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## The Leucocytic Response of Swine to Stilbestrol and a Progesterone-Estradiol Combination

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# The Leucocytic Response of Swine to Stilbestrol and a Progesterone-Estradiol Combination

B. N. DAY, J. L. LASLEY, H. E. ADDLEMAN, AND L. F. TRIBBLE

The cellular components of the blood have been studied for many years in connection with various diseases of man and domestic animals. These investigations have revealed that many disorders in the normal physiology of animals are accompanied by simultaneous changes in the types and proportions of circulating cells in the vascular system. Extensive application of this knowledge has been made in the medical field as a means of studying both normal physiological processes and pathological conditions.

Few physiological studies have been made of the blood components of swine. Little is known of the normal circulating level of leucocytes in swine produced under current management conditions. Further, investigations made on the cellular components of the blood would enhance the establishment of an accepted average blood cell count for swine. This information would be of value to the research investigator and to the veterinary clinician in the interpretation of variations observed in the cellular components of blood samples.

Sex differences exist in many performance traits. The direct causes of these variations are not clearly understood. Additional knowledge regarding the cause and effect relationships between sex hormones and performance traits may provide insight into the complexities of the growth processes. Observations made on the cellular components of the blood may serve as a common denominator *for measuring variations in both hormone secretion rates and growth*. The existence of a close association among these factors would provide a useful tool in the selection of genetically superior animals and, in addition, would be of assistance in the clarification of the effect of sex hormones on the rate and type of growth of swine.

The primary objectives of the present studies were (1) to obtain additional data on the number and proportion of circulating leucocytes in swine (2) to determine the leucocytic response of swine to various exogenous hormones and to mechanical stress (3) to study the relationship between variations in the circulating leucocytes and individual differences in certain economic traits.

## LITERATURE REVIEW

Previous investigations made on the number and kinds of circulating leucocytes in swine have been designed, primarily, to determine average values for

\*This work was conducted in cooperation with the Regional Swine Breeding Laboratory, AHRD, ARS, USDA.

these cellular components of the blood which are characteristic of the species. These studies have revealed that leucocyte counts made on blood samples collected from the peripheral circulatory system are subject to extreme fluctuations even under apparently homeostatic conditions. Age and sex have been found to be contributing factors to this variation.

Palmer (1917) stated that differential leucocyte counts were made in twenty-five examinations of the blood of normal pigs between the ages of two and forty-two days. The total leucocyte count for young animals averaged 12,328 as compared to 18,533 in older pigs. A sex difference was noted also which showed a higher leucocyte count in the males than females. Young males showed an average leucocyte count of 12,857 as compared with 11,800 in females. Older males had 1,184 more leucocytes per cubic millimeter of blood than did older females. Young males showed higher counts of lymphocytes and monocytes and fewer neutrophils than older males. However the differences between sexes in differential counts were not statistically significant.

Luke (1953) examined a series of 110 blood samples comprising 30 bacon-type pigs eight to nine months of age and 30 sows. The total leucocyte count for pigs averaged 13,747 as compared to an average of 15,900 for the group of 30 sows. The average percentages of lymphocytes in the blood samples collected from pigs and sows were 71 and 63, respectively. In 53 samples collected from newly born pigs, only 38% of the leucocytes were classified as lymphocytes.

Romic (1948), in a study of the blood of 120 pigs, reported that more lymphocytes and neutrophils were present in females than in males, but a higher concentration of monocytes was found in males. No sex difference was detected in total leucocyte number, number of eosinophils, or number of basophils. The average total leucocyte count of these pigs was 18,902.

Studies pertaining to the leucocyte numbers in swine that were made prior to 1926 have been reviewed by Scarborough (1931). The averages for 140 counts revealed that the total number of leucocytes in swine was 15,820 per cubic millimeter of blood, with a normal range of about 8,000 to 20,000. The approximate normal averages for the differential counts were: lymphocytes, 52.1%; polymorphonuclears, 39.0%; large mononuclears and transitionals, 3.3%; eosinophils, 4.5%; and basophils, 1.2%. Young pigs were reported to have 20 to 50% more leucocytes than adult animals which is in contrast to the findings of Palmer (1917) and Luke (1953).

The leucocytic response of pigs to exercise has been investigated by Palmer (1917a). Shortly after exercise, the following changes in differential counts were noted; lymphocytes and eosinophils decreased by an average of 12.6 and 2.3%, respectively; neutrophils increased an average of 15.1%; a slight increase occurred in the mononuclears and a slight decrease was found in mast cells. The extremes of the response for lymphocytes and polymorphs occurred at an average of four hours following exercise.

## EXPERIMENT I. Leucocytic Response to Stilbestrol and Mechanical Stress in Boars, Barrows, Gilts, and Spayed Gilts.

### *Experimental Procedure*

A total of 64 pigs of a Landrace x Poland first cross were studied. The experiment was designed to select all pigs on test from eight litters with each litter being represented by 2 boars, 2 barrows, 2 gilts, and 2 spayed gilts. The only exceptions to the basic experimental design occurred when a control gilt for one litter and a control barrow for a different litter were not represented in the experiment and a spayed gilt for one litter and a barrow for another litter were not available for the treated group. The experimental animals were assigned to a total of 8 pens of 8 animals each according to litter number and sex class.

One lot of animals of each sex class was fed a basal ration and replicated lots of animals were fed the same ration plus stilbestrol at a level of 0.25 mg. per pound of feed.

The average body weight of the pigs at the time the blood samples were taken was approximately 160 pounds. All animals had been fed the experimental rations in dry-lot for a period of approximately 3 months. All blood samples were collected during the month of February. The pigs had not been subjected to any undue excitement for several days prior to the initiation of the present investigation.

Blood samples were collected by holding the pig by the snout with a hog catcher while the distal end of the tail was severed and a 5 to 10 cc. sample of blood was collected. A second sample was collected one hour after the initial blood sample. All pigs were bled between 7:30 A.M. and 1:30 P.M.

Duplicate dry smears for the leucocyte differential count were made immediately after the sample was collected. The differential counts were made under oil immersion and 100 cells were counted on each slide. Total leucocyte counts were made by diluting the blood to a ratio of 1:19 with a 1% solution of hydrochloric acid and then determining the concentration of cells in this solution with a hemocytometer. Other data collected in this study were backfat thickness, growth rate, and efficiency of gain.

### *Results and Discussion*

This investigation of the influence of stilbestrol on circulating leucocytes was made in conjunction with a study of the effect of stilbestrol on growth and carcass characteristics of swine as reported by Tribble, *et al.*, (1958). It was felt that any variations induced in performance and body composition by feeding stilbestrol may be associated with similar changes in leucocytes since sex differences have been reported for all of these traits. It was also felt that the exogenous stilbestrol would possibly increase any existing sex difference in leucocytes due to the presence or absence of female sex hormones. The magnification of these differences would then make a study of the association between the level of leucocytes and rate and efficiency of gain between sexes more definitive.

Mechanical stress was applied to all animals in an effort to measure the influence of sex hormones on the animals' ability to adjust to adverse conditions. Variations between sexes in ability to maintain a homeostatic level may also be exhibited as differences in economic traits. One measurement that is available to determine the degree of hormone response to stress is changes in the numbers and kinds of circulating leucocytes. A variety of stress inducing agents will cause an increase in the secretion rate of adrenocorticotrophic hormone (ACTH) by the anterior lobe of the pituitary. ACTH will then bring about increased activity of the adrenal cortex with a subsequent increase in production of adrenocortical hormones. Hormones produced by the adrenal cortex will then induce changes in the kinds and numbers of circulating leucocytes. The general leucocytic reaction to physical stress is an increase in the total number of leucocytes and the percentage of neutrophils with a decrease in the proportion of circulating lymphocytes and eosinophils.

TABLE 1-INFLUENCE OF STILBESTROL ON THE AVERAGE NUMBER OF CIRCULATING LEUCOCYTES IN PIGS OF DIFFERENT SEXES, BEFORE AND AFTER MECHANICAL STRESS

Treatment Sample	Control		Stilbestrol	
	Initial	1 Hour	Initial	1 Hour
Gilts	16,281	19,419	16,612	19,162
Spayed gilts	17,569	19,838	17,956	19,631
Boars	19,882	20,444	19,569	19,125
Barrows	17,938	18,750	18,319	18,762
Average	17,917	19,613	18,114	19,170

### *Total Number of Leucocytes*

As shown in Table I, the total number of leucocytes per  $\text{mm}^3$  of blood in the initial sample was slightly higher in the stilbestrol-treated barrows, gilts, and spayed gilts than observed in the control animals. Boars receiving stilbestrol showed a slightly lower leucocyte count than did the control boars. Differences found between the average number of leucocytes per  $\text{mm}^3$  of blood for the control and stilbestrol-treated lots were not statistically significant, however. Generally, differences between spayed and intact gilts were not significantly different and the trend observed was in the opposite direction to that existing for the animals receiving stilbestrol.

Mechanical stress applied to the experimental animals during the process of collecting the initial blood samples induced a slight increase in the average number of leucocytes per  $\text{mm}^3$  of blood. This increase was observed in all lots of animals although these differences were not significant. Stilbestrol had no apparent effect on the degree of leucocytic response to mechanical stress. Females showed a larger increase in the number of leucocytes after stress than did males. However, on further analysis of these data, it was found that this difference in response was likely not one that was due to sex difference. Correlations were com-

puted between the initial count and the change and number of white cells from zero to one hour. The calculations revealed that a negative association existed between the two bleedings which was statistically significant. Animals having the lowest number of white cells initially showed the largest response to mechanical stress which appears to explain more adequately the increased leucocytic response observed in gilts and spayed gilts.

TABLE 2—INFLUENCE OF STILBESTROL ON THE PERCENTAGE AND NUMBER OF CIRCULATING LYMPHOCYTES IN PIGS OF DIFFERENT SEXES BEFORE AND AFTER MECHANICAL STRESS

Treatment Sample	Control		Stilbestrol	
	Initial	1 Hour	Initial	1 Hour
Percentage of Lymphocytes				
Gilts	72.5	69.5	69.9	65.4
Spayed gilts	68.8	63.1	65.0	72.6
Boars	71.5	70.1	70.9	69.4
Barrows	69.8	68.1	73.4	63.6
Average	70.6	67.7	69.8	67.8
Number of Lymphocytes per mm <sup>3</sup> of Blood				
Gilts	11,800	13,542	11,584	12,507
Spayed gilts	12,087	12,605	11,667	14,324
Boars	14,467	14,402	13,810	13,698
Barrows	12,520	12,733	13,454	11,840
Average	12,718	13,320	12,629	13,092

TABLE 3—INFLUENCE OF STILBESTROL ON THE PERCENTAGE AND NUMBER OF CIRCULATING NEUTROPHILS IN PIGS OF DIFFERENT SEXES BEFORE AND AFTER MECHANICAL STRESS

Treatment Sample	Control		Stilbestrol	
	Initial	1 Hour	Initial	1 Hour
Percentage of Neutrophils				
Gilts	19.1	24.1	23.9	27.4
Spayed gilts	23.4	28.0	27.1	23.8
Boars	21.6	23.8	24.4	25.0
Barrows	26.4	27.2	22.1	30.0
Average	22.6	25.8	24.4	26.6
Number of Neutrophils per mm <sup>3</sup> of Blood				
Gilts	3,109	4,644	3,995	5,274
Spayed gilts	4,116	5,417	4,883	4,611
Boars	4,035	4,794	4,828	4,959
Barrows	4,718	5,140	4,025	5,717
Average	3,994	4,999	4,433	5,140

*Lymphocytes, Neutrophils, Eosinophils, Basophils, and Monocytes.*

Differential counts made on initial blood samples indicate that the addition of stilbestrol to the ration did not influence the proportion of lymphocytes,

TABLE 4—INFLUENCE OF STILBESTROL ON THE PERCENTAGE AND NUMBER OF CIRCULATING EOSINOPHILS IN PIGS OF DIFFERENT SEXES BEFORE AND AFTER MECHANICAL STRESS

Treatment Sample	Control		Stilbestrol	
	Initial	1 Hour	Initial	1 Hour
	Percentage of Eosinophils			
Gilts	3.5	1.8	2.2	3.4
Spayed gilts	2.8	2.9	3.8	3.1
Boars	4.2	2.9	2.9	3.1
Barrows	2.5	2.2	2.4	3.0
Average	3.2	2.4	2.8	3.2
	Number of Eosinophils per mm <sup>3</sup> of Blood			
Gilts	584	344	372	652
Spayed gilts	488	585	664	594
Boars	847	572	542	620
Barrows	456	414	438	568
Average	594	479	504	608

neutrophils or eosinophils in the blood. As shown in Tables 2, 3, and 4, the percentages and numbers of lymphocytes, neutrophils, and eosinophils per cubic millimeter of blood were essentially the same in the control and stilbestrol groups of pigs when the means were computed over sex classes. Gilts, spayed gilts, and boars fed stilbestrol had a consistently lower concentration of leucocytes and a higher percentage of neutrophils than did the controls, but the differences were small in magnitude and were not statistically significant. Assuming sex differences that have been reported for swine previously were due to variations in types and amount of sex hormones secreted, the addition of stilbestrol to the ration failed to stimulate this effect. Many variables may contribute to this disagreement of which dose level and type of hormone involved may be of major significance.

A comparison of the differential counts made on the initial and one-hour blood samples reveals that mechanical stress induced rather consistent changes in the concentration of the various white cell types in the blood. The percentage of circulating lymphocytes had decreased by approximately 2.5% in the one-hour blood sample. The percentage of neutrophils was increased by approximately the same amount. Control animals showed a slight decrease in the proportion of eosinophils following stress, but the one-hour sample of blood collected from animals fed stilbestrol showed an increase in the concentration of eosinophils (Table 4). When the degree of change in the number of eosinophils per mm<sup>3</sup> of blood was considered, it was found that the control and stilbestrol means were significantly different ( $P < .05$ ).

Counts made on the concentration of monocytes and basophils in the blood samples were extremely erratic. The cause of the wide range observed between the animals within treatments was not evident and it was felt that these data were of limited value in making any further comparative studies between treat-



TABLE 5-INFLUENCE OF STILBESTROL ON THE AVERAGE PERCENTAGE AND NUMBER OF CIRCULATING MONOCYTES AND BASOPHILS OF SWINE BEFORE AND AFTER MECHANICAL STRESS

	Control		Stilbestrol	
	Initial	1 Hour	Initial	1 Hour
Monocytes, %	3.2	3.4	2.4	2.0
Number of Monocytes/ mm <sup>3</sup>	568	782	444	391
Basophils, %	0.4	0.7	0.6	0.4
Number of Basophils/ mm <sup>3</sup>	72	137	108	77

ments. The values of these cellular types are presented in Table 5, but no further comparisons have been made between the control and stilbestrol groups of animals or between sex classes.

The number of lymphocytes per mm<sup>3</sup> of blood was found to be the least variable of the cellular type studied (Table 6). Coefficients of variations computed for the lymphocyte counts were approximately one-third as large as those computed from the eosinophil counts and were about one-half as large as the neutrophil coefficients.

TABLE 6-COEFFICIENTS OF VARIATION COMPUTED FOR LEUCOCYTE COUNTS ON INITIAL BLOOD SAMPLES

	Control	Stilbestrol
Number of Animals	32	32
Total number of Leucocytes	14.2	13.4
Number of Lymphocytes	21.0	17.1
Number of Neutrophils	33.5	35.0
Number of Eosinophils	63.8	70.6

### *Influence of Sex on the Kinds and Numbers of Leucocytes.*

A study of the influence of the sex of the animal on the circulating leucocytes was made by disregarding treatment and combining the original eight classes of eight animals each into four classes with 16 animals represented in each class.

#### *Total Number of Leucocytes.*

The average numbers of white blood cells per mm<sup>3</sup> of blood in the initial samples, with regard to the sex of the animals, are presented in Table 7. The differences existing among the mean total white cell numbers were statistically significant ( $P < .01$ ). Further tests of these differences showed that boars had a significantly higher number of white cells per mm<sup>3</sup> of blood than did barrows, gilts, or spayed gilts. No significant differences were found among the remaining means for the various sexes. Similar results have been reported by Palmer (1917).

TABLE 7-INFLUENCE OF SEX ON THE PERCENTAGE AND NUMBER OF CIRCULATING LEUCOCYTES - INITIAL BLOOD SAMPLES

	Gilts	Spayed Gilts	Boars	Barrows
Number of Animals	16	16	16	16
Number of Leucocytes*	16,447	17,762	19,725	18,128
Percentages of Cell Types				
Lymphocytes	71.2	66.9	71.2	71.6
Neutrophils	22.0	25.2	23.0	24.2
Eosinophils	2.9	3.2	3.6	2.4
Monocytes	3.1	4.4	2.0	1.6
Basophils	0.8	0.3	0.2	0.2
Number of Cells per mm <sup>3</sup> of Blood				
Lymphocytes*	11,692	11,877	14,138	12,987
Neutrophils	3,552	4,500	4,431	4,371
Eosinophils	478	576	695	447
Monocytes	638	779	411	299
Basophils	125	51	40	36

\*Differences among sexes significant at the 5% level.

Considerable variation was observed in the leucocytic reaction of boars and barrows to stress. The degree of leucopenia due to stress was nearly as large in some animals as was the amount of leucocytosis that occurred in other animals within the same sex class.

#### *Lymphocytes, Neutrophils and Eosinophils.*

The proportions of the total white cells in the initial blood samples that were classified as lymphocytes, neutrophils and eosinophils are presented in Table 7, according to the four sex classes.

The average number of lymphocytes per mm<sup>3</sup> of blood was significantly higher in boars than in the two female classes. However, this difference was due primarily to the larger total white cell counts in boars, for little difference was observed among the sex classes in the percentage of lymphocytes. In contrast to these results, Romic (1948) found more lymphocytes in the blood of female pigs than males.

The average numbers of neutrophils and eosinophils were found to be similar in male and female pigs. As shown in Table 7, the differences observed between the mean number of neutrophils and eosinophils were mainly the result of variation in the total number of leucocytes and not due to differences in the proportions of these cell types. Romic (1948) reported that female pigs had a larger number of neutrophils per mm<sup>3</sup> of blood than did males.

#### *Influence of Litter on Total Number of Leucocytes.*

The number of white cells in the initial blood samples was compared with respect to the 8 litter groups to determine if the concentration of white cells and the degree of response to physical stress were genetically influenced. No significant differences were found among litters in the mean number of leucocytes

per mm<sup>3</sup> of blood or the average degree of response to mechanical stress. The lack of significance in this comparison indicates that the heritabilities of these characteristics are probably low.

### *Relationship of Leucocyte Counts with 154 Day Weight and Backfat Thickness.*

Coefficients of correlation between the number of leucocytes per mm<sup>3</sup> of blood and the 154 day body weight of the pigs were computed on the basis of numbers of white blood cells in the initial and one-hour blood samples, and the change in the number of leucocytes occurring as a result of stress.

The degree of association found between the traits studied indicate that these measurements of blood components are of limited value in predicting rate of gain in swine. The correlation coefficients computed between the 154 day weights and the leucocyte data on a within sex basis varied between .16 and .23 and all lacked statistical significance.

## EXPERIMENT II. Leucocytic Response of Barrows to Stilbestrol and a Progesterone-Estradiol Combination

### *Experimental Procedure.*

In a later investigation, blood studies were made on 47 Poland China barrows. The experimental treatments employed were as follows: (I) control; (II) 6 mg. stilbestrol; (III) 166.7 mg. progesterone and 3.3 mg. estradiol benzoate; (IV) 500 mg. progesterone and 10 mg. estradiol benzoate. The exogenous hormones were implanted subcutaneously in the ear when the average weight of all pigs on experiment was approximately 100 pounds. Blood samples were taken from each animal at a weight of approximately 200 pounds.

An initial blood sample was collected by restraining the animal with a hog catcher and severing the distal portion of the tail. A second sample was taken four hours after the initial collection. Differential and total white cell counts were made on the blood samples according to the procedure outlined previously in the present report. Computations were made on the basis of lymphocytes, neutrophils, and eosinophils, representing the total number of leucocytes.

### *Results and Discussion*

#### *Total Number of Leucocytes.*

As shown in Table 8, the total number of leucocytes per mm<sup>3</sup> of blood was not influenced to a detectable degree by the hormonal treatments.

The average number of leucocytes per mm<sup>3</sup> of blood in the initial samples was 20,491 as compared to an average of 21,291 leucocytes per mm<sup>3</sup> in the four-hour blood samples. A slight increase in the total white cells occurred in all treatment groups following the application of mechanical stress with the exception of the lots of barrows receiving a high level of the progesterone-estradiol preparation which showed a slight decrease in the concentration of leucocytes.

TABLE 8-TOTAL NUMBER OF CIRCULATING LEUCOCYTES BEFORE AND AFTER MECHANICAL STRESS (EXPERIMENT II)

Treatment Level	Control	Stilbestrol 6 mg.	Progesterone-estradiol	
			Low	High
Number of Barrows	12	11	12	11
Number Leucocytes/mm <sup>3</sup> , 0 hr.	20,433	20,109	18,988	22,575
Number Leucocytes/mm <sup>3</sup> , 4 hr.	21,812	21,250	20,312	21,832
Difference 4 and 0 hr. samples	1,379	1,141	1,324	-743

*Number of Lymphocytes, Neutrophils and Eosinophils.*

The proportion of lymphocytes, neutrophils and eosinophils observed in the blood samples collected from barrows before and after stress are presented in Table 9. No statistically significant differences were found among the treatment groups in the percentages of the different cell types. A trend toward a lower percentage of lymphocytes and a higher proportion of neutrophils was observed in barrows implanted with stilbestrol or 10 mg. of estradiol in combination with progesterone. An indication of this treatment effect was found in both the initial and four-hour blood samples.

TABLE 9-PERCENTAGE OF LYMPHOCYTES, NEUTROPHILS AND EOSINOPHILS BEFORE AND AFTER MECHANICAL STRESS (EXPERIMENT II)

Treatment Levels	Control	Stilbestrol 6 mg.	Progesterone-Estradiol	
			Low	High
Number of barrows	12	11	12	12
0 Hour Sample				
Lymphocytes, %	65.1	59.5	65.7	61.3
Neutrophils, %	29.2	34.1	28.8	30.2
Eosinophils, %	5.7	6.4	5.5	8.4
4 Hour Sample				
Lymphocytes, %	62.6	58.7	63.3	61.9
Neutrophils, %	31.5	35.9	31.9	31.2
Eosinophils, %	5.9	5.3	4.8	6.9

Small, but rather consistent, decreases in the percentages of lymphocytes and eosinophils, and increases in the percentage of neutrophils, were observed in the four-hour blood samples. Although small differences existed among the treatment groups in the type of leucocytic response to the mechanical stress, there did not appear to be any indication that animals receiving the exogenous hormones were more capable, or less capable, of adjusting to adverse conditions as measured by changes in the types of circulating leucocytes.

The numbers of lymphocytes, neutrophils and eosinophils per mm<sup>3</sup> of blood are presented in Table 10. The differences found among the treatment groups in the average number of the different types of leucocytes were not statistically significant with one exception; the number of eosinophils per mm<sup>3</sup> of blood in the initial sample. Barrows administered the high level of the pro-

TABLE 10-AVERAGE NUMBER OF LYMPHOCYTES, NEUTROPHILS AND EOSINOPHILS BEFORE AND AFTER MECHANICAL STRESS  
(EXPERIMENT II)

Treatment Levels	Control	Stilbestrol 6 mg.	Progesterone-Estradiol	
			Low	High
Number of barrows	12	11	12	11
	0 Hour Sample			
Lymphocytes/mm <sup>3</sup>	13,194	11,895	12,370	13,743
Neutrophils/mm <sup>3</sup>	6,080	6,935	5,598	6,924
Eosinophils/mm <sup>3*</sup>	1,158	1,274	1,014	1,899
	4 Hour Sample			
Lymphocytes/mm <sup>3</sup>	13,464	12,413	12,662	13,293
Neutrophils/mm <sup>3</sup>	7,072	7,694	6,710	7,018
Eosinophils/mm <sup>3</sup>	1,269	1,136	942	1,518

\*Significantly different at the 0.05 probability level.

gesterone-estrogen combination had a larger number of circulating eosinophils than did the other lots of barrows. The direct cause of this difference is not apparent as the group of barrows receiving a lower level of the same hormonal combination had the lowest number of eosinophils. A trend toward a higher concentration of eosinophils in the blood samples of barrows receiving the high level of progesterone and estradiol was also observed in the four-hour sample, but the differences were not statistically significant.

The differences observed between the four-hour and initial blood samples in the number of lymphocytes, neutrophils, and eosinophils, are similar to the variations found in the percentage values of these cells. In general, the number of lymphocytes and eosinophils decreased, whereas the number of neutrophils increased.

The average numbers of lymphocytes, neutrophils and eosinophils per mm<sup>3</sup> of blood in the initial sample for all animals employed in this experiment were 12,780; 6,361 and 1,325, respectively. Coefficients of variation computed from these data show that the eosinophil counts were the most variable—followed in the order of decreasing variability by neutrophils and lymphocytes, with the lymphocyte counts being considerably less variable than either the eosinophil or neutrophil counts. The ranges of the number of the various cell types counted in the initial blood samples were: lymphocytes 7,140 to 17,030; neutrophils, 2,135 to 13,560; and eosinophils, 330 to 3,840.

As shown in Table 11, correlation coefficients were computed between the leucocyte measurements in the initial blood sample and economic traits. The leucocyte counts were not significantly associated with rate of gain and carcass characteristics, with one exception. A significant relationship was found between backfat probe and percentage of eosinophils. Other correlations were computed which approached significance at the 5% level. Of these, the relationship of percentage of eosinophils with loin eye area and the correlation of percentages of neutrophils and lean cuts are of major interest. It is felt these data suggest that

TABLE 11-CORRELATION COEFFICIENTS BETWEEN ECONOMICS TRAITS AND LEUCOCYTE NUMBERS (EXPERIMENT II)

	Total Number of Leucocytes	Per Cent Lymphocytes	Per Cent Neutrophils	Per Cent Eosinophils
Rate of gain	-.03	.01	.00	.03
Backfat Probe	-.01	.24	-.25	.33*
Lean Cuts, %	.26	-.03	.28	.16
Loin eye area	.08	.06	-.02	-.26

\*Significant at the 5% level.

additional study with the development of new procedures for measuring the level of circulating leucocytes may result in the development of a means of adequately predicting carcass merit in the live animal.

### SUMMARY AND CONCLUSIONS

Initial blood samples were collected from 110 pigs approaching or at market weight which provided information on the number and proportion of circulating leucocytes in swine. Average number of leucocytes per cubic millimeter of blood was 19,051. Differential counts showed that approximately two-thirds of the leucocytes of swine are lymphocytes, about one-fourth are neutrophils, 4 to 5% eosinophils, 2 to 3% monocytes, and less than 1% basophils.

Comparisons were made in the numbers and proportions of circulating leucocytes of boars, barrows, gilts, and spayed gilts. The only statistically significant difference found among the sex means was in the number of lymphocytes per mm<sup>3</sup> of blood. Boars had a significantly higher count of lymphocytes than did barrows, gilts or spayed gilts.

Supplemental female sex hormones had little, if any, influence on the level and type of circulating leucocytes in swine with two possible exceptions. A higher level of circulating eosinophils was found in barrows implanted with a high level of progesterone and estradiol benzoate. Also, animals fed stilbestrol exhibited a significantly different response to mechanical stress than did the control pigs.

Mechanical stress applied by collecting an initial blood sample from each animal induced a decrease in the proportion of lymphocytes and eosinophils and an increase in the percentage of neutrophils and the total number of leucocytes per cubic millimeter of blood. Animals receiving exogenous hormone showed no detectable variation from the control pigs in the type of leucocytic response induced by stress. Also, the change in cell numbers following stress was not associated closely enough with performance traits to be used for prediction purposes. However, it is felt that a more pronounced reaction such as that produced by the injection of stress-inducing hormones should be obtained before a conclusive evaluation is made of the possible association between performance traits and the pituitary-adrenal cortex-leucocyte complex.

Associations were computed to determine if a close relationship existed between economic traits and the various blood components studied. Carcass characteristics and rate of gain were not significantly correlated with the number and kind of circulating leucocytes. The percentage of eosinophils was found to be significantly correlated with backfat thickness in one phase of the investigation. Other correlations that approached significance suggested a tendency for variations in the concentration of leucocytes to be associated with differences in carcass meatiness.

As has been reported for other species, considerable variation was observed within treatment groups for all types of leucocyte counts made in these studies. It seems apparent that this variation may have operated in a manner that would conceal real differences existing between experimental groups in the number and kinds of leucocytes.

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