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GONADOTROPIC POTENCY OF MULE DEER PITUITARIES
AND THE POSSIBLE CORRELATION WITH THE
REPRODUCTIVE CYCLE

by

KENNETH GRIESER

B.S., University of Idaho, 1953.

Presented in partial fulfillment of the requirements
for the degree of Master of Arts

MONTANA STATE UNIVERSITY

1955

Approved by:

Ludwig G. Bertram
Chairman, Board of Examiners

Andon B. Castle
Dean, Graduate School

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INTRODUCTION

A study involving female mule deer reproduction was initiated in September, 1953, by the Department of Zoology and Wildlife Research Unit of Montana State University. The deer were to be collected from the National Bison Range at Moiese, Montana. Howard S. Sears, a graduate student in Wildlife Technology was to collect the deer and conduct a study on the ovaries and reproductive tract. The author assisted Sears in the field collections and obtained material for this study.

There is in the literature considerable material dealing with pituitary gonadotrophic potency in birds and mammals. However, to my knowledge nothing has been done in this respect with deer. Therefore, it was felt that a study of this type, involving female mule deer, might lead to a more complete understanding of the overall reproductive process in this animal.

Many mammals have their breeding season in the spring or summer under the stimulus of increasing light intensity. The undomesticated ungulates of North America generally experience their period of sexual activity in the fall, when, if the light stimulus is involved, the animals

must be reacting to a diminution of light (Marshall, 1937). Evidence presented by Yeates (1949) indicates that these animals definitely do react to decreasing light if first subjected to a period of increased light. Wislocki (1943) suggests that in the deer, " . . .The annual hypophysis cycle has its onset much earlier than has been suspected from the sexual behavior and that, as in other animals, light, acting in spring or summer, probably serves as the initial exteroceptive stimulus." He further states that the withdrawal of light may be an active factor in precipitating the terminal phase (or rut) of the ungulate cycle. Studies involving pituitary potency of female sheep (Kammlade et. al., 1953) seem to support Wislocki's assumption that the hypophysis reacts to summer light stimulus. Kammlade found that anestrus pituitaries (from the summer months) were higher in total gonadotrophic potency than those collected during estrus. This was also found to be true in the sow (Nalbandov, 1953).

As previously mentioned, the pituitary of the deer has never been studied to determine its gonadotrophic potency in relation to the various seasons and reproductive phases. Therefore, the present study was initiated, first, to determine the unfractionated gonadotrophic potency of the female mule deer pituitaries for each month and incidentally, to correlate, if possible, the obtained potency levels with the phases of the reproductive cycle and with the changes in light and temperature. A second objective was to compare and contrast the pituitary potencies of the different age class i. e. fawns, yearlings and adults.

MATERIALS AND METHODS

The deer, from which the material for this study was obtained, were collected according to a schedule previously worked out with Sears (1955). On each collection date, an attempt was made to kill a fawn, a yearling and an adult. After each animal was selected and shot, it was dragged to the vehicle where an autopsy was performed (Sears, 1955). The time required for a complete autopsy was approximately two hours. Occasionally the animals were killed at a considerable distance from the car. In such cases, we either packed our equipment to the deer, or dragged the deer to the car. In any event, such procedures were time consuming and prevented the prompt removal of the pituitaries. The glands were removed from each animal as soon as possible, at least within three hours after death.

Fresh weights of 14 pituitaries were taken in the field. The fresh glands were trimmed as much as possible before the field weights were taken. However, the connective tissue capsule could not be removed without damaging the gland. The glands were promptly placed in an 8 dram vial of absolute acetone. Later, in the laboratory, they were placed in fresh acetone and put under refrigeration. All glands were stored in this manner until they were to be powdered for assay. Acetone

dry weights of the whole pituitary were obtained. Before the weights were taken, the capsule of connective tissue was removed. Each pituitary was powdered in a small agate mortar directly before it was to be assayed. The powder from each gland was weighed and the weight recorded. It was found that on the average 1 mg. of dry powder was equivalent to 6.63 mg. of fresh pituitary. The fresh weights of the rest of the pituitaries collected were then calculated by multiplying the powdered weights by 6.63.

A total of 61 pituitaries was collected and assayed. This total is made up of 22 adults (2 years or older), 19 yearlings (12-24 months) and 20 fawns (1 week to 11 months).

Tables I, II and III give all pertinent data, such as collection dates, age, hog dressed weights and pituitary weights. The ages and body weights were obtained from Sears's (1955) original data sheets. It will be noted that the powdered weights of the pituitaries range from 4 to 30 milligrams lighter than the whole weights. This weight loss can be attributed to the removal of additional connective tissue during the powdering process. A negligible amount of powder was lost in handling and weighing.

In many cases it was necessary to pool two or more pituitaries. On account of limited time, it was felt that for the adults and yearlings, 2 samples per month during the breeding season would be sufficient. It was

necessary to pool the fawn pituitaries collected during an entire month and sometimes for two months, in order to have enough powder for injecting a reliable number of test animals.

The assay animals were intact 20 to 21 day old female white mice, the stock of which came originally from the Microbiological Institute of the U.S. Public Health Service, Hamilton, Montana. It was fully realized that the intact immature animal is much less desirable for the assay of gonadotrophins than hypophysectomized immature animals. However, we did not have the equipment nor time to perform hypophysectomies, nor did we have the financial resources to purchase such animals. Therefore, it was decided that, with certain precautions, the results yielded by the immature intact mouse would be adequate for this study.

The best security in such a situation is the use of numerous controls. Therefore, each test animal was represented by one or two litter mates in the control group. In striving for uniformity of size and condition, each litter was reduced to six animals at the time of birth. The controls were given dummy injections of saline, equivalent to the amount received by the test animals. An attempt was made at all times to have from 8 to 10 test animals for assay with a corresponding number of controls. However, this objective could not always be realized because of a lack of a sufficient amount of pituitary powder.

Whole pituitary powder suspensions were made with 2.5 mg. of whole pituitary powder per cc. of sterile physiological salt solution. These

suspensions were prepared immediately before the first injection and refrigerated between injections. Each experimental animal received subcutaneously one half cc. of this suspension twice daily for four days. The total dose was 10 mg. of pituitary powder. This total dose was arrived at after several preliminary tests using December killed buck pituitaries and excess powder from series 2 and 5.

Autopsies of test and control mice were performed at the end of the fifth day following the initial injection. Body weights were obtained with a dietary scale graduated to 1 gram. The weights of uteri and ovaries were obtained to the nearest 0.1 mg. with a Roller-Smith torsion balance. All connective tissue was carefully dissected away from the uteri and ovaries before they were removed. The oviducts were not removed from the uterus. Precautions were taken to prevent drying of tissues before weights were secured. Observations were made on whether the vaginal orifice was open or closed.

Typical ovaries and uteri from each series were preserved in A. F. A. for histological sectioning. These were later sectioned and stained with hematoxylin and eosin.

The responses obtained are all expressed as percent of increase of the experimental over the control. This was calculated by first reducing the average uterine and ovarian weights to percent of the mean body weight. The percent figures obtained were put in the following

formula:

$$100x \frac{\text{exp. wt.} - \text{control wt.}}{\text{control wt.}} = \text{percent of increase.}$$

In order to have a known standard with which to compare the results obtained from the deer pituitaries, a response curve of graded dosages was established for Pregnant Mare Serum (P.M.S.) and Follicle Stimulating Hormone (F.S.H.) from sheep pituitaries.

Average weather temperatures for the entire collection period were secured from the records kept at the Headquarters Building of the National Bison Range. The average temperature was computed for the first half and last half of each month. Data on available light and percent of possible sunshine for each month was obtained from the U. S. Weather Bureau at Missoula, Montana. The Missoula Station is located approximately 30 miles south of the Bison Range. Information on the temperature and light is presented in table IV and figure 1.

TABLE I.
COLLECTION DATES, AGES, FIELD DRESSED BODY WEIGHTS,
AND PITUITARY WEIGHTS OF FAWN DEER.

Deer No.	Coll. Date	Age. (mo.)	Body Wt. (lbs.)	Pituitary Wt. (In mg.)			
				Acetone Dry	Powder	Fresh (act.)	Fresh (Calc.)
61	6-11-54	1 wk.	6.0	26.6	21.7	-	143.8
64	7-11-54	1-2	17.0	33.4	33.0	-	218.8
67	8-16-54	2-3	30.0	40.0	34.4	281.0	228.1
69	9-22-54	3-4	50.0	41.4	38.4	254.0	254.6
6	10- 3-53	4-5	42.5	24.0	20.6		136.6
11	10-24-53	4-5	39.0	19.0	16.0		106.1
14	10-21-53	4-5	40.0	25.9	21.2		140.5
16	11- 7-53	4-5	35.0	31.8	26.4		163.1
20	11-14-53	4-5	41.0	32.4	29.0		192.3
23	11-21-53	5-6	32.0	32.6	26.8		187.7
26	11-27-53	5-6	33.0	31.9	24.0		159.1
28	12- 5-53	5-6	35.0	25.6	20.1		133.2
30	12-11-53	5-6	28.0	32.0	23.6		156.5
35	12-17-53	6-7	17.0	28.0	19.8		131.3
37	12-22-53	6-7	28.0	26.6	17.8		118.0
41	1- 9-54	6-7	34.0	27.4	21.8		144.5
44	1-22-54	6-7	38.0	27.4	23.0		152.5
50	3-22-54	8-9	43.0	32.4	27.4		181.7
54	4-11-54	9-10	45.0	38.4	35.2		233.4
58	5-15-54	10-11	51.0	54.6	52.2	351.0	346.0

TABLE II.
COLLECTION DATES, AGES, FIELD DRESSED BODY WEIGHTS,
AND PITUITARY WEIGHTS OF YEARLING DEER.

Deer No.	Coll. Date	Age (Mo.)	Body Wt. (lbs.)	Pituitary Wt. (In mg.)			
				Acetone Dry	Powder	Fresh (act.)	Fresh (Calc.)
59	6-11-54	11-12	53	74.2	63.2	371.6	419.0
63	7-10-54	12-13		85.6	71.4	477.4	473.4
66	8-16-54	13-14	70	58.2	56.2	508.6*	372.6
70	9-22-54	14-15	76	87.6	79.2	318.0*	525.3
4	10- 3-53	15-16	73	33.2	28.8		190.9
7	10-16-53	16-17	72	41.8	36.8		244.0
10	10-24-53	16-17	84	72.8	50.8		336.8
13	10-31-53	16-17	72	38.4	35.6		236.0
17	11- 7-53	16-17	66	57.6	42.5		281.8
19	11-14-53	16-17	82	49.0	33.8		224.1
24	11-27-53	17-18	78	60.8	54.6		362.0
25	11-27-53		79	61.6	52.4		347.4
27	12- 4-53	17-18	79	60.2	52.4		347.4
33	12-16-53	18-19	65	57.4	46.8		310.3
34	12-16-53	18-19	70	52.2	43.4		287.7
38	12-22-53	19-20	65	42.8	38.0		251.9
42	1-22-54	20-21	75	54.6	40.2		266.5
43	1-22-54	20-21	68	49.4	40.0		265.2
52	4-11-54	22-23	75	89.0	73.8		489.3
55	5-14-54	23-24	69	99.0	88.6	506.2	587.4

* These weights were not used in calculating the fresh wt. because they are apparently in error.

TABLE III.

COLLECTION DATES, AGES, FIELD DRESSED BODY WEIGHTS,
AND PITUITARY WEIGHTS OF ADULT DEER.

Deer No.	Coll. Date	Age (yrs)	Body Wt. (lbs.)	Pituitary Wt. in mg.			
				Acetone Dry	Powder	Fresh (act.)	Fresh (calc.)
5	10- 3-53	6-7	105.5	91.0	69.2		458.8
8	10-16-53	4-5	84.0	81.8	62.0		411.1
9	10-24-53	4-5	101.0	86.4	82.0		543.6
12	10-31-53	2-3	90	88.0	81.8		542.3
15	11- 7-53	2-3	80	55.6	47.4		314.3
18	11-14-53	2-3	82	70.0	50.0		331.5
21	11-21-53	3-4	94	68.2	62.2		412.4
22	11-21-53	4-5	93	94.8	88.0		583.4
31	12-12-53	7-8	81	118.2	-		-
32	12-12-53	4-5	67	105.0	-		-
36	12-22-53	2-3	78	72.8	57.6		381.9
39	1- 9-54	2-3	84	79.2	63.8		423.0
40	1- 9-54	2-3	84	78.0	62.2		412.4
45	2-13-54	2-3	92	67.6	58.8		389.8
46	2-14-54	6-7	77	122.9	94.6		627.2
48	3-13-54	8-9	70	108.0	87.8		582.1
49	3-22-54	10	77	118.4	96.4		639.1
53	4-11-54	2-3	90	93.6	72.0		477.3
56	5-14-54	2-3	94	187.6	167.5	1155.0	1110.5
60	6-11-54	2-3	90	143.8	129.0	802.0	855.3
62	7-10-54	2-3	87	125.6	111.2	735.6	737.2
65	8-16-54	3-4	90	117.2	100.0	687.4	663.0
68	9-22-54	2-3	98	53.8	32.6*	535.0	-

* Approximately 10 mg. of this powder was lost, therefore, the fresh wt. could not be calculated.

TABLE IV.

WEATHER DATA DURING THE STUDY PERIOD
NATIONAL BISON RANGE, MOIESE, MONTANA AND MISSOULA, MONTANA

Month	Av. Bi-Weekly temperature °F	Av. Day length		% of possible sunshine
		hrs.	min.	
June 1954	55.5			
June 1954	59.6	15	44	50
July 1954	67.7			
July 1954	66.3	15	27	79
Aug. 1954	65.3			
Aug. 1954	59.5	14	9	64
Sept. 1954	56.7			
Sept. 1954	53.3	12	40	72
Oct. 1953	50.2			
Oct. 1953	44.1	10	53	66
Nov. 1953	37.9			
Nov. 1953	38.5	9	28	47
Dec. 1953	34.4			
Dec. 1953	35.0	8	35	21
Jan. 1954	33.1			
Jan. 1954	18.8	9	6	22
Feb. 1954	26.7			
Feb. 1954	40.4	10	15	38
Mar. 1954	29.9			
Mar. 1954	32.9	11	55	53
Apr. 1954	43.4			
Apr. 1954	44.5	13	30	49
May 1954	51.1			
May 1954	56.4	15	4	65

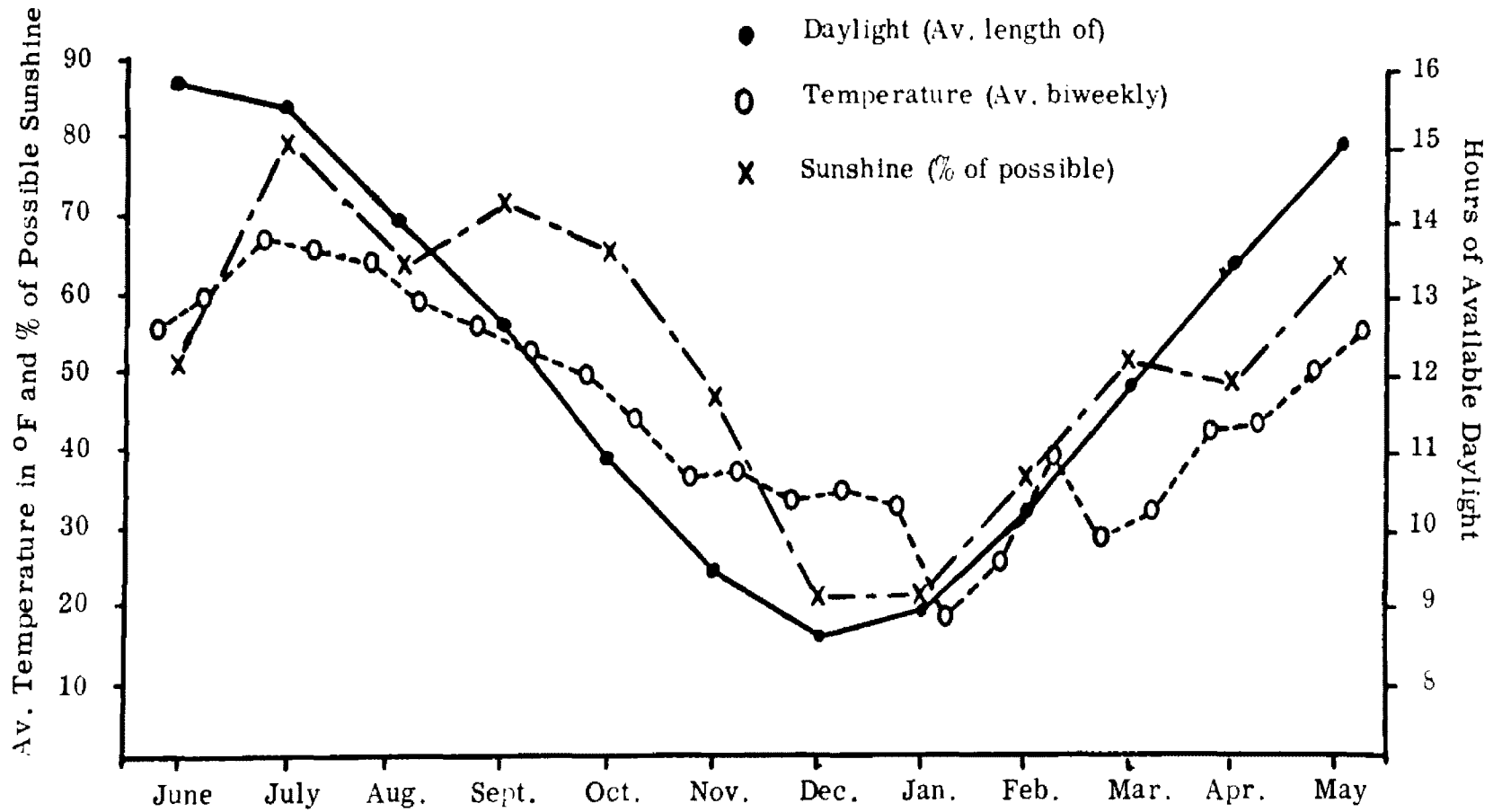


Figure 1. Weather conditions 1953-54, National Bison Range, Moiese, Montana and Missoula, Montana.

RESULTS

The results of bioassay from the three age groups are summarized in tables V, VI and VII. (The complete data are presented in the appendix.) The collection dates, ages and reproductive activities (inactive, pregnant or lactating) of the female deer are included in the tables for easy reference. Reproductive activity is not included in table V since all of the fawns would be classed as inactive. Each table also includes the ranges in uterine weights of the experimental and control mice. Parturition generally occurs in late May and early June on the Bison Range. Therefore, June was selected as the logical starting point for the tables and figures.

It was found that when using weight increase as a criterion, the uterus in the immature intact mouse is more sensitive to small quantities of unfractionated gonadotrophic hormones than the ovary. This agrees with the findings of Levien and Tyndale (1937). In the majority of cases, the ovarian weight increase was negligible. Even histological examination of typical experimental ovaries failed to demonstrate a significant increase in follicular development. The vaginal orifice response is unreliable (Frank and Berman 1939) and difficult to put into quantitative terms. Therefore, the gonadotrophic potency is determined primarily as the percent of uterine weight

increase of experimental immature mice over the uterine weights of immature intact controls.

Fawns:

The bioassay results obtained from the fawn pituitaries are presented in table V. The general pattern of the results for this group is a high response (187 percent increase in uterine weight) from the pooled August and September fawns and no response from the January animals. The peak response is preceded by a fairly low level (36 percent uterine weight increase) which was induced by the pooled June and July fawns. After the August-September peak, the fawn pituitaries show a gradual decrease in their ability to induce a uterine weight increase in the assay animals. According to the standard used here this decreased ability would mean a decrease in gonadotrophic content of the pituitaries. The results show an 88 percent response for October, 98 percent for November, and 11 percent for December. The February fawn was accidentally shot in the head causing complete destruction of the pituitary. Therefore, it is not known how long the low January level was maintained. The pooled March and April fawn pituitaries demonstrated a rise in gonadotrophic potency by inducing an 84 percent uterine weight increase in the assay mice. The pituitary from the May fawn also indicated a rising gonadotrophic potency. The actual values representing percent of uterine weight increase for the March-April

and May fawns series cannot be considered as highly significant.

These two series had to be conducted with only 4 and 3 assay animals which is not enough for a reliable sample. However the tests were conducted and the results presented here because they do seem to indicate that the gonadotrophic potency has increased from the low January level.

Yearlings:

The yearling deer, like the fawns, have pituitaries which are highest in total gonadotrophic potency during the summer months. Also, like the fawns, the yearlings are minimal in pituitary potency by January.

It should be pointed out that the May fawn was approximately 11 months old and June yearling was one year old. Therefore, the yearling group begins where the fawns leave off. The June yearling caused a slightly higher uterine response (61%) than did the May fawn, indicating a gradual build up to the summer peak. The July yearling was not a typical animal in that her gonads contained testicular tissue (Sears, 1955). The response obtained from this atypical yearling was a 157% uterine weight increase and also a 90% ovarian weight increase. The peak uterine response of 177 percent was obtained from the August yearling pituitary. The decline following the August peak is not as marked in the yearlings as in fawns. However, there are also more yearling samples than fawn samples. The response produced by the September

yearling was down to 95%. From this point the responses fluctuate somewhat. The animals taken in the first half of October elicited an 82% response; those from the last half were up to 98%. Complete data were not obtained from the early November yearlings, therefore, these animals cannot be considered. The pituitaries from the last half of November were quite low in potency causing only a 45% uterine weight increase. This response was followed by one of 102% caused by the early December pituitaries. The pituitary glands from the yearlings collected in late December demonstrated a drop to 40 percent. The January yearling failed to produce any response in the test mice. Attempts to collect yearling deer during February and March were not successful, therefore these months are blank. These months are indicated by long broken lines on figure 2. The pituitaries from the April and May yearlings indicate that the potency is again beginning to rise. The April uterine response was 17% and the May pituitary produced a 33% response.

Adults:

Data from the adult pituitary tests are summarized in table VI. According to the results presented here the adult pituitary cycle differs considerably from that followed by the fawns and yearlings. The adult pituitaries show an intermediate gonadotrophic potency from May to early November. They reach a peak potency in late

November and then undergo a gradual decrease until February and March when they cause no response at all.

The Uterine weight increases induced by the summer adult pituitaries generally range from 73% to 87%. Two exceptions to this were a July response of 16% and a late October one of 129%. The peak potency was reached rather sudden in that the responses jumped from 74% during the first half of November to 205% in the last half. Following the peak, the decline in gonadotrophic potency was more gradual than the rise. The uterine responses induced by the adult pituitaries from the first and last halves of December were 165 and 126% respectively. The response caused by the pituitaries from the January adults was still up to 100%. However, by the middle of February, the adults failed to elicit any response in the assay animals. Series ten involving the April adult pituitary was not completed. The pituitary suspension for this series was contaminated in some way and all of the test mice died on the third day of the injection period. As mentioned previously, the response elicited by the May adult pituitary (79%) falls within the approximate range which was maintained throughout the summer.

Figure 3 shows the graded dosage curves for Follicle Stimulating Hormone (F.S.H.) and Pregnant Mare Serum (P.M.S.). From these

curves the response equivalents were obtained for 10 mg. of deer pituitary powder. The values obtained do not mean that this is the amount of F.S.H. in 10 mg. of deer pituitary. It only means that the response elicited by this amount of unfractionated pituitary is the same as the response from the indicated amount of F.S.H. and P.M.S. The response elicited by unfractionated pituitaries in the assay animals is not caused by a single hormone, but there may be an augmentation or even inhibiting effect (Evans, Korpi, Pencharz and Simpson, 1936) of several hormones. Tables V, VI and VII show the estimated amounts of F.S.H. and P.M.S. that will give the same response as that obtained from the unfractionated deer pituitaries.

The highest uterine response induced by the adult deer pituitaries was a 205% weight increase in late November. This is equivalent to the response elicited by 1.94 mg. of F.S.H. and .85 mg. of P.M.S.. The highest yearling response occurred in July and equaled 177% uterine weight increase. This is equivalent to the increase caused by 1.73 mg. of F.S.H. and .83 mg. of P.M.S. The peak fawn response was a 187% increase in the August-September series which was equivalent to 1.80 mg. F.S.H. and .84 mg. of P.M.S.

In comparing the gonadotrophic activity of the three age groups (see figure 2) we find that the fawns and yearlings follow approximately the same pattern. They reach their peak potency during July and

August and undergo a gradual decline until January. The adult, however, maintains, a fairly constant level from May to early November, and rises to a rather sudden peak in the later part of November. This high level declines rather slowly through December and January. The potency of the yearling and fawn pituitaries have reached the minimum level by January while adults do not reach this level until February.

TABLE VII.
ASSAY RESULTS FROM ADULT PITUITARIES

Ser. No.	Deer No.	Coll. Date	Rep. Act.	No. of Mice		Av. Body Wt. (gms)		Av. Ovary Wt. (mg)		Av. Uterus Wt. (mg)		Range in Uterine Wt.		% of Wt. Increase		Uterine Resp. Equiv.		
				exp.	con.	exp.	con.	exp.	con.	exp.	con.	exp.	con.	ovary	uterus	F.S.H. (mg)	P.M.S. (mg)	
12	60	6-11-54	1.	*	10	10	11.9	11.5	3.4	3.68	16.67	9.3	10.3-22.0	8.0-12.6	0	73.7	.92	.72
13	62	7-10-54	1.		10	10	12.8	11.5	3.8	3.68	12.09	9.30	9.2-21.0	8.0-12.6	0	16.2	.53	.44
14	65	8-16-54	1.		9	12	11.1	11.6	3.7	4.53	21.80	12.10	15.3-35.0	8.2-19.0	0	87.5	1.02	.76
15	68	9-22-54	1.		3	6	11.8	11.9	3.9	4.95	22.46	12.30	20.4-23.8	10.0-17.0	0	84.3	1.00	.76
1	5 8	10-3-53 10-16-53	1. 1.		9	17	11.0	11.8	4.07	4.09	21.65	11.70	16.2-26.4	7.4-16.4	0	81.9	.97	.75
2	9 12	10-24-53 10-31-53	1. 1.		5	8	11.3	12.0	3.60	3.70	22.88	11.60	12.4-34.2	5.2-13.4	0	129.5	1.34	.79
3	15 18	11-7-53 11-14-53	1. 1.	*	9	17	11.0	10.8	4.2	4.09	20.7	11.7	15.3-23.6	7.4-16.4	0	74.0	.93	.73
4	21 22	11-14-53 11-21-53	1. 1.		11	12	10.4	10.5	3.72	3.38	32.2	10.5	19.4-43.2	7.6-12.4	11	205.9	1.94	.85
5	31 32	12-12-53 12-12-53	P. P.	*	7	10	9.9	9.6	3.83	2.94	19.3	7.08	12.6-26.0	3.6-8.4	13	165.7	1.63	.81
6	36	12-22-53	P.		5	5	12.3	11.2	4.3	4.4	21.8	8.8	16.3-39.9	7.2-12.4	0	126.9	1.32	.79
7	39 40	1-9-54 1-9-54	P. P.		10	17	11.3	10.8	3.96	4.09	24.6	11.7	16.2-37.0	7.4-16.4	0	100.9	1.12	.77
8	45 46	2-13-54 2-14-54	P. P.		10	13	12.5	12.4	4.02	3.9	14.2	14.6	9.2-21.6	10.6-19.2	2	0	0	0
9	48 49	3-13-54 3-22-54	P. P.		10	13	12.7	12.4	3.85	3.9	12.56	14.6	10.2-15.6	10.6-19.2	0	0	0	0
11	56	5-14-54	P.		10	8	11.7	12.3	4.33	4.32	24.75	14.5	12.3-32.0	8.4-29.6	0	78.8	.96	.74

* lactating (l.), inactive (i.) and pregnant (p.).

TABLE VI.

ASSAY RESULTS FROM YEARLING PITUITARIES

Ser. No.	Deer No.	Coll. Date	Rep. Act.	No. of Mice	Av. Body Wt. (gm.)		Av. Ovary Wt. (mg.)		Av. Uterus Wt. (mg.)		Range in Uterine Wt.		% of Wt. Increase		Uterine Resp. Equiv. F.S.H. (mg.) P.M.S. (mg.)		
					exp.	con.	exp.	con.	exp.	con.	exp.	con.	ovary	uterus			
25	59	6-11-54	1.*	6	5	12.6	12.0	3.91	3.82	33.7	12.5	16.6-20.2	9.2-12.4	0	61.4	.84	.89
26	63	7-10-54	1.	6	9	11.5	11.0	3.20	4.13	33.7	12.5	25.4-41.3	10.6-16.8	90.0	157.5	1.56	.81
27	66	8-16-54	1.	5	8	11.7	11.9	4.86	4.43	28.6	10.5	23.6-35.8	7.8-16.0	11.2	177.3	1.73	.83
28	70	9-22-54	1.	7	11	11.6	11.2	4.31	4.41	22.1	10.9	15.0-33.8	8.0-14.2	0	95.9	1.08	.76
16	4	10-13-54	1.	6	7	11.5	11.2	3.56	4.77	20.66	11.0	14.6-25.6	7.6-15.8	0	82.6	.98	.76
17	10	10-24-53	1.	8	8	9.9	11.9	4.25	4.43	17.45	10.51	12.6-23.8	7.8-16.0	0	98.9	1.11	.77
19	24	11-27-53	1.	10	9	11.7	10.5	3.79	3.68	17.6	10.9	12.6-26.2	9.6-15.4	0	45.6	.73	.60
20	25	11-27-53	1.	4	6	14.1	13.0	4.45	4.25	25.67	11.9	19.3-34.0	9.6-16.2	0	102.2	1.12	.77
21	33	12-16-53	P.	10	11	11.0	11.3	4.36	4.38	17.96	13.2	13.4-24.6	7.8-17.6	0	40.5	.70	.58
22	34	12-22-53	P.	7	6	12.6	13.1	4.4	4.4	12.71	14.5	10.8-15.6	9.2-22.2	0	0	0	0
23	42	1-22-54	P.	7	7	11.5	10.5	3.98	4.54	11.2	8.7	10.2-11.8	6.8-12.1	0	17.4	.55	.46
24	43	4-11-54	P.	8	11	11.7	11.2	4.02	4.41	15.2	10.9	12.5-17.4	8.0-14.2	0	33.0	.65	.55

* inactive (1.) and pregnant (p.) .

TABLE V.

ASSAY RESULTS FROM FAWN PITUITARIES

Ser.	Deer No.	Coll. Date	No. of Mice exp.	Av. Body Wt. exp. (gms)	Av. Ovary Wt. exp. (mg)	Av. Uterine Wt. exp.	Range in Uterine Wt. exp. cont.	% of Wt. Increase ovary uterus	Uterine Resp. Equiv. F.S.H.(mg) P.M.S.(mg.)
35	61 64	June July	5	12.10	3.44	18.88	15.4-23.0	0	.67
36	67 69	Aug. Sept.	7	10.10	3.67	23.03	17.2-36.4	0	1.80
29	6 11 14	Oct.	5	10.60	3.42	21.70	12.0-43.0	0	1.02
30	16 20 23 26	Nov.	9	11.44	3.47	19.61	10.6-36.7	0	1.11
31	28 30 35 37	Dec.	6	11.90	3.71	15.4	12.4-21.2	0	.58
32	41 44	Jan.	4	11.25	2.95	11.70	8.8-13.4	0	0
33	50 54	March April	4	10.60	4.15	20.9	15.6-24.2	4.7	1.00
34	58	May	3	11.16	2.90	15.30	13.3-17.8	0	.83

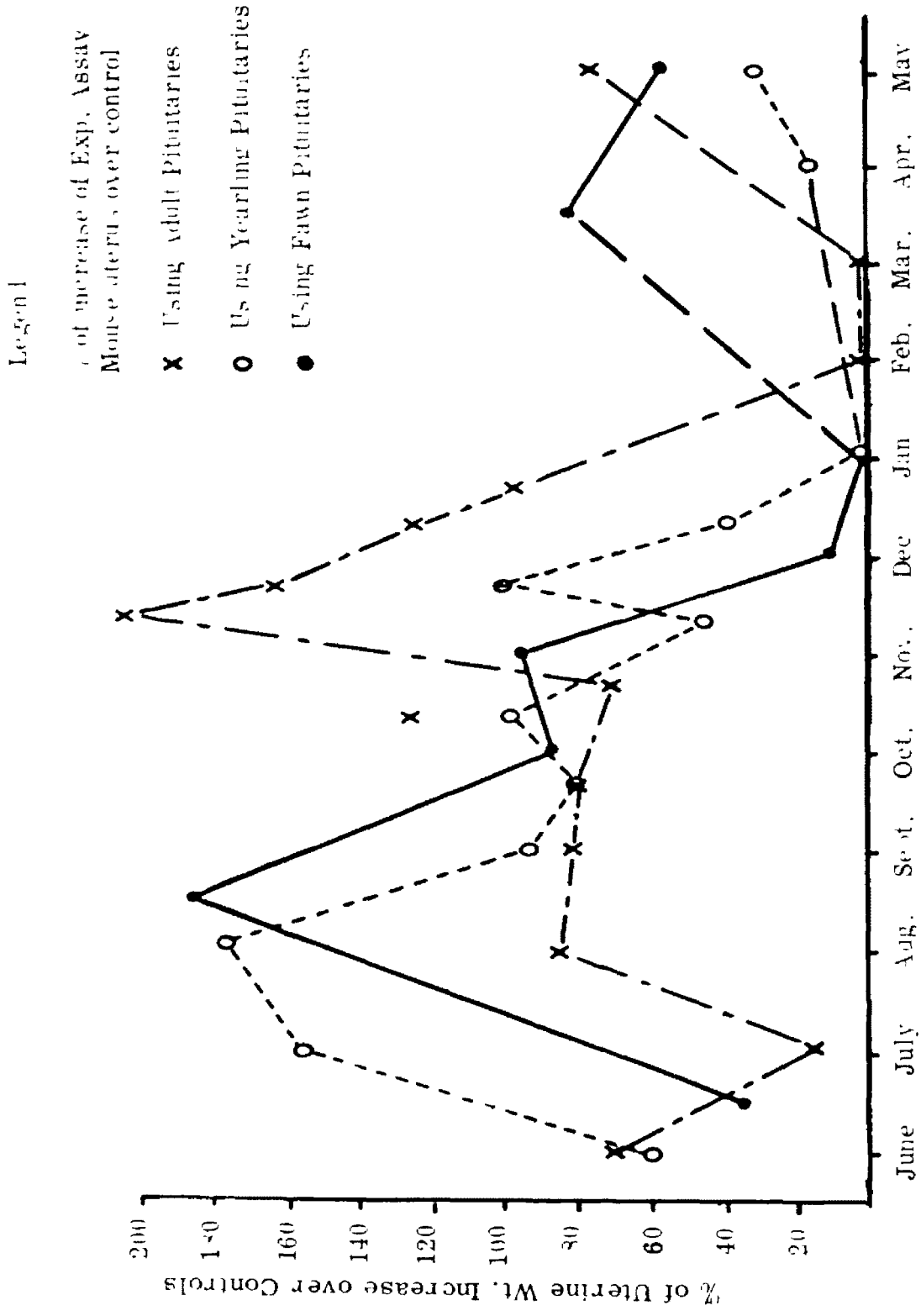


Figure 2. Bioassay of Deer Pituitaries Using Intact 20-21 Day old Female Mice.

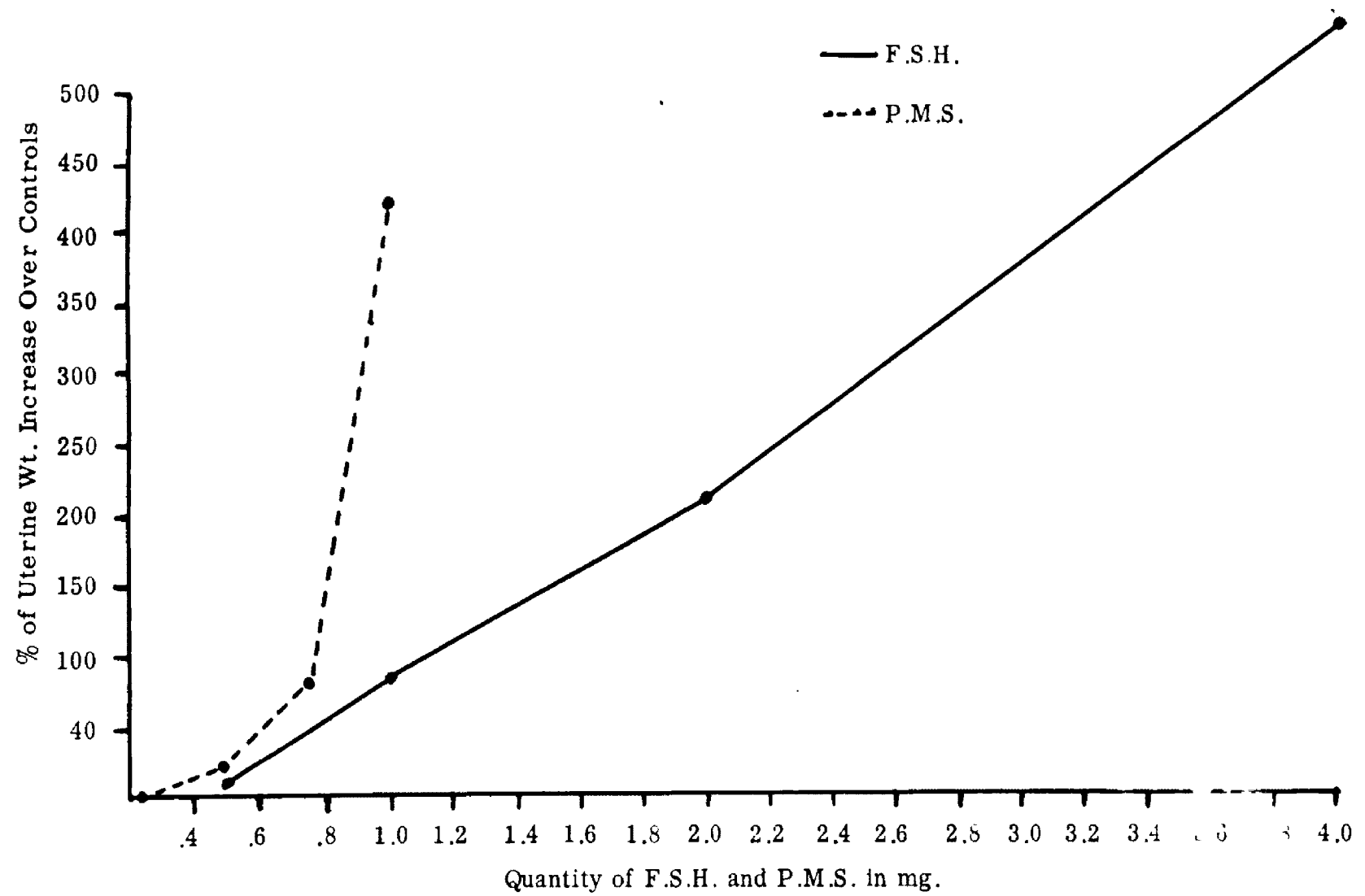


Figure 3. Bioassay of Sheep F.S.H. and P.M.S. Using intact 20-21 day old female mice.

DISCUSSION

A brief account of the general reproductive pattern of the deer herd in question is necessary before the present findings can be duly considered.

The fawns do not normally breed during their first season. Breeding mule deer fawns have been reported by Crane and Jones (1935), and Shantz (1943). One yearling doe, i.e. in its second year of life, collected in January 1954, carried a single corpus albicans in one of her ovaries. This structure was associated with a rupture site on the surface of the ovary. According to Sears it is thought probable that this deer ovulated as a fawn but did not implant an embryo. In the present collections, we failed to find any evidence that would indicate ovulation had occurred in the fawn group. Sears found that 16% of the yearling animals collected between January and June were non-gravid. Therefore, approximately 84% of the animals bred at 17 to 19 months of age. In the adult group all animals collected after December were pregnant.

The breeding season probably extends through November and into December. The first corpus luteum was found by Sears in an adult ovary collected November 14. The first identifiable embryo was recovered on December 12. Parturition occurs in late May and early

June. The adults with fawns lactate throughout the summer and into the fall. From the limited information we collected on lactation, it appears that the fawns are weaned sometime in November.

It seems well established that the anterior pituitary produces three gonadotrophic factors, two of which we are primarily concerned with here. One of these substances stimulates growth of the ovarian follicles and is called the follicle-stimulating hormone (F.S.H.). The second factor, called luteinizing hormone stimulates the development of the corpus luteum after ovulation and also maintains the interstitial cells of the gonads (Greep, VanDyke and Chow, 1941).

Fawns:

The peak of gonadotrophic potency for this group occurs approximately $1\frac{1}{2}$ months after the highest average temperature and the largest amount of available light intensity. (Compare figures 1 and 2) It is difficult to ascertain exactly when the highest potency occurs since it was necessary to pool the June and July collections. (see page 4). The June animal was approximately 1 week old when collected. The pituitary from this young animal may have diluted the total response for the series. Likewise, in the August-September series, one cannot be certain which animal contributed the most toward the response obtained. However, on the basis of the yearling results it is assumed that the peak occurs in August. This assumption is also based on the

ovarian activity (See Sears, 1955). Figure 4 shows the relationship between follicle number (2mm or more in size) and gonadotrophic potency. The average number of follicles per deer is highest at the same time that the gonadotrophic potency is high. The pituitary seems to be under the stimulus of light and/or possibly temperature, and is producing primarily F.S.H. This point will be discussed further under the yearling group.

The decline in the potency curve during the fall months appears to parallel the descending light and temperature curves. When the light and temperature curves are at their minimum levels, the fawn pituitaries failed to yield a response. In March, April and May when the days are growing warmer and longer the pituitary potency shows a gradual increase. The follicular numbers in the fawn ovaries follow a pattern very similar to that of the pituitary. It therefore appears that the hypophyseal cycle of the fawn, which is not interrupted by ovulation or pregnancy, varies with the seasonal changes in light and temperature.

Yearling:

The yearlings, demonstrated a peak gonadotrophic potency in August. These results are in close agreement with those from the sheep (Kammlade et. al., 1952). The female sheep pituitary maintains a relatively high gonadotrophic potency during July and August

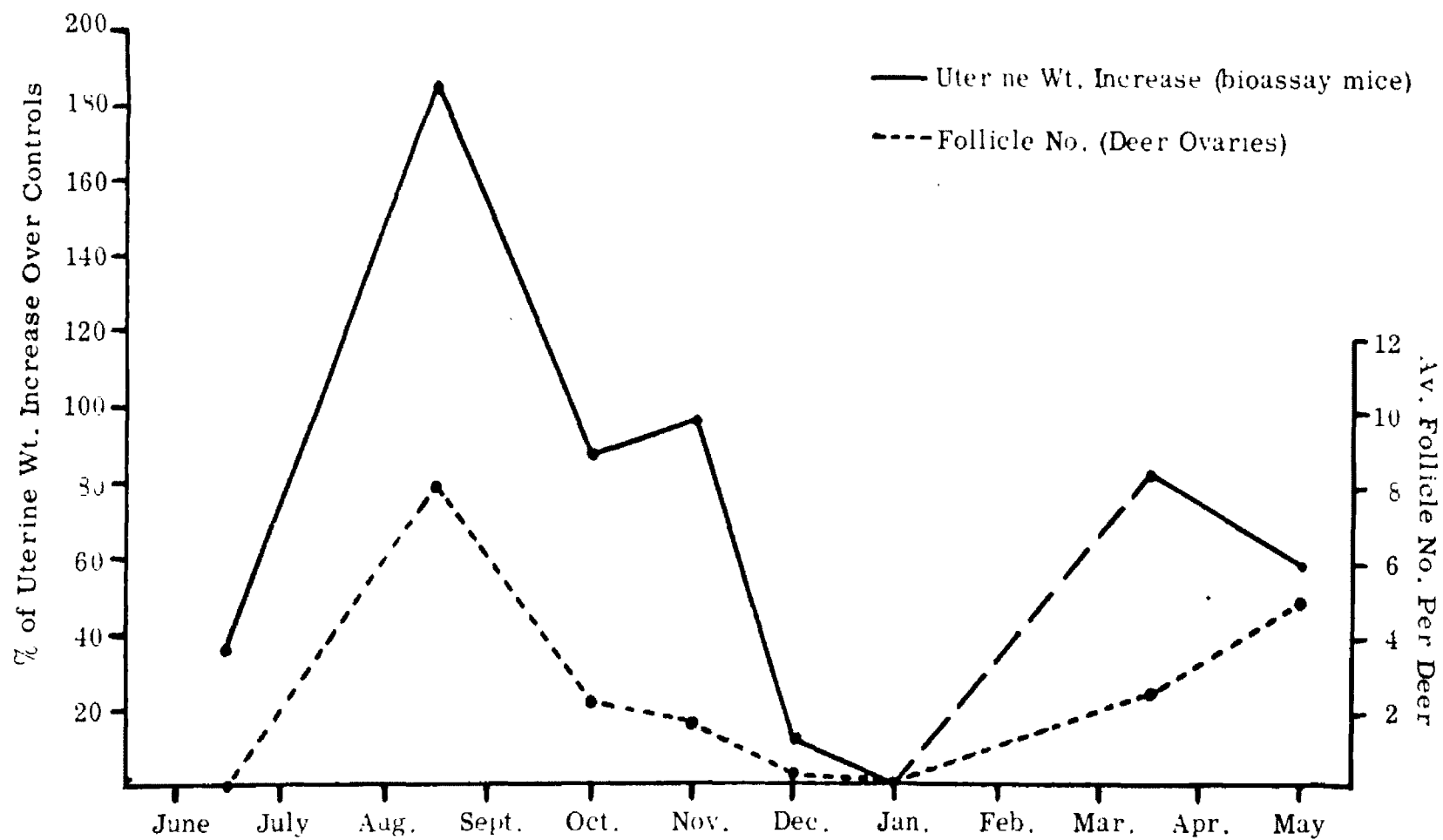


Figure 4. Bioassay of Fawn Pituitaries and Av. Follicle No. in the deer ovaries (ovarian analysis from Sears, 1955).

when it is in anestrus. The female sheep normally ovulates in October. In order for ovulation to occur, a correct ratio between the amount of follicle stimulating hormone and luteinizing hormone seems to be essential (Turner, 1949, p. 475). The anestrus sheep seem to lack this essential hormone ratio. If however, the anestrus animal is given injections of L.H., ovulation will occur. (Nalbandov, unpublished from Kammlade et. al., 1952). This would indicate an L. H. deficiency in the necessary F.S.H.-L.H. ovulation ratio. Greep et. al., (1942) and Fevold (1941) state that F.S.H. alone, while capable of causing follicular development, does not cause estrogen secretion in the absence of L.H. Kammlade et. al., (1952) suggest that the anestrus ewe is unable to secrete estrogen due to a lack of L.H. This assumption is based on the distinct castrate appearance of the uterine epithelium during anestrus.

The assay techniques employed in this study indicate total gonadotrophic potency only. Therefore, no definite statements can be made about the concentrations of the different gonadotrophic hormones in the deer pituitary. On the bases of the findings in the sheep, which has a reproductive pattern very similar to the deer, it is assumed that the yearling deer pituitary produces only minimal amounts of L.H. and large amounts of F.S.H. during July and August. This assumption is supported to some degree by the ovarian observations

of these deer. According to Sears (1955) there were more follicles in the yearling ovaries during the anestrus than during the breeding season. The follicular number and gonadotrophic potency for the yearling group is shown in figure 5. Although the gonadotrophic potency indicates a peak in August, the follicular number did not reach a peak until September. The samples for these months consist of only one yearling per month, therefore this difference may be due to individual variation. At any rate, the August follicle number is higher than anytime during the breeding season. The higher follicle number would indicate an increased stimulation by F.S.H. Kammlade et.al., (1952) found that the sheep ovary contained more follicles during estrus than anestrus. This difference is probably due to the fact that Sears considered only the follicles measuring 2 mm or more and Kammlade recorded all follicles of 1 mm or larger. Both workers agree that the ovaries are as active during anestrus as during estrus when using average follicle diameter as the criterion.

As the breeding season approaches, the pituitaries show a decreased gonadotrophic response in the assay animals. The August and September collections were more than a month apart; therefore, it is not known whether the drop is gradual or abrupt. Kammlade (1952)

found an abrupt drop on the first day of heat in the sheep. The decreased potency could be caused by decreasing light stimulus, hence a decrease in F.S.H. production. Yeates (1949) has shown that light very definitely has an influence on the breeding period of the sheep. Yeates discovered that by providing additional light artificially during the winter months and decreasing the duration during the summer, the sexual cycle could be reversed. These animals came into estrus and mated in May when the controls were in anestrus. The exact manner in which the light affected the hormone output is not known. If it is true that light stimulates the production of F.S.H., then it might follow that the removal of the stimulus would reduce the F.S.H. production. Another possible cause of a decrease in the potency of F.S.H. might be the inhibiting action of estrogen. This inhibition of F.S.H. production or release of estrogen was observed by Clark (1935) and later by Meyer and Biddulph (1941). It was recently confirmed by Byrnes and Meyer (1951) who demonstrated that very small amounts of estrogen are capable of causing this inhibition.

The factors causing L.H. production and release are still uncertain. Fevold, Hisaw and Greep (1936) state that the first effect of estrogen on the anterior lobe of the pituitary is to increase the secretion of L.H.. In the rat, both F.S.H. and L.H. appear to be essential for estrogen secretion by the ovary. (Fevold, 1941). Let

us again return to an earlier assumption that L.H. is minimal during July and August. If this assumption is false, and L.H. is being produced, then the ovaries should also be producing estrogen. This estrogen should in turn inhibit the F.S.H. production and increase the L.H. production. Unless this hormone interaction is a very slow process (lasting at least 2 or 3 months) the deer would ovulate much earlier than it normally does. The fact that the sheep ovary is capable of ovulating much earlier has been pointed out. Therefore, it seems that in order for the yearling deer to maintain a high titer of F.S.H. during July and August, the L.H. and estrogen hormone outputs would be minimal or completely lacking. Proceeding on this assumption it does not seem probable that estrogen could be the causative factor in the initial release of L.H. . Some other factor appears to be operating to initiate the production of this hormone by the pituitary. Could it be that this initiating factor is the reducing light stimulus? If this were to cause the first L.H. production, the secretion could then be maintained by ovarian estrogen. Again one can only speculate on the time at which this initial release of L.H. might occur. The uterine response in the assay animals declines until the later half of October, at which time the response is slightly higher. A remote possibility may be that this slight increase in response is caused by a synergistic effect of F.S.H. and L.H., therefore,

indicating a rise in L.H.. Unfortunately, results were not obtained from the early November yearlings which may have produced an even greater response because of the suggested increasing L.H.. The yearlings taken November 27th produced a lower response in the assay animal than the October yearlings. Kammlade et.al., (1952) found the sheep pituitary to be lower in total potency on the first day of heat than at anytime during June, July, August or September and also lower than on the succeeding days of the cycle. This low point was followed by a gradual linear rise until by the 17th day of the cycle the potency was back to the anestrus level. Nalbandov (1953) also found that the pituitaries of sows in heat were low in total gonadotrophic potency. The two yearlings (no. 24 and 25) which made up this November 27th series were carrying corpora lutea in their ovaries (Sears, 1955) which would indicate fairly recent ovulation. A possible explanation for the low total gonadotrophic potency during heat has been suggested by Kammlade et. al., (1952) and Nalbandov (1953). These men believe that the pituitary of an animal in heat produces primarily L.H., which would produce a smaller response in the end organs of the assay animals than would F.S.H. This assumption fits well with the findings that follicular estrogen inhibits F.S.H. secretion and stimulates L.H. secretion.

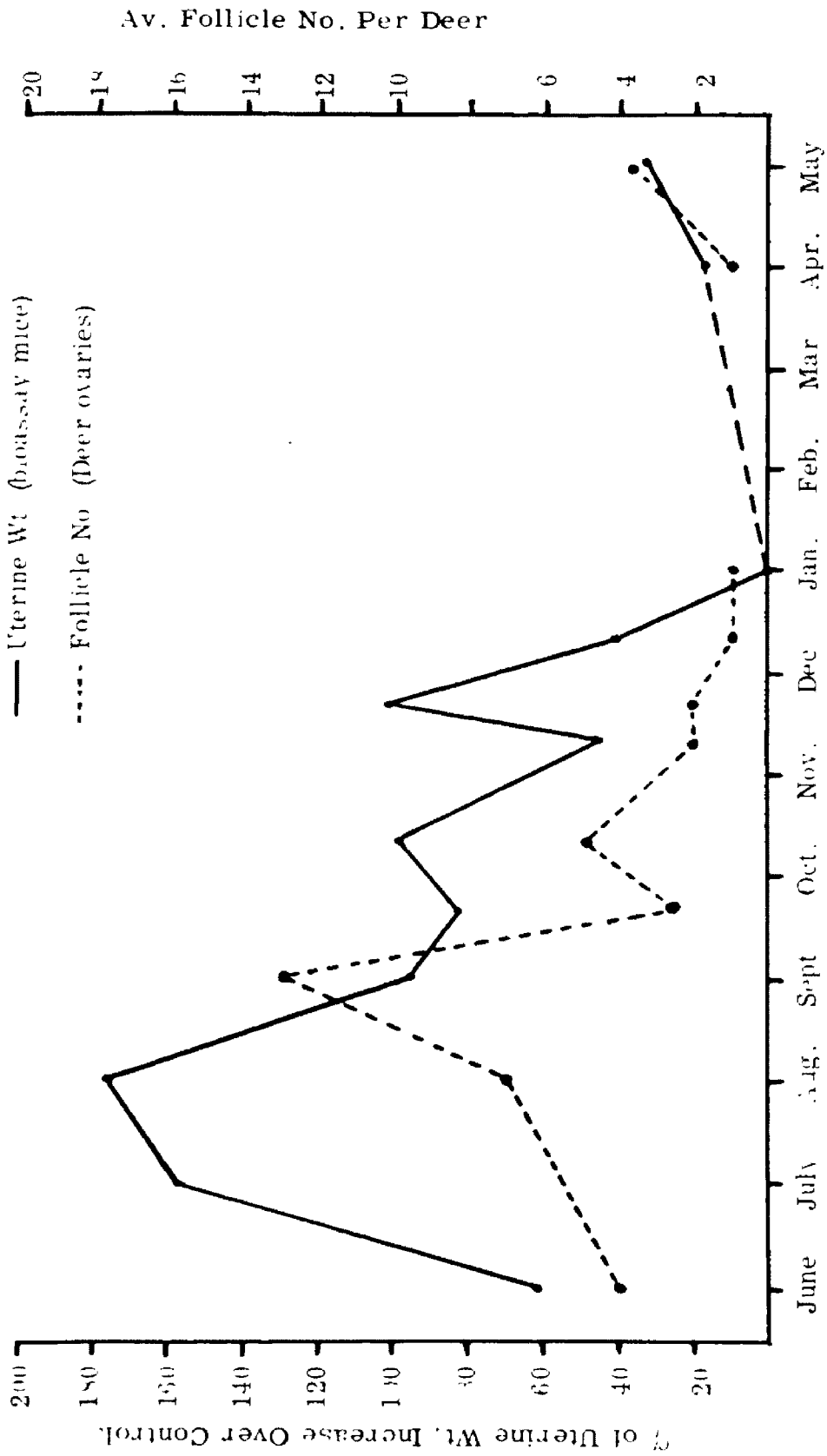


Figure 7. Biocassay of Yearling Pituitaries and Follicle Numbers in the Deer Ovaries (Ovarian analysis from Sear, 1955)

Series 20, representing the first half of December, consisted of one pregnant animal. The potency of this animal was higher than the previous series. This could possibly be caused by a synergistic effect of F.S.H. and L.H.. Nalbandov and Casida (1940) report that test rats showed predominant luteinization in their ovaries when implanted with pituitary powder from cows in early stages of pregnancy, indicating that L.H. is present at this time.

The last half of December is represented by three yearling pituitaries which were pooled to make a series. Animals No.'s 33 and 34 were pregnant and number 38 was not pregnant. The results obtained from this series are much lower than those obtained from the adults in early pregnancy (see below). Whether or not this animal (no. 38) would have bred later on is not known. However, her ovaries contained no follicles greater than 2 mm. nor did they contain any evidence of earlier ovulation (old corpora of ovulation)(Sears 1955). Possibly, the non-breeding fall yearlings follow the same or somewhat similar pituitary cycle as the fawns. If this is true and No. 38 is a non-breeding animal, one might expect this deer to be nearly as low in potency as the fawns at that time. Therefore, deer no. 38 may have caused a lower response than would have been obtained from the assay of No's. 33 and 34 only. The same factors may have been operating in the January series (22). Deer No. 42 was not pregnant and No. 43 was

carrying one embryo. The April and May yearlings will be discussed with the adult group.

Adults:

The uterine response elicited in the assay animals by the adult pituitaries, collected during the summer months, was only intermediate. The summer reproductive activity of the adults differs from that of the fawns and yearlings in that they lactate from June to October or November. It has been established that a hormone secreted by the anterior lobe of the pituitary is essential for the initiation and maintenance of lactation (Riddle, 1938). Bates, Lahr and Riddle (1935) observed that prolactin decreased the true ovarian tissue of fowl by 25% in the resting ovary and 55% in the active ovary. The oviducal size of these fowl was also reduced. Bailey (1950) states that prolactin inhibits the light induced gonad increase in the white crowned sparrow. According to Riddle (1938) and Mavromati (1950) the lactogenic hormone inhibits the secretion of gonadotrophins by the pituitary. It therefore seems that the lactating adults produce only an intermediate amount of gonadotrophins as compared with the summer collected fawns and yearlings. This reduced production is possibly caused by the inhibiting effect of the lactogenic hormone on the deers pituitary. This intermediate gonadotrophic production seems to be enough to cause a peak in the number of follicles in the adult

ovary (see figure 6).

No reason can be found to explain the very low response obtained from the July adult. Other than this, the summer adults produced a relatively uniform response until the last of October. At this time the response was higher than at anytime during the summer. The yearling also demonstrated a small increase at this time. As postulated on page 33 for the yearling, it seems likely that this adult increase in the assay animals could also be caused by a rise in L.H.. This increased uterine weight in the assay animals occurred in spite of the fact that these adults were still lactating.

The early November series (3) shows a 55 percent decrease from the late October level. Deer Number 15 appeared to be lactating and the ovaries did not contain corpora lutea. Number 18, which was pooled with number 15 to make up series 3, was not lactating and her ovaries contained two corpora lutea (Sears, 1955). Deer number 18 might be expected to be low in potency in the light of the finding from the sheep and pig (see page 35). Possibly deer number 15 tended to increase the response somewhat, but that is difficult to determine from the present data.

Series 4 gave a much greater response than any previous adult series (figure 2). Both deer involved in this series carried corpora lutea in their ovaries (Sears, 1955). These deer could have been

pregnant, but if so, we did not recover the fetuses. The smallest fetuses recovered were collected December 16. These were a pair of twins with crown-rump measurements of 8.5 mm and 11.5 mm.. A single fetus was taken December 11 which measured 13 mm. crown-rump. Therefore, it seems possible that the smaller embryos could have been missed in the field analysis of the adult uteri.

All adults collected from December to May were pregnant. The peak November potency was followed by a gradual decrease through December and January and by February no response was obtained. The response for March was also zero. No data were obtained for the April adult. The May response indicates an increase in pituitary gonadotrophic output. The April yearling which was pregnant, indicated a slight increase.

This general pattern (a high pituitary gonadotrophic potency at the onset of pregnancy followed by a gradual regression reaching a minimum during mid-pregnancy then rising again as parturition approaches) is in agreement with the findings in other animals. Nalbandov and Casida (1940) report that a steady and a significant decrease in gonadotrophic activity of glands from cows takes place from early to later pregnancy. Their findings indicate the presence of L.H. during early pregnancy and primarily F.S.H. during late pregnancy. Nalbandov and Casida, state that, the decrease in gonadotrophic potency may be explained by the assumption that placental

estrogen either inhibits the formation of gonadotrophic hormones in the pituitary or causes an increased rate of excretion of the hormones by the gland. The average decline in follicular development of the pregnant cow favors the first. Both adult and yearling deer show a decline in follicle number as pregnancy progresses. Bates, Riddle and Lahr (1935) also report a higher potency during early rather than late pregnancy for the cow. Witschi and Riley (1940) report that in the human the pituitary rapidly loses potency during the early pregnancy and slowly regains it toward the end.

A comparative summary of the uterine weight increase induced in the assay animals by the three age groups is demonstrated in figure 7. The fawn and yearling results are quite similar, therefore, they were pooled to make a single curve. The June-July results for the fawns were averaged with the June yearlings. Also, the August-September fawn results were averaged with the July yearlings. When two sets of results were available for a single month, the average of the two is represented.

Two peaks in uterine response are demonstrated in figure 7. One peak in July and August represented by the fawns and yearlings is assumed to be due to F.S.H. The second peak in November and December induced by the adults is assumed to be due to synergistic reaction of L.H. and F.S.H. during early pregnancy.

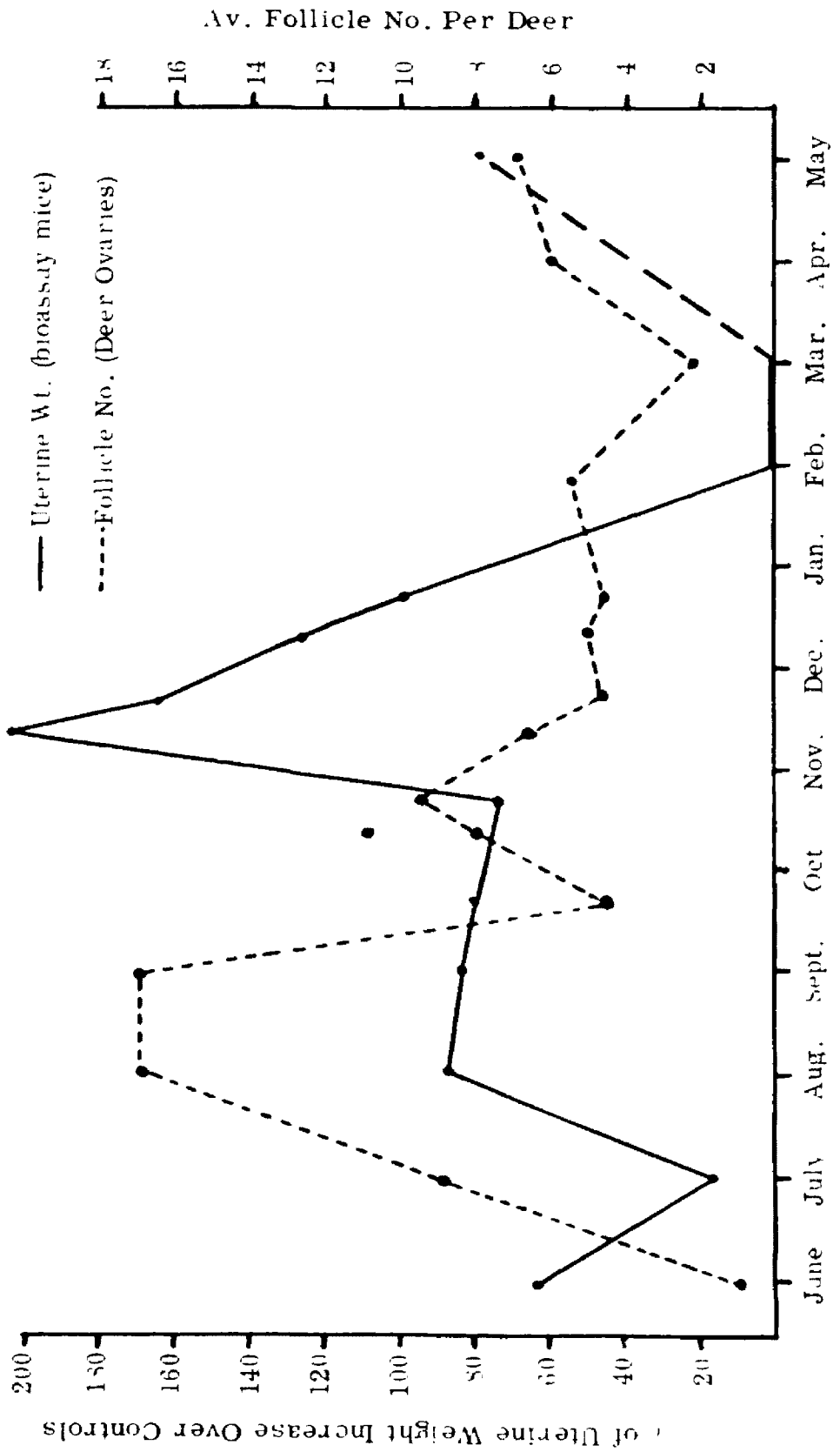


Figure 6. Bioassay of Adult Pituitaries and Av. Follicle No. in Deer Ovaries (Ovarian analysis from Sears, 1955).

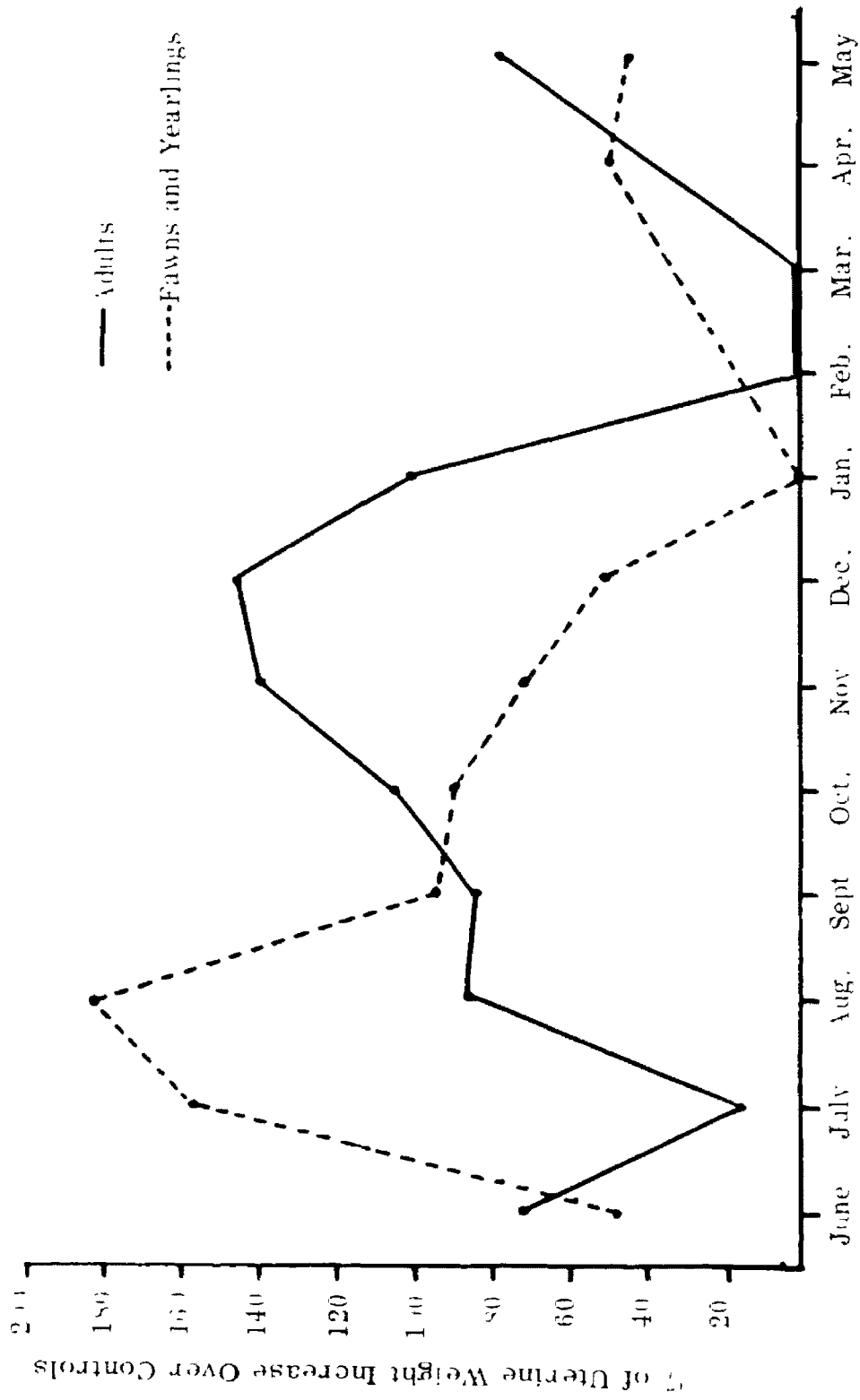


Figure 7. Summary of Uterine Wt. Increases Induced in Assay Mice by Fawn-Yearling and Adult Pituitaries

SUMMARY

A total of 34 mouse bioassay tests was conducted with unfractionated female mule deer pituitaries from 22 adults, 19 yearlings, and 20 fawns, collected from each month of the year.

The results presented were based on the percent of uterine weight increase of experimental intact immature mice over the uterine weight of the normal controls.

The results appear to indicate that the summer fawns and yearlings have pituitaries with a high total gonadotrophic potency. The potency drops to an intermediate level during the fall months. During the winter months, the potency appears to be very low and as spring progresses the potency demonstrates a gradual increase.

The lactating, summer collected, adults indicated only an intermediate gonadotrophic potency. A peak level is reached by the adult deer in November. This peak was followed by a gradual decline, becoming very low in late winter. As parturition approaches, the potency increases, and returns to the summer level by May.

The suggestion is made that the fawn-yearling peak in gonadotrophic potency may be due primarily to follicle stimulating hormone; and the adult high may be due to the synergistic action of luteinizing and follicle stimulating hormones.

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APPENDIX

This appendix contains complete data that was recorded at autopsy of experimental and control immature intact mice.

B. W. = Body weight of mice in grams.

O. W. = The weight of both mouse ovaries in mg.

U. W. = Uterine weight in mg.

V. O. = Vaginal orifice

x - indicates that the lumen was patent

- indicates that the lumen was closed

The number at the bottom of each column, under the line, is the arithmetic mean.

Series 1

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
8.0	3.0	16.2	-
11.5	4.2	24.4	x
11.5	3.8	21.6	-
11.5	4.2	26.4	x
12.0	4.8	20.0	x
11.5	4.8	25.8	x
10.5	3.4	21.2	x
11.5	3.7	21.3	x
11.0	4.8	18.0	x
<u>11.0</u>	<u>4.07</u>	<u>21.65</u>	

Control 1, 3, 7.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
13.5	4.4	11.4	-
10.0	4.4	11.0	-
10.0	4.6	11.4	-
10.0	4.8	12.2	-
11.5	4.6	14.0	-
11.5	3.4	9.2	-
11.5	3.4	13.2	-
8.5	3.8	7.4	-
10.5	4.8	16.4	-
12.5	4.6	16.0	-
10.5	3.2	8.8	-
10.0	2.6	8.8	-
11.0	5.0	14.2	-
10.5	4.4	11.0	-
11.5	4.4	13.4	-
10.0	3.4	9.2	-
10.5	3.8	11.4	-
<u>10.8</u>	<u>4.09</u>	<u>11.7</u>	

Series 2.

12.5	3.8	30.2	x
9.0	3.0	12.4	-
14.0	3.8	34.2	x
10.5	4.2	16.4	x
10.5	3.2	21.2	x
<u>11.0</u>	<u>3.60</u>	<u>22.88</u>	

Controls 2.

14.5	5.0	13.0	x
13.5	4.8	12.2	-
11.0	2.8	12.2	x
16.5	3.8	13.0	x
10.5	3.6	9.0	x
14.0	3.2	13.4	-
10.0	3.2	5.2	-
10.5	3.6	6.8	-
<u>12.06</u>	<u>3.75</u>	<u>10.6</u>	

Series 3.

11.0	3.6	20.2	x
10.5	4.6	18.0	x
11.5	3.4	23.6	x
12.0	4.6	23.2	x
11.5	3.8	18.0	x
8.5	5.6	23.0	x
12.0	3.6	21.8	x
11.0	3.2	22.8	x
11.0	5.6	15.6	x
<u>11.0</u>	<u>4.2</u>	<u>20.7</u>	

Controls 3.

Controls same as Series 1.

Series 4.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
10.5	4.4	37.4	x
13.0	4.2	42.0	x
10.0	4.2	43.2	x
9.5	2.4	25.0	x
10.5	3.6	32.0	x
10.0	3.2	35.0	x
9.5	3.0	19.4	x
9.5	4.0	26.0	x
11.0	4.8	28.6	x
8.5	3.6	28.0	x
<u>12.5</u>	<u>3.6</u>	<u>37.4</u>	x
10.4	3.72	32.2	

Controls 4.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
10.0	3.8	12.4	-
10.5	3.2	10.0	-
12.0	3.8	10.0	-
11.5	3.8	10.2	-
10.5	2.6	11.4	-
10.0	2.6	9.4	-
10.5	2.4	11.0	-
10.5	3.6	11.4	-
10.0	2.8	12.4	-
9.5	3.6	7.6	-
11.5	4.0	9.2	-
<u>9.5</u>	<u>4.4</u>	<u>12.4</u>	-
10.5	3.38	10.6	

Series 5.

11.5	4.8	18.8	x
8.0	3.6	12.4	-
10.0	4.8	26.0	x
11.5	3.2	21.2	x
9.5	3.6	22.0	x
8.5	3.4	12.6	-
<u>10.5</u>	<u>3.4</u>	<u>22.0</u>	x
9.93	3.83	19.3	

Controls 5.

11.0	3.6	8.4	-
10.5	2.8	7.8	-
9.5	2.8	7.6	-
9.5	3.4	5.8	-
9.0	2.6	5.8	-
10.5	3.0	8.2	-
9.5	4.2	8.4	x
9.5	3.4	8.2	-
9.0	1.6	3.6	-
<u>8.5</u>	<u>2.0</u>	<u>6.0</u>	-
9.65	2.94	7.08	

Series 6.

11.5	3.8	15.8	x
15.0	4.6	39.8	x
11.5	5.4	18.2	x
12.0	3.4	18.0	-
<u>11.5</u>	<u>4.4</u>	<u>17.2</u>	x
12.3	4.3	21.80	

Controls 6.

11.0	3.8	7.8	-
12.5	5.0	12.2	-
11.0	4.0	7.2	-
11.5	5.6	10.0	-
<u>10.0</u>	<u>3.6</u>	<u>7.2</u>	-
11.2	4.4	8.8	

Series 7.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
11.0	5.8	26.8	x
10.0	3.2	29.6	x
11.0	3.0	16.2	x
11.5	4.6	26.4	x
11.5	4.6	25.7	x
12.0	5.0	37.0	x
12.0	2.0	22.0	x
11.0	2.8	17.3	x
12.0	4.2	23.4	x
11.0	4.4	22.9	x
<u>11.3</u>	<u>3.96</u>	<u>24.6</u>	

Controls 7.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
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Same as Series 1.

Series 8.

11.5	2.4	9.2	x
14.0	4.7	21.6	x
12.0	5.4	14.0	-
13.0	5.6	15.4	x
13.5	3.8	16.6	-
11.5	2.4	13.0	-
13.5	4.8	12.0	x
11.5	3.3	15.8	x
12.0	4.8	11.6	-
13.0	3.0	13.3	-
<u>12.5</u>	<u>4.02</u>	<u>14.25</u>	

Controls 8, 9.

14.0	4.8	19.0	x
11.0	4.0	10.6	-
12.5	4.3	13.2	-
11.5	3.2	11.0	-
12.0	3.4	12.5	x
13.5	5.2	17.4	-
13.5	4.0	16.4	-
11.5	2.3	15.1	-
11.0	3.4	11.8	x
12.5	3.5	14.6	-
12.5	3.8	16.0	-
13.0	5.6	19.2	x
<u>12.5</u>	<u>3.6</u>	<u>13.9</u>	-
<u>12.4</u>	<u>3.9</u>	<u>14.67</u>	

Series 9.

13.5	2.4	12.4	-
11.5	3.6	10.2	-
12.5	4.8	11.0	x
13.5	2.8	10.6	x
12.5	4.4	11.2	-
13.0	3.3	15.0	-
11.0	3.2	10.8	x
13.5	3.4	13.8	x
12.5	3.8	15.0	-
<u>14.0</u>	<u>6.8</u>	<u>15.6</u>	x
<u>12.7</u>	<u>3.85</u>	<u>12.56</u>	

Controls 9.

Same as Series 8.

Series 11.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
12.0	3.4	24.6	x
13.5	3.8	12.8	x
11.5	4.8	21.1	x
14.0	3.6	17.4	x
13.0	3.4	24.4	x
12.5	3.8	24.0	x
12.5	5.3	21.2	x
12.5	5.2	42.0	x
12.0	4.2	31.4	x
14.0	5.8	28.6	x
<u>11.7</u>	<u>4.33</u>	<u>24.75</u>	

Controls 11.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
12.0	5.0	11.8	x
11.5	4.2	10.6	-
12.0	3.8	8.4	x
12.5	3.6	13.4	-
14.0	5.8	24.6	x
12.0	3.4	16.4	x
13.0	4.0	17.0	-
11.5	4.8	13.8	x
<u>12.3</u>	<u>4.32</u>	<u>14.5</u>	

Series 12.

12.0	3.5	10.6	x
14.0	3.4	16.3	-
11.5	3.4	22.0	x
10.5	3.6	20.0	x
13.0	3.6	18.3	x
11.5	3.2	12.8	-
11.5	4.2	20.6	-
10.5	3.2	12.8	-
12.0	2.4	16.6	-
12.5	4.2	16.7	-
<u>11.9</u>	<u>3.47</u>	<u>16.67</u>	

Controls 12, 13.

12.5	3.6	8.0	-
13.0	4.8	12.6	x
10.0	4.2	8.4	-
12.0	3.0	9.4	-
11.5	4.6	8.7	-
12.0	4.4	10.2	-
12.0	3.4	8.4	-
10.0	2.8	8.4	-
10.5	4.0	8.6	x
11.5	2.0	10.6	-
<u>11.5</u>	<u>3.68</u>	<u>9.3</u>	

Series 13.

13.5	4.6	21.0	-
13.6	3.2	9.5	-
13.5	3.5	11.8	-
12.5	4.6	11.4	-
14.0	4.2	13.4	-
12.5	4.2	12.4	-
13.5	3.8	11.6	x
10.0	2.6	9.2	-
13.5	4.6	10.8	x
12.5	3.2	9.8	x
<u>12.85</u>	<u>3.85</u>	<u>12.09</u>	

Controls 13.

Same as Series 12.

Series 14.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
10.0	3.2	19.8	x
12.5	3.6	15.8	x
13.0	3.8	18.4	x
12.0	3.9	18.4	x
13.5	3.2	23.9	x
14.5	5.2	27.6	x
10.5	3.5	17.3	x
11.0	3.5	20.2	x
13.5	3.8	35.0	x
<u>11.16</u>	<u>3.74</u>	<u>21.8</u>	

Controls 14.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
11.0	4.4	9.6	-
10.0	4.7	10.4	-
13.5	3.6	19.0	x
10.5	3.6	10.2	-
13.5	5.6	15.2	-
11.0	4.0	12.6	-
11.0	4.0	11.2	-
12.0	5.3	10.2	-
9.5	3.8	8.2	-
13.5	5.3	10.8	-
11.5	5.3	13.6	-
13.0	4.8	14.2	-
<u>11.6</u>	<u>4.53</u>	<u>12.1</u>	

Series 15.

11.0	3.4	20.6	x
9.5	3.7	23.0	x
15.0	4.6	23.8	x
<u>11.83</u>	<u>3.9</u>	<u>22.46</u>	

Controls 15.

15.5	5.6	17.0	-
11.0	4.0	11.0	x
11.0	5.4	11.0	x
14.0	5.3	14.6	-
10.0	4.6	10.2	-
10.0	4.8	10.0	-
<u>11.91</u>	<u>4.95</u>	<u>12.30</u>	

Series 16.

10.0	3.8	25.6	-
14.5	3.4	20.2	x
13.5	3.5	24.8	-
11.0	3.0	10.4	-
10.0	3.5	19.4	x
10.0	3.5	19.4	x
10.0	4.2	14.6	-
<u>11.5</u>	<u>3.56</u>	<u>20.66</u>	

Controls 16.

11.0	4.6	12.8	-
14.0	4.6	13.4	-
9.5	4.4	10.0	-
10.5	5.6	8.8	-
10.5	5.2	7.6	-
10.5	5.2	7.6	-
9.5	4.0	8.6	-
13.5	5.0	15.8	-
<u>11.21</u>	<u>4.77</u>	<u>11.0</u>	

Series 17.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
12.0	3.4	19.4	-
10.5	4.2	18.2	x
9.0	3.4	12.6	x
10.0	4.6	17.6	x
11.0	5.4	23.8	x
8.5	5.3	14.6	x
9.0	4.6	15.2	-
9.5	5.3	18.2	x
<u>9.93</u>	<u>4.52</u>	<u>17.45</u>	

Controls 17.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
10.5	3.2	7.8	-
10.5	5.3	10.7	-
14.0	5.3	16.0	-
10.5	4.6	9.4	x
11.0	4.7	10.2	-
11.5	4.2	12.6	-
10.5	4.6	7.8	-
9.0	3.6	9.6	-
<u>11.93</u>	<u>4.43</u>	<u>10.51</u>	

Series 19.

13.5	4.6	22.3	x
10.5	3.4	14.2	x
13.0	3.6	21.4	x
11.5	4.2	16.2	x
13.0	3.4	19.3	x
11.0	5.0	15.4	x
10.5	4.7	12.6	x
10.0	2.8	13.6	-
11.5	2.8	14.8	x
<u>12.5</u>	<u>3.4</u>	<u>26.2</u>	x
<u>11.7</u>	<u>3.79</u>	<u>17.6</u>	

Controls 19.

13.5	3.4	11.2	-
10.0	3.8	11.4	-
10.5	4.6	9.8	-
9.5	4.0	9.6	-
11.0	3.5	15.4	-
10.5	3.5	10.3	-
10.5	3.6	11.2	-
10.5	3.8	10.0	-
9.0	3.0	9.6	-
<u>10.55</u>	<u>3.69</u>	<u>10.94</u>	

Series 20.

14.0	4.4	34.0	x
14.0	3.7	30.2	x
15.0	4.8	19.3	x
13.5	5.3	19.4	x
<u>14.1</u>	<u>4.45</u>	<u>25.67</u>	

Controls 20.

12.0	3.8	9.6	-
13.0	4.6	16.2	-
15.0	4.2	14.6	-
13.0	4.8	10.2	-
13.0	3.5	10.6	-
12.5	4.6	10.2	-
<u>13.08</u>	<u>4.25</u>	<u>11.9</u>	

Series 21.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
11.0	4.6	16.4	-
11.0	3.2	14.8	x
13.0	4.0	16.4	x
12.5	5.0	17.6	x
10.0	4.8	13.4	x
12.0	5.2	17.6	x
11.5	4.8	17.4	x
13.0	3.8	24.6	x
13.5	4.6	23.4	x
12.5	3.6	18.0	x
<u>11.0</u>	<u>4.36</u>	<u>17.96</u>	

Controls 21.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
14.0	3.8	17.6	-
13.0	4.2	12.5	x
12.0	3.8	14.6	x
11.0	4.6	16.2	x
11.5	4.6	16.6	-
10.0	4.8	12.6	x
11.0	5.2	11.9	-
10.0	5.0	9.8	-
11.5	4.2	13.7	-
11.0	4.2	12.2	-
10.0	3.8	7.8	-
<u>11.36</u>	<u>4.38</u>	<u>13.2</u>	

Series 22.

13.0	4.4	14.7	-
12.0	4.6	11.8	-
12.0	4.4	10.8	-
14.0	3.6	13.3	-
13.0	5.2	11.4	-
12.0	3.6	15.6	-
12.5	5.0	11.4	-
<u>12.64</u>	<u>4.4</u>	<u>12.71</u>	

Controls 22.

14.0	5.4	22.2	-
13.0	4.2	13.7	-
13.5	4.2	11.4	-
12.5	3.4	9.2	-
13.0	4.2	14.4	-
13.0	5.0	16.4	-
<u>13.16</u>	<u>4.4</u>	<u>14.55</u>	

Series 23.

11.5	4.0	11.8	x
13.0	3.8	11.8	x
11.0	4.2	10.8	-
11.5	4.2	10.4	x
11.0	4.2	12.2	-
11.0	3.5	10.2	-
11.5	4.0	11.2	x
<u>11.5</u>	<u>3.98</u>	<u>11.2</u>	

Controls 23.

10.0	5.0	7.7	-
11.5	3.8	9.4	-
10.5	4.4	8.4	-
11.0	6.0	12.2	-
10.0	4.0	6.8	-
10.5	4.6	9.2	-
10.5	4.0	7.6	-
<u>10.57</u>	<u>4.54</u>	<u>8.74</u>	

Series 24.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
11.0	2.8	14.6	-
13.0	4.6	16.5	x
12.5	4.0	17.2	x
10.5	2.6	12.5	-
13.0	3.6	14.6	x
9.0	4.2	13.7	x
12.0	6.2	17.4	x
13.0	4.2	15.4	x
<u>11.75</u>	<u>4.02</u>	<u>15.23</u>	

Controls 24, 28.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
12.0	4.7	9.2	-
13.5	4.8	11.8	x
9.0	5.2	12.4	x
12.5	3.4	12.4	x
10.0	3.8	8.0	-
10.5	4.8	11.4	-
10.5	3.2	9.5	-
11.5	4.3	11.6	-
12.0	6.6	14.2	-
11.0	3.2	8.2	-
11.5	4.6	11.4	x
<u>11.27</u>	<u>4.41</u>	<u>10.91</u>	

Series 25.

13.5	4.0	20.2	x
13.0	3.6	17.4	x
11.5	3.5	17.8	-
11.5	3.4	16.6	-
14.0	4.6	18.2	x
12.5	4.4	17.6	x
<u>12.66</u>	<u>3.91</u>	<u>17.96</u>	

Controls 25.

14.5	4.8	10.4	-
11.5	3.5	12.4	-
11.5	4.2	10.6	-
11.5	2.8	9.2	-
11.0	3.8	10.4	-
<u>12.0</u>	<u>3.82</u>	<u>10.6</u>	

Series 26.

11.0	8.0	32.4	x
8.5	10.0	25.4	x
12.0	6.4	41.3	x
10.0	6.0	38.2	x
14.5	9.5	33.4	x
13.5	9.3	31.6	x
<u>11.58</u>	<u>8.20</u>	<u>33.71</u>	

Controls 26.

9.5	3.6	10.6	-
14.0	4.2	16.8	-
12.5	4.2	11.4	-
14.0	3.8	13.6	-
9.5	3.8	11.2	-
11.0	4.6	14.7	-
9.0	4.2	10.6	-
10.0	4.0	11.8	-
9.5	3.8	11.8	-
<u>11.0</u>	<u>4.13</u>	<u>12.50</u>	

Series 27.

13.5	5.2	28.0	x
10.5	5.4	25.4	x
11.0	5.0	22.6	x
12.0	3.4	31.3	x
11.5	5.3	35.8	x
<u>11.7</u>	<u>4.86</u>	<u>28.62</u>	

Controls 27.

10.5	3.2	7.8	-
10.5	5.3	10.7	-
14.0	5.3	16.0	-
10.5	4.6	9.4	x
11.0	4.7	10.2	-
11.5	4.2	12.6	-
10.5	4.6	7.8	-
9.0	3.6	9.6	-
<u>11.93</u>	<u>4.43</u>	<u>10.51</u>	

Series 28.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
11.5	4.0	21.0	x
10.0	4.9	20.8	x
13.0	4.2	33.8	x
13.5	3.8	21.3	x
12.0	4.9	26.8	x
10.0	2.8	15.0	x
11.5	5.6	16.6	x
<u>11.64</u>	<u>4.13</u>	<u>22.18</u>	

Series 29.

11.5	3.3	18.8	x
11.5	4.8	43.0	x
10.5	3.2	15.2	x
10.0	3.2	19.5	x
9.5	2.6	12.0	x
<u>10.6</u>	<u>3.42</u>	<u>21.70</u>	

Series 30.

10.0	3.4	19.4	x
11.0	2.6	14.6	x
16.5	4.8	36.7	x
10.0	3.8	21.2	x
12.5	3.6	13.6	x
13.5	3.4	19.6	x
10.5	3.5	19.8	x
9.5	2.4	10.6	-
9.5	3.8	21.0	x
<u>11.44</u>	<u>3.47</u>	<u>19.61</u>	

Series 31.

13.0	3.8	12.6	x
14.0	3.6	15.6	-
11.0	3.8	12.4	-
11.5	4.3	21.2	-
11.0	3.4	14.6	-
11.0	3.4	16.0	-
<u>11.9</u>	<u>3.71</u>	<u>15.40</u>	

Controls 28.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
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Same as Controls 24.

Controls 29.

11.5	4.0	9.8	-
12.5	5.2	17.4	x
10.5	3.2	13.2	-
12.5	3.4	7.2	-
12.0	3.6	16.3	-
10.0	4.0	11.2	-
<u>11.5</u>	<u>3.40</u>	<u>12.51</u>	

Controls 30, 34.

17.0	4.8	16.4	-
8.0	3.0	5.6	-
14.5	4.8	16.4	-
11.5	5.4	10.6	-
10.0	3.4	6.8	-
14.0	3.4	12.0	-
10.0	3.5	6.8	-
10.5	2.8	7.8	-
10.5	4.8	11.4	x
8.5	4.4	5.0	-
<u>11.45</u>	<u>4.03</u>	<u>9.88</u>	

Controls 31.

13.0	4.8	20.0	-
13.0	5.3	17.4	-
12.0	4.6	16.8	-
10.5	3.4	11.2	-
11.0	4.6	11.0	-
13.0	4.2	19.4	-
12.5	5.2	9.0	-
11.5	3.8	11.6	-
11.5	3.5	9.8	-
<u>12.00</u>	<u>4.37</u>	<u>14.02</u>	

Series 32.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V.O.</u>
12.0	3.2	13.4	-
11.5	3.8	12.6	x
10.5	2.4	12.0	-
11.0	2.4	8.8	-
<u>11.25</u>	<u>2.95</u>	<u>11.70</u>	

Series 33.

11.0	3.4	15.6	x
9.5	3.4	20.0	x
10.5	5.6	24.2	x
11.5	4.2	23.8	x
<u>10.6</u>	<u>4.15</u>	<u>20.90</u>	

Series 34.

13.5	3.5	14.8	x
11.0	2.8	17.8	x
9.0	2.4	13.3	-
<u>11.16</u>	<u>2.9</u>	<u>15.30</u>	

Series 35.

14.5	4.2	13.6	x
9.5	3.2	18.4	x
13.5	3.8	23.0	x
13.5	3.2	19.0	x
9.5	2.8	15.4	-
<u>12.10</u>	<u>3.44</u>	<u>18.88</u>	

Series 36.

13.5	3.2	36.4	x
11.5	3.6	32.0	x
11.5	2.4	17.2	x
11.5	4.6	26.0	x
11.5	4.2	33.4	x
14.0	4.2	31.4	x
10.0	3.5	20.2	x
<u>10.5</u>	<u>3.67</u>	<u>28.08</u>	

Controls 32.

<u>B.W.</u>	<u>O.W.</u>	<u>U.W.</u>	<u>V. O.</u>
11.0	3.3	13.4	-
11.5	3.6	17.4	-
11.0	4.0	11.6	x
10.5	3.5	10.2	-
7.5	3.5	7.6	-
10.5	2.6	12.8	-
<u>10.33</u>	<u>3.41</u>	<u>12.16</u>	

Controls 33.

9.5	3.4	10.2	x
9.0	3.3	7.6	-
11.0	4.8	11.8	-
8.5	3.4	8.4	-
13.5	4.9	17.4	x
<u>10.3</u>	<u>3.96</u>	<u>11.08</u>	

Controls 34.

Same as Series 30.

Controls 35.

10.5	3.5	14.6	-
12.5	3.5	9.6	-
14.0	4.0	15.2	-
11.0	3.0	8.4	-
14.5	4.4	23.8	x
<u>12.5</u>	<u>3.68</u>	<u>23.8</u>	

Controls 36.

13.0	3.6	11.2	-
12.5	4.6	10.4	x
9.5	3.2	6.8	-
12.5	4.0	10.8	-
10.5	3.4	9.4	-
12.5	4.2	15.6	-
9.5	3.7	10.3	-
11.5	5.3	10.8	-
<u>11.4</u>	<u>4.00</u>	<u>10.66</u>	