TABLE I.
Distribution of P^{32} in Incubated Eggs.
(Approximately 5 mg KH_2PO_4 containing 20μc P^{32} injected into yolk 5th day of incubation.)

Age of egg	Embryo		Allantois and amnion		Yolk		Albumen		Shell and membrane.
	Wt in g	% of total activity	Volume in cc	% of total activity	Volume in cc	% of total activity	Volume or wt	% of total activity	% of total activity
6	0.4	3	13.5	64	20	29	17 cc	2	2
7	0.8	6	14	52	20	37	14 "	2	1
8	1.4	10	13	53	22	36	12 "	1	1
9	2.2	18	15	50	21	29	11 "	3	1
10	3.1	28	11	29	26	39	10 "	3	1
11									
12	5.8	58	14	21	14	19	8 "	1	1
13	8.1	65	10.5	12	16	19	11.5 "	3	1
14	9.3	71	13	12	20	17	7 "	.7	1
15	13.3	73	13.5	13	14	12	5 "	.8	1
16	16.9	73	11	16	14	7	3 "	.6	1
17	18.1	76	6.5	17	14	7	1 g		.8
18									
19	25.5	93	3	3	14	4	1 "	.3	.3
20	27.4	90	7	4	9	5	.5 "	.4	.6

these various fractions was relatively constant throughout our period of observation from the 6th to the 14th day of incubation. Ninety per cent of the P^{32} in the allantoic and amniotic fluids was found in the H_2O soluble fraction, 9.4% in the trichloracetic acid precipitate and 0.6% in the ether soluble extract.

In the yolk nearly all of the activity was in the H_2O soluble fraction, only 1.1% was found in the ether extract.

In the albumen 80% of activity was found in H_2O soluble fraction and 20% in trichloracetic acid precipitate.

Summary. The radioactive isotope P^{32} was used as a tracer to follow the course of phosphorus combined as KH_2PO_4 and injected into the yolks of incubating eggs. The distribution of P^{32} within the various components of the eggs was determined daily throughout incubation. The distribution in ether soluble, water soluble, and trichloracetic acid precipitable fractions was determined for the fluid components of the eggs from the 6th to the 14th day of incubation. From the 6th day of incubation to the day of hatching, the P^{32} in the embryo increased from 3% to 90%.

16148

Changes in Plasma Inorganic Phosphorus of Dogs Under Postural Restraint.*

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During the course of some routine experiments the observation was made that, in unanesthetized dogs restrained in the supine position, the inorganic phosphorus concentration of whole blood and of plasma decreased to a low value of 30-60% of the original amount in 2 hours, after which it increased, in some instances attaining a level about

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TABLE I.
Average Plasma Inorganic Phosphorus Values in Dogs Restrained in the Supine Position.

Duration of restraint (min.)	Dog 1 (4 exp.)			Mean of 4 dogs (17 exp.)		Mean of all samples		
	M	S	A	M	A	M	S	A
0-9	4.36	0.34	100.	4.05	100.	4.07	1.10	100.
10-29	3.51	0.30	81.	3.52	87.	3.45	0.94	85.
30-59	2.38	0.35	55.	3.45	85.	2.58	0.62	63.
60-119	2.26	0.55	52.	2.23	61.	2.22	0.80	55.
120-239	2.55	0.94	58.	2.46	67.	2.23	1.20	55.

M Mean values in mg %.

S Standard deviation; S equals $\sqrt{\frac{\Sigma(X-M)^2}{n-1}}$

A Per cent of initial concentration.

equal to the original value.

Blood phosphate changes in normal animals have frequently been observed upon administration of substances such as insulin or epinephrine[1,2] or upon the ingestion of glucose,[3] but we have found in the literature no report of changes in blood phosphate in animals which, other than being restrained on their backs, were allowed to remain under normal conditions.

In view of these facts and since the phosphate changes we had observed were so marked and consistent, we investigated further the occurrence of this phenomenon under two different conditions of postural restraint: first, that in which the animals were tied in the supine position for 2 to 4 hours and second, that in which they stood in a Pavlov stock for the same period of time.

Methods. In the first group of experiments blood samples were drawn from the femoral vein immediately after the dogs were laid on the table and again at 30, 60, 120, and 240 minutes later. In the second group the animals were placed in a Pavlov stock after a "control" blood sample was taken.

Immediately after being drawn the blood was delivered into cold centrifuge tubes containing dried sodium oxalate and surrounded by crushed ice, centrifuged, and the plasma kept chilled until the inorganic phosphorus analysis was completed.

Inorganic phosphorus was determined by the method of Lowry and Lopez[4] on the ice-cold trichloracetic acid filtrate of the chilled plasma. Total phosphorus, acid soluble phosphorus, and lipid phosphorus, (after extraction with alcohol-ether mixture) all were evaluated after sulfuric-nitric acid digestion (2.5 ml of 5 N sulfuric acid and 3 drops of concentrated nitric acid) by addition of 2 ml of 2.5% ammonium molybdate solution and 3 ml of 1% fresh ascorbic acid solution. Exactly 20 minutes after addition of the ascorbic acid the developed color was measured in a Klett-Summerson photoelectric colorimeter, using filter No. 66. Blanks and standards were always done parallel to unknowns and duplicate samples were analyzed whenever possible.

Plasma glucose was estimated by a modification of the Somogyi method reported by Nelson.[5]

Results and Discussion. Marked decreases in plasma inorganic phosphate averaging 52% of the initial level were observed in all but one of a total of 17 experiments done on 4 dogs in the first group. Table I shows the mean values of all 17 experiments as well as average results of 4 experiments done on Dog 1 as a representative example. The mean of

1 Perlzweig, W. A., Latham, Emily, and Keefer, C. S., PROC. SOC. EXP. BIOL. AND MED., 1923, **21**, 33.

2 Harrop, George A., Jr., and Benedict, E. M., *ibid.*, 1923, **20**, 430.

3 Harrop, George A., Jr., and Benedict, E. M., *J. Biol. Chem.*, 1924, **59**, 683.

4 Lowry, Oliver H., and Lopez, Jeanne A., *ibid.*, 1946, **162**, 421.

5 Nelson, Norton, *ibid.*, 1944, **153**, 375.

TABLE II.
Plasma Phosphorus Fractions and Glucose Content, Dog 4, Exp. 17.

Duration of restraint (min.)	Phosphorus (mg %)				Glucose (mg %)	% of initial conc.	
	TP	LP	TASP	IP		IP	Glucose
0*	14.9	11.1	3.28	3.04	98.4		
0-9	14.5	11.3	3.34	3.18	107.	100.	100.
10-29	—	—	—	—	—	—	—
30-59	13.8	11.3	2.76	2.68	107.	84.	100.
60-119	12.5	11.1	0.93	0.97	158.	31.	151.
120-239	12.3	11.1	1.24	1.04	158.	33.	151.

* This blood taken 60 minutes prior to restraining the dog.
TP Total Phosphorus.
LP Lipid Phosphorus.
TASP Total Acid Soluble Phosphorus.
IP Inorganic Phosphorus.

TABLE III.
Plasma Inorganic Phosphorus Concentration of Dogs Restrained in a Stock.

Duration of restraint (min.)	Inorganic Phosphorus (mg %)		% of initial conc.
	Dog 3	Mean of 4 dogs	
0	4.76	4.64	100.
1-29	4.28	4.08	88.
30-59	4.28	3.79	82.
60-119	4.28	4.04	87.
120-239	4.48	3.98	86.

all the samples, also included in this table, indicates significant decreases through the 60-119-minute period, after which the concentration remained constant or increased slightly in most of the dogs.

In 8 of the 17 experiments performed with the dogs restrained in the supine position a complete analysis of the plasma phosphorus fractions was done, and in 3 of these the plasma glucose levels were also followed. A complete analysis of one of these three experiments is presented in Table II. In this experiment the dog was allowed to remain unrestrained on the floor for about one hour and then tied in the supine position for 2 to 4 hours. The dog was quiet while on the floor but showed signs of unrest soon after being confined. The irritability increased markedly after the 60-minute period and continued until the animal was set free. In all 8 experiments the acid soluble and total phosphorus paralleled the changes in the inorganic fraction, while lipid phosphorus remained quite constant during an entire experiment. In general, increases in plasma glucose were observed at about the same time or immediately after decreases in plasma inorganic phosphorus were noted.

Our animals, although trained to lie quietly on their backs, at times exhibited signs of emotional disturbances and fatigue. A study of the decreases in the obviously excited animals and in the apparently quiet ones revealed an average low value of 40% of the original concentration in the former group compared to 69% for the latter. This difference, when studied statistically, is not significant, the probability at the 60-119-minute period being 17%. Analysis of a larger number of experiments might show a more significant difference between the 2 groups.

The supine position is not a natural one for the dog and restraint in such a position might possibly be the cause of physiological changes in the animal, either directly because of the peculiar posture or indirectly because of aroused emotional status due to fatigue. In 4 experiments dogs restrained in a standing position in a stock did not offer any resistance or become excited to any noticeable

extent. In these experiments, the results of which are presented in Table III, there was a slight decrease in plasma inorganic phosphate, the greatest decrease occurring at the 30-59-minute period and amounting to 13% of the initial value.

It is not possible from the data obtained to derive any conclusions concerning the exact cause of these changes occurring in the plasma phosphorus concentration of restrained dogs. The principal factors appear to be those of posture and of emotional disturbance. This paper does not attempt to establish the relative contribution of each of these factors to the phosphorus decrease; its purpose is to report the observation of this phenomenon so as to bring it to the attention of those investigators whose experiments may be affected by its occurrence.

Summary. 1. A decrease in the plasma inorganic phosphate concentration amounting to 30-60% of the initial concentration was observed in dogs restrained in the supine position. This change was also evident in the total acid soluble and the total phosphorus fractions, while the lipid phosphorus fraction remained constant during an entire experiment.

2. The decrease in inorganic phosphate was accompanied by an increase in plasma glucose.

3. Dogs restrained on their backs exhibited a greater decrease than those restrained in a stock.

16149

Effect of Salicylate and Tripelennamine Hydrochloride (Pyribenzamine) on the Arthus Reaction and on Bacterial Allergic Reactions.*

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The tissue damage that results from the union of certain antigens with their corresponding antibodies may be prevented by non-specific agents.[1,2] In the case of the positive tuberculin reaction, cachexia and fever appear to inhibit the expected reactions.[2] As yet, however, little attention has been directed to the inhibition of necrotizing allergic reactions by therapeutic agents. The more transient reactions of classical anaphylaxis are found to be prevented effectively by the Mosnier compounds which have striking anti-histamine properties.[3,4] Urticarial reactions to specific allergens or to histamine can be inhibited by many of these synthetic compounds, of which the most widely used in this country are diphenhydramine hydrochloride (Benadryl) and tripelennamine hydrochloride (Pyribenzamine). Clinically, hayfever and analogous reactions have been benefitted by these agents.[4] These substances, however, have not been demonstrated to be beneficial in the "bacterial" type of allergic response, or in the local Arthus reaction, although these allergies may be important in the pathogenesis of many diseases. Reubi[5] reported that 2-(N-phenyl-N-benzyl-aminoethyl)-imidazoline (antistine) prevented the occurrence of the kidney damage that usually results when duck anti-kidney serum is in-

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1 Mitchell, A. G., Wherry, W. B., Eddy, B., and Stevenson, F. E., *A. J. Dis. Child.*, 1928, **36**, 720.

2 Gay, F. P., and Associates, *Agents of Disease and Host Resistance,* Springfield, Charles C. Thomas, 1935.

3 Feinberg, S. M., *J. A. M. A.*, 1946, **132**, 703.

4 Friedlander, S., *Am. J. Med.*, 1946, **1**, 174.

5 Reubi, F., *Helvetia Medica Acta,* 1946, **13**, Supp. XVIII.