Severe undernutrition in growing and adult animals

14.* The shafts of the long bones in pigs

BY C. W. M. PRATT AND R. A. McCANCE

Department of Anatomy and Medical Research Council Department of Experimental Medicine, University of Cambridge

(*Received* 25 *June* 1964—*Accepted* 13 *July* 1964)

The effects of undernutrition on the shafts of growing mammalian bones are not well known. Long bones grow relatively slowly in girth as compared with their more rapid growth in length so that the usual short-term experiments produce little or no change in the cortical bone. Jackson (1925) made no comment on the effects of inanition upon the shafts of growing bones of animals although Nicolaeff (1923) reported osteoporotic changes in the bones of famine-stricken children.

It has already been shown that there is some growth in length of the bones during undernutrition (Dickerson & McCance, 1961; Pratt & McCance, 1964), but the former authors showed that during prolonged undernutrition bones acquired approximately the proportions proper for their age rather than for their general size by a relatively greater increase in width than length. The nature of this increase in width will now be described and will be followed by an account of the changes in the shaft during rehabilitation.

EXPERIMENTAL

Forty-two pigs were used in these experiments. They were derived originally from a Large White × Essex cross. The undernourished animals were weaned at 7–14 days of age and were fed and housed as described by McCance (1960). The food, had they been given enough of it, would have given excellent rates of growth. The undernourished animals were killed at intervals up to 59 weeks, by which times they weighed from 3 to 8 kg. The animals were rehabilitated on the lines described by Mount, Lister & McCance (1963). Animals were killed at stages up to full recovery. The well-nourished control animals were derived from the same stock and often from the same litters as the undernourished animals.

The limb bones were removed and X-rayed and the femur was sectioned and stained as previously described (Pratt & McCance, 1960). The process of rehabilitation was also followed by a series of radiological examinations under Halothane (BP) anaesthesia.

RESULTS

Radiological appearances

Normal pigs. The femur of a pig 3 weeks old weighing 3·7 kg (Pratt & McCance, 1964, Pl. 1 *a*) had a short cylindrical shaft with an hourglass-shaped marrow cavity.

* Paper no. 13: *Brit. J. Nutr.* (1964), **18**, 587.

The cortical bone was therefore thickest in the centre of the shaft. The periosteal surface was fuzzy. The marrow cavity was filled with a dense reticulum of spongy bone.

In a pig 4 weeks old weighing 7·23 kg (Pratt & McCance, 1964, Pl. 1*d*) the shaft had increased both in external diameter and cortical thickness, the latter mostly on the posterior aspect. There had also been a simultaneous increase in the overall diameter of the marrow cavity, with a greater extension posteriorly. Where cortical bone had previously been, only occasional, radially directed trabeculae persisted. The reticulum of spongy bone had tended to disappear from the central portion of the shaft.

Pigs during the early stages of undernutrition. The X-rays of the femurs of three pigs which had been undernourished for 6 (Pratt & McCance, 1964, Pl. 1*b*), 7 or 8 weeks respectively showed that there had been an increase in width during this period. The periosteal outline had also become more sharply defined and the cortex thinner than in normal bones of the same size but the trabeculae in the medulla looked unchanged.

Pigs undernourished for long periods. Pigs which had been undernourished for more than 2 months (Pratt & McCance, 1964, Pl. 1*c*) differed little among themselves. The shaft of the femur then had a tubular appearance with thin, very dense walls and with sharply defined periosteal and medullary outlines. There was a tendency, particularly in some bones (Pratt & McCance, 1964, Pl. 2*a*), for the anterior border to become convex, instead of linear as in a bone of the same size from a normal animal. There was also some variation in the arrangement of the few persisting trabeculae in the medulla. The original hour-glass shaped regions of reticular trabeculae could usually still be made out, and these trabeculae appeared particularly dense. The peripheral part of the marrow cavity, which had been originally occupied by cortical bone, contained only a few trabeculae, which were coarse and linear. All the other long bones presented similar appearances.

Rehabilitated pigs. Three pigs which were X-rayed after being rehabilitated, one for 4 weeks (Pratt & McCance, 1964, Pl. 2*b*), and two for 5 weeks, showed that little change had occurred in the shaft by this time, though the posterior periosteal aspect had become fuzzy, and a few, new, less dense trabeculae could be seen in the marrow cavity. These passed across the cavity, buttressing the walls.

Three animals X-rayed at later stages, one at 9 and the others at 10 and 11 weeks after the commencement of rehabilitation (Pratt & McCance, 1964, Pl. 2*c*), showed that by these times a considerable increase in the width of the shaft had occurred. This had resulted in a thickening of the cortical part of the bone but with little or no enlargement of the marrow cavity. The cortex of the undernourished state persisted, being easily distinguished by its greater density, and in places was already separated from the newly formed shaft. The buttressing trabeculae in the medulla were more numerous.

One animal X-rayed after 14 weeks of rehabilitation showed no further important changes, but two other animals X-rayed after 14 and 15 weeks respectively showed an external thickening on the anterior wall of the shaft, which gave it a convex outline.

This latter feature was seen to be more pronounced when one of these animals was X-rayed 5 weeks later (Pl. 1 *b* and *d*), by which time the shaft had taken on a curvature in the anteroposterior plane with the convexity lying anteriorly. Also, when this bone was compared with a normal bone of exactly the same length (Pl. 1 *a* and *c*) it was found that the shaft of the rehabilitated bone was wider, the anterior wall was thinner (2 mm as compared with 5 mm), the extremities of the shaft were wider and the points of tendinous attachment were more prominent. The overall appearance was that of a heavier and ill-modelled bone. The femur from another animal, which was not X-rayed until it was killed after rehabilitation had been going on for 19 weeks, showed similar changes, though the curvature of the shaft was not so great.

The animal whose bone had shown no changes of curvature at 14 weeks was almost indistinguishable from its size control at 38 weeks, but the others still showed to a varying degree the deformities already described. The two animals which had had some curvature of the shaft retained this, and the walls of the shaft were very much thicker than in a 'size' control. Two other pigs, killed after 36 weeks of rehabilitation, also showed all these changes.

Whereas the changes in the femur were usual but variable in degree, those in the humerus appeared earlier and were observed in all seven pigs which had been rehabilitated for 9 weeks (Pl. 2 *a*) or longer (Pls. 2 *b* and *c* and 3 *b*). The distal portion of the humerus became curved with the convexity presenting posteriorly. This gave a knuckle-like appearance in undernourished animals (Pls. 2 *b* and 3 *b*) which was absent in the size controls (Pls. 2 *d* and 3 *a*). The shaft was wider in undernourished animals (Pl. 3 *d*) than in size controls (Pl. 3 *c*). In the early stages the cortical bone was narrower in the undernourished animals (Pl. 3 *d*) than in the size control (Pl. 3 *c*). The cortex which had been built up during undernutrition persisted in the marrow cavity for up to 19 weeks after the commencement of rehabilitation (Pl. 3 *b*) and traces were seen even later (Pl. 2 *c*), and the position of the persistent cortex showed that the growth in width had been equally great on the anterior and posterior aspects.

Histological appearances

Normal pigs. A transverse section of the shaft of a well-nourished pig 4 weeks old was found to consist of three types of bone (Fig. 1 *A* and Pl. 4 *a* and *c*), a central mass of trabeculae of endochondral bone, a zone of cancellous tissue, whose trabeculae were arranged circularly, and an outer ring of cancellous bone with radially directed trabeculae.

The central mass of endochondral bone (the 'medullary bone') occupied what was to become the marrow cavity, and the marrow tissue at that time was restricted to the spaces between the trabeculae. These contained remnants of the cartilaginous matrix which had been part of the original cartilage model. The finely fibred bone deposited on these vestiges had been laid down during foetal life.

The intermediate zone, the 'foetal circumferential zone', consisted of trabeculae whose cores were formed by basophilic loosely fibred bone. The vascular spaces within

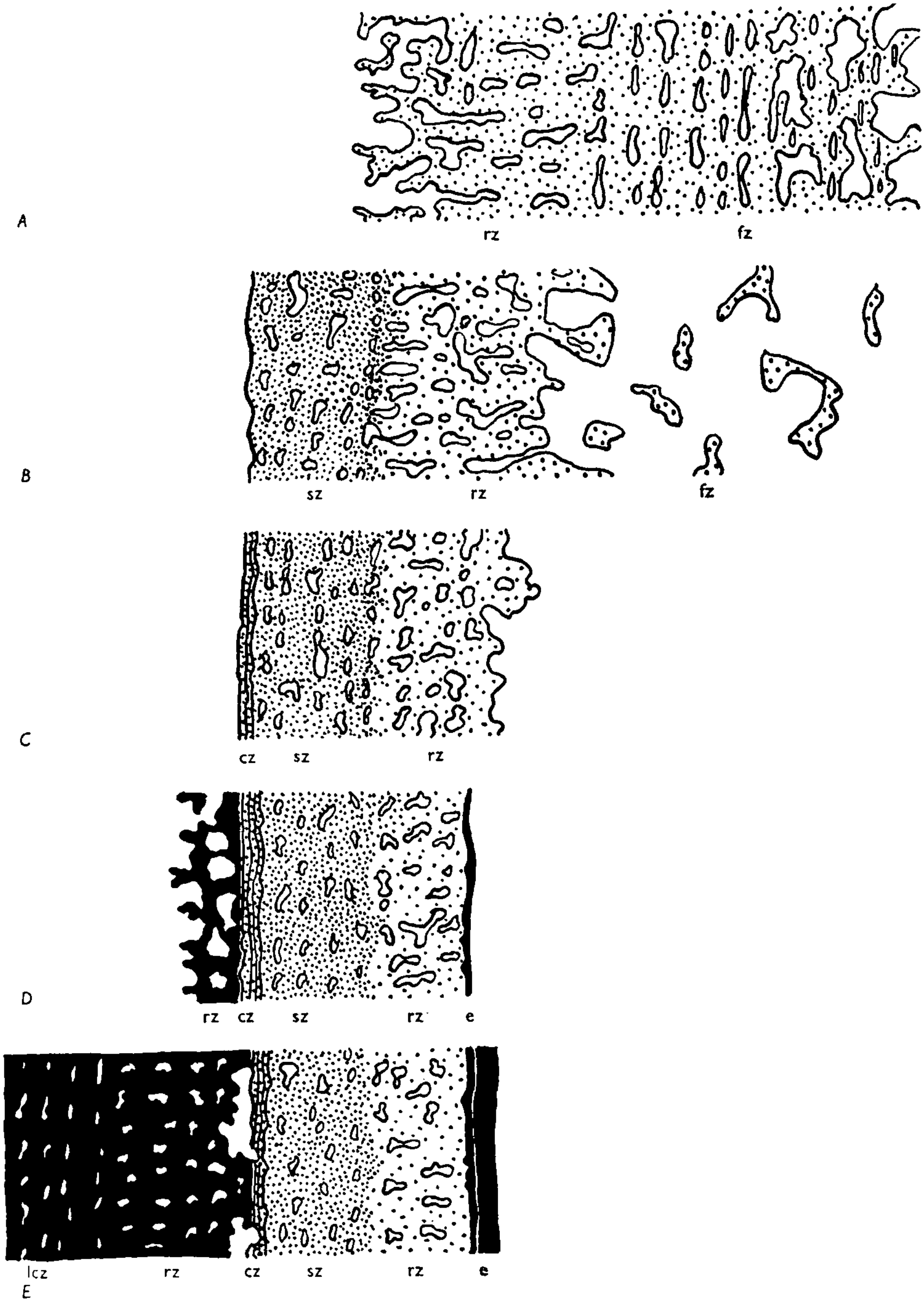


Fig. 1. For explanation see opposite page.

this zone were lined by eosinophilic densely fibred bone. These were what are usually termed the primary osteons and in this area had elongated profiles and were circumferentially arranged. This zone was formed during foetal life and was the only 'cortical' bone found in newborn pigs.

The outermost ring (the 'radial zone') had a structure similar to that found in the bone inside it except that the trabeculae were arranged predominantly in a radial manner and the primary osteons had a more circular profile. The outer aspect of this zone had an irregular appearance, for the radial trabeculae were being formed about bundles of collagen fibres passing from the fibrous periosteum into the bony matrix. This zone had been formed during postnatal life.

With subsequent growth the remains of the foetal shaft were lost as the marrow cavity appeared and enlarged. Later there was a return to the formation of circumferentially arranged trabeculae which ultimately came to constitute the entire shaft. Haversian systems, or secondary osteons with circumferential lining lamellae, were infrequent and were confined to bones from fully grown animals.

Pigs undernourished for periods up to 2 months. Three pigs came into this category and all presented a similar appearance (Fig. 1 *B* and Pl. 4*d*). The medullary bone which had been present at the beginning of the experiment was greatly reduced and only a few trabeculae remained. The foetal zone of circumferential trabeculae had disappeared and numerous osteoclasts were present on the medullary aspect of the shaft, which now consisted of the persisting radial zone together with some recently deposited periosteal bone. The latter had been laid down during the early period of undernutrition and formed about one-quarter of the total thickness of the cortex. It was easily distinguished from the radial zone inside it by the abrupt change to its irregular circumferential pattern of trabeculae with vascular channels smaller than those in the adjacent radial zone. This newly deposited bone, which will be referred to as the 'semi-compact bone', was also distinguished by a reduction in the amount of the 'loosely fibred' bone within it (Pl. 5*a*) and its increased basophilia. Osteoblasts

Legend to Fig. 1.

Fig. 1. Changes, shown semi-diagrammatically, that occurred in the cortical bone of the pig femur during undernutrition and on rehabilitation. Sparse stipple indicates bone formed before the commencement of undernutrition. Dense stipple indicates the bone formed during the period of undernutrition. Black indicates bone formed during rehabilitation. The periosteal surface lies to the left. cz, compact zone; e, endosteal bone; fz, foetal circumferential zone; lcz, late circumferential zone; rz, radial zone; sz, semi-compact zone.

A. Normal bone at 3–4 weeks of age. Some of endosteal aspect is not included. Note the commencement of resorption of the bone of the foetal shaft.

B. Early effects after 6 weeks of undernutrition. Note the loss of the bone of the foetal shaft, and the deposition of recently formed bone forming the semi-compact zone.

C. Late effects after 1 year of undernutrition. Note that about one-half of the thickness has been formed during the period of undernutrition. The compact zone is now present and contains cement lines.

D. Early changes after 4 weeks of rehabilitation. New bone has been deposited periosteally and endosteally.

E. Changes after 10 weeks of rehabilitation. Further periosteal deposition (not all is shown) and endosteal deposition has occurred. (The shaft present during the period of undernutrition is beginning to separate, following the appearance of resorption spaces.)

were found within its vascular spaces and in the subperiosteal tissues. The outer aspect was no longer ragged but had an irregular scalloped appearance.

Pigs undernourished for long periods. Fifteen pigs were undernourished for periods extending from 10 weeks up to 1 year. There were certain features common to all (Pl. 4*b* and *e* and Fig. 1 *C*) which included a narrow outer zone of 'compact bone,' the fibres of which were densely packed and arranged longitudinally (Pl. 5*b*). This compact bone contained one to eight smooth cement lines (Pl. 4*e* and Pl. 6*c*), the numbers of which appeared to vary with the duration of the undernutrition and consequently from animal to animal. The cement lines, which were not always visible on the posterior aspects of the bone, were derived from amorphous seams which had become buried by further depositions of subperiosteal compact bone. Two animals were killed while this process was taking place and active osteoblasts were present in the subperiosteal tissues. These two animals had also shown abortive processes of growth in the growth cartilage (Pratt & McCance, 1964). Examination of other bones showed that the number of subperiosteal cement lines appeared to correspond to the number of transverse plaques found in the metaphysis resulting from the abortive growth processes in the growth cartilage (Pratt & McCance, 1964).

The thickness of the cortical bone varied between 0·5 and 1·5 mm and was determined in part by the amount of intermittent periosteal growth which had taken place during undernutrition. The compact zone thus became slightly thicker with increase in age. The semi-compact bone inside it was, however, of similar thickness in all animals irrespective of the length of time that they had been undernourished, indicating that most of the periosteal activity had been confined to the early weeks of undernutrition. Both zones were wider anteriorly and this form of growth was responsible for the convex anterior border in some radiographs. This, however, is not the pattern of growth of the femur in normal pigs, in which maximum deposition occurs on the posterior aspect, as shown by the trabecular pattern in the well-nourished controls, and by the isotope studies of Comar, Lotz & Boyd (1952). The thickness of the cortical bone depended also to some extent upon the amount of medullary erosion, for some of the radial zone always persisted, though to a variable extent, as it was progressively but very slowly removed during the whole period of undernutrition. Only the anterior parts of the radial zone persisted in some animals. This erosion process was carried out by osteoclasts lying on the medullary surfaces. There was no evidence of any formation of bone on the endosteal surface of the shaft though this was a feature of normal bones.

Thirteen out of eighteen of the undernourished pigs, including the one undernourished for only 6 weeks, showed areas of increased porosity in the cortex of their femurs which may be termed 'osteoporotic' (Pl. 5*b*). Here the vascular channels were characterized by their large size owing to failure to complete the formation of primary osteons. These vascular spaces were lined with little or none of the dense parallel-fibred bone which is usually found here in both bones of undernourished animals (Pl. 5*a*) and in bones from size controls. There was no evidence of resorption being the cause of this state of affairs, for the walls were smooth, and osteoclasts were never seen there. The spaces were confined to the ring of semi-compact bone and the outermost layer of the radial zone; they did not extend throughout it and were usually confined to the posterior parts. Their location in the shaft indicates that they were a part of the growth that took place during the earlier weeks of undernutrition.

'Osteosclerotic' changes, decreasing the porosity of areas in the cortex, were also found in the bones of undernourished animals. Within the primary osteons of the semi-compact zone, and more rarely of the persisting radial zone in older animals, new bone formation reduced the diameter of the vascular channels and these were recognizable in sections stained with haematoxylin and eosin (Pl. 6*a*) by the basophilic resting cement lines which marked the limits of the tissue and also appeared within it, giving it a lamellated appearance. In silver-impregnated sections (Pl. 6*b*) the cement lines appeared as fibre-free zones, and the bone lying between the cement lines resembled that lining primary osteons, though usually the fibres were less dense. These osteosclerotic channels in no way resembled secondary osteons, which are always limited by a wavy reversal line. The osteosclerotic changes were often present side by side with the osteoporotic ones and the two tended to vary inversely. The scarcity of the former in animals undernourished for less than 4 months suggests that the osteosclerotic process was a late one. Similar osteosclerotic appearances were found occasionally in areas of old bone which had not been remodelled in well-nourished pigs.

Rehabilitated pigs. In an animal which had been rehabilitated for 4 weeks there was evidence of renewed periosteal activity (Fig. 1*D* and Pl. 6*c* and *d*) which had given rise to a fuzzy outline in X-ray photographs. Large numbers of active osteoblasts were to be seen in the subperiosteal tissues, but new bone formation was virtually restricted to the posterior aspect and sides, where radially directed trabeculae were forming around coarse collagen bundles. These bundles were anchored to the first bone formed during rehabilitation, which was a narrow zone of compact bone, found on all aspects, and separated by a cement line from the same type of bone formed during undernutrition. This latter persisted unaltered for some time and a thin layer of bone was subsequently deposited on its medullary surface, the two being separated by a resting cement line. Active osteoblasts lined this medullary surface but no osteoclasts were to be seen.

In a pig which had been rehabilitated for 11 weeks (Fig. 1*E*) the new bone which had formed during this period reached a considerable thickness over the whole external surface and, though the cortex was still narrower than in the size control, the trabecular pattern was indistinguishable, the more recent trabeculae being circumferential and the older ones radial. The vestige of the shaft which had been formed during the period of undernutrition persisted more or less intact, but it had been separated here and there, particularly posteriorly, from the newly formed shaft, by the development of large resorption spaces, lined with osteoclasts.

In two pigs which were killed after having been rehabilitated for 19 weeks, the bone formed during this period presented somewhat different appearances. In one it was indistinguishable radiologically and histologically from that of a size control, whereas in the other, which had shown considerable changes in X-ray photographs of both its femur and humerus, many of the vascular spaces were large, so giving a patchy porosity to the bone. This appearance was similar to the one described earlier in this paper in bones of undernourished animals due to a failure to fill in the primary osteons in the proper way, for there was no evidence that osteons had been widened by resorption. In some places there had been attempts to fill in these spaces, and a resting cement line separated this second deposition from the initial deposit. In both pigs the vestiges of the shaft of the undernourished state were still visible anteriorly and remnants could be seen elsewhere in the marrow cavity.

In four other pigs killed after periods of between 30 and 43 weeks of rehabilitation the cortical bone was indistinguishable from that of the bone of a normal animal of the same weight. No vestiges of the earlier shaft remained.

DISCUSSION

Undernutrition

Osteoblastic activity was not suppressed even by the severe degree of undernutrition used in these experiments, but the slow rate of bone formation altered the trabecular pattern. Internal remodelling also continued so that most of the bone laid down in foetal life was removed. Consequently the bones of these animals had thin-walled shafts, the inner half having been laid down before undernutrition began and the outer half during the period of undernutrition. The matrix of the bone formed during undernutrition was histologically distinct. Chemically, however, it was not, for Dickerson & McCance (1961) found that the collagen nitrogen:total nitrogen ratio was the same as that in healthy animals of the same weight. They also found, however, that the shaft as a whole had a high calcium:collagen ratio, and microradiographs (W. R. Lee, personal communication) indicate that the whole thickness of the shaft was involved and not merely the bone formed during the period of undernutrition.

The thin-walled bones of undernourished animals were brittle when dried, but they did not fracture during life. Investigations of the mechanical properties of these bones, moreover, showed that the breaking stress was unaltered, Young's modulus was increased and the strain at elastic limit was decreased (McCance, Dickerson, Bell & Dunbar, 1962). It is tempting to ascribe some of these changes to the increased calcification of the matrix.

The osteoporotic areas in the bones of undernourished animals are difficult to understand. Similar areas have been reported in the bones of growing animals which have had the limb denervated (Allison & Brooks, 1921). The undernourished animals had adrenal glands enlarged up to three or four times the size of those in their weight controls (McCance, 1960). The failure of bone matrix formation in the osteoporosis of Cushing's syndrome (Sisson, 1956) and the rarefaction found experimentally to follow prolonged cortisone administration (Storey, 1958) have been attributed to the anti-anabolic activity of the increased amounts of glucocorticoids, but this explanation, if the correct one, cannot account for the osteosclerotic regions.

The intermittent periosteal activity in the severely undernourished animals recalls that shown by the growth cartilage. The periosteal activity, which was often confined to one aspect, may have followed the latter, which would result in changes in tension on the fibrous periosteum. It is unlikely that they both were responses to common stimuli and took place simultaneously as there was no evidence of a generalized osteoblastic reactivity.

Rehabilitation

Periosteal growth did not become active until rehabilitation had been in progress for several weeks and in this way resembled the changes in the growth cartilage which were described by Pratt & McCance (1964). The former did not seem to depend upon the reappearance of osteoblasts, for these were present in large numbers in the marrow cavity and collected on aspects of the bone which as yet showed no signs of new bone formation. Subperiosteal deposition of bone may not have become active until the recovery of the muscles. This would result in an increase in the tensile forces which are known to initiate osteogenesis in vitro (Glücksmann, 1942).

Between the 9th and the 19th weeks of rehabilitation, however, there is evidence that the weight of the animal outstripped the mechanical reserves of the femur and humerus. It is interesting to note that McCance *et al.* (1962) found that the breaking stress of the humerus of rehabilitated animals, whose average age was 26 weeks, was less than that found in size controls. The bone removed by the excessive internal remodelling during undernutrition was not replaced on rehabilitation, though transverse trabeculae were formed in the marrow cavity at this time. Hence during the 2nd month of rehabilitation the overall diameter of the bones was the same as in those of the size controls but the walls of the shaft were thinner. It would seem that in an attempt to attain greater mechanical stability the shaft grew as rapidly as it could, which involved growth all round the circumference which was similar in pattern to that observed during undernutrition, but both differed from the normal pattern in which growth is eccentric (Payton, 1932; Comar *et al.* 1952). This atypical growth led to a curvature of the shaft of the femur and a knuckle-like appearance in the humerus which persisted once it had formed. The muscular activity at this time also led to the formation of prominent points of attachment which usually only appear when growth has ceased. Investigations are continuing to determine if these deformities persist indefinitely.

General considerations

No records have been found in the literature describing detailed studies of the effect of undernutrition and rehabilitation on bone structure and it appears that none were made before this series of observations on pigs and cockerels began (Pratt & McCance, 1960, 1961, 1964). The interpretation of the findings is difficult and not likely to be complete until more is known about the fundamental effects on cellular metabolism of undernutrition and age, and of full feeding after undernutrition. Much undoubtedly depends upon the relative growth priorities of muscle, bone and fat during these periods of limited and unlimited feeding. The observations made previously indicate that basically the reactions of both cockerels and pigs are the same, but that the detailed effects of undernutrition and particularly of rehabilitation differ considerably because the response observed depends upon the stresses, which are set up locally, and these are not always the same. This generalization applies within each species also, for there was a considerable measure of difference between the response of the femur and humerus to undernutrition and to rehabilitation in some animals. This being so, it is probable that no two species will react in quite the same way to this kind of undernutrition. Rats should not be expected to give the same results, and above all, these findings should not be taken as more than suggestive evidence about the effects of prolonged periods of undernutrition in growing children.

SUMMARY

1. Twenty-six pigs were undernourished from early in suckling so that they weighed from 3 to 8 kg when they were a year old. Normal pigs at this age weigh about 170 kg. Eight were then rehabilitated and killed at intervals up to 43 weeks.

2. The whole appendicular skeleton was X-rayed and the femurs were sectioned transversely and longitudinally.

3. By the 8th week of undernutrition the width of the shaft had been increased by the deposition of a layer of semi-compact bone which was structurally distinct from the bone previously formed.

4. After 8 weeks any further increase in the width of the shaft resulted from the deposition of compact bone. The osteoblastic activity, as in the growth cartilage, was intermittent and gave rise to circular strata separated by cement lines.

5. Many of the undernourished animals had 'osteoporotic' areas in the cortex of the femur which have been attributed to a failure in the formation of primary osteons.

6. All of the undernourished animals had areas of 'osteosclerosis' resulting from constructive filling in of the primary osteons. This process increased with the period of undernutrition and varied inversely in amount with the 'osteoporotic' change.

7. Osteogenesis was active by the 4th week of rehabilitation but only a small amount of bone had been deposited periosteally or endosteally by that time.

8. By the 9th week of rehabilitation the humerus and usually the femur differed radiologically from the corresponding bones of size controls by having wider shafts, increased curvature and thinner cortices.

9. The shaft of the bone formed during undernutrition persisted for some time as a vestige in the marrow cavity.

The authors acknowledge the helpful criticism of Professor J. D. Boyd in whose department the radiological and histological investigations took place. They are also indebted to Terry Cowen for his care of the animals and also to J. A. F. Fozzard and Miss J. Minns for radiological assistance, to J. F. Crane and G. A. H. Oakes for photographic assistance and to R. Smith and Miss G. Easy for technical assistance. The authors are grateful to L. W. Hall of the Veterinary School who anaesthetized the animals for radiology, and to Dr E. M. Widdowson for wise advice.

REFERENCES

Allison, N. & Brooks, B. (1921). *Surg. Gynec. Obstet.* **33**, 250.

Comar, C. L., Lotz, W. E. & Boyd, G. A. (1952). *Amer. J. Anat.* **90**, 113.

Dickerson, J. W. T. & McCance, R. A. (1961). *Brit. J. Nutr.* **15**, 567.

Glücksmann, A. (1942). *J. Anat., Lond.*, **76**, 231.

Jackson, C. M. (1925). *The Effects of Inanition and Malnutrition upon Growth and Structure.* London: J. A. Churchill Ltd.

McCance, R. A. (1960). *Brit. J. Nutr.* **14**, 59.

McCance, R. A., Dickerson, J. W. T., Bell, G. H. & Dunbar, O. (1962). *Brit. J. Nutr.* **16**, 1.

Mount, L. E., Lister, D. & McCance, R. A. (1963). *Brit. J. Nutr.* **17**, 407.

Nicolaeff, L. (1923). *Pr. méd.* **2**, 1007.

Payton, C. G. (1932). *J. Anat., Lond.*, **66**, 414.

Pratt, C. W. M. & McCance, R. A. (1960). *Brit. J. Nutr.* **14**, 75.

Pratt, C. W. M. & McCance, R. A. (1961). *Brit. J. Nutr.* **15**, 121.

Pratt, C. W. M. & McCance, R. A. (1964). *Brit. J. Nutr.* **18**, 393.

Sisson, H. A. (1956). *J. Bone Jt Surg.* **38** B, 418.

Storey, E. (1958). *J. Bone Jt Surg.* **40** B, 558.

EXPLANATION OF PLATES

PLATE 1

(*a*) Lateral radiograph of the femur of a normal pig aged 14 weeks (weight 36·8 kg).

(*b*) Lateral radiograph of the femur of a pig (weight 30·4 kg) which had been undernourished for 37 weeks and then rehabilitated for 19 weeks. Compare with (*a*) which is of similar length but the shaft is narrower. Note that in the former bone the cortical bone is thinner than in (*a*) and the shaft has developed a curvature.

(*c*) Anterior radiograph of bone shown in (*a*).

(*d*) Anterior radiograph of bone shown in (*b*). Compare with (*c*). Note that it has a wider shaft with very much more pronounced prominences for muscle attachments.

PLATE 2

(*a*) Lateral radiograph of the humerus of a living pig (weight 25·9 kg) which had been previously undernourished for 57 weeks and then rehabilitated for 9 weeks. Note the vestige of heavily calcified tissue which formed the shaft of the undernourished state and the less dense new bone formed subperiosteally in the distal part of the shaft.

(*b*) Lateral radiograph of the same bone as in (*a*) but after 15 weeks of rehabilitation (weight 63·6 kg). The vestige is becoming separated from the walls of the shaft.

(*c*) Lateral radiograph of the same bone as in (*a*) and (*b*) but after 43 weeks of rehabilitation (weight 208 kg). Traces of vestige are still present (arrowed).

(*d*) Lateral radiograph of the humerus of a normal pig aged 31 weeks (weight 150 kg). Compare with (*c*) which is of similar length but has a wider shaft and a curvature of the distal portion of the shaft which is not present in this bone.

PLATE 3

(*a*) Lateral radiograph of the humerus from the same normal pig as in Pl. 1 (*a*).

(*b*) Lateral radiograph of the humerus from the same undernourished pig as in Pl. 1 (*b*). Compare with (*a*). Note that the shaft is wider, there is a curvature in the distal part, and the vestige (arrowed) persists in the marrow cavity.

(*c*) Anterior radiograph of the same bone as (*a*).

(*d*) Anterior radiograph of the same bone as (*b*). Compare with (*c*). Note the wider shaft.

PLATE 4

Photomicrographs of transverse sections through the mid-shaft of the femur of normal and undernourished pigs. A, anterior aspect; c, compact zone; e, endochondral bone; f, foetal circumferential zone; l, cement line; m, marrow cavity; o, lacuna for osteocyte; P, posterior aspect; p, periosteum; r, radial zone; s, semi-compact zone; v, vascular space.

(*a*) Control, aged 4 weeks (weight 6·83 kg). Note the endochondral bone in the marrow cavity. Long's method.

(*b*) Undernourished, aged 39 weeks (weight 5·0 kg). Compare with (*a*), the length control, note the thinner cortex. Long's method.

(*c*) Control, same bone as (*a*) showing the zones of cortical bone. Haematoxylin and eosin.

(*d*) Undernourished, aged 8 weeks (weight 3·7 kg). Showing changes which have occurred during the early weeks of undernutrition. Note the recently formed zone of semi-compact bone. Haematoxylin and eosin.

(*e*) Undernourished, aged 28 weeks (weight 5·2 kg). Note the more compact subperiosteal bone and the cement line. Haematoxylin and eosin.

PLATE 5

Photomicrographs of transverse sections through the mid-shaft of the femur of an undernourished pig aged 15 weeks (weight 4·03 kg). Key as in Pl. 4.

(*a*) Shows the usual structure; note the arrangement of fibres in the bone. Long's method.

(*b*) Shows an 'osteoporotic' area with large vascular spaces substituting for the semi-compact zone. Compare with (*a*). Long's method.

PLATE 6

Photomicrographs of transverse sections through the mid-shaft of the femur of undernourished and rehabilitated pigs. Key as in Pl. 4.

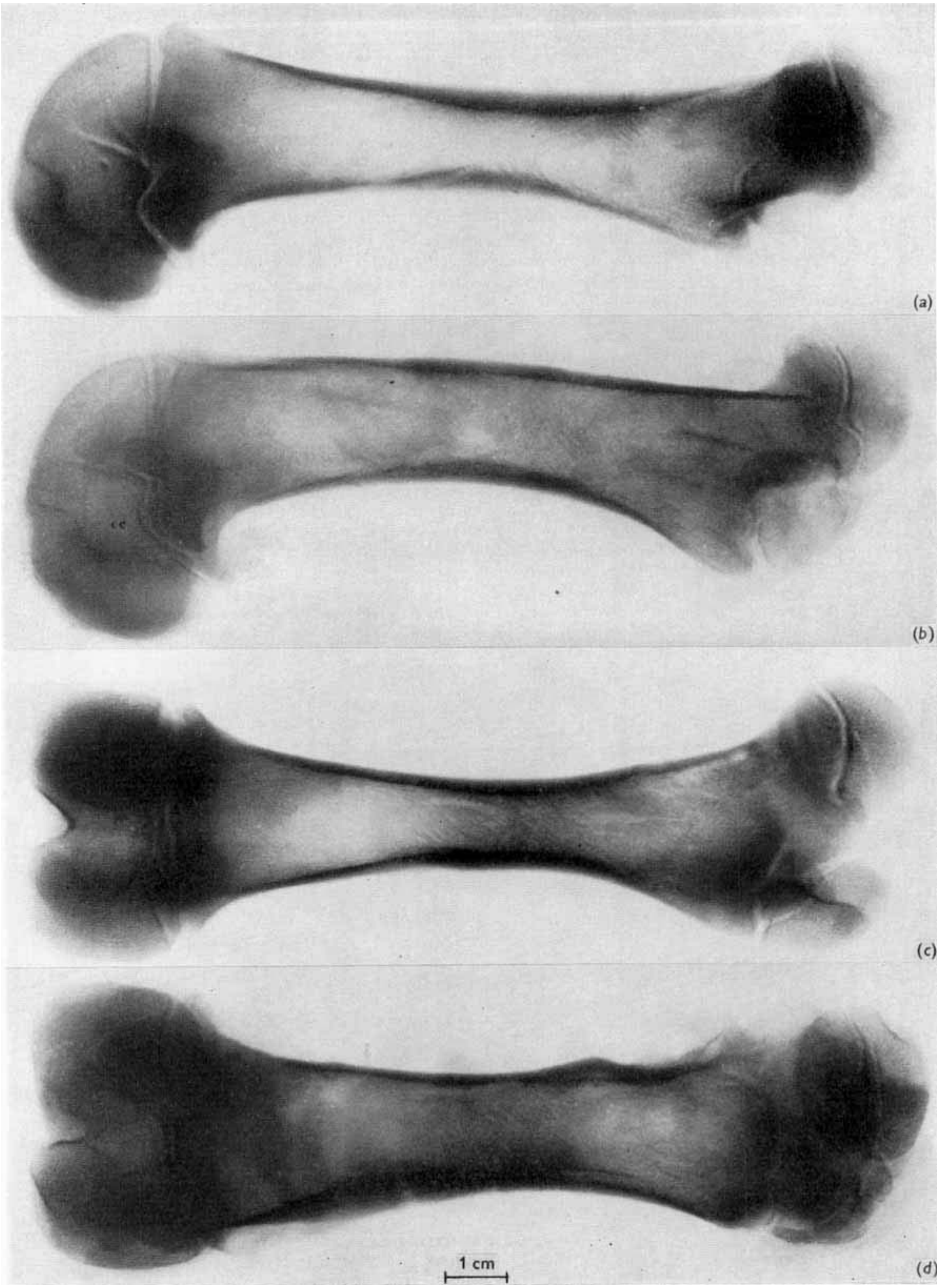
(*a*) Undernourished, aged 39 weeks (weight 5·0 kg), showing 'osteosclerotic changes'. Haematoxylin and eosin.

(*b*) Same bone as (*a*) showing the fibrous structure of the bone deposited within the vascular spaces. Note the fibre-free cement lines which indicate the original boundary of the vascular space. Long's method.

(*c*) Undernourished 51 weeks and then rehabilitated for 4 weeks (weight 7·0 kg). Showing the recovery of periosteal bone formation on the anterior aspect. Large numbers of darkly staining osteoblasts can be seen surrounding the newly formed trabeculae. Six cement lines are present in the compact zone on this surface. Haematoxylin and eosin.

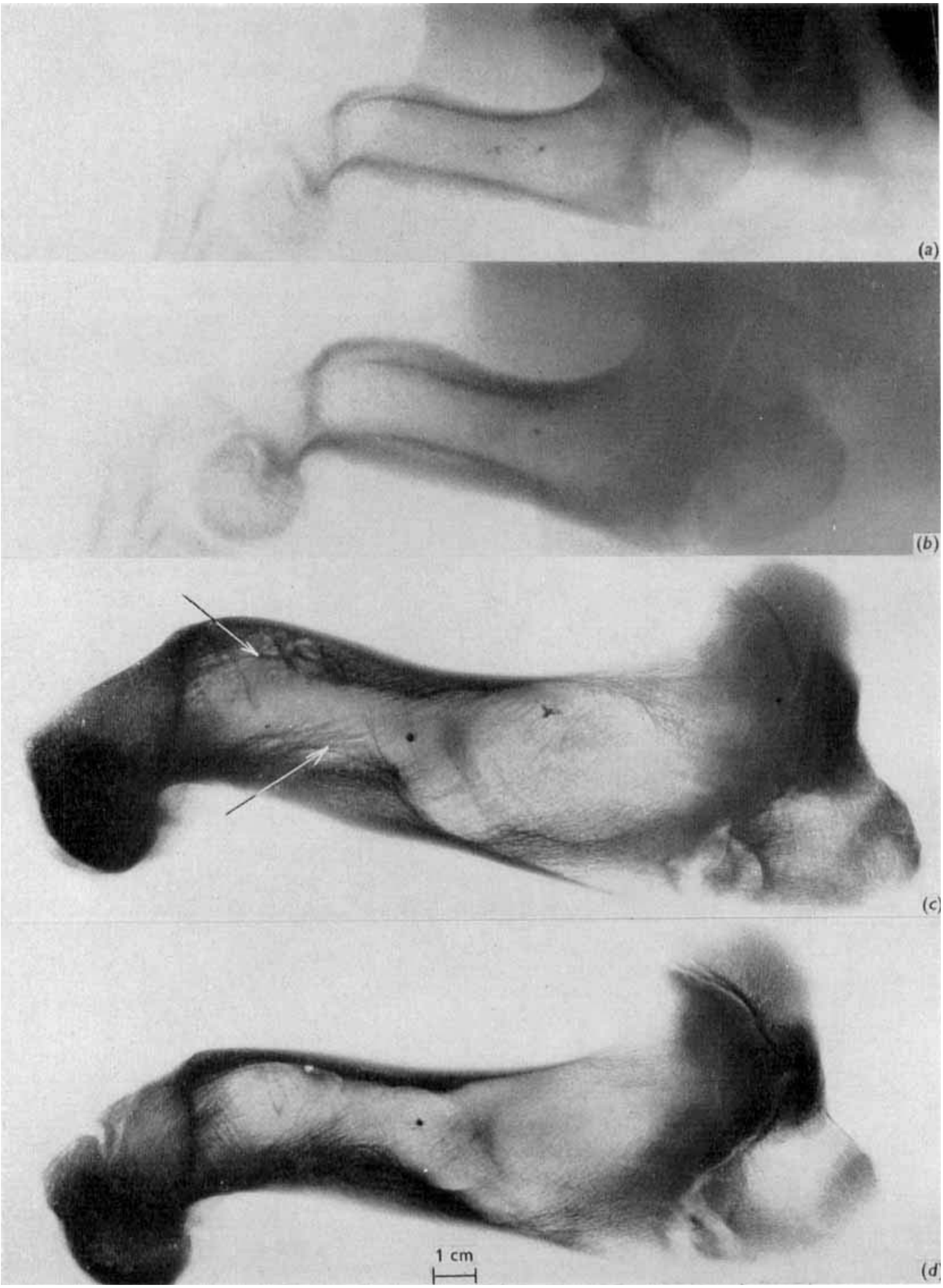
(*d*) Same bone as (*c*) but posterior aspect where the periosteal deposition is greater. Note the cement line separating the newly formed bone from the shaft of the undernourished state. There are no cement lines in this latter bone. There has been a slight deposition of bone on the endosteal surface (arrowed). Haematoxylin and eosin.

Printed in Great Britain

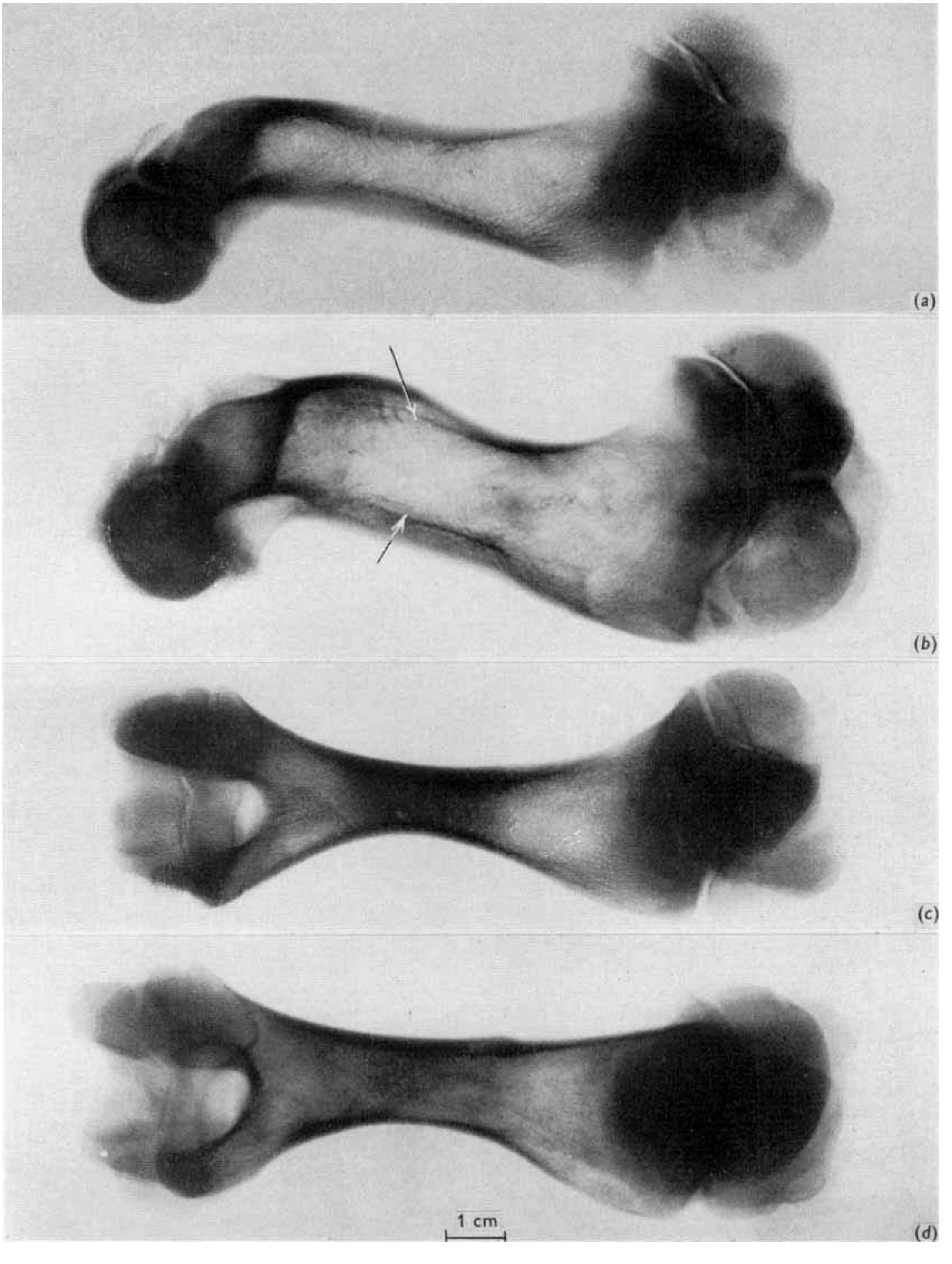


C. W. M. PRATT AND R. A. McCANCE

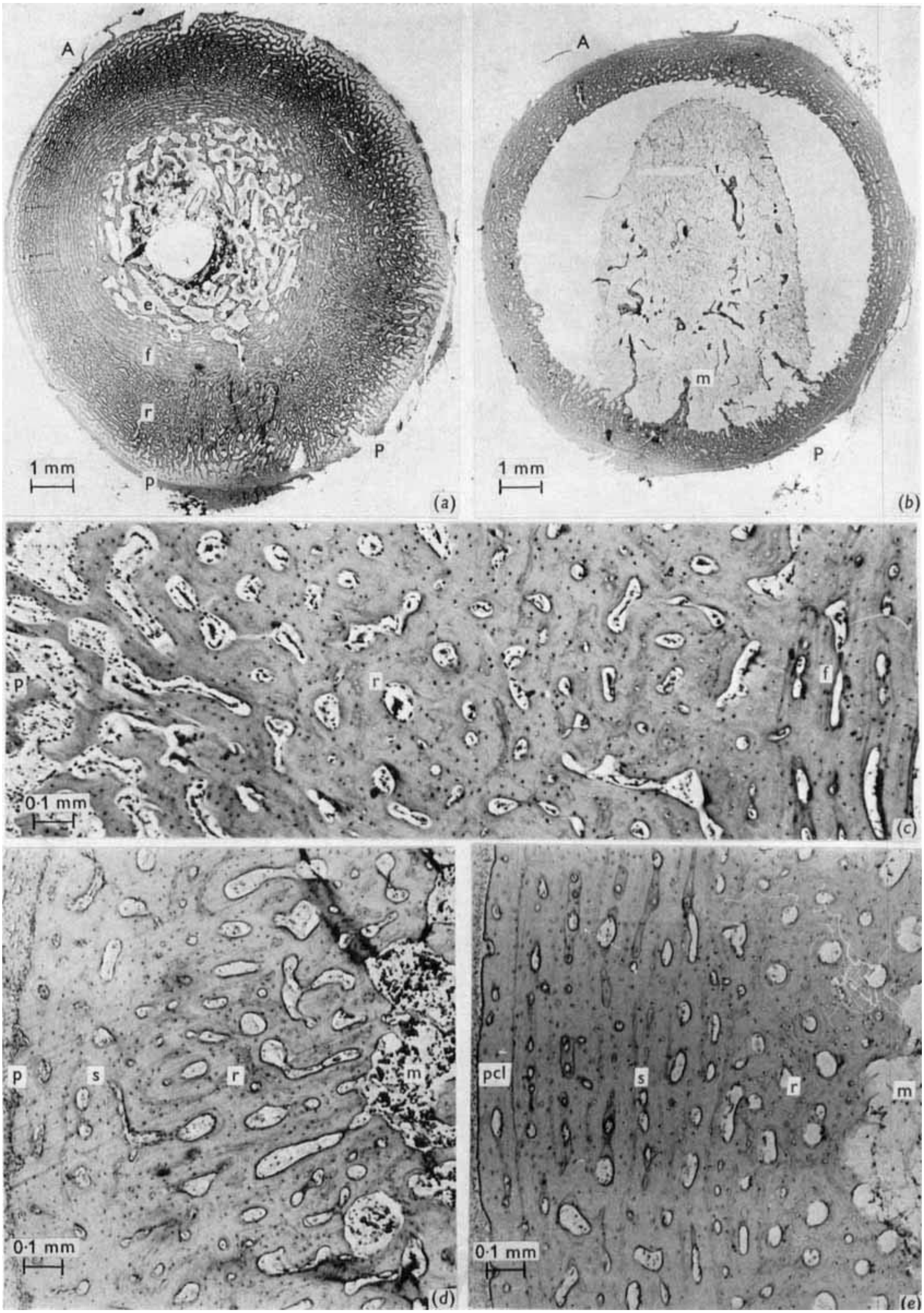
(*Facing p.* 624)



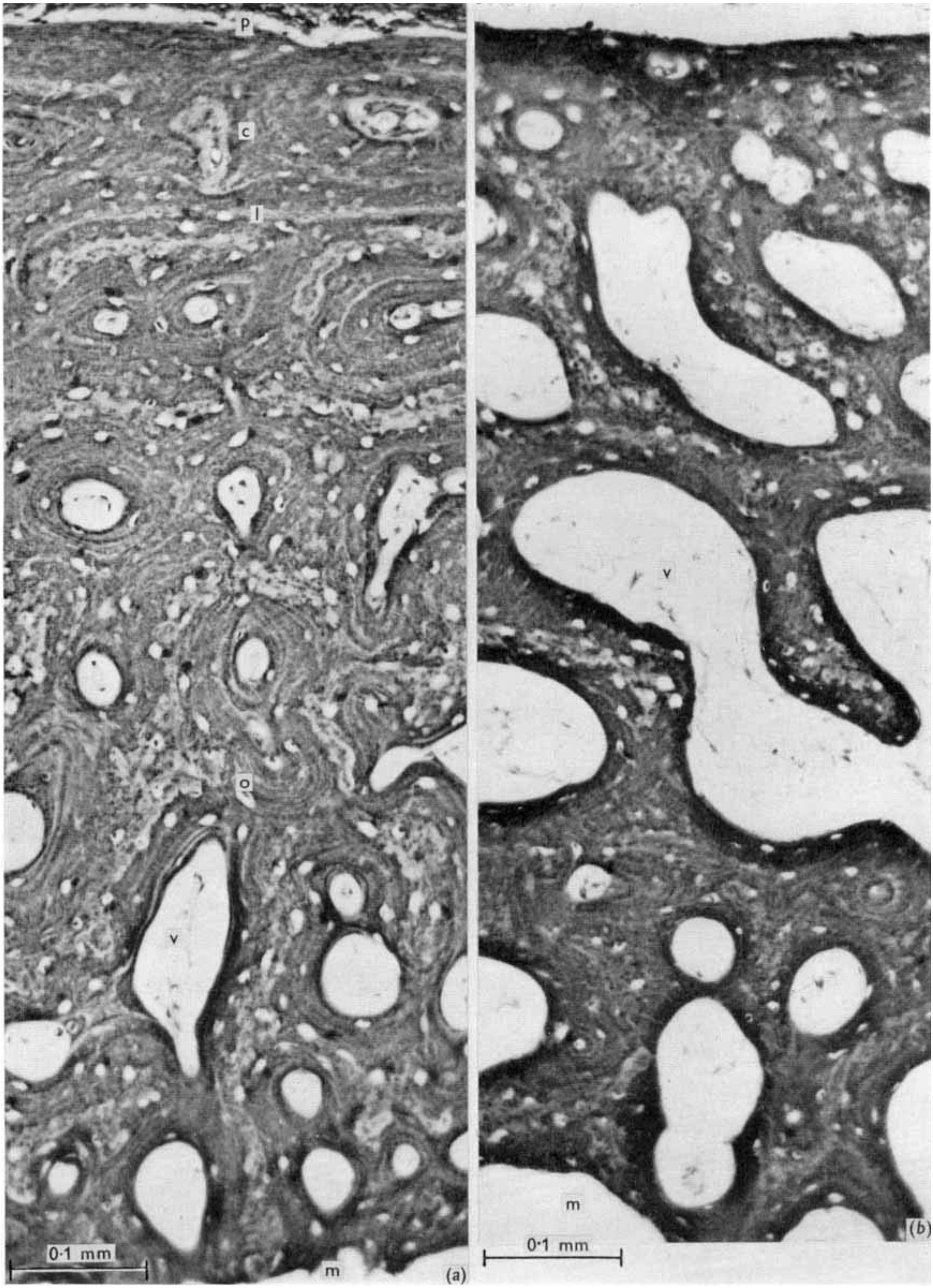
C. W. M. PRATT AND R. A. McCANCE



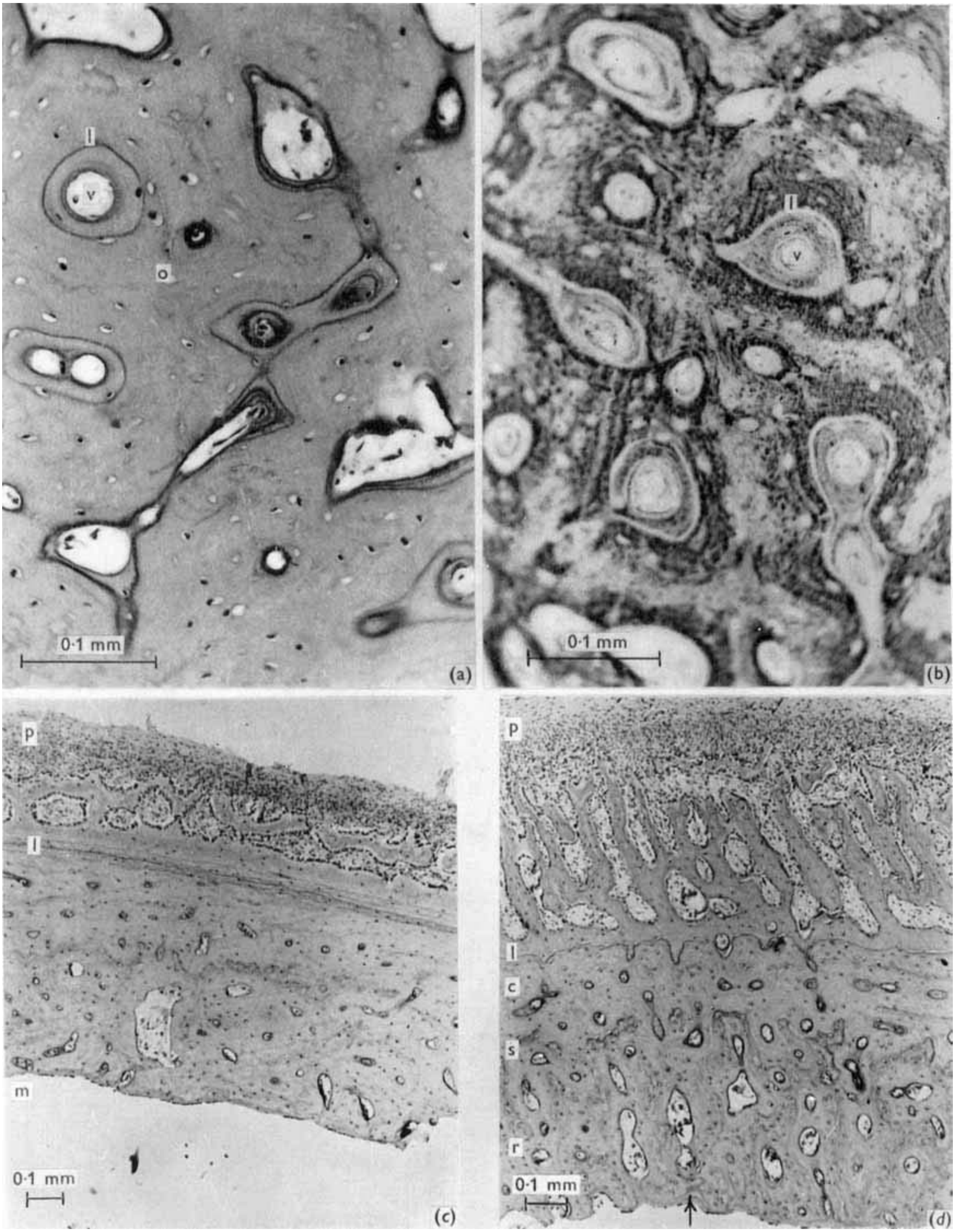
C. W. M. PRATT AND R. A. McCANCE



C. W. M. PRATT AND R. A. McCANCE



C. W. M. PRATT AND R. A. McCANCE



C. W. M. PRATT AND R. A. McCANCE