THE USE OF RADIOACTIVE PHOSPHORUS IN THE DIAGNOSIS OF AVASCULAR NECROSIS OF THE FEMORAL HEAD

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Mechanical factors have now been excluded as the main cause of non-union in fractures of the femoral neck: the development of efficient methods of internal fixation has removed this source of non-union and the chief remaining cause is disturbance of the circulation of blood to the capital fragment. The incidence of avascular necrosis of the femoral head appears to depend upon normal variations in the vascular pattern and also upon the severity of the vascular injury (Kolodny 1925, Wolcott 1943, Tucker 1949). It is possible that delayed vascular occlusion may play a part; but the vascular damage is usually sustained at the time of the original injury, although it may perhaps be increased by inadequacy of immobilisation, violence of manipulation and by the effects of some forms of internal fixation.

It is a striking fact that there may be a mass of avascular bone in the body without any means of distinguishing it clinically or radiographically from living bone. This is true particularly during the first few months after fracture of the femoral neck with vascular occlusion; there is no reliable clinical evidence as to the viability of the femoral head. Eyre-Brook and Pridie (1941), in reporting a series of such fractures, noted that avascular necrosis of the capital fragment did not develop when the fracture-shaft angle was greater than 37 degrees; but apart from this observation there is little evidence on which the diagnosis can be based and each fracture is an enigma. The surgeon must wait for several months, or even for several years, before the fate of the head can be predicted with certainty.

THE RELATIONSHIP OF PATHOLOGICAL TO RADIOGRAPHIC CHANGES IN AVASCULAR NECROSIS OF BONE

The first result of loss of blood supply to bone is death of the cellular components—osteoblasts, osteoclasts and marrow cells. Calcified intercellular substance is not a living tissue—it is the inert product of osteoblasts—and for this reason, death of bone does not produce immediate change in its radiographic density. After injury, and the disuse that follows injury, there is usually deossification of living bone; and since dead bone cannot share in this vital process differences in radiographic density may be observed when dead and living bone are compared. The contrast in density depends largely upon the degree of immobilisation that is maintained, and to a lesser extent upon the local reactions of bone to injury. Now that internal fixation is used so generally, and early functional activity is encouraged, such atrophy of disuse is minimised and there is therefore no certain contrast, and perhaps no contrast at all, between living and dead bone.

Axhausen (1922) showed that when bone was avascular the organic matrix gradually lost its strength and resistance. Under the pressure of weight-bearing the degenerated trabeculae were liable to fracture and compression, and bone dust accumulated in the marrow spaces. Such collapse of cancellous bone is the usual cause of true increase of the density of avascular bone—a change that becomes evident radiographically from three months to three years after injury. Moreover, like other dead tissues, dead bone may attract calcium salts from surrounding fluids thus accounting for increase in density when the bone is not crushed and compressed but retains its original contour.

In time, an inflammatory response develops in adjacent living tissue: the avascular fragment is invaded by granulation tissue; the bone matrix is eroded by osteoclastic components; and new bone is laid down on the remaining trabeculae. Such regeneration is,

of course, slow. Phemister (1939) said that "substitution of the dead portion by the living bone may gradually take place and in from one and a half to three years it may be complete." This slow process of revascularisation with erosion of trabeculae can be traced in radiographs by the extending area of reduced density during which time there is an increasing tendency to collapse under the strain of weight-bearing.

THE PROBLEM OF EARLY DIAGNOSIS OF AVASCULAR NECROSIS

The fate of the femoral head is usually determined at the time of injury; and, if there is complete loss of blood supply, every attempt to restore normal function must fail. How then can a reliable indication of the vascularity of the femoral head be gained within a few days of fracture? At first, attempts were made to determine whether or not there were living cells by removing bone with a trephine and studying the cell structure; but this never gave conclusive evidence when carried out within a few days of injury. Supravital staining of bone and marrow cells was also attempted but it proved to be unreliable. Attention was therefore directed to measures by which the arterial supply to bone could be demonstrated. The injection of non-toxic chemicals and dyes, such as fluorescein, was rejected by reason of the difficulties of detection and the lack of sensitive reaction. The method finally selected was the intravenous injection of radioactive isotopes which are carried by the blood stream to all tissues with a normal blood supply. Demonstration of the isotope in a given tissue proved that the circulation was normal, or at least that there was absorption from neighbouring vascular tissues. After fracture of the neck of the femur, comparison of concentration of the isotope in the femoral head with that of a control bone such as the trochanter in which the blood supply was known to be normal, gave an indication of the residual vascularity of the femoral head. Theoretically, vascularity could be assessed most accurately if the isotope was retained in the blood stream so that the radioactive content of a unit mass of bone would be an accurate index of the active vascular bed in the bone. Such an isotope is not yet readily available. In this investigation radioactive phosphorus was used—a safe and readily available isotope which is a normal metabolite showing a tendency to accumulate in bone.

Radioactive phosphorus has the same physical and chemical properties as ordinary phosphorus but is distinguished from it by the emission of β rays which can be detected. The half-life of radioactive isotope P^{32} is 14.3 days which is long enough for our purpose. Moreover, the rate of radioactive decay is constant under all conditions; the β rays are absorbed by the tissues, 0.1 millimetres of tissue absorbing about 90 per cent. of the rays (Hamilton and Soley 1939). By placing the ashed tissue near a Geiger counter the radioactivity of the sample can be measured accurately. The number of counts recorded from 1 gramme of tissue in one minute gives an assessment of the content of P^{32} in the tissue. It is necessary to subtract a predetermined number of counts, known as the "background count," due to the recording of cosmic rays on the Geiger counter.

For a given dose of P^{32} the amount of radioactive phosphorus deposited in a unit quantity of bone is variable and seems to depend on the vascularity of the bone, its composition and the time interval between injection and assay. Since the relative proportions of bone, red marrow and fatty marrow vary in the individual, and from one area of bone to another, it will be appreciated that the amount of radioactive isotope accumulating in bone in a given time will vary according to the tissue composition. Red marrow contains more phosphorus than fatty marrow and both these components are difficult to separate from the bone tissue. For this reason, small variations are to be expected between any two specimens.

METHOD OF INVESTIGATION

Radioactive phosphorus was obtained in the form of disodium phosphate in aqueous solution. It was diluted with normal saline and sterilized by autoclaving. Investigations

were first undertaken in rabbits. The dose employed was 50 microcuries injected into an ear vein. Operations on the neck of the femur and ligamentum teres were performed before injection of the isotope. Two hours after injection, a fragment was removed from the femoral head and also from a neighbouring bone the blood supply of which was known to be undisturbed. The details of the experiments, which were undertaken by Professor H. L. Sheehan, will be understood from three representative examples.

Experiment 1—Under anaesthesia the femoral neck was divided close to the trochanter and 50 microcuries of radioactive phosphorus in 0.8 millilitre of disodium phosphate were injected into an ear vein. Both femoral heads were excised two hours later and assayed:

	Femoral head subjected to operation	Normal femoral head as control
Weight of bone	0.626 gm.	0.461 gm.
Total weight of ash	0.245 gm.	0.168 gm.
Ash for assay	0.166 gm.	0.165 gm.
Count per minute	621	757
Count/gm. wet bone/min. . . .	1450	1675

Ratio of isotope in the femoral head on the side of operation,
as compared with normal control bone, 1 : 1.

Experiment 2—Under anaesthesia the right femoral head was dislocated and the ligamentum teres divided. One hour later 50 microcuries of radioactive phosphorus in 0.8 millilitre of disodium phosphate solution were injected into an ear vein. Two hours later both femoral heads were excised and assayed:

	Femoral head on the side of operation	Opposite femoral head as control
Weight of bone	0.383 gm.	0.583 gm.
Weight of ash	0.143 gm.	0.180 gm.
Ash for assay	0.134 gm.	0.169 gm.
Count per minute	509.6	549.2
Count/gm. wet bone/min. . . .	1420	1005

Ratio of isotope in the femoral head on the side of operation,
as compared with normal control bone, 1 : 1.

Experiment 3—Under anaesthesia the right femoral head was separated from its blood supply by division of the femoral neck and division of the ligamentum teres. The head was replaced in the acetabulum, the wound closed, and 50 microcuries of radioactive phosphorus in 0.8 millilitre of disodium phosphate were injected into an ear vein. Both femoral heads were excised two hours later and assayed:

	Femoral head on the side of operation	Normal femoral head as control
Weight of bone	0.389 gm.	0.323 gm.
Total weight of ash	0.149 gm.	0.166 gm.
Ash for assay	0.143 gm.	0.121 gm.
Count per minute	152	1300
Count/gm. wet bone/min. . . .	407	5540

Ratio of isotope on the side of operation, as compared with normal
control bone, 1 : 14.

These three experiments indicate that basal fracture of the femoral neck alone, and division of the ligamentum teres alone, does not alter the vascular supply to the femoral head in rabbits; but that when all vascular connections to the femoral head were severed

TABLE I
ANALYSIS OF THIRTEEN FRACTURES OF THE NECK OF THE FEMUR INVESTIGATED BY
INJECTION OF RADIOACTIVE PHOSPHORUS

Patient	Initials and age of the patient in years	Type of fracture	Date of operation	Dosage of radioactive isotope P^{32} in c.c.	Time between injection of isotope and removal of bone sample	Radioactive emanation per minute/gramme		Ratio of isotope content Head: Trochanter	Clinical result
						Head	Trochanter		
1	M. H. 69	Basal neck	9. 9.47	135	4 hours	722	730	1 : 1	Viable head. Union
2	M. E. J. 70	Subcapital	16. 9.47	287	3 hours	97.9	*	1 : 16	Avascular necrosis
3	M. C. 83	Mid/cervical	7.10.47	150	2 hours	63	680	1 : 10.9	Avascular necrosis
4	A. W. 68	Subcapital	13. 1.48	100	30 minutes	324	204	3 : 2	Viable head. Union
5	S. K. 65	Subcapital	19. 1.48	100	5 minutes	134	225	1 : 1.6	Viable head. Pin migration
6	E. P. 78	Subcapital	4. 2.48	110	25 minutes	32.2	97.7	1 : 3	Viable head. Poor pinning. Non-union
7	A. B. 59	Subcapital	9. 2.48	120	45 minutes	37.5	18.5	2 : 1	Viable head. Union
8	E. C. 70	Subcapital	16. 2.48	200	1 hour	1265	1385	1 : 1	Viable head. Union
9	M. M. 68	Subcapital	16. 2.48	175	1 hour	593	336	2 : 1	Viable head. Extrusion of pin
10	W. H. 69	Subcapital	22. 3.48	175	90 minutes	188	182	1 : 1	Viable head. Union
11	W. J. 54	Subcapital	3. 5.48	165	1 hour	40.7	218	1 : 5	Segmental avascular necrosis
12	E. M. C. 48	Mid/cervical	4. 5.48	150	1 hour	444	196	2.3 : 1	Viable head
13	R. M. 18	Slipped epiphysis	4. 5.48	150	2 hours	164	325	1 : 2	Viable epiphysis

* In this case a sample was not removed from the trochanter, and a control the time interval between administration of the isotope and removal of was not therefore available. But on consideration of the dosage of P^{32} , and the bone, the *minimal* count to be expected would give a ratio of 1:16.

by division of both these structures, absorption of radioactive material dropped to a very low level so that the ratio of content in the femoral head on the operated side was only one-fourteenth that of bone with a normal blood supply.

The detection of some radioactive phosphorus in the dead head may appear to contradict the very principles involved; but it is explained adequately by absorption of radioactive phosphorus from the surrounding haematoma and synovial fluid. For this reason negative counts on the Geiger counter are unlikely to be obtained even when the bone is completely avascular. Nevertheless if a bone sample is removed from the centre of the bone within an hour of administration of the isotope, absorption from the surrounding tissue fluids should be minimal.

The method applied to human cases—In humans the isotope was injected into the antecubital vein, the usual dose being from 150 to 200 microcuries. A sample of bone was

FIG. 1

FIG. 2

Case 2. M. E. J. There is radiographic evidence of avascularity of the femoral head in the increasing penetration of the nail through the relatively dense head (Fig. 2); but this evidence became available only after two years. At the time of the original operation the content in the femoral head of radioactive phosphorus injected intravenously was estimated at one-sixteenth that of normal bone, thus proving avascularity.

removed from the head of the femur after reduction of the fracture and the insertion of a guide wire by which to direct the three-flanged nail used for internal fixation. A quarter-inch drill hole was made, concentric with the guide wire, down to the fracture line, preferably in the posterior and inferior quadrant of the neck; and a calibrated three-sixteenths inch trephine, six inches in length, was passed along the drill hole and hammered or rotated into the proximal fragment. The position of the trephine was checked radiographically in two planes before it was driven into the head, and radiographs were also taken after final insertion, in order to confirm that the sample was obtained from the proximal fragment. The fragment of bone thus secured was sprayed immediately with saline or water to remove surface blood, and dried on a piece of gauze. A piece of cancellous bone was also removed from the greater trochanter by a similar technique. Both specimens were assayed for radioactive content.

Specimens weighing from 100 to 150 milligrammes were usually obtained. The specimen was weighed accurately, ashed, spread evenly on a small paraffin-coated paper tray, and the radioactive content assayed by a Geiger counter. The pulses of the counter were recorded on a scale-of-hundred unit (Rotblat *et al.* 1948). Each specimen was examined on three occasions for a period of twenty minutes and the average reading was calculated. By further calculation the activity per gramme of wet tissue per minute was determined and the counts of the two samples were compared. As a rule the ratio of radioactive content of the bone under investigation in relation to normal bone was approximately 1:1; but variations from 1:3 to 3:1 were accepted as evidence of a normal blood supply.

Results of investigation—Table I summarises the observations in a series of patients investigated by this method. It is not suggested that these are more than preliminary findings. It is of course possible that patients who do not yet show clinical evidence of avascular

FIG. 3

FIG. 4

Case 3. M. C. Radiographs twelve months and eighteen months after operation, in which the nail was at first placed perfectly in the middle of the head, leave no doubt that the head is avascular. But this radiographic proof was gained only after twelve months—which was too late. At the time of the first operation, when radioactive phosphorus was injected into the antecubital veins, the femoral head took up only one-tenth of the content of bones with a normal blood supply. Laboratory evidence of death of bone can be gained within hours instead of months or years.

necrosis of the femoral head may develop signs of it in future years. But it is noteworthy that, in this early stage, when a closely comparable content of the radioactive isotope was found in the femoral head and the trochanter (with ratios of 1:1, 3:2, 2:1 and 1:2) the fractures united quickly and there was no clinical or radiographic evidence of avascular necrosis; whereas when the ratio of isotope content between the femoral head and trochanter was 1:16 or 1:10, there was undoubted clinical evidence of avascular necrosis; and in case 11 where there was a ratio of 1:5 there was evidence of localised segmental necrosis.

Dangers and technical difficulties—The possibility of complications arising from the administration of radioactive phosphorus calls for careful consideration. Radioactive phosphorus may depress the activity of the haemopoietic system and indeed it is used in the

treatment of polycythaemia vera. But the dosage used to inhibit haemopoiesis (4 to 12 millicuries) is from twenty to sixty times greater than that used in this investigation (150 to 200 microcuries). We have seen no evidence of injury to the cellular components of the blood. The precise dosage of radioactive substances needed to stimulate carcinogenesis is unknown but we believe that in this investigation we have always been far below the critical level. Finally, although radioactive substances have been proved to cause mutation of genes in animals, and may no doubt have similar effect in humans, this can have little application to the aged patients we have studied.

Drilling of the femoral neck and head may damage the bone sufficiently to impair the efficiency of the Smith-Petersen nail. This risk can be reduced by careful technique, particularly if the sample is taken from the posterior and inferior quadrant of the neck and head. The greatest danger is that the trephine may damage the remaining vascular supply to the head. The most important vessels are the postero-superior retinacular arteries and the foveolar vessels. Any instrument driven into the head in the postero-superior quadrant, or into the foveolar region, may well injure the remaining patent vessels; but if these zones are avoided the investigation can be carried out with safety.

DISCUSSION

In an effort to reduce the amount of radioactive material to a minimum, dosages ranging from 287 to 100 microcuries of P^{32} were tried. If the readings are to be significant they must be considerably greater than the background count. A satisfactory reading was obtained in case 1 with a dose of 135 microcuries; but a dose of 120 microcuries in case 7 gave a reading that was too close to the background count to be of much value. Moreover, in case 1, four hours elapsed between injection of the isotope and removal of the sample and this long interval may be responsible for a relatively high reading in an avascular femoral head. There can be little doubt that 150 microcuries is the minimal dosage if local concentration in the bone tissues is to be examined adequately.

Time factors were also varied in an effort to determine the optimal interval between the injection of isotope and the removal of samples; the intervals varied from five minutes to four hours. In animal experiments it was found that the femoral head contained some radioactive material even when it was completely isolated from its attachments, and this has been attributed to absorption from the surrounding synovial fluid and blood. It was thought that the shorter the period of contact of the avascular fragment with the medium containing the isotope, the smaller would be the radioactive content of the avascular fragment. The interval was reduced to five minutes in one case, and to twenty-five minutes in another, but it seems probable that one hour is the optimal interval.

One of the disadvantages of this method of investigation is that only one small area of the femoral head is obtained for examination. It is known that segmental necrosis of the femoral head may occur and the method described may fail to demonstrate such regional infarction. It is unwise to remove more than one sample from the head because of the risk of damaging the blood vessels that remain. Moreover the technique requires radioactive material and expensive recording equipment. It is submitted, however, that laboratory evidence of death of the femoral head may be gained within a few hours; whereas clinical evidence, equally conclusive, is delayed until after many months.

It is hoped that the method will be investigated by others and that, if confirmed, it may become an accepted routine in the treatment of intracapsular fractures of the neck of the femur when the viability of the head is in doubt. It may even be possible to succeed in the attempts we have made to construct a small Geiger counter which can be inserted into the femoral head at the time of the original operation so that the surgeon will know at once whether the femoral head is dead or alive and will be able to modify his treatment accordingly.

SUMMARY

1. The pathology of avascular necrosis of bone and its relationship to the radiographic changes are considered briefly.
2. The inadequacy of radiographic examination in the early diagnosis of avascular necrosis is stressed.
3. Methods of early assessment of the residual vascularity of a bone mass are discussed.
4. Details of the principles, methods, and results of using radioactive phosphorus in the diagnosis of avascularity of bone are described.
5. The dangers and technical difficulties of using radioactive substances are discussed.
6. The possibility is envisaged that surgeons may soon be able to determine at the time of emergency operation whether the femoral head is dead or alive.

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