the fibres used (about 8–9 μ diameter, the internodal distance being less than 1 mm). The experiments were carried out at room temperature (20–23°C). Under these conditions the isolated nerve fibres remain excitable and action potentials can be recorded for several hours after dissection.

Figures *a*–*b*–*c*–*d* illustrates the results of one of the experiments. Each of the photographs is obtained by the superimposition of the potentials elicited by 10 successive stimuli, cathodic shocks of brief duration (about 60 μs). Record *a* shows the local potential changes due to shocks of strength equal to 0·5 threshold. After the initial shock–and capacitative–artifacts, a lasting polarization (electrotonic potential) can be observed which declines in an approximately exponential way. If the strength of the shock is now increased to about 0·82 threshold–Figure *b*–a hump in the decaying polarization potential appears, corresponding to a local response which develops above the passive electrotonic potential. In this record a fluctuation in the amplitude of the local response is evident; the size of the potentials generated by shocks of the same strength vary within a wide range. This fluctuation is even more evident if the potentials are produced by shocks of higher intensity (0·85 threshold in Figure *b*). Most of the potentials so elicited are only slightly higher than those of Figure *b*, whereas two of them have grown into huge humps which illustrate particularly well the phenomenon of the local response. In Figure *d* the strength of the shock was slightly above threshold, and propagated action potentials arising after various delays can be seen. A potential change is also seen in this record which propagated away although it did not grow to a full spike at the directly stimulated node. Apparently, the local response of this node of Ranvier stimulated by electrotonic spread the adjacent node, whose excitability was higher.

A comparison of these results with those already quoted from amphibian medullated fibres, non-medullated invertebrate axons and striated muscle fibres, reveals a fundamental similarity of the events which after cathodic stimulation lead to the onset of propagated potentials in all the excitable tissues which have so far been studied.

J. DEL CASTILLO-NICOLAU

Department of Physiology, University College, London, October 30, 1951.

Zusammenfassung

Lokale Potentiale, welche für die Entstehung fortgeleiteter «Spikes» verantwortlich sind, konnten an einzelnen Ranvierschen Schnürringen motorischer Fasern des Mäuse-Ischiadicus nachgewiesen werden.

Diese Befunde stützen die Ansicht, dass die Entstehung fortgeleiteter Potentiale in allen erregbaren Membranen auf ähnliche Weise stattfindet.

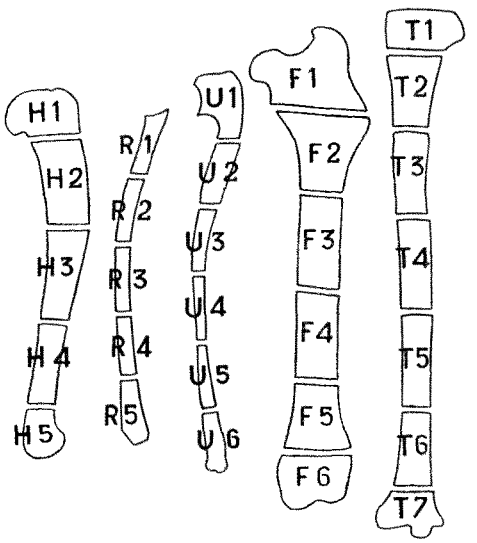
Distribution of Radiophosphorus in the Long Bones of Adult Rabbits[1]

The earliest experiments made with radiophosphorus[2] have shown that, immediately after administration of the isotope, the specific activity of the epiphyses of the long bones is always found to be higher than that of the diaphyses.

Several authors[1] add that the specific activities of the phosphorus are reduced to uniform values after some days or a few weeks.

As far as we can ascertain, this latter statement rests on experiments made with growing animals, and its general validity should not be accepted before experiments are made with fully grown animals.

Since it has been shown that bone marrow[2] and periosteum[3] have a high phosphorus specific activity, care must be taken to deal with bone tissue completely freed from the soft tissues.



This figure shows how the bones were divided. *H* = humerus; *R* = radius; *U* = ulna; *F* = femur; *T* = tibia.

Furthermore, it seems advisable to estimate the specific activities, not just in the shaft as compared with the epiphyses, but in as many portions of the long bones as is compatible with accurate measurements.

Procedure. Three fully grown rabbits (skeletal state checked by X-ray examination), weighing an average of 4·800 kg, were injected subcutaneously with carrier-free P 32 in solution made up in normal saline at pH 7 (supplied by Atomic Energy Research Establishment, Harwell). The one to be sacrificed 6 days afterwards received 2 mC in two days. The two others received 4 mC in three days.

The bones were cleaned with a knife and divided with a band-saw into portions as indicated by the Figure. These portions were boiled for one hour, digested for 24 hours at 37° in a 1 per cent. solution of papain renewed after 12 hours, boiled again for a few minutes, dried, and extracted in chloroform in a Soxhlet apparatus for 24 hours. They were then dried, weighed and dissolved in nitric acid. An aliquot was used for radio-assay while the remainder of the solution was used for chemical determination of phosphorus according to the method of Fiske and Subarow.

Results. The bones, before going to nitric acid, ap-

[1] This work was supported in part by grants from the Fonds National de la Recherche Scientifique of Belgium.

[2] L. A. HAHN, G. C. HEVESY, and E. C. LUNDSGAARD, Biochem. J. *31*, 1705 (1937).– M. J. L. DOLS, B. C. P. JANSEN, G. J. SIZOO, and G. J. VAN DER MAAS, Nature *142*, 953 (1938).—G. C. HEVESY, H. B. LEVI, and O. H. REBBE, Biochem. J. *34*, 532 (1940).

[1] M. L. MANLY and W. F. BALE, J. Biol. Chem. *129*, 125 (1939). R. S. MANLY, H. C. HODGE, and M. L. MANLY, J. Biol. Chem. *131*, 293 (1940).

[2] W. D. ARMSTRONG and C. P. BARNUM, J. Biol. Chem. *172*, 199 (1948).

[3] C. P. LEBLOND, G. W. WILKINSON, L. F. BELANGER, and J. ROBICHON, Amer. J. Anat. *86*, 289 (1950).

peared under the binocular microscope, to be completely cleaned of articular cartilage and of soft tissues.

The specific activities have been expressed in number of counts per minute per mg of phosphorus. Since the interest lies in the comparison of various portions of the bones at various intervals, an arbitrary value of 100 has been assigned to the middle fifth of the humerus in the three animals and all the measures have been reduced accordingly.

SPECIFIC ACTIVITIES / DAYS	6	31	76		6	31	76		6	31	76
H 1	344	286	315	R 1	90	101	145	U 1	110	186	249
H 2	211	160	140	R 2	47	44	86	U 2	72	98	154
H 3	100	100	100	R 3	81	51	144	U 3	56	63	119
H 4	90	83	94	R 4	65	65	152	U 4	62	76	181
H 5	135	161	194	R 5	169	359	379	U 5	57	65	174
								U 6	135	142	358

SPECIFIC ACTIVITIES / DAYS	6	31	76		6	31	76
F 1	156	271	263	T 1	288	346	302
F 2	115	176	155	T 2	221	142	190
F 3	77	91	81	T 3	141	108	150
F 4	78	89	99	T 4	151	97	151
F 5	101	155	180	T 5	126	132	127
F 6	107	274	219	T 6	137	104	217
				T 7	294	184	179

Specific activities at various time intervals of the portions of the bones as indicated by the Figure.

The results are tabulated (Table) and the Figure gives the key to the symbols indicating the portions of the bones.

Discussion. Large doses of radiophosphorus were used in order to recover a fairly good activity in the smallest portions of the bones, even after 76 days. With such doses, radiation damage is a theoretical possibility, although it should be pointed out that the animals did not lose weight during the experiment.

The number of animals is too small to allow comparison between them of corresponding portions of bones at various time intervals. The only conclusion which may be drawn from the results as a whole is that, at least up to the 76th day, the phosphorus specific activities of the epiphyses and of the diaphyses do not show any tendency to be reduced to uniform values.

We have used fully grown animals, and the discrepancy between our results and those reported up to now is thus easy to explain; it shows that the two sets of experiments, the previous ones with growing, and ours with adult animals, must be clearly distinguished from each other.

It is suggested that in the discussion of results obtained with adult animals, due consideration be given to the studies of WEIDMAN and ROGERS[1]. These authors have found that the cancellous femoral bone of the adult rabbit femur contains less calcium and more nitrogen than the cortical bone. Similar figures had been reported by STROBINO and FARR[2] who had found that a minimum value for nitrogen and a maximum for ash existed at the longitudinal midpoint of the long bones of cows and oxen. In connection with these values, let us recall here that COHN and GREENBERG[3] had been led to assume that the organic phosphorus metabolism may be an important factor in the mineralization of the bone.

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[2] L. J. STROBINO and L. E. FARR, J. Biol. Chem. *178*, 599 (1949).
[3] W. E. COHN and D. M. GREENBERG, J. Biol. Chem. *130*, 625 (1939).

Addendum: Since this article was submitted to the Editors, three papers[1] have appeared on closely related subjects. RUTISHAUSER and MAJNO have confirmed that cancellous bone is less mineralized than compact bone in man. PERROTTET and DUCKERT have found the same difference in rabbits. AMPRINO has presented the first results of a radioautographic analysis of the distribution of labelled Ca and P in bones which might lead to an understanding of the data recorded here.

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Institute of Anatomy, University of Louvain, Belgium, November 18, 1951.

Résumé

Dans les os longs du lapin adulte, les activités spécifiques du phosphore des épiphyses et des diaphyses ne manifestent pas, au cours d'une période d'observation de 76 jours, cette tendance à se réduire à des valeurs uniformes qui avait été enregistrée dans les expériences utilisant des animaux en croissance.

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Sur l'action des esters amides polyphosphoriques de l'aneurine sur la glycolyse par le sang lavé

Nous avons précédemment rapporté[1] la faible activité cocarboxylasique des esters amides polyphosphoriques de l'aneurine (E.A.P.P.). Nous pouvions dès lors nous demander si la présence des deux chaînes au moins triphosphoriques que nous fixons par valence chimique sur la molécule d'aneurine[2] ne conférait pas à ces corps d'autres propriétés biochimiques. Nous avons, dans cette voie, entrepris des recherches préliminaires en réalisant la chaîne de glycolyse constituée par du sang humain lavé 3 fois avec du liquide de Ringer alcalin et mis ensuite en suspension dans une solution de Ringer additionnée de glucose (2 g pour 1000), de pyruvate de sodium (1 g pour 1000) et de chlorure de magnésium (0,005 g pour 1000).

Nous portons dans plusieurs matras 40 cm³ de cette suspension de globules correspondant à 20 cm³ de sang total initial. Deux des matras sont utilisés tels quels (témoins), et nous ajoutons aux autres une certaine quantité de cocarboxylase ou d'E.A.P.P. Ces derniers ont été utilisés avant séparation des polyphosphates minéraux[3] (liqueur totale) ou après séparation de ces derniers corps au moyen du roussinate de sodium (esters purifiés).

Après avoir effectué les prises d'essai nécessaires aux dosages, nous établissons une atmosphère d'azote et nous portons les matras au thermostat à 37°. Nous agitons 1 à 2 heures et nous effectuons ensuite de nouvelles prises d'essai en vue des dosages. Ces recherches préliminaires étant destinées à reconnaître si ces corps avaient ou non une action biochimique sur le déroulement des phénomènes de glycolyse, nous avons utilisé dans ce premier travail des méthodes de dosage qui nous fournissaient seulement des résultats globaux.

Nous dosons les substances réductrices fermentescibles aldosiques par la méthode de HAGEDORN-JENSEN[4].

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