Discussion. When one contrasts the "specific radioactivities" of Case 4 with those of the other 3 cases it becomes apparent that fairly large amounts of P^{32} have been retained by the various tissues at the end of the second day after administration. Apparently, however, a large part of the quantity leaves the tissues between the second and eleventh day. That retained is apparently specific for each type of tissue since the "specific radioactivities" of each tissue of the 3 cases were similar and were independent of the total amount (within the limits listed) of P^{32} administered and the other factors mentioned above. The quantities of phosphorus appearing in the tissue during the first few days probably take part in "carbohydrate" metabolism while those retained for longer periods of time may be involved in "nucleoprotein" and "phospholipid" metabolism.⁵

13064 P

Photoelectric Microdetermination of Calcium in Serum.

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A new microcolorimetric method has been developed for the determination of calcium by direct precipitation, in samples of 0.2 cc or less of serum. Although work is now in progress on a simplification of the technic and the use of smaller samples for analysis, the following procedure has given, for some time, such satisfactory results (accuracy within $\pm 2\%$) as to warrant its present description.

Procedure. In test tubes $(10 \times 75 \text{ mm})$, to 0.2 cc samples of serum are added 1.0 cc of water and 0.2 cc of saturated ammonium oxalate. Two *blank* samples, with water in place of serum, are treated in exactly the same way. After standing overnight, 0.2 cc of 0.1% Triton N E (Röhm and Haas) is added to each tube,¹ the contents stirred, then poured into a pyrex sintered glass filter funnel (No. 2 F, Buchner type) held by a rubber stopper in the mouth of a 500 cc suction flask. Three successive portions of 0.8 cc of 2% ammonia water are pipetted into the tube, agitated to pick up any remaining crystals of CaC₂O₄, then poured upon the filter. A receiving tube $(7 \times 7/8^{"})$ is placed in the suction flask under the stem

⁵ Tuttle, L. W., Erf, L. A., and Lawrence, J. H., J. Clin. Invest., 1941, **20**, 57. ¹ Alter, C. M., and Thomas, D. S., Jr., Ind. Eng. Chem., Anal. Ed., 1940, **12**, 525.

of the filter funnel. The vacuum is turned off, while the walls of the precipitation tube are washed with 1 cc of hot 1 N H₂SO₄ which is then poured on the filter and allowed to stand for one minute. Suction is reapplied and the acid is caught in the receiving tube. The precipitation tube is discarded. The walls of the filter funnel are then washed with 1 cc and 2 cc portions of hot 1 N H₂SO₄, allowed to stand for one minute as above.

The 4 cc of acid in the receiving tube are transferred to a 50 cc volumetric flask, with 3 washings of 3 cc of water. To the 2 blanks, 1 and 2, which have been washed and filtered in exactly the same way as the serum samples, in the 50 cc flasks there are added 1 cc and 2 cc, respectively, of freshly prepared 0.000625 N $Na_2C_2O_4$ (1 cc of 0.125 N diluted to 200 cc). To each flask there is then added 1 cc of 0.0018 N Ce $(SO_4)_2$ (9 cc 0.1 N diluted to 500 cc in 1 N H₂SO₄). After mixing, the flasks are allowed to stand for 30 minutes at room temperature. One cc of freshly prepared 0.5% KI is added to liberate free iodine,² allowed to react for 60 seconds, then water is added to approximately 40 cc volume. The contents are brought to temperature of 25°C, and 0.5 cc of 2% starch in saturated NaCl solution is slowly added, with gentle mixing by rotation. Water is added to volume, and 15 cc portions of the blue solution are read at 25°C in the Evelyn colorimeter,³ with a No. 600 filter in place.⁴ The center setting is obtained with a pure water tube set at 100.

The readings are plotted on semi-logarithmic paper (K. and E. No. 358-51) with abscissa units of micro-equivalents oxalate per liter from 0 to 35, and ordinate units of galvanometer readings from 10 to 100. A straight line is drawn between the readings for the two *blanks 1* and *2*, representing 12.5 and 25 micro-equivalents per liter, respectively, of oxalate. The oxalate values for the serum samples found by interpolation of the galvanometer readings on this line, when multiplied by 0.25 or 0.5 give directly Ca as milli-equivalents per liter, or mg per 100 cc, respectively, in the original serum samples.

Results. Samples of human serum analyzed as above (C) and by gasometric analysis (G) of 1 cc samples directly precipitated,⁵ in terms of mE. Ca per liter serum, gave the following typical results:

Sample	1	2	3	4	5	6	7	8	9	10
G	4.83	4.97	4.51	4.90	5.06	4.84	5.28	4.92	5.12	5.12
C	4.80	5.00	4.35	4.80	5.20	4.81	5.23	4.86	5.07	5.17

² Groák, B., Biochem. Z., 1929, 212, 47.

³ Evelyn, K. A., J. Biol. Chem., 1936, 115, 63.

⁴ Sendroy, J., Jr., J. Biol. Chem., 1939, 130, 605.

⁵ Van Slyke, D. D., and Sendroy, J., Jr., J. Biol. Chem., 1929, 84, 217.

The average deviation of the micro (0.2 cc) colorimetric results from the macro (1 cc) gasometric values was $\pm 1.5\%$, with extreme limits of -3.6 and +2.8%.

13065 P

Electro-Encephalographic Diagnosis of Extradural and Subdural Hemorrhage.

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It has been our belief that abnormal waves produced by pressure lesions upon the surface of the brain, such as extradural or subdural hemorrhage, etc., would differ from those wherein the brain was actually damaged or destroyed by laceration, contusion or tumor invasion. In order to determine these facts, we have studied, to date, 30 experimental animals, retesting them for 4 months, and 3 clinical cases of verified acute and subacute hematomas.

In 3 patients with verified subdural hematoma, 2 were subacute and one was acute; the electro-encephalographic diagnosis demonstrated delta waves of 1 cycle per second, with a voltage of 100 microvolts, combined with a rapid wave of 16 cycles per second, and a voltage of 20 microvolts. This pattern was localized over the hemorrhage. Following the surgical removal of these hemorrhages, the waves immediately returned to normal in the acute case. Six months later the 2 subacute cases showed normal waves. In the acute case the test was repeated within 12 hours after the removal of the hemorrhage. The high beta waves had disappeared and the 1 and 2 cycle delta waves of 100 microvolts had changed to 2 to 4 cycle deltas of 75 to 150 microvolts. This type of wave was found in all leads.

The normal electro-encephalogram of the rabbit, as obtained through the intact skull, was established by 150 recordings. No. 5 gelatine capsules, covered with animal membrane, were filled with citrated rabbit's blood. In one-half of these capsules thorotrast was added to make the capsule opaque to the roentgen ray. This addition of thorotrast did not affect the electrical waves. Under aseptic pro-