulls (above 6 years of age) than with young bulls (under 6 years of age). With mature bulls, 3.83 services were required whereas with young bulls, 2.28 services were required per conception.

Morgan and Davis (1938) reported that virgin heifers required more services per conception than cows from 2 to 9 (inclusive) years of age. However, little difference was observed in number of services required per conception in cows of agesfrom 2 to 12 (inclusive) years with the exception of 10-year old cows. They also reported that the conception rate was the highest with young bulls under 2 years of age. However, a very little difference was noted in the number of services per conception with bulls from 2 to 8 (inclusive) years of age. Further, in an analysis of the data on the mating of bulls and cows of varying ages these investigators concluded that the number of services required per conception was the smallest when 2-year-old bulls were mated with 2-year-old cows.

Bartlett and Perry (1939) reported that heifers required more services per conception than cows. They observed that a higher conception rate was generally obtained with young bulls. A tendency for young bulls settling the heifers at a higher rate than mature bulls was also noted.

Bowling, Putnam and Ross (1940) reported that heifers required a significantly greater number of services for conception than cows. They also found that less services per conception were needed by heifers when bred to bulls under 4 years of age as compared to older bulls. A gradual decline in the breeding efficiency of bulls with increasing age was also noted.

According to Erb, Wilbur and Hilton (1940) breeding efficiency was highest in 1 and 2-year-old bulls. Thereafter, a gradual decline in breeding efficiency with advancing age resulted.

Hilder et al. (1944) in a study on the Bureau of Dairy Industry herd at Beltsville, Md., observed that the number of services per conception was higher for heifers than for cows. These investigators also noted a gradual decline in the breeding efficiency of bulls with increasing age. However, the 7-year-old group was found to be an exception to this observation.

Tanabe and Salisbury (1946) studied the effect of age (1-12 years) of bull on conception rate and found that young bulls of the age group 1-3 years had the highest breeding efficiency.

(g) Month and Season of Breeding

Miller and Graves (1932) in a study of breeding records of 22 Holstein and Jersey bulls at Beltsville, Md., found that on an average the breeding efficiency (based on conception regardless of whether females reproduced normal live-calves) was lowest in July, August, September. This lowered breeding efficiency in summer months was attributed to high environmental temperature.

Morgan and Davis (1938) in a study of records of 5 dairy breeds at the University of Nebraska, reported that the percent of conceptions was the lowest in August (35.9%) and September (35.14%) but it was highest in December (51.61%). These investigators also noted a higher percent of conceptions in July.

Erb et al. (1940) in an examination of the records of the Purdue University dairy herd for a 20-year period found that breeding efficiency based on calving results was highest in May (74.3%) and lowest in August (58.2%).

Seath and Staples (1941) in an analysis of the records of the North Louisiana experimental (Bangs free) herd and the Louisiana State University herd located in South Louisiana reported that maximum number of services per conception were required during summer months. In the experimental herd, conception rate was highest during winter and second highest in fall, whereas, in the University herd, conception rate was highest in the fall and second highest conception rate was noted in winter season. Later, Seath, Staples and Neasham (1943) in a study of breeding records of the Louisiana State University dairy herd and the North Louisiana Experiment Station herd, reported that conception rate was the highest in fall and winter and lowest in spring and summer season.

Phillips et al.(1943) in an analysis of 1,135 matings from Beef Shorthorn and Milking Shorthorn bulls found that the breeding efficiency expressed as the percentage of fertile matings, was the highest in April (59.6%) and the lowest in August (40.8%). The breeding efficiency in summer season (July-September) was found to be the lowest of all seasons.

According to Hilder et al. (1944) the number of services per conception was much higher in July and August than other months of the year. It was also higher during September, February and March. During the fall season however, a sharp decline in the number of services per conception was observed. The lowest conception rate obtained during midsummer was attributed in part to high environmental temperatures.

Trimberger and Davis (1945) in an analysis of the breeding records for the Ayrshire, Guernsey, Jersey and the Holstein females of the University of Nebraska dairy herd found that of all the seasons, the conception rate as determined by actual pregnancy examinations per rectum was the lowest in summer. The number of inseminations required per conception was the highest in August and was highly significant.

Ellenberger et al. (1946) found little or no relationship of season of the year with conception rate under Vermont conditions.

Mercier and Salisbury (1946) reported a highly significant monthly variation in the fertility (percent of non-returns) of bulls of New York Artificial Breeders' Co-op, Inc.

Mercier and Salisbury (1947a) in a study of conception rate of cattle under natural breeding conditions in Eastern Canada found that the conception rate expressed as percent of successful services was the lowest during winter and spring and highest during summer and fall. The differences in the conception rate between seasons were significant. These investigators concluded that light (length of day light) is an important factor in the breeding efficiency of cattle.

Mercier and Salisbury (1947b) in an analysis of the artificial breeding records of about 125,000 cows and 71 bulls in New York State reported that the breeding efficiency expressed as percent of non-returns was the lowest in winter. A significant correlation between the breeding efficiency and the length of daylight was also reported. Effect of season on the breeding efficiency was more readily noticed in the younger (under 4 years of age) and older (over 8 years of age) cattle than in the mature cattle (4 to 8-year-old).

Schultze et al. (1948) of Nebraska State reported that the over-all conception rate expressed as percent of non-returns was the highest in June and was the lowest in August. Spring season was found to be the best for conception rate and summer season the poorest. The percent of non-returns for spring, winter, fall and summer seasons was found to be 59.36, 57.63, 56.90 and 52.48, respectively. In an analysis of the same data by age of semen (1 to 4 days), these investigators obtained no seasonal difference in the rate of decline of percent of non-returns.

Lewis (1948b) in a study of breeding records of Holstein and Guernsey bulls of Michigan Artificial Breeders' Association reported that the percentages of non-returns with these bulls were poorer in winter and summer. The percent of non-returns for Holstein breed was found to be highest in March and April (spring) whereas for Guernsey bulls, breeding efficiency was highest in October and November (fall season).

Erb et al. (1952) in an analysis of breeding records of Guernsey, Jersey and Holstein bulls used by Northwest Co-op Breeders, Mt. Vernon, Washington, reported that the average non-return rate was the lowest in January and highest in September. Breeding efficiency showed a definite pattern in that the percent of non-returns was lowest in January to April, followed by a gradual increase to the highest level in September, October and November.

Sweetman (1953) using dairy bull semen up to 80-hour-old under Alaska conditions, noted that the percent of non-returns on all 1st and 2nd services was the highest in fall (59.1%), whereas, it was the lowest in

winter (49.2%). The percent of non-returns for summer was 54.2% whereas it was 52.9% for spring.

(h) Time of Insemination in Relation to Time of Ovulation

Kufarev (1935) reported that the second half of estrus was the optimum time for insemination.

According to Kirillov (1937), in 60% of the cows that were bred at the beginning of heat, re-breeding was necessary whereas in only 26% of the cows of the same herd that were bred 18-24 hours after the beginning of heat, re-breeding was required.

Herman (1939) inseminated cows 4 to 12, 12 to 24, 24 to 48 and 48 to 60 hours following the first signs of heat and noted that the average number of inseminations required per conception was 1.72, 2.19, 2.13 and 2.70, respectively. Thus, a high breeding efficiency was observed when cows were bred during "active heat". Herman, therefore, concluded that for good results cows should be inseminated within 12 hours after the first signs of heat.

Bartlett and Perry (1939) obtained highest rate of conception when inseminations were done during full and late estrus. The periods of 8-12 hours and of 12-24 hours from the time of onset of heat were considered by these investigators as full and late estrus, respectively.

Trimberger and Davis (1943) in a study involving 295 dairy cows and heifers found that when females were inseminated at the middle of estrus or when bred artificially at the middle of estrus and rebred in 24 hours, the conception rate was much higher than when inseminated at the start of estrus or at 12 hours or more after the end of estrus.

Barrett and Casida (1946) observed a little difference in the percentage of conception when cows were inseminated from 3 to 25 hours from the onset of heat. On the contrary, when cows were inseminated less than 3 hours or when inseminated 25 or more hours after the onset of heat, lower conception rate resulted.

Trimberger (1948) found that when females were inseminated more than 6 hours but less than 24 hours before ovulation, very high rates of conception were obtained. But when the females were bred after ovulation, very low conception rates resulted.

Brewster et al. (1940) found that the average time of ovulation from the end of the estrus was 13.57 ± 0.63 hours. Heifers ovulated on an average 3.04 hours earlier than cows. No significant difference between the age of animal and the rate of sperm travel was noted. The minimum time required for sperm to reach the fallopian tube in mature cows was 5 hours and 30 minutes as compared to 4 hours and 15 minutes in heifers.

VanDemark et al. (1951) using a more refined technique found that the motile spermatozoa was transported from cervix to ovarian portion of oviduct in 2.5 minutes after insemination.

(i) Time of Breeding after Calving

In a study of 79 cows having fairly good breeding efficiency, Hofstad (1941) reported that when the cows were bred before the 60th day following parturition, the percentage of abortions, cases of metritis, dystocias and retained placentae were relatively high and conception rates were relatively low as compared with the cows bred after the 60th day of calving.

The results suggest that the breeding of the cow before the 60th day after calving should be discouraged.

VanDemark and Salisbury (1950) in a study involving 1,674 pregnancies (pregnancies confirmed by calving) found that on an average, when cows were first bred 1-20, 21-40, 41-60 and 61-80 days after calving, the percentages of services resulting in conceptions (reproductive efficiency) were 35.0, 41.0, 44.0 and 50.8 respectively. With a further increase in post-partum interval to 1st service, the average reproductive efficiency increased and reached the maximum (57.8%) when the cows were first bred 101-120 days after calving. Thereafter, the reproductive efficiency showed a decline and reached the level of 46.3% when the post-partum interval to first service was 201 or more days.

(j) Skill and Experience of Technicians and Inseminators

Perry and Bartlett (1945) have pointed out that the skill of the technician is an important factor in relation to conception rate. Dickensheet and Herman (1949) have quoted Peterson who reported that experience of the inseminators does have a great influence on the conception rate. According to Peterson, efficiency of the inseminators was found to reach the peak with an experience of 7 months.

(k) Herd Management

Herd management has been shown to affect conception rate. In the efficient herds, poor management has been reported to have required 1.90 services as compared to 1.74 services per conception in the satisfactorily managed herds (Dickensheet et al. 1949).

(1) Breeding Efficiency by Artificial vs. Natural Method

For the success of artificial breeding program the breeding efficiency obtained with artificial insemination must compare favorably with that by natural service.

The number of artificial services per conception reported by various investigators varies from about 1.6 to 2.07. According to Davis and Trimberger (1937), Swanson and Herman (1941b) two services by artificial breeding were required per conception. Cole and Winters (1939) reported that 1.77 artificial services were required per conception. Henderson (1939) required 1.91 inseminations per conception. Trimberger et al. (1945) reported that 1.95 inseminations per conception were required. These investigators also reported that 60.25% of the females conceived to the 1st services, 18.31 percent to the 2nd, 7.82 percent to the 3rd service, 4.23 percent to the 4th, 3.91 percent conceived with 5 or more services and 5.48 percent of the females were found to be sterile. Tanabe and Salisbury (1946) in a study of the records of the New York Artificial Breeders' Cooperative Association found that on the average 2.07 services were required per conception by artificial insemination. Schaefer (1948) in a study of breeding records of 23,141 cows reported that 1.53 services by artificial breeding were required per conception.

The number of natural services per conception reported by various investigators varies from about 1.5 to 2.2. Morgan and Davis (1938) found that on an average, females of all ages required 2.21 natural services per conception. In a study of 20 year period breeding data of Missouri Station herd, Herman (1939) reported that on the average 1.53 natural

services were required per conception. Later, in a study of breeding data of 2 year period, Herman found that 1.66 services were required per conception when half the herd was bred naturally. The other half herd which was bred artificially required 1.59 services per conception. Dokudovskii (1934) found a little variation in conception rate between the natural and artificial breeding. Erb et al. (1940) in a study of breeding records of Purdue University Dairy herd reported that 1.56 natural services were required per conception. Perry and Bartlett (1945) quoted Larsen (Veterinarian in Denmark) who reported that the rate of conception by artificial breeding (1.68) was as good as by natural breeding.

From the literature cited above, it can be said that the artificial breeding is as efficient as the natural breeding from the standpoint of breeding efficiency.

(B) EXPERIMENTAL PROCEDURE

The "Final" summary cards were used to arrange and tabulate the breeding data in order to study the influence of various factors on percent of non-returns (percent N.R.) as per outline mentioned below. The factors studied were age of semen, bull, breed, month and season of year, registered and non-registered females.

(1) Yearly percent of non-returns for total current services (1, 2, 3, 4, 5 and over) to all females, by bull and by: (a) age of semen (1, 2, 3, 4, 5 day-old), (b) irrespective of age of semen

(2) Yearly percent of non-returns for total current services (1, 2, 3, 4, 5 and over to all females, by breed and by: (a) age of semen (1, 2, 3, 4, 5 day-old), (b) irrespective of age of semen

(3) Yearly percent of total current services (1, 2, 3, 4, 5 and over) for each age of semen, by breed

(4) Yearly percent of non-returns for total current services (1, 2, 3, 4, 5 and over) to all dairy females regardless of breed, by: (a) age of semen (1, 2, 3, 4, 5 day-old), (b) irrespective of age of semen

(5) Percent of non-returns for total current services (1, 2, 3, 4, 5 and over) to all females, by breed, by month of breeding and by: (a) age of semen (1, 2, 3, 4, 5-day-old), (b) irrespective of age of semen

(6) Percent of non-returns for total current services (1, 2, 3, 4, 5 and over) to all females, by breed, by season of breeding and by: (a) age of semen (1, 2, 3, 4, 5-day old), (b) irrespective of age of semen

In the present investigation, the months were grouped into seasons as follows:

Fall season:	September, October and November
Winter season:	December, January and February
Spring season:	March, April and May
Summer season:	June, July and August

(7) Yearly percent of non-returns with 1-day-old semen for 1st service registered females and for 1st service non-registered females by bull (Holstein)

(8) Mean and the distribution (standard deviation) of total current services (1, 2, 3, 4, 5 and over), by breed, for each date of semen collection for a bull.

The results obtained in the present investigation were tested for significance at the 5% level of probability and at the 1% level of probability. In this manuscript, the 5% level of probability will be referred as 5% level and 1% level of probability will be referred as 1% level.

(C) RESULTS

Table I shows the yearly percent of non-returns on total current services (1, 2, 3, 4, 5 and over) by bull, and by: (a) age of semen, (b) irrespective of age of semen. From the Table, it can be seen that the yearly percent of non-returns for both dairy and beef bulls decreased as the age of semen advanced from 1 to 5 days. Few exceptions, however, were noted. For example, in bull 123 (code number), the percent of non-returns for 5-day old semen was 1% greater (49-48) than that for 4-day-old semen. This is more likely to be due to less number of total services with 5-day-old semen.

Bulls were found to vary widely in the amount of decline of semen fertility (percent of non-returns) for day-to-day storage of semen. In bull 126, the percent of non-returns declined 1% (71-70) from 1-day to 2-day-old semen, 5% from 2-day to 3-day-old semen, 11% from 3-day to 4-day-old semen and 1% from 4-day to 5-day-old semen. On the contrary, in bull 128, the percent of non-returns declined 10% from 1-day to 2-day-old semen, 10% from 2-day to 3-day-old semen, 7% from 3-day to 4-day-old semen and 3% from 4-day to 5-day-old semen. A wide variation was also noted between bulls in the average daily decline of fertility of semen

TABLE I

YEARLY PERCENT OF NON-RETURNS BY BULL AND BY AGE OF SEMEN

Bulls**	Age of Semen in Days										Total	
	1		2		3		4		5			
	Services Number	N.R.* %	Services Number	N.R. %	Services Number	N.R. %	Services Number	N.R. %	Services Number	N.R. %	Services Number	N.R. %
106	646	70	690	69	235	57	30	43	8	50	1,609	67
107	454	70	883	64	846	58	291	52	30	43	2,504	61
111	473	73	959	71	392	60	62	63	17	65	1,903	69
113	584	72	558	64	145	58	15	53	7	29	1,309	66
115	1,063	73	1,540	62	734	47	99	49	22	55	3,458	62
116	468	72	608	64	383	61	92	51	19	58	1,570	65
117	430	71	873	65	485	51	139	50	22	55	1,949	62
121	1,550	71	2,313	71	1,578	67	667	62	80	66	6,188	69
122	799	66	1,138	56	949	47	394	37	57	40	3,337	53
123	499	74	949	68	1,114	59	650	48	70	49	3,282	62
125	460	71	1,051	65	740	58	175	43	52	46	2,478	62
126	755	71	1,488	70	1,364	65	726	54	125	53	4,458	66
127	559	62	1,183	58	627	47	119	41	33	45	2,521	55
128	1,120	69	1,681	59	1,159	49	354	42	59	39	4,373	57
130	851	75	1,540	76	2,060	72	1,237	69	143	60	5,831	72
131	419	61	422	50	153	40	27	44	4	0	1,025	53
135	580	71	650	60	257	49	51	43	29	41	1,567	61
136	1,217	74	1,909	68	1,897	59	829	52	71	51	5,923	64
140	362	74	505	71	297	58	125	62	12	75	1,301	68
141	534	71	912	63	581	54	157	54	42	55	2,226	62
143	1,701	69	2,456	56	1,035	48	125	36	37	43	5,354	58
144	1,808	69	2,556	58	2,139	51	810	42	193	44	7,506	57
145	526	74	986	73	635	70	199	68	90	56	2,436	71
146	126	75	211	61	74	55	16	31	2	50	429	63
147	769	68	1,201	64	577	54	72	57	18	61	2,637	63
148	534	73	1,055	63	675	52	144	40	25	68	2,433	61
149	46	67	95	62	78	28	12	25	2	0	233	49
150	371	69	905	57	594	49	205	37	33	58	2,108	55
151	514	77	1,034	69	1,018	65	483	59	35	71	3,084	67
152	1,329	74	1,997	65	1,444	58	510	47	100	59	5,380	63
155	209	66	335	60	136	61	25	40	11	36	716	61
156	644	72	954	71	558	66	107	64	21	52	2,284	69
157	1,233	72	2,505	68	1,239	60	222	59	45	62	5,244	67
158	1,258	62	1,888	52	988	44	158	41	47	45	4,339	53
159	99	78	135	70	72	72	10	70	8	50	324	72
160	448	74	758	65	454	64	76	53	22	50	1,758	66
161	857	69	1,432	63	1,691	56	772	50	97	39	4,849	59
162	488	66	934	56	666	43	172	31	28	29	2,288	52
163	1,242	72	1,963	68	1,017	59	131	53	54	57	4,407	66
164	847	72	1,331	68	587	61	101	52	21	67	2,887	67
165	182	73	453	72	227	64	20	60	18	50	900	69
166	340	70	691	62	343	53	44	52	6	17	1,424	61
167	134	70	305	68	153	55	23	39	5	40	620	67
168	197	70	271	60	154	47	43	53	3	33	668	59
169	45	67	91	58	23	48	6	67	7	57	172	59
170	20	85	38	66	22	64	5	40	2	100	87	69
405	265	50	457	35	221	33	42	38	8	38	993	39
410	303	66	557	61	364	52	165	36	30	47	1,419	56

410	303	68	357	61	304	52	109	56	41	41	1,[illegible]	[illegible]
412	472	61	1,002	54	746	46	181	47	37	51	2,438	52
416	62	71	121	67	79	42	17	35	2	50	281	59
417	367	74	904	64	721	62	339	55	43	63	2,374	64
418	266	66	354	47	176	44	32	41	8	38	836	52
421	164	63	291	58	126	53	23	57	8	38	612	58
422	363	71	639	61	199	58	50	44	36	56	1,287	63
423	517	68	1,081	61	727	59	256	51	41	61	2,622	61
424	277	57	665	52	487	49	263	38	46	41	1,738	50
425	562	68	951	65	884	61	463	59	70	47	2,930	63
426	280	67	786	60	533	51	200	44	31	48	1,830	57
429	182	59	291	53	149	44	42	52	5	100	669	53
430	498	62	981	63	644	56	217	53	35	60	2,375	60
431	404	67	863	65	643	56	210	44	34	44	2,154	60
432	391	66	687	55	460	43	160	39	9	44	1,707	53
433	42	71	64	59	28	46	3	33	1	100	138	60
434	347	79	664	70	430	55	195	51	54	50	1,690	65
435	527	72	1,129	71	628	65	162	62	40	68	2,486	69
436	122	67	246	62	146	51	35	66	7	100	556	61
437	242	63	554	55	306	44	109	36	20	45	1,231	52
438	260	73	430	72	169	63	18	56	15	27	892	70
439	148	58	361	47	251	35	117	50	33	42	910	45
440	102	75	160	68	108	56	22	59	4	50	396	66
441	313	59	546	47	379	42	95	35	18	56	1,349	50
442	215	71	416	69	351	67	218	56	22	55	1,222	66
443	216	68	464	65	366	58	139	58	32	47	1,239	62
444	53	52	109	64	79	41	14	57	2	50	257	56
445	339	67	694	63	654	57	346	51	72	49	2,105	59
446	130	70	301	74	233	64	113	60	9	67	786	68
447	62	74	111	47	97	29	35	46	9	33	344	46
448	64	63	122	54	102	67	7	29	3	33	298	59
449	84	77	193	74	169	69	76	62	3	100	545	71
601	17	65	27	67	19	58	3	100	2	50	68	65
602	23	74	33	58	20	30	1	0	1	100	78	55
603	341	63	453	57	264	52	73	48	15	53	1,146	57
605	272	70	462	64	344	57	77	43	26	46	1,181	62
606	7	71	17	71	13	69	3	33	-	-	40	68
608	33	79	50	66	7	57	6	67	1	0	97	69
609	225	74	387	71	270	65	86	65	5	40	973	69
612	136	68	147	59	71	45	19	53	4	0	377	59
615	194	65	303	57	168	60	71	72	20	50	756	61
616	313	62	568	56	484	48	161	45	14	50	1,540	53
618	318	63	515	63	348	62	65	54	16	50	1,262	62
619	175	65	346	66	268	54	115	52	10	50	914	60
620	198	70	520	63	466	61	203	55	18	50	1,407	62
621	32	72	35	60	17	35	4	25	1	0	69	57
792	44	60	118	75	92	72	26	58	7	71	287	73
793	110	75	363	71	338	61	216	60	31	52	1,058	65
794	178	74	514	69	435	68	246	61	52	63	1,425	68
795	86	60	371	69	304	66	119	66	20	60	900	67
796	29	52	46	59	48	40	27	52	13	46	163	50
797	189	77	552	74	478	65	248	65	56	64	1,523	70
798	154	72	377	75	295	66	112	63	20	65	958	70
799	146	68	387	59	304	45	173	43	54	43	1,044	53
801	42	74	414	64	420	53	225	55	52	48	1,153	58
805	33	55	246	51	348	43	126	35	23	65	776	45
806	11	73	148	55	157	59	48	46	10	40	374	56
808	34	74	338	66	328	63	141	69	26	58	867	65
809	67	82	523	61	649	51	363	50	41	37	1,643	55
810	48	79	372	67	388	63	196	57	41	51	1,045	64
812	25	72	205	54	225	50	103	40	17	29	575	50
813	25	80	278	62	241	57	88	58	24	46	656	60

* Non-returns ** 100 Series: Holstein; 400 Guernsey; 600 Jersey; 800 Red Dane; 700 Angus.

(percent of non-returns). For example, the average daily decline of semen fertility for bull 126 was 4.5% $(\frac{71-53}{4})$, whereas, it was 7.5% for bull 161.

Bulls varied widely in the ability of semen to maintain fertility (percent of non-returns) during storage for 5 days. In bull 130, the yearly percent of non-returns for 1-day-old semen was 75%, whereas, it was 60% for 5-day-old semen. Thus, there was a drop of 15 percent of non-returns from 1-day to 5-day-old semen. On the other hand, in bull 161, the yearly percent of non-returns for 1-day-old semen was 69%, whereas, it was 39% for 5-day-old semen, resulting in a drop of 30 percent of non-returns from 1-day to 5-day-old semen.

There was a great variation in over-all (i.e. regardless of age of semen) yearly percent of non-returns between bulls in a breed and between bulls irrespective of breed. In Holstein breed, for example, the over-all percent of non-returns for bull 162 was 52%, whereas, it was 72% for bull 130. The over-all percent of non-returns for Holstein bull 145 was 71%, whereas, it was only 39% for the Guernsey bull 405.

Table II shows the yearly percent of non-returns for total current services (1, 2, 3, 4, 5 and over) by breed and by age of semen in days. In Figure 1 is presented, by breed, the effect of age of semen on yearly percent of non-returns based on total current services (1, 2, 3, 4, 5 and over). From the Table II and Figure 1, it is clear that the yearly percent of non-returns in Holstein, Jersey, Red Dane and Angus breed, declined for day-to-day storage of semen. The same was true in Guernsey breed except that the percent of non-returns with 5-day-old semen was slightly

TABLE II

YEARLY PERCENT OF NON-RETURNS BY BREED AND BY AGE OF SEMEN

Breed	Age of Semen in Days									
	1		2		3		4		5	
	Number Services	% N.R.	Number Services	% N.R.	Number Services	% N.R.	Number Services	% N.R.	Number Services	% N.R.
Holstein	29,790	70.52	48,432	64.12	32,595	57.13	10,730	51.96	1,832	51.31
Guernsey	8,641	67.02	17,224	60.57	11,695	54.06	4,364	50.00	785	51.34
Jersey	2,284	66.51	3,863	61.69	2,761	56.36	887	53.33	133	47.37
Red Dane	285	74.74	2,524	61.49	2,756	54.21	1,290	52.09	234	47.44
Angus	936	72.01	2,708	69.76	2,294	62.42	1,167	59.13	253	56.92

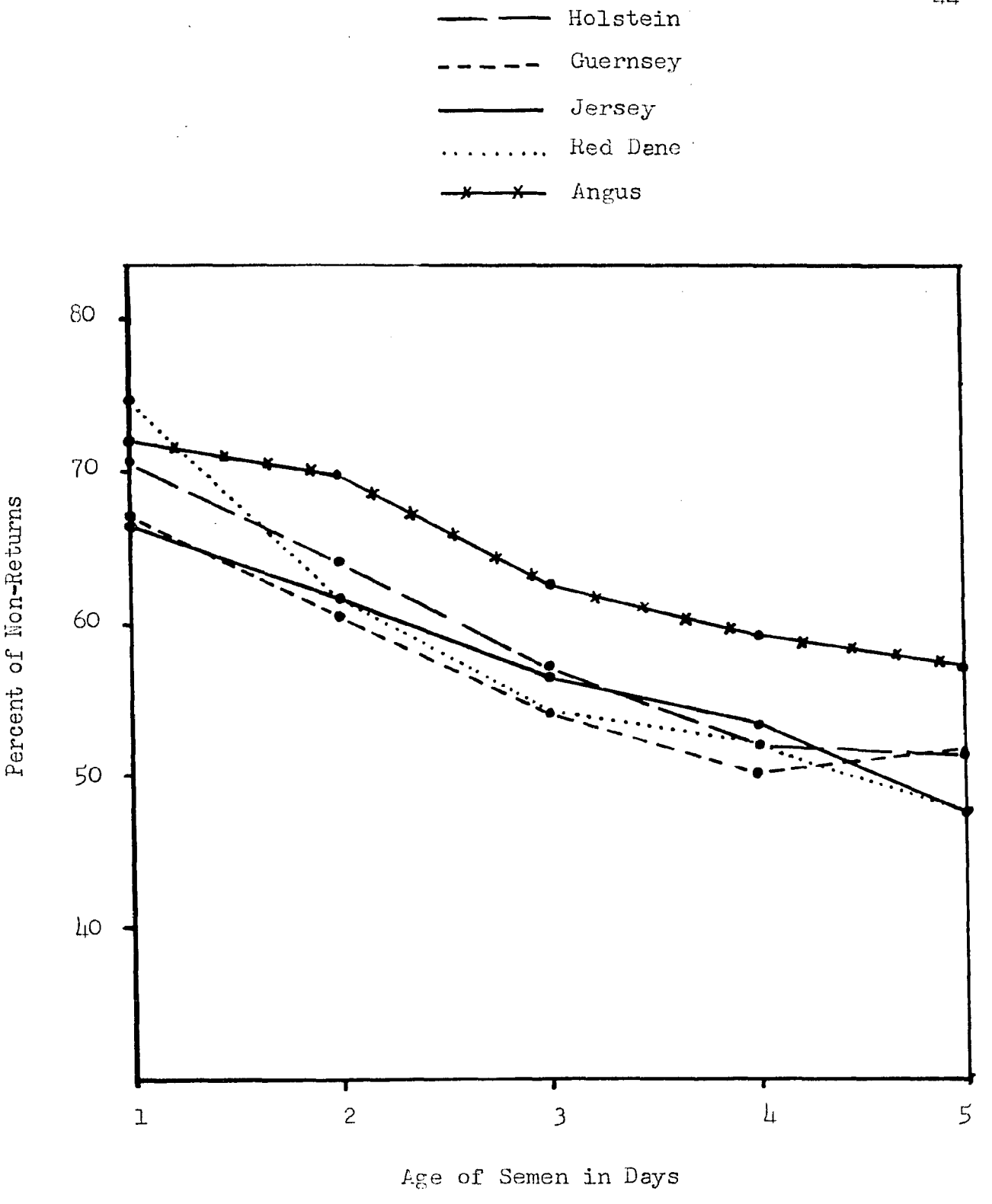


Fig. 1. The effect of age of semen on yearly percent of non-returns, by breed.

higher (51.34-50.00 = 1.34%) than that with 4-day-old semen. This difference of 1.34 percent, however, was not statistically significant (5% level).

In Holstein and Guernsey breeds, highly significant differences (1% level) were noted in the yearly percent of non-returns between 1 and 2-day, 2 and 3-day, and between 3 and 4-day-old semen, but not between 4 and 5-day-old semen. The difference in percent of non-returns between 4 and 5-day-old semen was not statistically significant (5% level). In Jersey and Red Dane breeds, highly significant differences (1% level) were noted in the yearly percent of non-returns between 1 and 2-day and between 2 and 3-day-old semen, but not between 3 and 4-day and between 4 and 5-day-old semen. The difference in percent of non-returns between 3 and 4-day and between 4 and 5-day-old semen was not statistically significant (5% level). In Angus breed, there was a highly significant difference (1% level) in yearly percent of non-returns between 2 and 3-day-old semen but not between 1 and 2-day, 3 and 4-day, and between 4 and 5-day-old semen. The percent of non-returns between 1 and 2-day, 3 and 4-day and between 4 and 5-day-old semen was not statistically significant (5% level).

From Table II it can be stated that there was a wide variation between breeds in the average daily decline of semen fertility (percent of non-returns) during storage of semen. The average daily decline of semen fertility up to 5 days of storage of semen was the highest ($\frac{74.74-47.44}{4}$=6.83%) in Red Dane, whereas, it was the lowest (3.77%) in Angus breed. In other words, the ability of semen to maintain fertility during the storage period up to 5 days was the highest in Angus breed and was the lowest in Red Dane.

Table III shows by breed, the difference in percent of non-returns between 1 and 2, 2 and 3, 3 and 4, and between 4 and 5-day-old semen. The percentages of non-returns are based on total current services (1, 2, 3, 4, 5 and over) for the period of one year. From the Table it can be stated that in each breed there was a variation in the amount of decline of percent of non-returns for day-to-day storage of semen. For example, in Red Dane, the percent of non-returns decreased 13.25% from 1 day to 2-day-old semen, 7.28% from 2 day to 3-day-old semen, 2.12% from 3 day to 4-day-old semen and 4.65% from 4 day to 5-day-old semen. A wide variation between breeds in the amount of decline of percent of non-returns for day-to-day storage of semen was also noted. In Jersey, the decline of percent of non-returns from 1 day to 2-day-old semen was 4.82% (66.51-61.69) and that from 2-day to 3- day-old semen was 5.33%. On the other hand, in Red Dane, the decline of percent of non-returns from 1-day to 2-day-old semen was 13.25% and that from 2-day to 3-day-old semen was 7.28%.

Table IV shows the yearly percent of non-returns for total current services (1, 2, 3, 4, 5 and over) by breed irrespective (regardless) of age of semen. From the Table, it is clear that the yearly percent of non-returns, regardless of age of semen, was the highest for Angus breed whereas it was the lowest for Red Dane. Holstein was the 2nd highest whereas Jersey ranked the third highest in yearly percent of non-returns. Statistical analysis revealed highly significant differences (1% level) in yearly percent of non-returns between Angus breed and Holstein, Angus and Jersey, Angus and Guernsey and between Angus and Red Dane. Highly significant differences (1% level) in percent of non-returns were also

TABLE III

DIFFERENCE IN PERCENT OF NON-RETURNS BETWEEN 1 AND 2, 2 AND 3, 3 AND 4, 4 AND 5-DAY-OLD SEMEN (BY BREED)

Breed	Age of Semen in Days			
	1 - 2 % N. R.	2 - 3 % N. R.	3 - 4 % N. R.	4 - 5 % N. R.
Holstein	-6.40	-6.99	-5.17	-0.65
Guernsey	-6.45	-6.51	-4.06	+1.34
Jersey	-4.82	-5.33	-3.03	-5.96
Red Dane	-13.25	-7.28	-2.12	-4.65
Angus	-2.25	-7.34	-3.29	-2.21

TABLE IV

YEARLY PRECENT OF NON-RETURNS BY BREED REGARDLESS OF AGE OF SEMEN

Breed									
Holstein		Guernsey		Jersey		Red Dane		Angus	
Services	N.R.	Services	N.R.	Services	N.R.	Services	N.R.	Services	N.R.
Number	%	Number	%	Number	%	Number	%	Number	%
123,379	62.57	42,709	58.84	9,928	60.37	7,089	57.02	7,358	65.63

noted between Holstein and Jersey, Holstein and Guernsey and between Holstein and Red Dane. Similarly, the difference in percent of non-returns between Jersey and Guernsey and between Jersey and Red Dane was highly significant (1% level). There was also a highly significant difference (1% level) in the yearly percent of non-returns between Guernsey and Red Dane breeds.

Table V shows by breed, the yearly percent of total services (1, 2, 3, 4, 5 and over) for each age of semen. From the Table it can be stated that the yearly percent of total services for each age of semen in Holstein and Jersey breeds was almost the same. In Guernsey, the percent of total services for each age of semen was nearly the same as in Holstein and Jersey except that for 1-day-old semen, it was about 3 - 4% less. In Angus and particularly in Red Dane, the percent of total services for 1-day-old semen was markedly low as compared to other breeds. The percent of total services for 2-day-old semen in these breeds was also low as compared to other breeds.

Table VI shows the yearly percent of non-returns by age of semen in days, regardless of breed. Data for Angus breed are not included in Table VI. The percentages of non-returns are based on total current services (1, 2, 3, 4, 5 and over) to all females. From the Table it is clear that the percent of non-returns decreased as age of semen advanced. The decline in percent of non-returns from 1 day to 2-day-old semen, 2 day to 3-day-old semen, 3 day to 4-day-old semen, and from 4 day to 5-day-old semen was 8.54, 6.85, 4.66 and 0.70 percent, respectively. Statistical analysis revealed a highly significant difference (1% level) in the percent of

TABLE V

YEARLY PERCENT OF TOTAL SERVICES BY BREED FOR EACH AGE OF SEMEN

Breed	Age of Semen in Days									
	1		2		3		4		5	
	Services (Number)	Percent of Total Services	Services (Number)	Percent of Total Services	Services (Number)	Percent of Total Services	Services (Number)	Percent of Total Services	Services (Number)	Percent of Total Services
Holstein	29,790	24.15	48,432	39.26	32,595	26.42	10,730	8.70	1,832	1.49
Guernsey	8,641	20.23	17,224	40.33	11,695	27.38	4,364	10.22	785	1.84
Jersey	2,284	23.01	3,863	38.91	2,761	27.81	887	8.93	133	1.34
Red Dane	285	4.02	2,524	35.60	2,756	38.88	1,290	18.20	234	3.30
Angus	936	12.72	2,708	36.80	2,294	31.18	1,167	15.86	253	3.44

TABLE VI

YEARLY PERCENT OF NON-RETURNS BY AGE OF SEMEN REGARDLESS OF BREED (DAIRY)

Age of Semen in Days									
1		2		3		4		5	
Services	N.R.	Services	N.R.	Services	N.R.	Services	N.R.	Services	N.R.
Number	%	Number	%	Number	%	Number	%	Number	%
41,000	69.59	72,043	63.05	49,807	56.20	17,271	51.54	2,984	50.84

non-returns between 1-day and 2-day-old, 2 day and 3-day-old, and between 3-day and 4-day-old semen. The difference in percent of non-returns between 4-day and 5-day-old semen was not statistically significant (5% level).

The over-all yearly percent of non-returns on total current services (1, 2, 3, 4, 5 and over), for dairy bulls under study was 61.4%, whereas, it was 61.53% for all bulls, including beef bulls.

Table VII shows by breed, the percent of non-returns on total current services (1, 2, 3, 4, 5 and over), by month of breeding, regardless of age of semen. From the Table, it can be seen that for Holstein breed, the percent of non-returns was the highest in October. The percent of non-returns was also high in September as compared to other months. May was found to be the month of lowest percent of non-returns for Holstein. Statistical analysis showed a highly significant difference (1% level) in percent of non-returns between October and May.

In Guernsey breed, the percent of non-returns was the highest in September. It was also high in April and October as compared to other months. The percent of non-returns was lowest in August. The difference in percent of non-returns between September and August was highly significant (1% level).

In Jersey breed, the percent of non-returns was highest in April and was also higher in October and September as compared to other months. The percent of non-returns was lowest in August. The difference in percent of non-returns between April and August was highly significant (1% level).

TABLE VII

PERCENT OF NON-RETURNS BY BREED BY MONTH REGARDLESS OF AGE OF SEMEN

Month of Breeding	Breed									
	Holstein		Guernsey		Jersey		Red Dane		Angus	
	Number Services	% N.R.	Number Services	% N.R.	Number Services	% N.R.	Number Services	% N.R.	Number Services	% N.R.
January	12,763	60.46	3,893	59.36	1,088	61.21	571	54.29	911	62.13
February	11,478	62.07	3,967	57.95	1,146	57.59	726	58.40	923	65.55
March	11,892	63.26	3,455	59.04	769	58.52	651	57.30	955	68.90
April	11,486	62.81	3,206	61.29	932	66.52	628	62.10	770	75.19
May	13,518	60.25	4,546	58.01	1,146	57.16	891	54.88	937	67.66
June	12,159	62.27	5,199	58.80	1,000	58.30	829	57.06	166	60.24
July	10,219	62.45	4,060	56.65	882	58.50	799	56.95	232	64.66
August	8,183	63.16	3,471	56.38	601	55.57	336	52.08	372	63.44
September	7,280	65.49	2,995	61.44	604	65.40	419	56.80	206	57.77
October	7,497	66.97	2,519	60.90	600	66.50	406	64.29	406	62.81
November	7,286	63.18	2,456	59.57	560	64.11	310	54.19	549	64.48
December	9,618	62.05	2,942	58.74	600	59.50	523	54.68	931	61.55

In the Red Dane breed, the percent of non-returns was the highest in October and was also high in April compared to other months. The percent of non-returns was the lowest in August. The difference in percent of non-returns between October and August was highly significant (1% level).

For the Angus breed, the percent of non-returns was the highest in April and was the lowest in September. The difference in percent of non-returns between April and September was highly significant (1% level).

Table VIII shows the difference in percent of non-returns between 1 and 2 day, 2 and 3 day, 3 and 4-day-old semen for the month of highest (October) and lowest (May) breeding efficiency (percent of non-returns) in the Holstein breed. The percentages of non-returns are based on total current services (1, 2, 3, 4, 5 and over). The number of total current services for 5-day-old semen was less and hence the amount of decline in percent of non-returns from 4-day to 5-day-old semen is not shown in Table VIII.

From the examination of Table VIII, it can be stated that the amount of decline in percent of non-returns from 1 day to 2-day-old semen was 4.82% (8.33 - 3.51) greater in May than that in October. However, the amount of decline in percent of non-returns from 2 day to 3-day-old semen in October and May was about equal. Similarly, the amount of decline in percent of non-returns from 3 day to 4-day-old semen in October and May was nearly the same.

Table IX shows by breed, the percent of non-returns on total current services (1, 2, 3, 4, 5 and over), by season of breeding, regardless of age of semen.

TABLE VIII

DIFFERENCE IN PERCENT OF NON-RETURNS BETWEEN 1 AND 2, 2 AND 3, 3 AND 4-DAY-OLD SEMEN FOR MONTH OF HIGHEST AND LOWEST BREEDING EFFICIENCY IN HOLSTEIN BREED

Month of Breeding	Age of Semen in Days		
	1 - 2 % N. R.	2 - 3 % N. R.	3 - 4 % N. R.
October	-3.51	-6.70	-5.46
May	-8.33	-6.46	-4.86

TABLE IX

PERCENT OF NON-RETURNS BY BREED BY SEASON REGARDLESS OF AGE OF SEMEN

Season of Breeding	Breed									
	Holstein		Guernsey		Jersey		Red Dane		Angus	
	Number Services	% N.R.	Number Services	% N.R.	Number Services	% N.R.	Number Services	% N.R.	Number Services	% N.R.
Fall	22,063	65.23	7,970	60.69	1,764	65.36	1,135	58.77	1,161	62.70
Winter	33,859	61.45	10,802	58.67	2,834	59.39	1,820	56.04	2,765	63.07
Spring	36,896	62.01	11,207	59.27	2,847	60.59	2,170	57.70	2,662	70.29
Summer	30,561	62.57	12,730	57.45	2,483	57.71	1,964	56.16	770	63.12

In Figure 2 is shown by breed, the effect of season of breeding on the percent of non-returns (regardless of age of semen) based on total current services (1, 2, 3, 4, 5 and over).

From Table IX and Figure 2, it is clear that for Holstein breed, the percent of non-returns was the highest in fall and was the lowest in winter season. Statistical analysis of the data revealed that the difference in percent of non-returns between these two seasons was highly significant (1% level). Highly significant differences (1% level) in percent of non-returns were also noted between winter and summer and between fall and spring seasons.

In the Guernsey breed, the percent of non-returns was the highest in the fall and was the lowest in summer. Statistically, the difference in percent of non-returns between the fall and the summer was highly significant (1% level). There was no significant difference (5% level) in percent of non-returns between winter and summer.

In the Jersey breed, the percent of non-returns was the highest in fall and was the lowest in summer. The difference in percent of non-returns between fall and summer was highly significant (1% level).

In the Red Dane breed, the percent of non-returns was the highest in fall and was the lowest in winter season. However, the difference in percent of non-returns between fall and winter was not statistically significant (5% level).

In the Angus breed, the percent of non-returns was the highest in spring and was the lowest in fall. The difference in percent of non-returns between spring and fall was highly significant (1% level). There were no

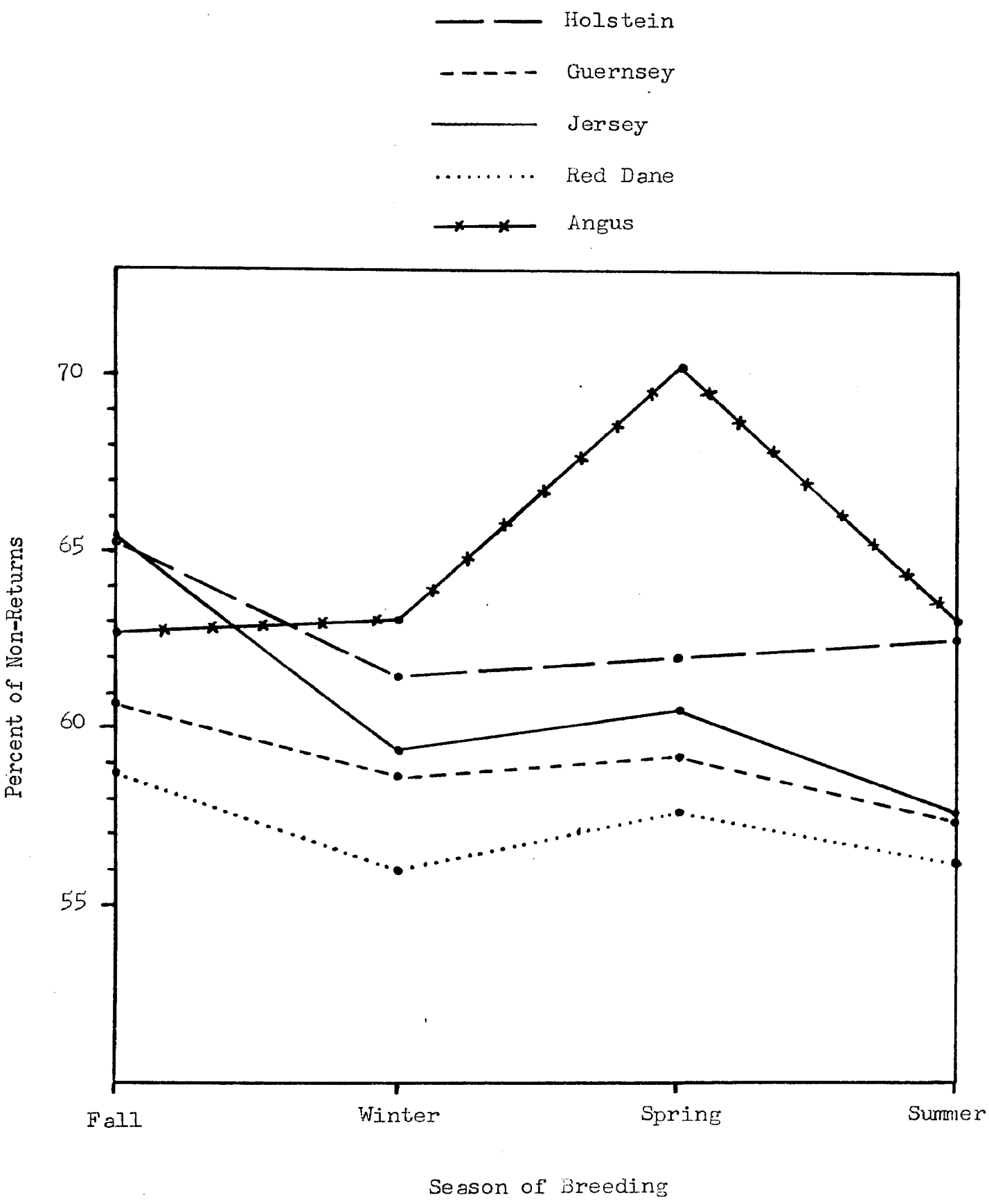


Fig. 2. The effect of season of breeding on the percent of non-returns (regardless of age of semen).

significant differences (5% level) in percent of non-returns between summer and fall and between winter and fall.

Table X shows the difference in percent of non-returns between 1 and 2-day, 2 and 3-day and between 3 and 4-day-old semen for the season of highest (fall season in both the breeds) and lowest (winter in Holstein, summer in Guernsey) breeding efficiency (percent of non-returns) in Holstein and in Guernsey breed. The percentages of non-returns are based on total current services (1, 2, 3, 4, 5 and over). The number of total current services for 5-day-old semen was less and hence the amount of decline in percent of non-returns from 4 day to 5-day-old semen is not shown in Table X.

From the examination of Table X, it can be stated that for Holstein the amount of decline in percent of non-returns from 1-day to 2-day-old semen was 0.45% (6.14-5.69) greater in winter season than that in fall season. Similarly, the decline in percent of non-returns from 2-day to 3-day-old semen was 0.73% greater in winter, whereas, the decline in percent of non-returns from 3-day to 4-day-old semen was 2.26% greater in winter than that in fall season.

For Guernsey breed, the amount of decline of percent of non-returns from 1-day to 2-day-old semen was 0.93% greater in summer season than that in fall season. Similarly, the decline in percent of non-returns from 3-day to 4-day-old semen was 3.82% greater in summer than that in fall. On the other hand, the amount of decline in percent of non-returns from 2-day to 3-day-old semen was 2.79% less in summer than that in fall season.

TABLE X

DIFFERENCE IN PERCENT OF NON-RETURNS BETWEEN 1 AND 2, 2 AND 3, 3 AND 4-DAY-OLD SEMEN FOR SEASON OF HIGHEST AND LOWEST BREEDING EFFICIENCY

Season of Breeding	Age of Semen in Days		
	1 - 2 % N. R.	2 - 3 % N. R.	3 - 4 % N. R.
		(A) Holstein Breed	
Fall	-5.69	-6.54	-3.11
Winter	-6.14	-7.27	-5.37
		(B) Guernsey Breed	
Fall	-5.60	-8.64	-2.20
Summer	-6.53	-5.85	-6.02

Table XI shows the yearly percent of non-returns based on first current services for 1-day-old semen in the registered and non-registered females.

Statistical analysis of the data presented in Table XI, showed a highly significant difference (1% level) in the percent of non-returns between registered and non-registered females in favor of registered females. The weighted average of percent of non-returns with 1-day-old semen was 10.9% (79.36 - 68.46) greater for the registered females than for the non-registered females.

In Table XII are presented the mean and distribution (standard deviation) of total current services (1, 2, 3, 4, 5 and over), by breed, for each date of semen collection for a bull. The mean of total current services for each date of semen collection for a bull was the highest in Holstein breed and was the lowest in Jersey breed.

(D) DISCUSSION

Willett et al. (1940) reported that with proper precautions in handling the semen and diluent, the percent of non-returns resulting from diluted semen stored up to 4 days is as satisfactory as that obtained with semen used on the day of semen collection. According to Henderson (1946) with good quality semen that is properly cooled and diluted, no significant decrease in fertilizing capacity should result up to 4 days of storage. Anderson (1945) reported that conception rate from the use of semen stored up to 72 hours was equal to that of fresh semen.

TABLE XI

YEARLY PERCENT OF NON-RETURNS FOR 1-DAY-OLD SEMEN
IN REGISTERED AND NON-REGISTERED FEMALES

Bull	Registered Females		Non-Registered Females	
	Services Number	N.R. %	Services Number	N.R. %
106	83	81	325	69
107	26	73	255	71
111	35	83	237	74
113	42	83	367	71
115	81	78	583	72
116	52	85	230	69
117	30	73	229	69
121	171	79	811	71
122	78	76	424	61
123	33	67	267	81
126	37	84	378	69
127	29	62	300	59
128	105	77	581	67
130	29	76	430	72
131	53	81	217	53
135	79	70	286	72
136	111	74	615	70
140	31	68	172	78
141	27	89	326	70
143	151	81	918	68
144	181	84	1,009	66
145	47	77	254	69
147	50	68	423	70
151	32	75	260	73
152	130	85	747	70
156	46	91	338	67
157	86	80	684	70
158	91	66	701	61
160	40	83	243	70
161	53	87	483	67
163	100	88	709	67
164	74	89	440	67
Weighted Average	-	79.36	-	68.46

TABLE XII

MEAN AND DISTRIBUTION OF TOTAL CURRENT SERVICES FOR EACH DATE OF SEMEN COLLECTION FOR A BULL
(BY BREED)

	Breed				
	Holstein	Guernsey	Jersey	Red Dane	Angus
N*	1302	756	247	142	123
Mean	95	57	41	50	59
Std. Deviation	47	31	20	19	26

* Number of items or frequency.

Erb et al. (1950) reported that the average of percent of non-returns with 1-day-old semen was equal to that of 2-day-old semen.

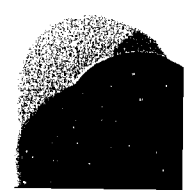
The results obtained in the present investigation do not agree with the results reported (mentioned above) by Willett et al. (1940), Henderson (1946), Anderson (1945) and by Erb et al. (1950). Our results confirm the findings of Schultze et al. (1948), Schaefer (1948), and those of Sweetman (1953) in that the over-all percent of non-returns decreased as age of semen advanced. However, the amount of decline in percent of non-returns for day-to-day storage of semen was somewhat higher than that reported by these investigators.

In the present study, a great variation in the fertility (percent of non-returns) of bulls was observed. The result agrees with that reported by White et al. (1925) and Miller and Graves (1932). Bulls were found to vary widely in the amount of decline of semen fertility (percent of non-returns) for day-to-day storage of semen. The result is in agreement with that reported by Schultze et al. (1948) and Swanson (1951). From the data presented in Table I, it can be seen that in bull 126, the amount of decline in percent of non-returns (fertility) from 1-day-old to 2-day-old semen, 2-day-old to 3-day-old, 3-day-old to 4-day-old semen was 1, 5 and 11 percent, respectively. In bull 128, the amount of decline in percent of non-returns from 1-day-old to 2-day-old semen, from 2-day-old to 3-day-old semen was 10 and 10 percent, respectively. The results quoted above suggest that in bull 126 or in bulls showing similar decline of percent of non-returns for day-to-day storage of semen, efforts should be made not to use semen which is more than 2 days or 3 days old. On the

other hand, in bull 128 or in bulls showing similar decline of percent of non-returns for day-to-day storage of semen, efforts should be made to use semen as fresh as possible in order to secure higher percent of non-returns.

Schultze et al. (1948) reported that the over-all percent of non-returns for Guernsey breed was greater than that for Jersey breed. Erb et al. (1950) found no difference in percent of non-returns for Holstein and Guernsey. However, Jersey was found to have the lowest percent of non-returns. Mercier and Salisbury (1947) reported that the Guernsey bulls had a slightly higher fertility (percent of non-returns) than the Holstein-Friesian bulls. The results obtained in the present investigation do not agree with the results reported by Schultze et al. (1948), Erb et al. (1950) and Mercier and Salisbury (1947). On the contrary, our results confirm the findings of Mercier and Salisbury (1946) and Lewis (1948b) in that the percent of non-returns for Holstein breed was greater than that for the Guernsey breed. However, the difference in percent of non-returns between Holstein and Guernsey was much less than that reported by these investigators.

The yearly over-all (regardless of age of semen) percent of non-returns for Red Dane breed, in the present study, was the lowest of all the breeds (dairy and beef type) even though the percent of non-returns with 1-day-old semen was the highest in Red Dane. This might be attributed to some extent to the fact that the percent of total current services for 1-day and for 2-day-old semen was the lowest in Red Dane as compared to any other breed under study. Moreover, our results, presented in Table III,



showed that the amount of decline of yearly percent of non-returns from 1-day-old semen to 2-day-old semen was the largest (13.25%) in Red Dane as compared to other breeds. Hence, in order to obtain higher percent of non-returns, efforts should be made in Red Dane breed in particular, to use the semen as fresh as possible for breeding the females.

In Angus breed, the amount of decline in semen fertility (percent of non-returns) from 1-day-old semen to 2-day-old semen was 2.25% whereas the amount of decline in percent of non-returns from 2-day-old semen to 3-day-old semen was 7.34%. The results suggest that much higher conception rate (percent of non-returns) with Angus bulls could be secured by using semen which is 2-day-old or less than 2-day-old.

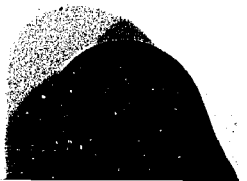
Of all the breeds under study, the breeding efficiency (percent of non-returns) was the highest in Angus breed. This, to some extent, might be due to greater ability of Angus breed compared to other breeds, to maintain the fertility of semen (percent of non-returns) during storage for 4 or 5 days (Table III).

Lewis (1948a) noted that the breeding efficiency (percent of non-returns) in Holstein bulls was the highest in March and April and was the lowest in December. In Guernsey, the percent of non-returns was the highest in November and lowest in July. The percent of non-returns in Guernsey was also high in October and March. Our results for Holstein breed showed that the percent of non-returns regardless of age of semen, was the highest in October and was the lowest in May. In Guernsey breed, the percent of non-returns was the highest in September and was the lowest in August.

Lewis (1948a) found that the percent of non-returns for Holstein breed was much higher during spring season as compared to other seasons. The percent of non-returns was the lowest in summer. However, the percentages of non-returns in summer (64.4%) and in winter (64.5%) for Holstein were almost equal. In Guernsey, the percent of non-returns was the highest in fall and was the lowest in winter. However, the percentages of non-returns in winter (57.8%) and in summer (57.9%) were almost equal.

In the present study, the percent of non-returns (regardless of age of semen) for the Holstein breed was the highest in fall and was the lowest in winter. The difference in percent of non-returns between winter and summer seasons was highly significant (1% level). A highly significant difference (1% level) in the percent of non-returns was also noted between fall and spring seasons in Holstein. In Guernsey breed, the percent of non-returns (regardless of age of semen) was the highest in fall and was the lowest in summer. However, the difference in percent of non-returns between summer and winter seasons was not statistically significant (5% level).

In the present investigation, the weighted average of percent of non-returns with 1-day-old semen was 10.9% greater for registered females than for non-registered females. Statistical analysis revealed a highly significant difference (1% level) in the percent of non-returns between registered and non-registered females. The higher percent of non-returns in registered females might be due to better management of the registered females. Poor management of the herds has been reported to affect adversely the conception rate. (Dickensheet et al. 1949).



Attempt was made to find out whether the difference in percent of non-returns between registered and non-registered females with 1-day-old semen was due, in part, to more use of fresh semen in the registered females. The breeding data showed that the percent of total services with 1-day-old semen was 26.44% in registered females as against 24.98% in the non-registered females. Thus, the use of 1-day-old semen in the registered females was somewhat greater than that in the non-registered females. However, the difference in percent of total services with 1-day-old semen in registered and non-registered females was not statistically significant (5% level). Similarly, the percentages of total services with 2-day-old semen in registered (39.35%) and in non-registered females (39.43%) were almost equal. The results indicate that factors other than age of semen, such as better feeding and better management (care, disease control, etc.) might be responsible for the higher percent of non-returns in registered females as compared to that in non-registered females.

(E) SUMMARY AND CONCLUSIONS

The breeding records of dairy and beef bulls of Michigan Artificial Breeders Association were analyzed starting from the current service date of June 1, 1951 to June 4, 1952, in order to study the influence of various factors on percent of non-returns. The factors studied were age of semen, bull, breed, month and season of breeding, registered and non-registered females in relation to percent of non-returns. The percentages

of non-returns were based on 90-day non-returns of all females with 1, 2, 3, 4, 5 and over services except in the study of registered and non-registered females, the percentages of non-returns were based on 1st service females owing to unavailability of data for 2, 3, 4 5 and over services to registered females. The investigation led to the following findings and conclusions:

(1) Influence of Bull

In general, the yearly percent of non-returns for both dairy and beef bulls decreased as age of semen advanced from 1 to 5 days. Hence, semen should be used as fresh as possible in order to secure higher percent of non-returns.

Bulls varied widely in the amount of decline of percent of non-returns for day-to-day storage of semen. A wide variation was also noted between bulls in the average daily decline of percent of non-returns up to 5 days of storage of semen. There was a great variation in over-all (i. e., regardless of age of semen) yearly percent of non-returns between bulls within a breed.

(2) Influence of Breed

The yearly percent of non-returns in Holstein, Jersey, Red Dane, and Angus breed, declined as the age of semen advanced from 1 to 5 days. Similar finding was also noted in Guernsey breed except that the percent of non-returns for 5-day-old semen was slightly higher (statistically not significant) than that for 4-day-old semen.

There was a wide variation between breeds in the average daily decline of percent of non-returns during storage of semen for 5 days. The

average daily decline of percent of non-returns up to 5 days of semen storage was the highest in Red Dane, whereas, it was the lowest in Angus breed. In each breed there was a variation in the amount of decline of percent of non-returns for day-to-day storage of semen. A wide variation between breeds in the amount of decline of percent of non-returns for day-to-day storage of semen was also noted. The decline of percent of non-returns for 1-day to 2-day old semen was the highest in Red Dane whereas it was the lowest in Angus. Of all the breeds under study, the yearly percent of non-returns, regardless of age of semen, was the highest for Angus breed and the lowest for Red Dane. Holstein ranked the second highest in yearly percent of non-returns, whereas Jersey ranked the third.

(3) Influence of Percent of Total Services for Each Age of Semen

In Red Dane, the percent of total services for 1-day old semen was markedly low compared to other breeds. This might, in part, explain the lowest percent of non-returns observed in Red Dane.

(4) Over-all Yearly Percent of Non-returns Regardless of Breed

The over-all yearly percent of non-returns on total current services (1, 2, 3, 4, 5 and over), for all bulls under study was 61.5%.

(5) Influence of Month of Breeding

For Holstein and Red Dane breeds, the percent of non-returns was the highest in October. For Jersey and Angus, the percent of non-returns was the highest in April whereas for Guernsey, the percent of non-returns was the highest in September.

For Holstein, the percent of non-returns was the lowest in May. For Guernsey, Jersey and Red Dane, the percent of non-returns was the lowest in August whereas for Angus, it was the lowest in September.

(6) Influence of Season of Breeding

In all the dairy breeds under study, the percent of non-returns was the highest in fall season whereas in beef breed (Angus), it was highest in spring. For Holstein and Red Dane, the percent of non-returns was the lowest in winter whereas for Guernsey and Jersey, it was lowest in Summer.

In Holstein, the amount of decline of percent of non-returns from 1-day to 2-day, 2-day to 3-day, 3-day to 4-day-old semen was greater in season of lowest percent of non-returns (winter) as compared to season of highest percent of non-returns (fall).

(7) Influence of Registered and Non-registered Females

The weighted average of percent of non-returns with 1-day-old semen was 10.9% greater for first service registered females than for first service non-registered females.

(8) Number of Females Inseminated with a Day's Semen Collection for a Bull

The number of females inseminated with the semen obtained from a day's collection for a bull was the highest in Holstein breed (95 females) and was the lowest in Jersey breed (41 females).

INTER-RELATIONSHIP OF CERTAIN SEMEN CHARACTERISTICS AND RELATIONSHIP BETWEEN VARIOUS SEMEN CHARACTERISTICS AND PERCENT OF NON-RETURNS

(A) REVIEW OF LITERATURE

According to Davis (1938) initial motility of the spermatozoa is one of the best, if not the best, single evidences of viability of semen.

Davis and Williams (1939) in a study on dairy bulls found a slightly significant correlation between volume of semen and initial motility of spermatozoa, volume of semen and concentration of spermatozoa per cubic mm., and between percentage of motility (initial) of spermatozoa and concentration of spermatozoa per cubic mm. Motility in this study was expressed as percentage of spermatozoa showing progressive motion.

Anderson (1940) found that infertile bulls produced less volume of the ejaculate.

Dougherty and Evalt (1941) reported little correlation of initial motility of sperm, percentage of abnormal spermatozoa and concentration, with breeding efficiency.

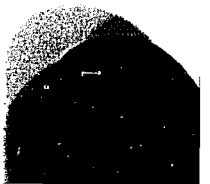
Swanson and Herman (1941a) in a study of semen from 10 dairy bulls used in the University of Missouri dairy herd reported that semen volume, concentration, pH of semen, percentage of abnormal spermatozoa and initial motility of spermatozoa were not correlated with the fertility of the bull. The semen characteristic most nearly correlated with fertility was the length of survival with vigorous motility when the semen was stored undiluted at 40°F. When separate ejaculates from the same bull were considered, initial motility was found to be roughly correlated with

viability in storage. Pregnancies, in this study, were determined by examination and by failure of the cows to come in heat after service.

In the study of semen from Arizona range beef bulls, Lasley and Bogart (1943) found no significant correlation of semen volume with concentration, initial motility rating, abnormal spermatozoa, percentage live spermatozoa in semen immediately after collection. Concentration of spermatozoa was highly correlated with initial motility rating but was negatively correlated with the percentage of abnormal spermatozoa. Motility (initial) rating was significantly correlated with the percentage of live spermatozoa immediately following collection.

Ejaculates containing larger amounts of semen were found to give the greatest fertility. Concentration of spermatozoa and the number of spermatozoa per ejaculate were highly correlated with fertility. The correlation between concentration of spermatozoa and fertility was curvilinear (concave type) in that the ejaculates containing 800,000 to 1,000,000 sperm per cubic millimeter gave the lowest fertility whereas ejaculates of lower and higher concentration gave greater fertility. Initial motility rating and the percentage of abnormal spermatozoa were not significantly correlated with fertility. Semen containing less than 50 percent live spermatozoa immediately following ejaculation, was of questionable fertility. But semen samples containing 50-90% of live spermatozoa, varied little in fertility.

The number of spermatozoa per insemination was significantly correlated with fertility. The number of live spermatozoa used per insemination was more closely related to fertility than the total number of spermatozoa.



In this study all the semem samples were given an initial motility rating of 2 or over 2.

Margolin et al. (1943) reported a significant correlation between longevity (duration of motility) of diluted semen samples stored in a refrigerator at 45°F and the conception rate. Conception was ascertained by palpation or by the calving of the cows served.

Lasley (1944) found a highly significant correlation between live and percentage of motile spermatozoa and between live and progressively motile spermatozoa in non-stored semen. Furthermore, a correlation between the percentage of motile spermatozoa and fertility was also noted.

The percentage of live spermatozoa in this study was determined by the opal blue-eosin staining method.

Swanson and Herman (1944) reported that concentration of sperm, abnormal sperm and pH of semen of dairy bulls were not significantly correlated with conception rate. Conception was determined either by calving, manual examination for presence of fetus or by failure of cow to return to heat within 90 days after insemination.

A highly significant linear correlation between viability of the sperm in storage and conception rate was found. There was also a significant curvilinear (convex type) relationship between motility rating of semen at the time of insemination and conception rate. With the increase of motility rating up to rating of 3, conception rate increased but with further increase in motility rating, conception rate showed a nonsignificant increase.

Laing (1945) found that bulls of higher fertility gave on the average more volume of semen than bulls of lower fertility. Laing suggested that although the fertility is adversely affected if a minimum requirement in quantity and quality of spermatozoa is not met, factors other than semen quality are in operation within the range of fertility from 1.0 to 3.5, which may vary breeding results irrespective of quality of semen.

Fertility in this study was measured by the number of services per conception based on diagnosis of pregnancy by rectal examination at about 50 days from the date of last service.

Mercier and Salisbury (1946) in an analysis of the combined data for the ejaculates of Holstein and Guernsey breed found highly significant correlation of concentration of spermatozoa with percent of non-returns. Percentage of abnormal spermatozoa and methylene blue reduction time were significantly correlated with percent of non-returns.

Ellenberger and Lohmann (1946) found that volume of ejaculate, concentration of spermatozoa, total number of spermatozoa per ejaculate of dairy bulls were not significantly correlated with conception rate. These investigators believed that there was little to be gained by continuing observations on duration of motility in storage beyond periods of from 3 to 7 days.

Madden et al. (1947) in a study on dairy bulls reported a highly significant correlation between initial motility and initial percentage of live sperm. However, this relationship was fairly variable. These investigators found no significant differences in the initial motility,

initial percentage of live sperm or longevity in storage between semen samples which resulted in conception and samples which did not.

Lewis (1948a) in a study on dairy bull semen found a significant correlation between semen volume and initial motility (percent of progressively motile spermatozoa) for Holstein bulls but correlation for Guernsey bulls was not statistically significant. Correlations between concentration and initial motility were highly significant for the Guernseys but were not significant for the Holsteins. Semen volume, concentration, total spermatozoa per ejaculate or initial motility of the semen shipped was not significantly correlated with percent of non-returns.

Percent of living sperm determined with eosine-aniline blue staining method was reported by Schaffer and Almquist (1949) of having a highly significant curvilinear regression with percent non-returns. However, the predictive value with relatively high quality semen was questionable.

Ludwick et al. (1948) using semen samples bred to 30 or more cows obtained a high correlation ($r = 0.84 \pm 0.03$) between incubation time (time for sperm to lose all activity when held at 100° F) and percent of non-returns. However, the correlation was lowered considerably when semen samples bred to fewer cows were used.

Erb et al. (1950) reported that a minimum of 30, first and second services were required before the conception rate (percent of non-returns) of a single semen sample could be accepted as a reasonable approximation of its fertilizing capacity. These investigators found no significant relationship of initial motility, concentration of sperm or initial pH with fertilizing capacity of semen from dairy bulls with acceptable breeding

efficiency. The correlation of the percentage of sperms alive at the time of semen collection and the fertilizing capacity was highly significant. Motility for each day of storage up to 6 days showed a highly significant correlation with percent non-returns. The correlation between initial motility and concentration was highly significant.

Branton et al. (1951) in an analysis of semen quality and breeding results of 100 semen samples found no significant correlation of initial motility of spermatozoa with percent of non-returns. But there was a highly significant correlation between concentration of the individual ejaculates with percent of non-returns. A still greater highly significant correlation was observed when the analysis was based on the means for the individual bulls. The numbers of motile spermatozoa per milliliter of diluted semen or per insemination showed a highly significant correlation with percent of non-returns.

Table XIII shows the correlation between semen characteristics reported by various investigators. Table XIV shows the correlation of semen characteristics with conception rate or percent of non-returns reported in the literature.

(B) EXPERIMENTAL PROCEDURE

Semen collection records (Standard Card No. 2) for bulls of various breeds, namely, Holstein, Guernsey, Jersey, Red Dane and Angus were used to find the arithmetic mean and standard deviation for various semen characteristics, by breed and regardless of dairy breed. The semen

TABLE XIII

CORRELATION BETWEEN SEMEN CHARACTERISTICS

Correlation Between	Davis et al. (1939)		Lasley et al. (1943)		Lasley (1944)	
	N+	r (Pearson)	N	r	N	r
Semen volume and sperm - concentration	224	+0.3553*	476	-0.035		
Semen volume and progressive motility of sperms of non-stored semen	224	+0.3783*				
Semen volume and abnormal - spermatozoa			176	-0.006		
Semen volume and percentage of sperms alive in non-stored semen			224	+0.075		
Sperm concentration and progressive motility of sperms of non-stored semen	264	+0.3095*				
Progressive motility of sperms in non-stored semen and percentage of sperms alive in non-stored semen					158	+0.696**

\+ Number of samples used in calculating the coefficient of correlation (r)
* Significant at 5% level of probability
** Significant at 1% level of probability

TABLE XIV

CORRELATION OF SEMEN CHARACTERISTICS WITH CONCEPTION RATE OR PERCENT OF NON-RETURNS

	Lasley et al. (1943)		Swanson et al. (1944)		Mercier et al. (1946)		Erb et al. (1950)		Branton et al. (1951)	
	N+	r	N	r	N	r	N	r	N	r
Semen volume	303	+0.909**			330	-0.033				
Sperm concentration			559	+0.63	328	+0.162**	590	+0.034	100	+0.2834**
Initial motility of sperm (non-stored semen)							187	+0.138		
Percentage of sperms alive (non-stored semen)							198	+0.212**		
pH			205	-0.18			373	+0.002		
Percent abnormal sperm			525	-0.12	280	-0.133*				
Viability in storage			475	+0.84**						
Motility of sperm in semen stored:										
for 3 days							187	+0.335**		
for 4 days							187	+0.317**		
No. of sperm per insemination					316	-0.096				

+ Number of samples used in calculating the coefficient of correlation (r).
* Significant at 5% level of probability.
** Significant at 1% level of probability.

collection and breeding data of dairy bulls, (regardless of dairy breed) were then subjected to correlation analysis so as to find the inter-relationship between various semen characteristics and the relationship of semen characteristics with percent of non-returns. The non-return rates (percent of non-returns) were based on 90-day non-returns of females with 1, 2, 3, 4, 5 and over current services regardless of age of semen. In view of the results reported by Ludwick et al. (1948), Erb et al. (1950), only the semen samples bred to 40 or more females were used to determine the correlation of semen characteristics with percent of non-returns. However, no such restriction of the number of services, was placed in determining the correlation between various semen characteristics

(C) RESULTS

Table XV shows the arithmetic mean and standard deviation (distribution) for various semen characteristics of dairy bulls. In this Table are also shown by breed (dairy and beef) the arithmetic mean and the standard deviation for certain semen characteristics, namely, semen volume, sperm concentration, percentage of sperms alive in non-stored semen and progressive motility of sperms in non-stored semen. The raw semen movement for Angus bulls is also included in the Table. From the Table, it can be seen that in dairy breeds, the semen volume was the largest in Guernsey and was the smallest in Jersey. Concentration of semen (sperm concentration) was highest in Jersey and it was lowest in

TABLE XV

MEAN AND STANDARD DEVIATION FOR SEMEN CHARACTERISTICS

Breed	No. of Samples	Semen Volume (mls.)		Sperm Concentration (per 1/1000 mm³.)		Sperms Alive in Non-Stored Semen %		Progressive Motility of Sperm in Non-Stored Semen %	
		Mean	Std. Dev.	Mean	Std. Dev.	Mean	Std. Dev.	Mean	Std. Dev.
Holstein	1304	6.9	2.11	1067	373	73.2	3.63	80.5	4.69
Guernsey	779	7.2	2.26	1727	397	72.8	3.50	80.6	3.48
Jersey	245	5.6	1.73	1689	360	73.0	4.24	80.6	3.69
Red Dane	142	6.2	1.45	1866	385	73.5	3.58	80.9	4.11
Dairy bulls (average)	2470	6.6	2.47	1625	386	73.1	3.65	80.6	4.21
Angus	123	5.3	1.30	1651	443	71.0	5.94	79.7	3.37

		Raw Semen Movement %	
		Mean	Std. Dev.
Dairy bulls	1204	81.3	9.9
Angus bulls	77	81.9	9.9

	3-Day-Old* Re-examined Semen			4-Day-Old Re-examined Semen				
	Progressive Motility of Sperm %				Sperms Alive %		Progressive Motility of Sperm %	
	No. of Samples	Mean	Std. Dev.	No. of Samples	Mean	Std. Dev.	Mean	Std. Dev.
Dairy bulls	860	35.0	8.9	830	36.5	8.6	33.9	9.3

* Semen sample re-examined 72 hours after collection.

Guernsey. However there was not much difference between dairy breeds in the percentage of sperms alive in non-stored semen and the progressive motility of sperms in non-stored semen.

The semen volume, sperm concentration, percentage of sperms alive in non-stored semen and the progressive motility of sperms in non-stored semen were greater in dairy bulls than in the beef bulls (Angus). However, the raw semen movement in dairy bulls was slightly less than that in beef bulls.

The percentage of sperms alive in 4-day-old re-examined semen and the progressive motility of sperms in 3-day- and 4-day-old re-examined semen showed a marked decline as compared to fresh semen examined soon after the collection.

In Table XVI are presented the r-values (coefficient of correlation) for the correlation between semen characteristics of dairy bulls. From Table XVI, it can be seen that there were highly significant (1% level) positive correlations between the following pairs of semen characteristics: concentration of semen and percentage of sperms alive in non-stored semen; percentage of sperms alive in non-stored semen and progressive motility of sperms in non-stored semen; raw semen movement and concentration of semen; raw semen movement and percentage of sperms alive in non-stored semen; raw semen movement and progressive motility of sperms in non-stored semen.

Highly significant (1% level) negative correlation was noted between volume of semen and concentration of semen and between volume of semen and percentage of sperms alive in non-stored semen.

TABLE XVI

CORRELATION BETWEEN SEMEN CHARACTERISTICS OF DAIRY BULLS

Correlation Between	No. of Samples	Simple Correlation Coefficient (r)
Volume of semen and concentration of semen	2470	-0.0856**
Volume of semen and percentage of sperms alive in non-stored semen	2470	-0.0531**
Volume of semen and progressive motility of sperm in non-stored semen	2470	+0.0442*
Concentration of semen and percentage of sperms alive in non-stored semen	2470	+0.0829**
Concentration of semen and progressive motility of sperm in non-stored semen	2470	+0.0430*
Percentage of sperms alive in non-stored semen and progressive motility of sperm in non-stored semen	2470	+0.2741**
Raw semen movement and volume of semen	1204	+0.0057
Raw semen movement and concentration of semen	1204	+0.4195**
Raw semen movement and percentage of sperms alive in non-stored semen	1204	+0.2698**
Raw semen movement and progressive motility of sperm in non-stored semen	1204	+0.1465**

* Significant at 5% level of probability.

** Significant at 1% level of probability.

Volume of semen was positively correlated with progressive motility of sperms in non-stored semen. The correlation was found to be statistically significant (5% level). A significant (5% level) positive correlation was also noted between concentration of semen and progressive motility of sperm in non-stored semen. However, there was no significant correlation between raw semen movement and volume of semen.

Table XVII shows simple correlation coefficients obtained for the correlation of characteristics of dairy bull semen with percent of non-returns. From the Table, it can be seen that there was highly significant (1% level) positive correlation between each of the following semen characteristics and percent of non-returns. However, the r-values (coefficient of correlation) obtained were small.

Progressive motility of sperms in non-stored semen

Raw semen movement

Concentration of sperms per 1/1000 cubic mm. of diluted semen

Percentage of sperms alive in 4-day-old re-examined semen.

The correlation between volume of semen and percent of non-returns was not statistically significant. Similarly, the correlation of the concentration of semen and that of percentage of sperms alive in non-stored semen, with percent of non-returns was not statistically significant. The progressive motility of sperm either in 3-day-old or 4-day-old re-examined semen was not significantly correlated with percent of non-returns.

TABLE XVII

CORRELATION OF SEMEN CHARACTERISTICS OF DAIRY BULLS WITH PERCENT OF NON-RETURNS

Semen Characteristic	No. of Samples	Simple Correlation Coefficient (r)
Volume of semen	1897	+0.0074
Concentration of semen	1897	+0.0138
Percentage of sperms alive in non-stored semen	1897	+0.0100
Progressive motility of sperm in non-stored semen	1897	+0.0884**
Raw semen movement	1000	+0.1950**
Concentration of sperm per 1/1000 mm.^{3} of diluted semen	1887	+0.1213**
Progressive motility of sperm in 3-day-old re-examined semen	860	-0.0059
Percentage of sperms alive in 4-day-old re-examined semen	830	+0.1262**
Progressive motility of sperm in 4-day-old re-examined semen	830	+0.0405

* Significant at 5% level of probability.
** Significant at 1% level of probability.

(D) DISCUSSION

Statistical analysis of the data in the present investigation showed a highly significant (1% level) negative correlation (r = -0.0856) between volume of semen and concentration of semen of dairy bulls. Contrary results were reported by previous workers. Davis et al. (1939) in dairy bulls obtained a significant positive correlation (Pearson r = +0.3553) whereas Lasley et al. (1943) in beef bulls found no significant correlation of semen volume with concentration of semen.

In the present study, a highly significant (1% level) negative correlation (r = -0.0531) was obtained between volume of semen and percentage of sperms alive in non-stored semen. The result obtained is not in agreement with that reported by Lasley et al. (1943) who found no significant correlation in beef bulls.

Davis et al. (1939) reported significant positive correlation between sperm concentration and progressive motility of sperms (Pearson r = +0.3095) in non-stored semen of dairy bulls. Our results confirm the finding of above investigators in that there was a significant position correlation between those characteristics.

In the present investigation, a highly significant (1% level) positive correlation (r = + 0.2741) was noted between percentage of sperms alive in non-stored semen and progressive motility of sperms in non-stored semen. Similar finding was also reported in beef bulls by Lasley (1944). However, the correlation coefficient reported by Lasley was much higher. (r = +0.696).

Lasley et al.(1943) in beef bulls reported highly significant correlation between volume of semen and conception rate. Mercier et al. (1946) in dairy bulls found no significant correlation between semen volume and percent of non-returns. The results obtained in the present investigation are not in agreement with that reported by Lasley et al. (1943). On the other hand, our results confirm the finding of Mercier et al. (1946) in that there was no significant correlation between semen volume and percent of non-returns.

Swanson et al. (1944) and Erb et al. (1950) found no significant correlation between sperm concentration and conception rate or percent of non-returns. On the contrary, Mercier et al. (1946) and Branton et al. (1951) noted highly significant (1% level) correlation between sperm concentration and percent of non-returns. The results obtained in the present study confirm the findings of Swanson et al. (1944) and Erb et al. (1950) since no significant correlation was obtained in our study between concentration of semen and percent of non-returns.

Erb et al. (1950) reported a highly significant (1% level) correlation between percentage of sperms alive in non-stored semen and percent of non-returns. However, in the present investigation no significant correlation was obtained between percentage of sperms alive in non-stored semen and percent of non-returns.

Mercier et al. (1946) found no significant correlation between number of sperm per insemination and the percent of non-returns. On the other hand, our results showed a highly significant (1% level) correlation

between concentration of sperm per 1/1000 cubic mm. of diluted semen and percent of non-returns.

The disagreements in the results reported by various investigators and also those obtained in the present investigation might be due to one or more factors mentioned below:

(1) Small number of samples

(2) Differences in methods used for determining various semen characteristics

(3) Restriction as regards minimum requirements of various semen characteristics (percentage of sperms alive, concentration of semen, etc.) before semen sample could be sent for insemination

(4) Restriction regarding the total number of services from a semen sample before it can be used in correlation study

(5) Type of animal (dairy or beef type)

In the present investigation, of all the semen characteristics under study, the raw semen movement showed the highest correlation with percent of non-returns. Percentage of sperms alive in 4-day-old re-examined semen ranked 2nd to raw semen movement in correlation with percent of non-returns. Concentration of sperms per 1/1000 cubic mm. of diluted semen ranked 3rd in correlation with percent of non-returns, whereas progressive motility of sperms in non-stored semen ranked 4th in correlation with percent of non-returns. The results obtained suggest that in Michigan Artificial Breeders Association, a semen sample meeting the minimum requirements of semen characteristics or combination of semen characteristics can possibly be judged with greater accuracy as regards its potential

ability for higher percent of non-returns from the following characteristics. These characteristics are listed in order of importance.

Raw semen movement

Percentage of sperms alive in 4-day-old re-examined semen

Concentration of sperms per 1/1000 cubic mm. of diluted semen

Progressive motility of sperms in non-stored semen

The concentration of semen and the percentage of sperms alive in non-stored semen may not be considered in judging the potential ability for higher percent of non-returns, of a semen sample meeting the minimum requirements of semen characteristics or combination of semen characteristics in Michigan Artificial Breeders' Association. This is because, in the present study there was no significant correlation between concentration of semen and percent of non-returns. Similarly, the correlation between percentage of sperms alive in non-stored semen and percent of non-returns was not significant.

In the present investigation, there was no significant correlation of progressive motility of sperms in 3-day-old re-examined semen with percent of non-returns. Similarly, there was no significant correlation of progressive motility of sperms in the 4-day-old re-examined semen with percent of non-returns. These results indicate that there is very little to be gained by continuing observations on progressive motility of sperms in 3-day and that in 4-day-old re-examined semen, so far as predicting the percent of non-returns is concerned.

In the present study, a highly significant (1% level) correlation of raw semen movement and progressive motility of sperms in non-stored

semen was noted. However the correlation coefficient (r value) was not high. Similarly, the correlation coefficient (r-value) for the correlation of raw semen movement and percent of non-returns was not high. The results suggest that the observations on progressive motility of sperms in non-stored semen be continued even though the observations on raw semen movement are being taken.

(E) SUMMARY AND CONCLUSIONS

The semen collection records of Michigan Artificial Breeders Association were studied from June 1, 1951 to May 31, 1952, with two objectives: (1) to determine the inter-relationship of certain semen characteristics and relationship between various semen characteristics and percent of non-returns for dairy bulls, (2) to find the mean and distribution (standard deviation) of certain semen characteristics of dairy bulls and of beef bulls.

For the study of correlation of semen characteristics with percent of non-returns, only the semen samples bred to 40 or more females were used. The percentages of non-returns were based on 90-day non-returns of females with 1, 2, 3, 4, 5 and over current services.

The investigation led to the following findings and conclusions:-

Semen Characteristics: Mean Values

(1) Of all the dairy breeds under study, the semen volume was the largest (7.2ml.) in Guernsey and was the smallest (5.6 ml.) in Jersey. Concentration of semen was the highest (1889 per 1/1000 cubic mm.) in

Jersey and was the lowest (1727 per 1/1000 cubic mm.) in Guernsey. There was not much difference between dairy breeds in the percentage of sperms alive in non-stored semen and the progressive motility of sperms in non-stored semen.

(2) The semen volume and concentration of semen were much greater (1.5 mls. and 174 per 1/1000 cubic mm., respectively) in dairy bulls than in Angus bulls. The percentage of sperms alive in non-stored semen and the progressive motility of sperms in non-stored semen were also greater (2.1% and 0.9%, respectively) in dairy bulls than the Angus bulls. However, the raw semen movement in dairy bulls was somewhat low (0.6%) as compared to that in beef bulls.

(3) The percentage of sperms alive in 4-day-old re-examined semen and the progressive motility of sperms in 3-day and in 4-day-old re-examined semen showed a marked decline as compared to fresh semen examined soon after the collection.

Correlation between Semen Characteristics

(4) A highly significant (1% level) positive correlation between the following pairs of semen characteristics was found.

Concentration of semen and percentage of sperms alive in non-stored semen ($r = +0.0829$).

Percentage of sperms alive in non-stored semen and progressive motility of sperms in non-stored semen ($r = +0.2741$).

Raw semen movement and concentration of semen ($r = +0.4195$).

Raw semen movement and percentage of sperms alive in non-stored semen ($r = +0.2698$).

Raw semen movement and progressive motility of sperms in non-stored semen ($r = +0.1465$).

There was a highly significant (1% level) negative correlation between volume of semen and concentration of semen ($r = -0.0856$) and between volume of semen and percentage of sperms alive in non-stored semen ($r = -0.0531$).

Volume of semen was significantly correlated with progressive motility of sperms in non-stored semen ($r = +0.0442$). A significant (5% level) positive correlation was also noted between concentration of semen and progressive motility of sperms in non-stored semen ($r = +0.043$). However, no significant correlation was found between raw semen movement and volume of semen.

Correlation Between Semen Characteristics and Percent of Non-Returns

(5) Highly significant (1% level) positive correlation was noted between each of the following semen characteristics and percent of non-returns. However, the r-values (simple correlation coefficient) obtained were small. Progressive motility of sperms in non-stored semen ($r = +0.0884$)

Raw semen movement ($r = +0.1950$)

Concentration of sperm per 1/1000 cubic mm. of diluted semen ($r = +0.1213$)

Percentage of sperms alive in 4-day-old re-examined semen ($r = +0.1262$)

The volume of semen, concentration of semen and the percentage of sperms alive in non-stored semen, were not significantly correlated with

percent of non-returns. Similarly, no significant correlation was noted between the progressive motility of sperms either in 3-day-old or in 4-day-old re-examined semen and percent of non-returns.

(6) The results (r-values) obtained suggest that a semen sample meeting the minimum requirements of semen characteristics or combination of semen characteristics at Michigan Artificial Breeders Association can possibly be judged with greater accuracy as regards its potential ability for higher percent of non-returns from the following characteristics listed in order of importance:

Raw semen movement

Percentage of sperms alive in 4-day-old re-examined semen

Concentration of sperms per 1/1000 cubic mm. of diluted semen

Progressive motility of sperms in non-stored semen

(7) Observations on progressive motility of sperms either in 3-day or in 4-day-old re-examined semen seem to be of very little use in predicting the percent of non-returns.

(8) The correlation of raw semen movement with progressive motility of sperms in non-stored semen ($r = +0.1465$) and that of raw semen movement with percent of non-returns ($r = +0.1950$) was highly significant (1% level). However, the r-values were not large. This, together with the fact that there was a highly significant correlation (1% level) between the progressive motility of sperm in non-stored semen and the percent of non-returns ($r = +0.0884$) suggests the desirability of continuing the observations on progressive motility of sperms in non-stored semen even though the observations on raw semen movement are being recorded.

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