groups. When other groups were maintained on diets containing 1% cholesterol for 6 months, the severity of atherosclerosis was increased, and inclusion of 10% safflower oil in the diets exerted no prophylactic effect. When cholesterol was removed from the diets, and the birds were maintained for an additional 12 months on either pellets alone, or pellets plus 10% safflower oil, there was no decrease in the level of aorta cholesterol and a slight decrease in aortic atherosclerosis.

1. Katz, L. N., Stamler, J., Pick, R., *Nutrition and Atherosclerosis,* Lea & Febiger, Philadelphia, 1958.

2. Clarkson, T. B., Prichard, R. W., Netsky, M. G., Lofland, H. B., *A.M.A. Arch. Path.,* 1959, v68, 143.

3. Lofland, H. B., Clarkson, T. B., *Circulation Research,* 1960, v7, 234.

4. Lofland, H. B., Prichard, R. W., Netsky, M. G., *Circulation,* 1959, v20, 10.

5. Searcy, R. L., Bergquist, L. M., *Clin. Chim. Acta,* 1960, v5, 192.

6. Clarkson, T. B., Lofland, H. B., *Circulation Research,* 1961, v9, 106.

7. Clarkson, T. B., *Proc. of Symposium on Drugs Affecting Lipid Metabolism,* Editors, S. Garattini and R. Paoletti, Elsevier Publ. Co., Amsterdam, 1961, 314.

8. Prichard, R. W., Clarkson, T. B., Goodman, H. O., Lofland, H. B., *Fed. Proc.,* 1962, v21, 97.

9. Lofland, H. B., Clarkson, T. B., Goodman, H. O., *Circulation Research,* 1961, v9, 919.

Received September 21, 1962. P.S.E.B.M., 1963, v112.

Effects of Starvation on the Cardiovascular System of the Chicken.* (27965)

JAMES A. VOGEL† AND PAUL D. STURKIE (Introduced by P. Griminger)
Physiology Section, Rutgers University, New Brunswick, N. J.

Most of the studies concerning the effects of acute starvation on the cardiovascular system of laboratory animals have been limited to blood pressure, heart rate and electrocardiography(1,2,3). Following starvation, blood pressure and heart rate drop and then usually stabilize at a low level. Partial starvation in humans for a 24-week period produced bradycardia, moderate hypotension and a marked reduction in cardiac output(4).

The effects of acute starvation in birds have received scant attention(8). The objectives of this experiment were to determine the effects of starvation on cardiac output, peripheral resistance, blood pressure and heart rate in chickens.

Methods and procedure. The experiment was conducted on 2 groups of White Leghorn males, 5 months and 15 months of age. Both groups were kept in cages. Feed was withheld from the birds for 8 days, and on the 9th day they were fed. Water was supplied *ad libitum.* Determinations on cardiac output and direct blood pressures were made 7 days before and 8 days after starvation. Cardiac output was determined by the dye dilution method(5), slightly modified. A Water's densitometer and Cardio-green dye were used, and the signal was recorded on a Recti-Riter. Direct blood pressure was determined by cannulation of the carotid artery and the use of a Statham pressure transducer and a Sanborn recorder. Indirect blood pressures were also determined daily, before and during starvation on the 5-month-old birds by the method of Sturkie *et al.*(6), utilizing an Infraton pickup, and the Visiton oscilloscope, or a Sanborn recorder when pulse rate was desired. Total peripheral resistance was calculated in arbitrary units from cardiac output and mean blood pressure.

Results. The daily changes in body weight, systolic blood pressure and heart rate

* Paper of Journal Series, N. J. Agri. Exp. Station. Supported by Public Health Service grant.

† Present address: Physiology Division, U. S. Army Med. Research & Nutrition Lab., Fitzsimons Gen. Hospital, Denver, Colo.

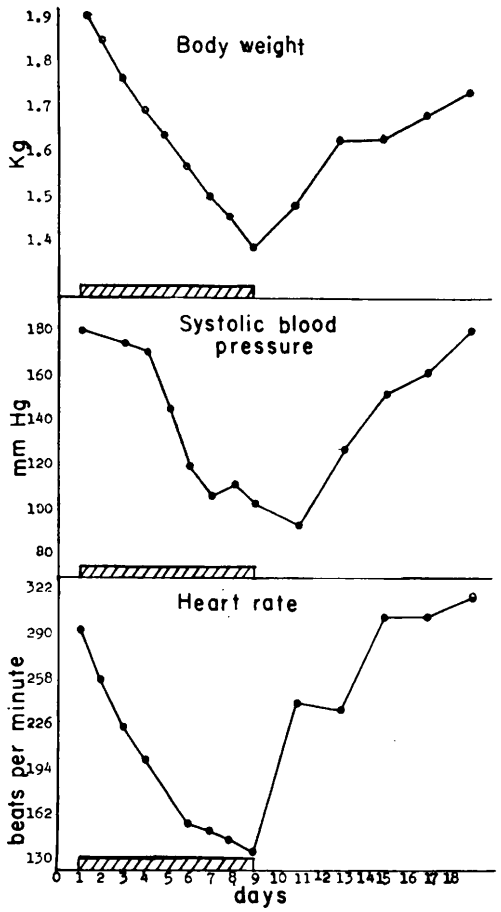

FIG. 1. Effect of an eight-day starvation period on 10 growing male chickens. Blood pressure and heart rate taken by indirect method.

of young males following starvation are illustrated in Fig. 1. Heart rate and body weight declined in a linear fashion after the first day of starvation. Heart rate on the last day of starvation was 133 per minute, a decrease of 55% from the pre-fasted level of 293. Body weight decreased 31% over the same period. Systolic blood pressure decreased slightly during the first 3 days of starvation and dropped rapidly thereafter, reaching a minimum level of 96 mm Hg 2 days after the starvation period was terminated. The pressure level then began to rise and returned to the pre-fasting level.

Heart rate and blood pressure when determined directly on the starved birds, restrained on their backs, were higher than corresponding values obtained with the indirect method (Table I, trial 1). In fact, heart rates after 8 days of starvation were only 13% less than the pre-starved values, but systolic blood pressure level exhibited a 31% decrease. Cardiac output decreased from 330 to 126 ml per minute, or a decrease of 62% on an absolute basis and 48% on a body weight basis. Total peripheral resistance following starvation increased approximately 100%.

The results of starvation of the older males (trial 2, Table I) exhibited the same trend, but the magnitude of change for some of the parameters was not as great. The drop in blood pressure was significant but less than for the younger birds. The extent of decrease in cardiac output, although statistically significant was considerably less than for the younger birds, and total peripheral resistance was not significantly influenced by starvation.

Discussion. Decreases in heart rate and blood pressure observed in chickens following starvation were also reported in starved dogs by Wilhelmj(1), but the magnitude of the changes was greater in chickens. The possible mechanisms involved in the blood pressure drop are not known and have received little attention. Whatever mechanisms are involved appear to be related to general level of metabolic activity and depletion of body tissues in the starved state.

Our data indicate that the decline in heart rate begins immediately with fasting and show that heart rate of the starved bird may be increased to near the normal level by excitement and handling attending the restraint and surgery involved in cannulation of an artery. Thus, the excitement brought about by handling may diminish vagal tone, increase cardiac sympathetic activity or both. Preliminary work in this laboratory reveals that atropine administered to the starved bird increases heart rate by abolishing vagal tone. Data from other sources tend to support this view. Carter and Drury(7) who produced bradycardia in pigeons by feeding a diet deficient in Vit. B_1, which is known to produce inanition, reported normal heart rates after administration of atropine or sectioning of vagi. A recent report(8) indicates

TABLE I. Effect of 8 Days of Starvation on Cardiovascular Parameters of Male Chickens, 5 Months (Trial 1) and 15 Months (Trial 2) of Age.

Measurement	Trial 1		Trial 2	
	Pre-starved	Starved	Pre-starved	Starved
(Number of birds)	(10)	(9)	(9)	(6)
Body wt, g	1961 ± 58†	1356 ± 74*	2425 ± 58	1893 ± 62*
Systolic BP, mm Hg	224 ± 9.7	139 ± 9.2*	206 ± 5.3	158 ± 5.1*
Diastolic BP, " "	168 ± 6.7	124 ± 6.7*	158 ± 3.7	132 ± 4.6*
Pulse pressure, " "	56 ± 8.1	14 ± 3.1*	48 ± 5.0	26 ± 2.1*
Mean BP, " "	189 ± 7.0	130 ± 7.6*	176 ± 3.3	142 ± 4.7*
Heart rate, per min.	342 ± 8.0	297 ± 23	303 ± 12.0	235 ± 13.8
Cardiac output				
ml/min.	330 ± 19	126 ± 22*	493 ± 27	327 ± 35*
ml/min./kg	170 ± 11	88 ± 10*	202 ± 8	173 ± 16
Stroke volume, ml	.97 ± 11	.40 ± .09*	1.65 ± .12	1.39 ± .11
Total periph. resistance				
TPR units	.59 ± .03	1.19 ± .13*	.37 ± .03	.45 ± .05
TPR units/kg	1.16 ± .08	1.65 ± .10*	.89 ± .05	.85 ± .06
Hematocrit	39 ± .90	44 ± .96*	44 ± 1.2	45 ± 2.7

* Difference between pre-starved and starved groups significant at .05 level.
† Mean ± S.E.

that starvation and not Vit. B_1 deficiency *per se* causes bradycardia in chickens.

Summary. Acute starvation of chickens produces a drop in heart rate, blood pressure and cardiac output. The decline in heart rate, which is influenced by excitement and handling of the bird, may be due to enhanced vagal tone.

1. Wilhelmj, C. M., Meyers, V. W., Milani, D. P., McDonough, J. R., Racher, E. M., McGuire, T. F., Waldmann, E. B., McCarthy, H. H., *Circulation Res.*, 1953, v1, 419.

2. Wilhelmj, C. M., Waldmann, E. B., McGuire, T. F., *Am. J. Physiol.*, 1951, v166, 296.

3. Smith, G. S., Mameesh, M. S., Johnson, B. C., *Fed. Proc.*, 1960, v19, 100.

4. Keys, A., Chap. 28, *Biology of Human Starvation*, U. Minn. Press, Minneapolis, 1950.

5. Sturkie, P. D., Vogel, J. A., *Am. J. Physiol.*, 1959, v197, 1165.

6. Sturkie, P. D., Durfee, W., Sheahan, M., *Poultry Sci.*, 1957, v36, 1160.

7. Carter, C. W., Drury, A. W., *J. Physiol.*, 1929, v68, proc. 1-2.

8. Griminger, P., Kloos, W., *5th Internat. Congress of Nutrition*, 1960, p14.

Received September 26, 1962. P.S.E.B.M., 1963, v112.

Preparation and Properties of an Aluminum Citrate-Endotoxin Complex from *Salmonella enteritidis*. (27966)

W. T. HASKINS, E. RIBI, M. LANDY, R. L. ANACKER AND K. C. MILNER
(Introduced by J. J. Munoz)
U. S. Dept. of HEW, Public Health Service, Nat. Inst. Health, National Institute of Allergy and Infectious Diseases, Rocky Mountain Laboratory, Hamilton, Mont.

Aqueous ether extracts containing the endotoxin of *Salmonella enteritidis* have previously been processed in various ways in order to reduce their content of protein and lipid. Treatments included fractional precipitation with ethanol and salts, extraction with boiling mixtures of chloroform-ethanol, and extraction with aqueous phenol. Resulting preparations contained as little as 0.4-0.5% nitrogen and about 3-4% total fatty acids (1).

Since these procedures were tedious and time consuming, other means were sought for preparing endotoxin with low nitrogen and fatty acid contents. In this study the nitrogen content was reduced by treating aqueous