

METABOLISM OF ^{32}P IN POULTRY

By

E.R.M. ABOU-HUSSEIN, M.A. RAAAFAT, I.M. EL-GINDI
AND M.S.M. SAMY*

Experiments were conducted to determine the rates of uptake and distribution of intramuscularly and intraperitoneally administered ^{32}P by cocks that had previously received phosphorus deficient and adequate diets. These studies indicated that the inorganic phosphorus in plasma was highly significant affected by its level in the diet. It was found that phosphorus level in the diet had very slight effect on its level in the soft tissues and organs tending to be the highest in the supplemented group and the least in the deficient one. The level of dietary phosphorus affected its content in bones. Bones in the supplemented group contained the highest level of phosphorus while that of the deficient one contained the least. The results of these experiments showed that the order of distribution of ^{32}P among tissues and organs of each group was not affected by the dietary phosphorus level. The mode of injection viz., intramuscularly and intraperitoneally had no effect on the distribution of ^{32}P among tissues and organs. Of the soft tissues tested, the liver contained the greatest amount of ^{32}P followed by the kidneys heart muscles and others, with the brain containing only very small amounts. These data indicated that the tissues and bones of the deficient group retained generally more ^{32}P than those of the other two groups, and those of the supplemented one retained the least. The standard specific activities of most tissues and organs tended to be high at times soon after tracer administration and then declined gradually thereafter, while that of bones increased at later times after injection.

Recently, the production of artificially radiocative phosphorus (^{32}P) has furnished a new experimental approach of phosphorus metabolism. Inoue *et al.* (1950) observed that ^{32}P was most abundantly by mice in bones, liver and blood, while the small intestines, spleen, kidney, testes and muscles contained minute amounts. Smith *et al.* (1951) found that the rate of ^{32}P uptake by swine tissues decreased in the following order : bile, liver, kidneys, thymus, lymphnode, heart, spleen tongue muscle, lung, stomach (smooth) muscle, gastrocnemius and biceps (skeletal) muscle and brain. Rosenfeld and Inoue (1950) Beath (1952) furnished a new experimental approach of phosphorus metabolism. Pointed out that the rate of ^{32}P uptake by adult rabbits tissues was bility of the cells to phosphate ion. Freeksen and Meissner (1953) found that the activity of ^{32}P in rabbits, blood decreased exponentially with the lapse of time up to 10 days after injection. After intravenous, intrapleural or subcutaneous injections the curves were similar in the shape, but after oral administration the activity in the blood reached its maximum only after the lapse of 2—3 days and then decreased. Their results showed little effect due to the route but a definite effect due to the lapse of time. Smith *et al.* (1951) observed that the rate of ^{32}P uptake by turkey tissues was greater in the liver than in the kidneys, heart, spleen, lungs, gastrocnemius and brain.

* Animal Production Department, Faculty of Agriculture, Cairo University,

Material and Methods

Thirty six Rhode Island Red cocks of about one year old were experimented on. They were divided into equal three groups. The first group received a ration containing the normal level phosphorus (0.6 gm phosphorus per cock daily) recommended by Morrison (15). The second group (supplemented one) was given the previous ration plus phosphate salt (to cover three times phosphorus requirements of cocks. The third group (deficient one) was fed on yellow corn alone which covered only half of phosphorus requirement of cocks. Water was supplied ad libitum. The composition of the diet of the three groups are given in the following :

Criteria	Group I	Group II	Group III
Corn grains %	20	20	100
Barley %	20	20	—
Wheat bran %	20	20	—
Rice bran %	20	20	—
Undecorticated cottonseed cake %	20	20	—
Quantity of the ration/cock daily gm.	100	100	100
Phosphorus per cock daily gm.	0.621	1.821	0.301

Two months before administrating radioactive isotope, the cocks were individually caged. On the sixty first day, immediatyl after the first feed intake, each cock in the three groups was injected with 20 Mc ³²P labeled sodium phosphate. Six cocks in each group were injected intramuscularly and the others intraperitoneally. Two cocks from each group but differing in the site of injection were killed and bled to death at 6, 12, 24, 48, 72 and 96 hours after injection. Blood was collected and some tissues were taken for radioactive assaw and chemical determination. The injectoin, precautions, and techniques involving the radioactive material were carried out according to Comar (1955), and Hansard *et al.* (1951).

Radioactivity was counted using Geiger muller counter. Preparation of samples of plasma for counting was carried out as reported by Kamal and Cragle (1962). Organs and tissues were prepared using the method of Smith *et al.* (1951). The samples used for radioassay were used also for phosphorus determination. The method of Fiske and Sabbarow (1925) was used for phosphorus estimation. The calculations of radioactivity were carried out according to Hevesy (1948), Sacks (1953), Comar (1955), and Overman and Clark (1960). Snedecor's book (1959) was consulted for statistical analysis.

Results and Discussion

1. *Radioactive and inorganic phosphorus in plasma :*

The results indicated that there was no effect for the route of injection on the course of the standard specific activity (S.A.) of plasma during the different intervals after injection. This is in agreement with that obtained by Freerksen and Meissner (1953) with rabbits. It can be noticed from table (1, 2) that the dietary phosphorus level had a significant effect on phosphorus concentration in the different parts of the body, being the lowest in the deficient group and the highest in the supplemented one. The average plasma phosphorus was 5.37, 6.87 and 3.89 mg/ 100 ml plasma for the cocks fed on the normal, supplemented and deficient ration respectively. There were highly significant differences between the normal group and either the supplemented or the deficient one. These results are in good agreement with those obtained by Abou-Hussein *et al.* (1968) with rams, Beeson *et al.* (1944) with lambs, Gibson *et al.* (1950), and Tillman *et al.* (1959) with cattle.

2. *Phosphorus distribution :*

It can be seen from Table (3) that phosphorus level in the diet had a very slightly effect on its level in the soft tissues and organs, tending to be the highest in the supplemented group and the least in the deficient one. These findings are in good agreement with those of Schneider and Steengock (1939) with rats, and Tillman *et al.* (1959) with cattle. In this connection Hansard and Plumlese (1954) reported that soft tissues are apparently maintained when necessary at the expense of bone minerals. The equilibrium existing between bone and soft tissues would probably require complete bone depletion before these tissues would materially be affected. It can be observed that there was a marked effect of dietary phosphorus on its content in bones. The supplemented group contained the highest level of phosphorus while the deficient one contained the least. The results indicate that the phosphorus content, in the diaphysis, is always higher than that of the epiphysis in the different groups. These results are in agreement with those found by Rosenfeld and Beath (1952) with rates, Smith *et al.* (1951) with swine, Smith *et al.* (1955) with sheep and Smith *et al.* (1956) with turkeys.

3. *Radioactive phosphorus retention :*

It was observed that the deficient group generally retained more ^{32}P than the other two groups while the supplemented one retained the least. These results are similar to those found by Allam (1966), Hansard and Plumlese (1954) with ^{45}Ca on rats and Abou-Hussein *et al.* (1964) with ^{52}Mn on rams. The retention of ^{32}P by bones was affected by the level of phosphorus in the diet. The phosphorus deficient group retained more ^{32}P in tissues organs, and bones than the normal one, while the supplemented group retained the least. This adaptation may be mediated through the postulated active transport system.

TABLE 1.—RADIOACTIVE AND INORGANIC PHOSPHORUS IN PLASMA OF COCKS
FED ON NORMAL AND SUPPLEMENTED RATIONS

Hours after injection	Normal group				supplemented group			
	Inorganic P mg/100 ml.	³² P Uc/ml.	Specific activity	Standard S.A.	Inorganic P mg/100 ml.	³² P Uc/ml.	Specific activity	Standard S.A.
6	5.33	0.00748	0.1403	9.296	7.03	0.00206	0.0293	2.080
12	5.68	0.00368	0.0648	4.130	7.01	0.00793	0.1131	8.486
24	5.04	0.00362	0.0647	3.660	6.75	0.00249	0.0369	3.137
48	4.93	0.00304	0.0616	3.836	6.28	0.00246	0.0361	3.194
72	5.00	0.00320	0.0640	3.348	6.88	0.00162	0.0235	1.924
96	6.10	0.00176	0.0288	2.040	6.86	0.00099	0.0144	1.428

TABLE 2.—RADIOACTIVE AND INORGANIC PHOSPHORUS IN PLASMA COCKS
FED ON NORMAL AND DEFICIENT RATIONS

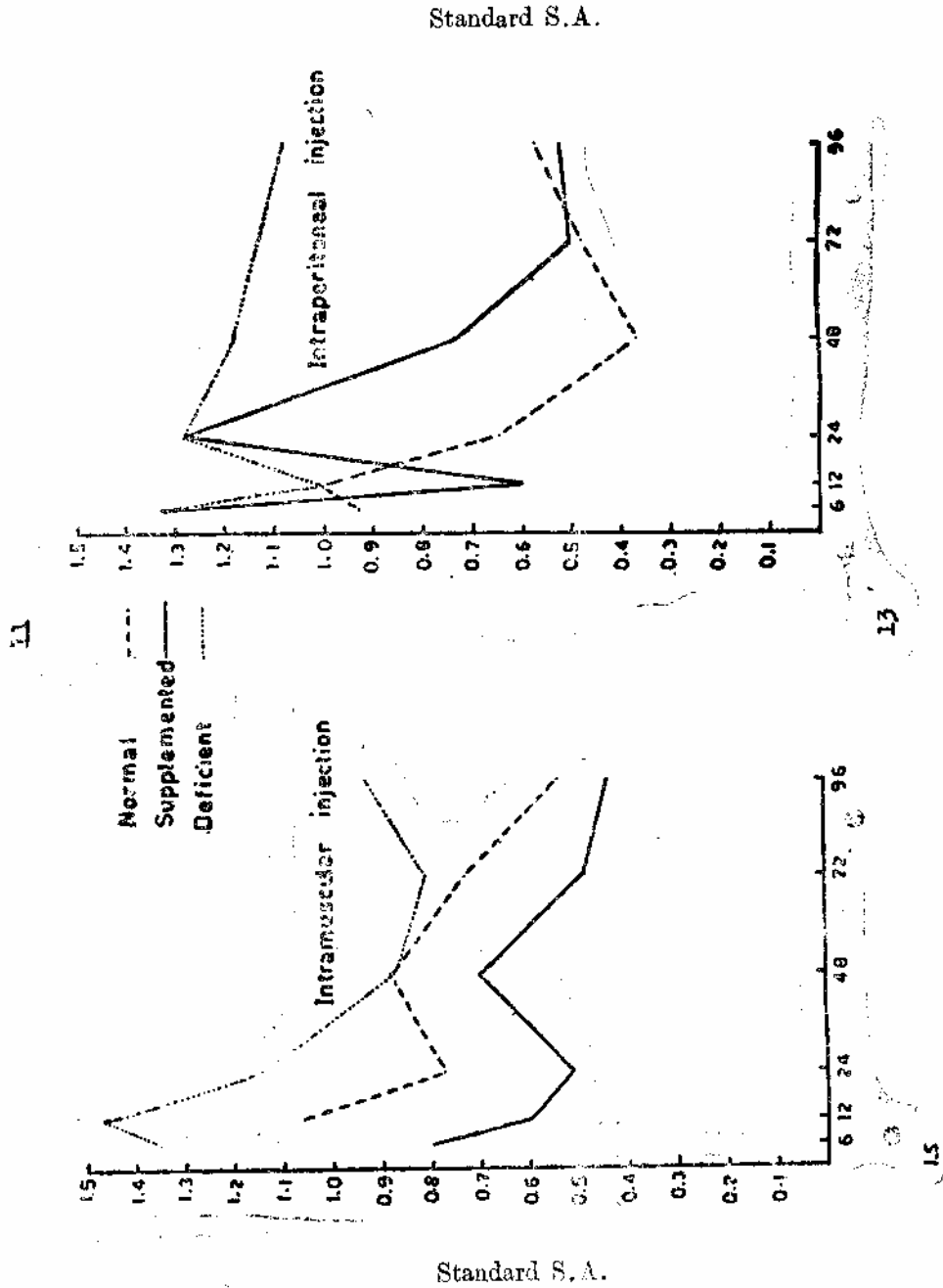
Hours after injection	Normal group				Deficient group			
	Inorganic P mg/100 ml.	³² P Uc/ml.	sepecific activity	Standard S.A.	Inorganic P mg/100 ml.	³² P Uc/ml.	sepecific activity	Standard S.A.
6	5.33	0.00748	0.1403	9.296	3.92	0.00567	0.1446	14.865
12	5.68	0.00368	0.0648	4.130	4.02	0.00704	0.1751	14.009
24	5.04	0.00326	0.0647	3.660	3.95	0.00311	0.0787	7.209
48	4.93	0.00304	0.0616	3.836	3.91	0.00189	0.0483	4.871
72	5.00	0.00320	0.0640	3.348	3.82	0.00228	0.0597	7.016
96	6.10	0.00176	0.0288	2.040	3.87	0.00270	0.0698	7.312

TABLE 3.—PHOSPHORUS CONTENT IN TISSUES AND ORGANS OF COCKS
FED ON RATIONS CONTAINING DIFFERENT PHOSPHORUS LEVELS

I T E M	mg. phosphorus/ gm. fresh weight		
	Normal	Supplemented	Deficient
Pectoral muscle	2.19	2.31	2.11
Gastrocnemius muscle	1.85	1.90	1.72
Kidneys	2.40	2.58	2.34
Liver	2.78	2.87	2.63
Testes	1.57	1.64	1.38
Heart	2.06	2.16	1.95
Spleen	2.69	2.90	2.53
Bile	1.75	1.89	1.55
Femur :			
Epiphysis	38.14	50.75	21.34
Diaphysis	75.22	80.34	68.75

4. Radioactive phosphorus distribution :

The results indicated that the level of dietary phosphorus did not influence the distribution of ^{32}P among tissues and organs. Moreover, the mode of injection viz. intramuscularly and intraperitoneally had no effect on the radioactive distribution. In this connection, Havey (1948), Rosenfeld and Beath (1952) reported that the uptake of phosphorus by a particular tissue may depend upon the permeability of the cells to phosphate ion. Some tissues, notably the liver, Kidneys, heart, spleen; showed a rapid uptake rate; while others particularly the testes, and gastrocnemius and pectoral muscle showed much slower rate (Figures 1—3). Within the three groups, the tissues can be arranged generally in the following order of decreasing radioactive phosphorus uptake rate : bile, liver, kidneys, heart, spleen; testes; gastrocnemius muscle and pectoral muscle. It can be noticed also that ^{32}P uptake by these tissues was the highest in the deficient group and the lowest in the normal one. These results are in harmony with those found by Cohn and Greenberg (1938) with rats, Smith *et al.* (1951) with swine, Smith *et al.* (1955), and Abou-Hussein *et al.* (1968) with sheep.



Hours after injection
Hours after injection
Fig. 1.—Standard S.A. for the liver of cocks at different intervals after injection.

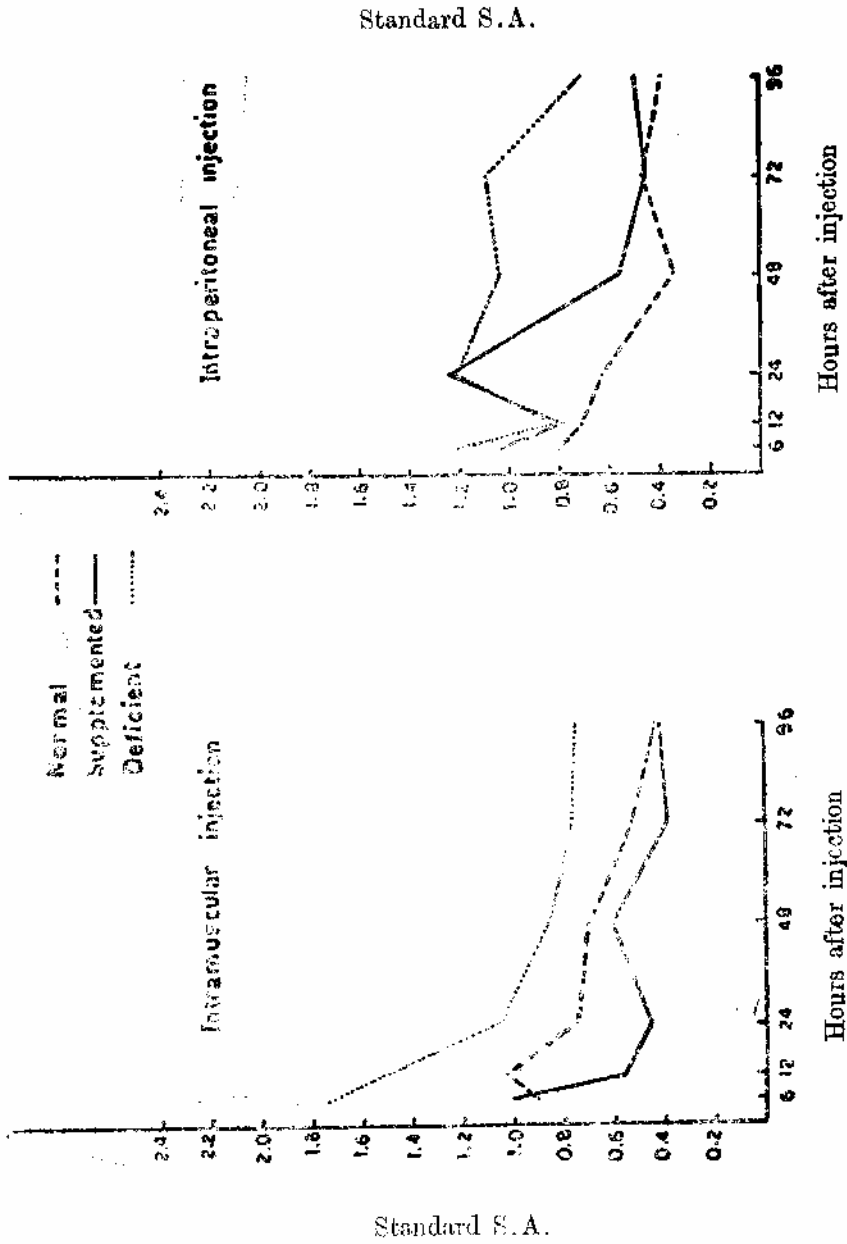


Fig. 2.—Standard S.A. for the kidney of cocks at different intervals after injection.

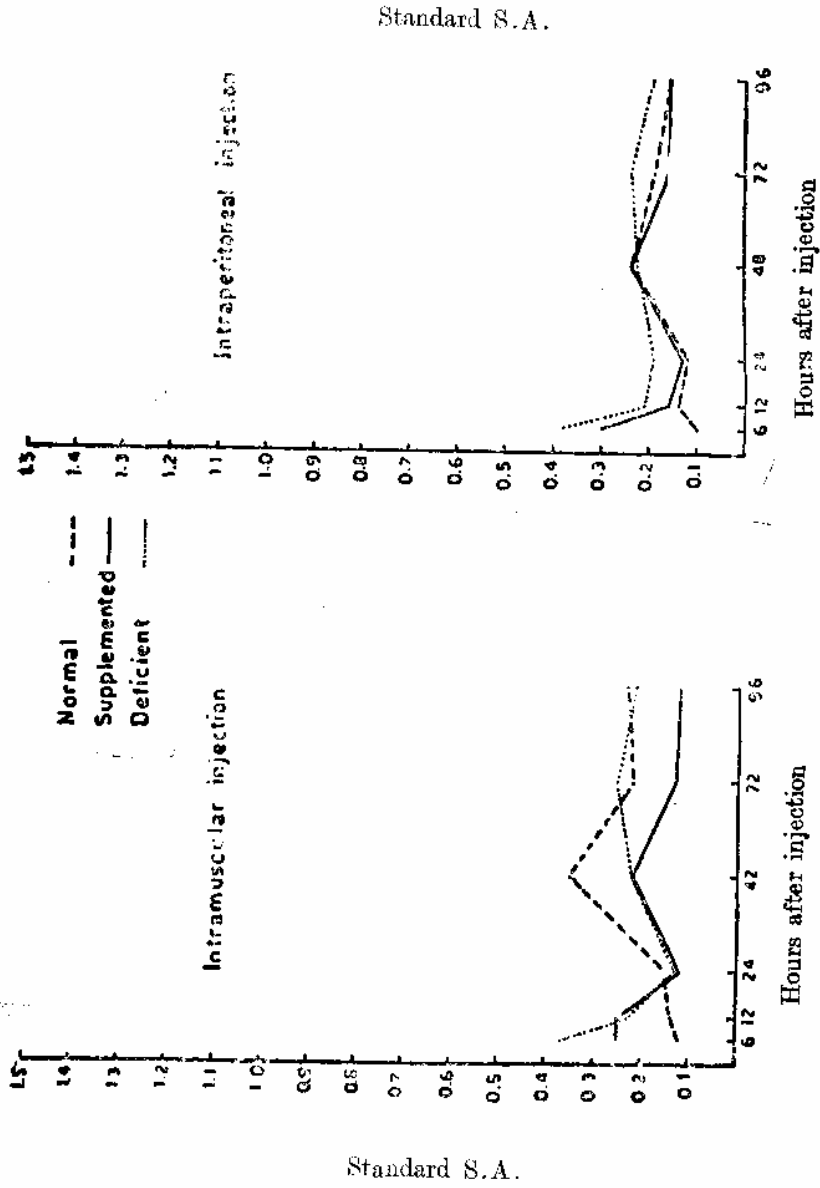


FIG. 3.—Standard S.A. for the pectoral muscle of cocks at different intervals after injection.

REFERENCES

- ABOU-HUSSEIN, E.R.M., EL-SHAHIE, M., and ALLAM, S.M. (1964).—“Study of manganese uptake by farm animals”. Middle Eastern Regional Radioisotope Center for the Arab Countries.
- ABOU-HUSSEIN, E.R.M., RAAFAT, M.A., EL-GINDI, I.M., and SAMY, M.S. (1968).—Metabolism of ^{32}P in sheep. *Pakistan J. of Sci. Res.* **20**, No. 1
- ALLAM, S.M. (1966).—Chemical and nutritional studies on calcium metabolism in farm animals. *M.Sc. Thesis, Fac. Agric.*, Cairo Univ.
- BEESON, W.M., JOHNSON, R.M., BOLIN, D.W., and HICKMANN, C.W. (1944).—The phosphorus requirement for fattening lambs. *J. Animal Sci.*, **3**, 63-70.
- COHN, W.E., and GREENBERG, D.M. (1933).—Studies of mineral metabolism with the aid of artificial radioactive isotopes. I. Absorption, distribution, and excretion of phosphorus. *J. Biol. Chem.* **123**, 185-198.
- COMAR, C.L. (1955).—“Radioisotopes in Biology and Agriculture”. Mc Graw-Hill Book Company, Inc. New York.
- FISER, C.H., and SUBBAROW, Y. (1925).—The colorimetric determination of phosphorus. *J. Biol. Chem.* **66**, 375-400.
- FREERIKSEN, E., and MEISSNER, J. (1953).—Animal experiments on the uptake and distribution of radioactive phosphate. *Ztschr. Ges. Exp. Med.*, **120**, 190-214. (*In Nutrition Abst.* **23**, 609, 1953).
- GIBSON, M.E., JR., GALLUP, W.D., and ROSS, O.B. (1950).—Forage composition in relation to phosphorus deficiency in range beef cattle in Southeastern Oklahoma. *Proc. Okla. Acad. Sci.*, **31**, 94-99. (*In Chem. Abst.*, **46**, 6283 h, 1952).
- HANSARD, S.L., COMAR, C.L., and PLUMLEE, E. (1951).—Radioisotope procedure with farm animals II—Quantitative administration and blood sampling technique *Nucleonic*, **9**, 38-46.
- HANSARD, S.L., and PLUMLEE, M.P. (1954).—Effect of dietary Calcium and phosphorus levels upon the physiological behaviour of calcium and phosphorus in rat. *J. Nutrition*, **54**, 17-31.
- HEVESY, G. (1948).—“Radioactive Indicators”. Interscience publishers. Inc., New York.
- INOUE, K., KIRUCHI, T., MIYAKE, T., WAKISAKA, G., and NISHIKAWA, M. (1950).—Some experiments on phosphorus. III—Biological tracer experiments. *Bull. Inst. Chem. Research, Kyoto Univ.*, **22**, 72-73. (*In Chem. Abst.*, **46**, 9448 d, 1952).
- KAMAT, T.H., and CRAGLE, R.G. (1962).—Significance of plasma ultrafiltrable ^{45}Ca and ^{32}P in milk synthesis *J. Dairy Sci.*, **45**, No. 1, 43-47.
- MORRISON, F.B. (1948).—“Foods and Feeding” 21st Edit., Morrison Publishing Co. Ithaca, N.Y.
- OVERMAN, R.T. and CLARK, H.M. (1960).—“Radioisotope Techniques”. Mc Graw-Hill Book Company, Inc. New York.
- ROSENFELD, I. and BEATH, O.A. (1952).—Distribution of ^{32}P in tissues of normal animals. *Proc. Soc. Exp. Biol. Med.*, **81**, 608-611. (*In Nutrition Abst.*, **23**, 609, 1953).
- SACKS, J. (1953).—“Isotopic Tracers in Biochemistry and Physiology”. Mc Graw-Hill Book Company, Inc. New York.
- SCHNEIDER, H. and STEENBOCK, H. (1939).—A low phosphorus diet and the response of rats to vitamin D_2 . *J. Biol. Chem.*, **128**, 159-171.

- SMITH, A.H., KLEIBER, M., MELVIN EDICK, A.L., ROBINSON, R.R. HERTMAN H. Ja. (1951).—Distribution of intravenously injected radiophosphorus (^{32}P) among swine tissues. *J. Animal Sci.*, **10**, 893-901.
- SMITH, A.H., KLEIBER, M., BLACK, A.L., LUICK, J.R., LABON, R.F., and WEBB, W.C. (1952).—Distribution of intravenously injected radioactive phosphorus ^{32}P among sheep tissues. *J. Animal Sci.*, **11**, 638-645.
- SMITH, A.H., KLEIBER, M., BLACK, A.L., and BAXTER, C.F. (1955).—Transfer of phosphate in digestive tract. I-Sheep. *J. Nutrition*, **51**, 507-527.
- SMITH, A.H., BOND, G.H., and WEGET, C.M. (1956).—Distribution of intravenously radiophosphate among turkey tissues. *Poultry Sci.*, **35**, 576-581.
- SNENDECOR, G.W. (1959).—“*Statistical Methods*”. Iowa State College, Press, Amer., Iowa.
- TILLMAN, A.D., BRETHOUR, J.R., and LANDSARD, S.L. (1959).—Comparative procedures for measuring the phosphorus requirement of cattle. *J. Animal Sci.*, **18**, 249-255.

تمثيل الفوسفور المشع في الدجاج

رفعت أبو حسين ، محمد على رافت ، ابراهيم الجندي ، محمد سامي

الملخص

استخدم في هذا البحث ستة وثلاثون ديكا رود ايلاند احمر وقسمت هذه الديوك الى ثلاث مجاميع متساوية وغذيت ديوك المجموعات الاولى والثانية والثالثة طوال مدة الدور التمهيدي الذي استمر لمدة شهرين على علائق عادية وعنية وفقيرة بالنسبة لعنصر الفسفور . وفي نهاية مدة الشهرين حقن كل ديك في الثلاث مجموعات بمقدار ٣٠ ميكروكوري فسفور مشع ف٢٠٣٢ . وقد حقن في كل مجموعة ستة ديوك في العضل والستة الباقية حقنت في التجويف البطني . ثم ذبح ديكان في كل مجموعة تختلفان في موضع الحقن على الفترات التالية ٦ ، ١٢ ، ٢٤ ، ٤٨ ، ٧٢ ، ٩٦ ساعة بعد الحقن . جمع الدم من كل ديك عند الذبح وقدر الفسفور الغير عضوي والمشع في البلازما . وكذلك أخذت عينات من بعض الاعضاء وانسجة معينة من كل ديك وذلك لتقدير الفسفور المشع . أما بالنسبة للفسفور الكلي فقد قدر في اعضاء وانسجة ثلاث ديوك مختلفة لكل مجموعة من المجموعات الثلاث .

وقد اوضحت النتائج ان الفسفور الغير عضوي في بلازما الدم يتأثر تأثرا معنويا بمستوى الفسفور في الغذاء فكان متوسطه ٦٨٧ ، ٣٧ ره ، - ٣٨٩ جم/١٠٠ سم ٣ بلازما في كل من المجموعات التي غذيت على علائق عنية وعادية وفقيرة في الفسفور على التوالي . كان لمستوى الفسفور في الغذاء تأثير طفيف جدا على مستواه في الاعضاء والانسجة المختلفة بالجسم . وان كان يميل للزيادة في المجموعة التي غذيت على عليقة غنية به وللقلة في المجموعة التي غذيت على عليقة فقيرة به وكان لمستواه في الغذاء تأثير واضح على مستوى الفسفور في العظم اذ احتوى عظم المجموعة التي غذيت على عليقة غنية بالفسفور على أعلى مقدار منه واحتوى عظم المجموعة التي غذيت على عليقة فقيرة في الفسفور على أقل مقدار منه .

ووجد أن موضع الحقن سواء في العضل أو التجويف البطني ليس له تأثير على توزيع الفسفور المشع بين الانسجة المختلفة وبعضها في المجاميع الثلاث . وقد أمكن ترتيب الانسجة والاعضاء المختلفة في المجاميع الثلاثة تبعا للنشاط النوعي المعرض للفسفور المشع في ترتيب تنازلي كالاتي :

« مرارة - كبد - كلية - طحال - خصيتين - عضلة الفخذ - عضلة الصلر » .

قسم الانتاج الحيوان (فرع التنفيذية) كلية الزراعة . جامعة القاهرة .

ولقد وجد أن المجموعة التي غذيت على عليقة فقيرة في الفسفور قد احتجزت أعلى كميات من الفسفور المشع (نشاط نوعي موحد) في أعضائها وأنسجتها المختلفة واحتجزت المجموعة التي غذيت على عليقة غنية في الفسفور أقل مقادير منه . وكان النشاط النوعي الموحد للأنسجة والأعضاء المختلفة في المجاميع الثلاث عاليا في الأوقات القريبة من الحقن بينما وجد أنه ازداد في العظام في الفترات البعيدة من وقت الحقن .