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## INTRODUCTION

Osteoporosis is the most common degenerative condition affecting the ageing lumbar spine. It involves a loss of bone substance and it may occur as a consequence of disease (Dent & Watson, 1966), <sup>a</sup> marked decrease in weight-bearing (Hall, 1976), or it may simply be considered as an inevitable accompaniment of ageing (Gitman & Kamholtz, 1965). It is more prevalent in females after the menopause than in males, and in white females than in black females (Boukhris & Becker, 1974). At age 65, X-ray comparison with a 'standard' suggests that 65.8% of females and 21.5% of males have osteoporosis. In women, the incidence increases by about  $8\%$  for each additional decade, whereas, in men, a large increase does not occur until after age 76 (Gitman & Kamholtz, 1965).

During childhood and early adult life, the 'cortical shell' of a lumbar vertebral body is thin and dense, while its cartilage-covered vertebral end plate surfaces contain multiple perforations (Warwick & Williams, 1973). Cancellous bone comprises about 66  $\%$  of all vertebral bone, and in the vertebral bodies it is arranged in irregular plates or trabeculae  $0.12{\text -}0.24$  mm thick. The trabeculae are oriented parallel to the lines of stress with 'vertical' and 'horizontal' components. In the anterior two thirds of the vertebral body, vertical trabeculae predominate, whereas, in the posterior one third, horizontal trabeculae are more evident, as in the pedicles (Singh, 1978).

According to Atkinson (1967, 1972), osteoporosis in the cancellous bone of vertebral bodies involves not only a general loss of bone substance, but there is a proportionately greater loss of the horizontal trabecular components, which act as supporting cross-braces. Both these changes would cause vertebral bodies to lose a considerable part of their ability to bear weight. Thus, normal weight transmission through the nucleus pulposus of the intervertebral disc causes micro-fractures and micro-calluses to appear in the subchondral bone, with an increase in the concavity of the disc-vertebral junction (Schmorl & Junghanns, 1971; Boukhris & Becker, 1974; Vernon-Roberts & Pirie, 1973; Hansson, 1977; Frymoyer & Pope, 1978). This phenomenon of inward bowing of the lumbar end plates is characteristic of the osteoporotic process. It has been measured, using a vertebral biconcavity index of mid-line vertical height divided by anterior vertical height, as in Figure <sup>1</sup> (Barnett & Nordin, 1959, Hurxthal, 1968; Arnold et al. 1970; Ericksen, 1974).

Osteoporosis is linked with a loss of stature due to shortening of the trunk (Dent, 1955), causing the crown-pubis distance (similar to sitting height) to become less than the pubis-heel distance (Dent & Watson, 1966). This decrease in trunk length



Fig. 1. Biconcavity index is calculated by dividing the central vertical vertebral body height by the anterior vertical vertebral body height.  $I = B/A$ .

may be due in part to the loss of bone substance and the change in vertebral body shape which may, in extreme cases, proceed to vertebral body wedging and collapse (Shapiro, 1975).

The purpose of the present study is to quantify changes in bone density and trabecular architecture which occur in the lumbar vertebrae of a general population with increasing age, and to relate these changes to age changes in vertebral body shape.

#### MATERIALS AND METHODS

The lumbar vertebral column was removed at post mortem from 93 adult subjects and stored in 10  $\%$  formaldehyde solution. Following careful mid-line section of the column, vertical measurements of anterior vertebral and mid-vertebral heights were recorded for all vertebrae to establish biconcavity indices for the whole lumbar spine (Fig. 1, Table 4). The third lumbar vertebra was then removed from each column, for measurements of bone density using sagittal sections. This vertebra was selected because of its symmetrical shape and because of its central position in the lumbar column. Following unsuccessful attempts to produce regular, uniformly thick sections, using a double-bladed hacksaw (Atkinson, 1967), a successful method of sectioning was developed as follows:

#### (1) Sectioning procedure

Two <sup>2</sup> mm sagittal slices of bone were sectioned from deep-frozen vertebrae, using a butcher's bandsaw (four teeth per cm of blade) suitably adapted by the incorporation of a specially designed guide to ensure both a clean vertical cut and a section of regular <sup>2</sup> mm thickness. The uniform thickness of each section was ascertained by nine separate measurements, centrally and around the perimenter, using dial vernier calipers (Fig. 2). Average thickness was  $2.04 \pm 0.17$  mm.

The first section was immediately adjacent to the mid-line, while the second section was 25 % of the distance from the mid-line to the lateral border of the vertebral body (this section is usually just medial to the junction of pedicle and vertebral body). The sections were immersed in  $5\%$  sodium hydroxide solution for 2 hours and washed with running water until cleared of all soft tissue.



Fig. 2. A <sup>2</sup> mm bone section from the L3 vertebral body, mounted in its prepared frame. The numbers indicate the positions where thickness measurements were made for each specimen.

## (2) Measurements of bone density

# (a) Dry weight per unit volume

In order to make comparable measurements of bone density by means of weighing, sections were allowed to dry at room temperature to a stable state. Following exposure in the air-conditioned laboratory atmosphere for 24 hours, no further weight loss was recorded by serial weighings at two hourly intervals. Sections were weighed, using a Fisher model 200 balance which is accurate to two decimal places of grams. Because sections were of equal and uniform thickness, an index of weight per unit volume was obtained by dividing the weight by the area of the section.

#### (b) Light meter measurements

Each dried section was mounted in <sup>a</sup> <sup>2</sup> mm thick opaque cardboard frame, using a plastic bonding material to fill in all spaces between the perimeter of the specimen and the frame, in preparation for light transmission measurement.

A measure of bone 'translucency' (the inverse of bone density) was obtained by fitting each specimen, in its frame, into a device which measured the amount of light passing through the specimen. The device, which was fully sealed, consisted of an even, standardised light source above the specimen and a large photo-electric cell (75 mm diameter, 0-42 V, <sup>1200</sup> mA) below the specimen and connected to <sup>a</sup> light meter which registered the amount of transmitted light. An index of light transmission per unit area of section was calculated.

The indices of weight and of transmitted light per unit area were grouped into age and gender categories for comparison of males and females in three adult age categories.

#### (c) Image analyser measurements

Measurements of bone density were made in two vertical strips of each <sup>2</sup> mm mid-line section of L3 from randomly selected examples of young and old adults: six young males (average age 27 years), six young females (average age 22 years) and six elderly males (average age 64 years) and six elderly females (average age 73 years). A centre line and <sup>a</sup> base line were traced as shown in Figure 3, and the two vertical strips were defined, each containing  $10\%$  of the anteroposterior extent of the section and located as shown in Figure 3.



Fig. 3. The two strips selected for image analyser measurements of bone area. (Each shaded strip is  $10\%$  of A.)



Fig. 4. The position of the marker lines over the L3 vertebral body, used to count the number of horizontal and vertical trabeculae.

The total area of each strip was measured by tracing around its perimeter and the area of the marrow spaces was established by tracing around each space contained within each strip. The difference between the total area of the strip and that of the spaces within it represents the area occupied by the bony trabeculae. An index was established and used as a measure of bone density.

Bone density index  $=$   $\frac{\text{total strip area} - \text{total clear space area}}{\text{total strip area}}$ 

# (3) Measurements of trabecular pattern

The trabecular pattern of each specimen (in the horizontal and vertical planes) was assessed by counting the bony intersections on three lines drawn across each specimen (Fig. 4). The numbers of trabeculae intersected by each line, and the length of each line, were measured. Results were expressed as the number of trabeculae per unit length of the line.

Age and gender category	Bone slice	Measures of bone density			
		weight Index of area*		light transmission Index of area* $=$ 'bone translucency index'	
		M	F	м	F
$20-35$ years (14 M, 15 F)	A в	$8.51 \pm 1.26$ $9.15 \pm 1.16$	$6.37 + 1.16$ $7.77 + 1.20$	$1.24 + 0.16$ $1.14 \pm 0.09$	$1.36 + 0.24$ $1.27 + 0.18$
$36 - 59$ years (15 M, 15 F)	A в	$6.91 + 1.69$ $7.75 + 1.45$	$6.50 + 0.98$ $7.41 + 1.30$	$1.41 + 0.16$ $1.35 + 0.16$	$1.52 + 0.16$ $1.39 \pm 0.17$
$60 + \text{years}$ (19 M, 15 F)	A в	$5.85 + 1.45$ $6.86 + 1.82$	$5.76 \pm 0.97$ $6.54 + 1.35$	$1.68 + 0.22$ $1.53 \pm 0.21$	$1.65 + 0.47$ $1.57 + 0.48$
thick.		Area represents volume in these indices of 'bone density', because all sections are uniformly 2 mm			

Table 1. Two indices of bone density:  $\frac{\log n}{\alpha r}$  and  $\frac{n \log n}{\alpha r}$  and  $\frac{n \log n}{\alpha r}$ 

M, males; F, females; A, central slice of bone; B, lateral slice of bone.

# Table 2. Index of  $\frac{area\ of\ bone\ tissue}{area\ of\ bone\ sections}$  for central and posterior bone sections, in males and females, for young and elderly adults



\* Area represents volume in these indices of 'bone density' because all sections are uniformly <sup>2</sup> mm thick.

M, males; F, females.

# (4) Concavity indices

Using the data of vertebral heights measured on all lumbar vertebrae in mid-line sections (Fig. 1), the 'concavity' index was calculated for each vertebral body by dividing the mid-vertebral height by the anterior vertebral height (Barnett & Nordin, 1959). Data were listed for males and females, separately, in three adult age groups (Table 4).

# RESULTS

## Bone density

Assessments of bone loss with increasing age were recorded in three indices of bone density. A clear trend of decline in the index of weight per unit volume, and <sup>a</sup> substantial increase in the bone translucency index with increase in age were found (Table 1). The age change in weight per unit volume in males and the increase in

# Table 3. The numbers of vertical and horizontal trabeculae per unit length for young, middle aged and elderly adult subjects.

(The horizontal trabeculae were counted in a mid-vertebral line only, while the vertical trabeculae were counted in a mid-vertebral line and just beneath the superior vertebral margin.)



light transmission were significant in both sexes ( $P < 0.0005$ ). The two indices correlated closely for both sexes ( $r = 0.97$  in males;  $r = 0.87$  in females).

The third index of bone density recorded image analyser measurements from young and old adult groups of both sexes. This index also showed a significant decline with increase in age ( $P < 0.0125$ ) for both areas measured in both sexes (Table 2).

All three indices confirmed the trend of loss in bone density with increase in age in both sexes.

## Trabecular pattern

Changes in the numbers of vertical and horizontal trabeculae in mid-line bone sections were recorded (Table 3; Figs. 5, 6), comparing young, middle aged and old adult groups of both sexes. Although a small decline in numbers of vertical trabeculae in old age was observed in both sexes, this trend was not statistically significant. On the other hand, there was a statistically significant decline in the numbers of horizontal trabeculae in old age in both sexes ( $P < 0.01$  in males;  $P < 0.0025$  in females).

# Concavity index

Age change in the concavity of the vertebral end plates for all lumbar vertebrae in both sexes was recorded (Table 4). Changes in the concavity index reflected a general trend to increased concavity in old age in both sexes. Considering the data for all lumbar vertebrae, the trend was significant ( $P < 0.01$  in males;  $P < 0.05$  in females). There was no significant change from LI through to L5 in terms of the concavity index. All lumbar vertebrae appeared to be similarly affected with increasing age.

In summary, the measures of bone density and architecture demonstrated a decrease in the amount of bone substance and a reduction in the numbers of horizontal trabeculae. These changes correlated well with the changes in the concavity



Fig. 5. The numbers of vertical trabeculae per unit line length, in males and females, in the mld-vertebral line and at the superior margin of the L3 vertebral body, as measured in the central bone slice. The data for the lateral slice parallel those for the mid-line slice. Standard deviations (see Table 3) are omitted from the graph for the sake of clarity. Age category: 4, young adults; 5, middle aged adults; 6, elderly adults.

# Table 4. Indices of mid-vertebral body height divided by anterior vertebral body height, for age and sex.

(The decline in index between young adults and old adults is significant, at the  $P < 0.01$  and  $P < 0.05$  levels for males and females, respectively.)







Fig. 6. The numbers of horizontal trabeculae per unit line length, in males and females, measured in the mid-vertebral line of the L3 vertebral body. The decrease is statistically significant in both sexes (see text). Standard deviations (see Table 3) are omitted from the graph for the sake of clarity. Age category: 4, young adults; 5, middle aged adults; 6, elderly adults.



Fig. 7. The manner in which vertical trabeculae may buckle under load if no transverse ties are  $\frac{1}{4}$ present (as on left) where effective length =  $l$ . If ties are present (right), effective length = and buckling load must be four times greater (Bell, Dunbar & Beck, 1967).

index ( $r = 0.99$  and  $r = 0.75$  for males and females, respectively, correlating the concavity index with the index of weight per unit volume).

#### DISCUSSION

All three measures of bone density, in the current study, indicate a loss of bone substance in old age. The index of weight per unit volume, and the index of area of bone/total area of section, which resemble traditional methods of estimating bone density (Bell, Dunbar & Beck, 1967) confirm, but are less sensitive than the bone translucency index, the bone loss which accompanies ageing. Because the sample of subjects in this study can be taken to be representative of the Western Australian community (Twomey, 1981) decrease in bone density can be regarded as a 'normal' accompaniment of ageing in the local community, but the definition of the critical level at which a decrease in bone density becomes 'osteoporosis' remains largely a clinical problem (Nordin et al. 1980).

An analysis of the mechanism by which decrease in bone density leads to changes in vertebral shape, with increased concavity and possible collapse of the vertebral end plate, has an important bearing on the problem of defining osteoporosis. The changes in trabecular structure demonstrated in this study are central to this analysis.

There is a reduction in both the number of trabeculae (particularly of horizontal trabeculae, Fig. 6) and in the thickness of the trabeculae in old age. Atkinson (1967), in a radiographic study of 68 subjects, found a considerable decrease in the number of transverse trabeculae over the age of 50 years, but considered that the vertical trabeculae increase in width. These findings accord with the subjective observations of Casuccio (1962). The present study finds a decrease in the numbers of both vertical and transverse trabeculae (particularly the latter) in old age. While measurements of trabecular thickness were not made, careful inspection could not confirm Atkinson's (1967) observation (with no supporting data) that vertical trabeculae increase in thickness in old age, in partial compensation for the loss of transverse trabeculae.

Atkinson (1967) argues that the reduction in the number of transverse trabeculae leads to fracture of supporting vertical trabeculae, resulting in end plate microfracture. The consequence of this is usually an increase in the concavity of the end plates in old age, although in extreme cases, collapse occurs.

Atkinson's interesting radiographic study (1967) of thin bone slices may be criticised in that it proved impossible in this study to obtain bone slices of consistent thickness, despite great care in using the methods described in his paper. However, the conclusions from the present study agree substantially with Atkinson (1967) as regards horizontal trabecular changes and lend further support to the view that the number and distribution of horizontal trabeculae is an important feature affecting the 'stiffness' and the strength of cancellous bone (Doyle, 1972; Pugh, Rose & Radin, 1973; Chalmers, 1973; Townsend, Rose, Radin & Raux, 1977). Euler's theory is an engineering concept used by Bell et al. (1967) and Doyle (1972) to explain the inherent strength of cancellous bone. The theory demonstrates that the rigidity of the structure is enhanced more by its geometry than its mass, and suggests that the strength of a column of bone (e.g. a vertical trabecula) is dependent upon the number of its reinforcing 'ties' (Doyle, 1972), or 'cross-braces' (Pugh et al. 1973). In the vertebrae, these are represented by the horizontal trabeculae, which are more effective in providing strength to a vertical strut that would be an increase in the diameter of the vertical strut (trabecula), as shown in Figure 7.

Young adult vertebral bodies are able to resist the vertical compressive effects of body weight because of their trabecular arrangement (Fig. 7).

As in the disc, the vertical compressive fiorces of body weight on a vertebra may be partly resolved in a horizontal direction (along the 'cross-braces'). The selective loss of horizontal trabeculae which occurs in old age might be expected to lead to collapse of some of the vertical trabeculae under the compressive load of body weight, which is usually maintained into old age, or even increased (Stenhouse, 1972). This would explain the observed loss in vertebral height and increased vertebral concavity in old age (Trotter & Gleser, 1951; Twomey, 1981). The collapse tends to be more marked below the nucleus pulposus, that part of the disc which is most efficient in transmitting loads. It is the area of bone immediately adjacent to the nucleus which shows the largest number of micro-fractures in old age (Vernon-Roberts & Pirie, 1973; Hansson, 1977).

The various observations of change in trabecular pattern, vertebral end plate concavity, and subjacent micro-fractures, strongly suggest a cause-and-effect relationship. Height loss is not so generally apparent at the vertebral rim. This, together with the increase in all horizontal vertebral dimensions in old age (Ericksen, 1974; Twomey, 1981), may be taken to suggest 'ballooning' of the vertebral body because horizontal trabecular cross-braces no longer fully support the perimeter of the vertebral body. This allows the 'waist' of the vertebra to spread. The height of the periphery of the vertebral bodies may be maintained, in part, by the traction effect of the anulus fibrosus at the vertebral rims.

This research poses the question as to why there is a selective loss of horizontal trabeculae in lumbar vertebral bodies. It is generally assumed that hormonal factors are of primary importance in osteoporosis of the elderly (Nordin et al. 1980) but mechanical factors are also known to influence bone structure, notably in young athletes (Jones et al. 1977) but also in middle aged men (Dalen & Olssen, 1974). The effect of the forces imposed on bone by exercise, causing cancellous bone to be organised parallel to the lines of stress, is well known (Dent & Watson, 1966). Exercise has also been shown to prevent osteoporosis in post-menopausal women (Aloia et al. 1978). Hormonal deficiencies alone would be expected to lead to a generalised loss of cancellous bone, and the selective loss of horizontal trabeculae suggests that mechanical factors may also be important. It may be postulated that the reduced activity of old age is responsible for the selective decline in the numbers of horizontal trabeculae, while the forces of body weight through the vertebrae are responsible for the persistence of vertical trabeculae.

#### **SUMMARY**

Old age is associated with a decline in bone density in lumbar vertebral bodies in both sexes, although the rate and amount of the decline is greatest in females. The bone translucency index method, described in this study, is a sensitive method of estimating bone density. The primary reason for this decline is the significant decrease in the number of transverse trabeculae of lumbar vertebrae in old age. It is postulated that the increase in vertebral end plate concavity and the increased horizontal dimensions of lumbar vertebral bodies in old age follows as a direct consequence of the selective loss of the transverse trabeculae.

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