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NUTRITIONAL FACTORS IN OSTEOPOROSIS

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OVERVIEW OF OSTEOPOROSIS

Osteoporosis is a multifactorial disorder in which the skeleton is sufficiently fragile so that it fractures when exposed to the mechanical forces and accidents that are a routine part of ordinary living. Nutrition is only one of several factors that influence bone strength. To understand adequately where nutrition fits in, it will be helpful, first, to provide a general description of the complex domain of osteoporotic fragility.

Fracture is almost always due to an interaction of bony fragility with injury. Fragility, in turn, is due not simply to decreased bone mass, but to accumulated fatigue damage and to critical trabecular disconnections, as well. Reduced bone mass also has many contributing causes, of which inadequate nutrition

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is one. Others include a genetically small skeletal program, gonadal hormone deficiency, physical inactivity, and many life-style, co-morbid, and pharmacologic agencies.

Nutritional factors important for bone health include calcium, phosphorus, protein, vitamins C, D, and K, and various trace minerals. Of these, calcium has been the most extensively studied. Intake of calcium may be inadequate for the obvious reason that it is low; however, even when statistically "normal," it may still be inadequate because of subnormal absorption (60) or greater than normal excretory losses.

Frailty and Injury

Almost all fractures, even those we term "low-trauma," occur as a result of some injury—the application of more force to the bone than it is able to sustain. Usually this is a result of a fall or the application of bad body mechanics. Although fracture incidence patterns differ somewhat from site to site, the risk of virtually all fractures rises with age, and all fractures contribute to the burden of illness, disability, and expense that the elderly (and society) bear. Hip fracture is perhaps the most serious of the fragility fractures, inasmuch as it carries an excess mortality, is expensive, and causes significant deterioration in quality of life for many of its survivors. It is, as well, a good example of the many interacting factors that constitute this fracture domain.

First, there is the fall itself. Normally, postural reflexes work to get the arms into position to break the force of the fall, or to swing the body so that it lands on the buttocks (or both). These reflexes almost always operate effectively in younger individuals, but commonly fail in the elderly. As a result, young people rarely strike the lateral portion of the trochanteric region of the hip when they fall, whereas the fragile elderly more commonly do so. Additionally, hip fracture is a particularly serious problem in undernourished elderly individuals who have less muscle and fat mass around the hip, and therefore less soft tissue through which the force of the impact can be distributed to a larger area of the lateral surface of the trochanter. The force of the impact, when falling from standing height, may well be sufficient to break even a healthy femur if that force is concentrated in a small enough impact area (117).

Nutrition enters into this region of the fracture domain primarily through its effect on maintenance of the soft tissue mass that serves to cushion the impact of falls. In some cases nutrition may also influence central nervous system processing time or contribute to the general feebleness that predisposes to falling. But energy dissipation by soft tissue at the point of impact is the major factor here.

Intrinsic Bony Strength and Fragility

Strength in bone, as in most engineering structures, is dependent upon its mass density, upon the arrangement of its material in space, and upon the intrinsic strength of its component material (particularly as influenced over long periods of use by the accumulation of unrepaired fatigue damage). All three factors play some role in most low trauma fractures, and it is not possible to say which may be the most important in any given case. Nevertheless, most of the effort in this regard in the past 30 years has been devoted to the measurement of bone mass and density, and hence much of what we know about bone strength in living individuals comes from our observation of this facet of the bone strength triad. The general consensus is that decreased bone mass produces a decrease in bone strength (26), but there is disagreement about how much of a strength reserve bone possesses and whether it usually takes more than a simple decrease in mass to produce a fragility fracture.

Whatever the final answer, it is an inescapable fact that most elderly individuals have bone mass values that are more than two standard deviations below the young adult mean and hence they are *all* at increased risk for fragility fracture. Why some older persons do fracture and some do not appears to be explainable, at least in part, by differences in bony architecture and in the effectiveness of repair of universally occurring fatigue damage. These factors and their interplay have been reviewed extensively elsewhere (53).

Briefly, individuals with compression fractures of the vertebrae have been found to have excessive loss of horizontal, cross-bracing trabeculae in their cancellous bone, whereas other individuals with the same overall degree of bone loss, but with the bracing trabeculae maintained intact, do not fracture. Women, particularly, are more prone to lose their horizontal trabeculae than are men, and this fact is probably the explanation for the 6:1 to 8:1 female:male sex differential in vertebral osteoporosis. Similarly, studies of elderly individuals with fractures of the femoral neck have shown localized failure of bone remodeling in the region concerned (40) as well as cytochemical abnormalities of osteocytes in the fracture region (33). The ultimate significance of such findings is not known, but they suggest a sluggishness of the bone remodeling process which, other things being equal, might have removed accumulated fatigue damage and better maintained the strength of the bony structures.

Nutrition enters into this portion of the fracture domain predominantly through its influence on bone density. However, trace nutrients such as certain vitamins (e.g. D and K), or the trace minerals such as manganese, copper, and zinc, may directly influence the remodeling process and hence affect bone strength through their impact on the repair of inevitable fatigue damage. However, little is known about these possibilities, and in most of the following discussion, the emphasis is on the nutritional factors that influence bone *mass*.

Bone Mass/Density

Bone mass and density are themselves influenced by many factors. The three most important, overall, are physical activity, gonadal hormones, and calcium intake. Manganese, zinc, copper, vitamin C, vitamin K, phosphorus, and protein are also essential for building a healthy skeleton, but, except for calcium, their effects are usually seen most clearly during growth. Once built, however, the skeleton tends to be relatively insulated from many subsequent nutritional deficiencies. In addition, a number of other factors also influence bone mass, such as smoking, alcohol abuse, and various drugs used to treat a variety of medical illnesses, as well as those illnesses themselves.

The effects of each of these factors are largely independent and therefore one cannot substitute for, or compensate for, the other. Thus, a high calcium intake will not offset the loss of bone that occurs immediately following menopause in women or castration in men. Nor will vigorous physical activity alter menopausal loss, for that matter. Similarly, physical activity will not compensate for an inadequate calcium intake. Neither will a high calcium intake offset the effects of alcohol abuse or smoking. Much of the apparent confusion in the bone field over the past 20 years could have been avoided if we had better understood that these factors, while interactive, are largely independent.

Finally, although much of the following discussion focuses on calcium, it is necessary to stress, what should perhaps go without saying, that calcium is not an isolated nutrient; it occurs in foods along with other nutrients, and diets low in calcium tend also to be nutritionally poor, generally (7). Thus, while it is necessary to deal with nutrients one at a time in an analysis such as this, the disorders in patients are likely to be more complex.

CALCIUM

The topic of calcium and osteoporosis has been reviewed in these pages by Avioli in 1984 (3) and again by Arnaud & Sanchez in 1990 (2). Cumming in 1990 published a metaanalysis (28) of all studies up through 1988 relating calcium intake and bone status and concluded that the evidence supported a positive relationship between the two variables. Given this prior work, this review focuses mainly on studies published since 1988. Interestingly, the quantity of such studies is surprisingly large and their quality, overall, is gratifyingly high. The majority view among members of the nutritional scientific community now appears to be that calcium intake, within the range of plausible intakes, has a positive influence on bone status (2, 9, 82, 96). There have also been dissenting views (71). However, while several questions remain unanswered, previous controversy can be said to be resolved, and the

results of disparate studies can be satisfactorily explained within a comprehensive model of bone metabolism.

In the discussion of published studies it is helpful to highlight certain methodological problems which inevitably are a part of studies of this topic and which may explain prior disagreement:

1. Multifactorial character of age-related change in bone mass and of bone fragility.
2. Differing calcium intake distributions in various study populations.
3. Inability in most types of studies to address the several causes of calcium deficiency; i.e. while calcium deficiency may be caused by low intake, it may also be due to decreased absorptive performance or high obligatory excretory loss.
4. Insufficient power to detect plausible population-level correlations, if they exist, in many published studies.
5. Failure in most studies of the postmenopausal period to recognize the special circumstances created by estrogen withdrawal in the few years immediately following hormone loss and, in the analysis of published reports, to separate studies on the basis of proximity to menopause.
6. Use of weak or inaccurate tools for estimating calcium intake in all studies except for intervention or metabolic studies, and the seeming failure to recognize this limitation.

1. Multifactorial character of change in bone mass and of bone fragility. Calcium, to the extent that it plays a role in bone health, is only one of many interacting factors (54, 55). Some of these factors remain unknown, or at least inadequately explored; hence it is difficult to control for them. While exercise, hormonal status, heredity, co-morbidity, medications, smoking, and alcohol consumption are all recognized in a general way, they are seldom adequately quantified in observational studies. Finally, as already noted, bony fragility, which underlies low-trauma fractures, is due only partly to reduced bone mass. While the nonmass fragility factors may yet prove to have nutritional correlates, little is known of these possibilities to date. Calcium deficiency, to the extent that it plays a role in this complex context, is postulated to have an effect only on bone mass. Hence the connection of calcium intake to fracture risk can be no stronger than the connection between bone mass and fracture.

2. Differing calcium intake distributions in various populations. Reported studies need to be interpreted against the national background of the individuals studied. To the extent that low calcium intake contributes to the osteoporotic fracture problem, one would expect to find the evidence most clearly presented in populations with intakes that span the range from low to

high, not in populations with predominantly high intakes. Beaton, in his McCollum lecture (11), dealt clearly with this problem for nutrients generally. For example, most of the reported studies from the Netherlands, where calcium intakes are comparatively high, have shown little or no relationship between calcium intake and bone mass (126, 127). The two studies of Elders et al. (37, 38) are comparative exceptions for Dutch studies.

In general, osteoporosis occurring in populations with high calcium intakes would be expected to have causes other than calcium deficiency, and one would not look to studies in such countries for evidence bearing on this question. Even the discrepancy between the hip fracture studies of Holbrook et al (67), who found a protective effect of high calcium intake, and of Wickham et al (130), who did not, may be due to differences in distribution of calcium intake between their respective populations. Mean calcium intake in the Wickham study was well into the upper tertile of intakes for the Holbrook study.

3. Inability in most studies to address the several bases for calcium deficiency. Calcium deficiency may be caused not only by low intakes but also by inefficient absorption (60) or by high obligatory losses, neither of which is quantified in most observational or even intervention studies. (For the most part, these causes of calcium deficiency can be satisfactorily measured only in metabolic studies.) The net result of ignoring them is a misclassification bias. Individuals classified as having high intakes may still be actually deficient if their absorptive or renal excretory performances are not appropriate for their intake. Conversely, other individuals with lower absolute intakes may be fully calcium-replete if their absorptive and excretory processes have adapted sufficiently. If these misclassifications occur equally in both directions (which seems unlikely from the evidence available), the result would be only a loss of power (see below). But if the misclassification of deficient subjects as high intake individuals dominates, the result will be a bias against finding a calcium effect.

4. Insufficient power. Power is a well-understood, if sometimes ignored, problem in studies testing an hypothesis. What has been little appreciated until recently is the relative magnitude of the calcium effect to be expected in observational studies in a free-living population, and the impact that that magnitude has on investigational power. For example, the longitudinal study of Riggs et al (114), widely quoted as showing no benefit from calcium, had a stated power to detect a population level correlation with a value in the range of 0.6 or higher. But even given zero errors in estimating either bone loss or calcium intake (which are not even remotely possible), Avioli & Heaney (4) calculated that the highest plausible population level correlation would be less than 0.4. Given the problems discussed below inherent in assessing calcium intake, it seems clear that the correlation likely to be detectable would have

been less than 0.30—far smaller than Riggs et al could reliably have detected. This is a problem to which Cumming (28) also referred in his metaanalysis.

5. Failure to recognize the special characteristics of the immediate postmenopausal period. It now seems increasingly clear that bone loss in the immediate postmenopausal period is due almost exclusively to loss of gonadal hormones. This point is discussed at greater length below. Its importance in the context of interpreting published studies lies in the fact that the effect of gonadal hormone loss is relatively short-lived. Studies that either concentrated upon or failed to exclude early postmenopausal women generally failed to find a calcium effect or underestimated its magnitude. On the basis of current understanding, this is exactly what would be expected, and the ineffectiveness of calcium at this time is not relevant to the need for calcium at earlier and later life stages.

In a cross-sectional investigation Elders et al (37), found habitual calcium intake to be a significant determinant of perimenopausal bone mass, but their data suggested that calcium probably exerted its effect by influencing peak bone mass, rather than by influencing menopausal bone loss. This conclusion is consistent with the findings in the study by Hansen et al (49): calcium intake was highly correlated with bone density both before and after menopause, but it had no effect on the quantity of bone lost across menopause. In their intervention study Elders et al (38), using daily supplements of 25 and 50 mmol of calcium in addition to a basal diet averaging about 29 mmol per day, found a dose-related, stepwise reduction in rate of bone loss before menopause, in the early postmenopause, and in the late postmenopause as well. However, in their early postmenopausal women, mean rates of bone change were still negative, even at the highest calcium intake (79 mm/day), though slightly less so than at lower intakes. These latter findings may reflect what Kanis & Passmore (71) have termed a pharmacologic effect of calcium, i.e. a suppression of the basic remodeling process itself, which thereby slows change in bone mass irrespective of its cause.

6. Use of weak and inaccurate instruments for assessing calcium intake. Most observational studies have assessed calcium intake at one or, rarely, two points in time, using a food frequency questionnaire (FFQ) limited in some studies to as few as seven food items. Others used one-day recall, three-day diaries, and a few, seven-day diaries. The accuracy of these assessments is a problem of underappreciated importance, at least among many workers in the bone health field, if not among dietitians and nutritional scientists. Only a few of the problems with assessing calcium intake by such tools can be touched upon here.

Even when one knows to the gram exactly how much of what foods went into an individual's mouth, chemical analysis invariably reveals a somewhat different figure from the entries in food database estimates for those same

foods. (This, of course, is because the nutrient content of food varies, lot-to-lot.) Charles (23) found, under metabolic ward conditions, a correlation of only 0.87 between database values for calcium content of various diets and the actually analyzed contents. This means that fully one fourth of the actual interindividual variation in intakes could not be explained by knowledge of the precise quantities of foods eaten. In similar studies performed in my own laboratory the correlation was better ($r = 0.98$), but it still was not perfect, and can never be. The problem becomes even more difficult when one moves from subjects studied under metabolic ward conditions to a free-living population, where we no longer can measure, but only can estimate, quantities consumed. Even seven-day diaries fall short of capturing adequately the full details of intake (54, 57). Given typical day-to-day variation in calcium intake, diaries extending as long as 13–17 days may be needed for an acceptably small error estimate. These are plainly impracticable for most studies.

FFQs, the most commonly used tool, are attractive for their simplicity and ease of administration, but are generally less accurate than multiple-day diaries. In any event, in several studies in which they are compared with other methods, they produced substantially higher intake estimates than did diet diaries taken at the same time in the same individuals. Bergman et al (13) reported that a FFQ produced more than 50% higher figures for some nutrients, notably calcium, than did a 3-day food record. The validation usually cited in published reports is that the FFQ value correlated with some other measure of calcium intake. For example, Musgrave et al (93) reported a correlation coefficient of 0.73 between a FFQ value for calcium intakes and a concurrently developed food record. But one would expect correlation. That is not the issue. What is at stake is substitution. It is this chain of substitutions (FFQ for multiple day food record; food record for actual quantities of foods consumed; data base values of foods consumed for actual food content) that degrades the estimate produced by the measures actually employed in most observational studies, whether cross-sectional or longitudinal.

A further problem is one-point sampling. Heaney et al (57) have documented the quite considerable extent to which individual intakes vary over time. To the extent that bone density is affected by calcium intake, it would be the integrated intake over many years which would be important. Yet, very few published studies have used multiple point sampling. Finally, there is the general failure to include, in estimates of intake, the calcium content of excipients in medications or supplements taken for some reason other than their (generally unrecognized) calcium content (57). The error so introduced will generally be small, but in perhaps 5–10% of middle-aged or elderly women it will result in a substantial misclassification bias, i.e. counting people as having low intakes when they are actually high.

What may be considered surprising in all this is the contrast between the great attention paid to the accuracy, sensitivity, and specificity of bone mass

Table 1 Categorization of studies of the relation of calcium intake to bone status in Caucasian women

		Investigator-controlled calcium intake?		
		No	Yes	Total
Women from 0-5 years postmenopausal excluded?	No	0/8	4/7	4/15
	Yes	11/16	12/12	23/28
	Total	11/24	16/19	27/43

measurements and the low accuracy of the means to quantify what is postulated to be the independent variable in the hypothesis being tested. What should not be surprising, therefore, is that some studies fail to support the hypothesis.

In summary, Table 1 presents a classification of the 43 relevant studies in Caucasian women published since 1987. To facilitate analysis, studies are categorized first by menopausal age of the subjects and then by degree of investigator control over, and/or knowledge of, calcium intake.

The Table shows that about 60% of the studies reported a benefit from calcium (27 of 43). But this simple tallying of successes and failures treats all studies equally and does not do full justice to the data. For reasons already discussed, studies in which calcium intake is not investigator-controlled are biased toward the null hypothesis. When such studies find an effect, therefore, the result is more convincing than when they do not. In a similar way, as also discussed, endogenous calcium released from bone in the wake of gonadal hormone loss substitutes for, or displaces, exogenous calcium. For that reason, studies performed within the first five years after menopause also tend to show no benefit of dietary and supplemental calcium.

The 4-way breakdown presented in the Table is the result of applying these two criteria. As can be seen, the proportion of positive studies rises with the rigor and salience of the scientific design. Twenty-three of 28 studies excluding early postmenopausal women were positive, as were 16 of 19 in which the investigators controlled the calcium intake. And without exception all 12 studies meeting both criteria showed a calcium benefit.

The Requirement for Calcium

Within cells, calcium is a nearly universal second messenger mediating such diverse responses as muscle contraction, mitosis, and secretion. Extracellularly, it is essential for blood coagulation and neuromuscular signal transmis-

sion. The cells themselves regulate the concentration of free calcium ions in their cytosol, keeping it at least three orders of magnitude lower than in the surrounding extracellular fluid (ECF). They do this both by pumping calcium up a concentration gradient out to the ECF and by sequestering calcium in intracellular vesicles, such as the sarcoplasmic reticulum in muscle. In turn, ECF [Ca^{2+}] is rigidly controlled at about 1.2 mM, mainly through the actions of parathyroid hormone, calcitriol, and calcitonin. The secretion of all three hormones is responsive directly or indirectly to changes in ECF [Ca^{2+}] levels.

The primary metabolic functions of calcium are as outlined, and if calcium were like most other nutrients, its requirement would be defined as the intake needed to sustain those functions. But true nutrient deficiency in that sense is essentially never encountered in humans. This is because the skeleton serves as a very large—essentially inexhaustible—reserve of calcium for these critical biochemical functions. Over the millennia of vertebrate evolution, the skeleton has acquired important mechanical functions as well. It is for these mechanical functions of the skeleton, rather than in relation to the metabolic activity of calcium, that the notion of a requirement has meaning. Apart from fat, which in addition to being an energy reserve, provides insulation for animals living in cold environments, bone is the only known instance of a nutrient reserve having an important function in its own right. In fact, the size of the reserve (i.e. bone mass), as has just been seen, is one of the main determinants of bone strength. Thus, unlike most other nutrients, the requirement for calcium is related to building and maintaining the largest reserve possible within the individual genetic program.

Primary Prevention: The Acquisition of Genetically Programmed Bone Mass

Calcium is a threshold nutrient (as, for example, is iron). Forbes et al (43) showed in growing rats that femur bone mass was a linear function of diet calcium content up to values of about 0.6%. Above that intake, bone mass remained constant. Long bone growth was not limited in these animals at subthreshold levels of calcium intake, but bone *mass* was. Current models of bone metabolism explain this effect by a relative excess of bone resorption on the subthreshold diets, resulting in a tearing down of some of the bone deposited so that its calcium could support the demands of continuing linear growth. The resulting bone is thus of normal external size and shape, but of increased internal porosity and/or decreased cortical and trabecular thickness.

No evidence suggests that dietary calcium itself affects the mineralization process so long as serum calcium levels are maintained, either during growth or later, although some data suggest that low calcium intakes may limit linear growth. This may occur in some growing animal models with very low calcium intakes, and has been reported for Scottish school children (78), in whom

addition of 12 oz of milk per day to the diet of one group resulted in substantially greater linear growth. But in such experiments it is not possible to tease apart the effects due specifically to calcium from the effects of the associated extra protein and energy intake. In general, it seems probable that the effect of low calcium intake on the growing skeleton is mediated largely through modulation of the balance between bone formation and bone resorption and is confined to an effect on bone density.

Clearly, while sufficient exogeneous calcium must be present to sustain density during growth and to maintain skeletal mass later in life, additional calcium above the threshold (whatever its value may be) will not produce more bone than is required either by the genetic program or by current levels of mechanical loading. The notion of a requirement is thus tied to this threshold. An individual's requirement would be the inflection point of the curve relating bone mass to intake, i.e. the threshold. A recommended dietary allowance (RDA) would be the corresponding point for a population. In both, further increases in intake produce no additional benefit or confer no additional protection. The following discussion therefore centers around new evidence in humans as to where the inflection point of the curve relating calcium intake to bone mass is located.

Matkovic assembled all studies, performed in healthy growing subjects, of calcium balances which had been published since 1922 and eliminated only those which tested altered intake or evaluated experimental foods or diets (83). He was able to find over 500 such studies, a number sufficiently large to detect threshold behavior if present. He and Heaney (85) analyzed these data by age group and found clear evidence of a balance threshold at all stages of growth from infancy up through the skeletal consolidation years (ages 18–30). The threshold calcium intake in children ages 2–8 was 35 mmol/day; in adolescents aged 9–17, 37 mmol/day; and in young adults aged 18–30, 24 mmol/day. These values are somewhat above the current RDAs for the ages concerned (20–30 mmol/day).

Johnston and colleagues, in a 3-year double-blind placebo-controlled trial in identical twins, found significantly augmented skeletal density in children receiving 17.5 mmol of supplemental calcium in addition to their normal diets (70). Mean intakes in the unsupplemented twins were between 21 and 22 mmol, already above the RDA for prepubertal children. Using a similar design, but a more modest supplement dose (8.75 mmol/day), Lloyd et al (80) found augmented bone gain in adolescent females, relative to unsupplemented controls whose intake averaged about 22 mmol/day. These findings thus confirm the conclusions drawn by Matkovic & Heaney in their metaanalysis (85).

Finally, Recker et al (108), in a four year longitudinal study of women aged 19–30, found continued augmentation of skeletal mass and density at total

body, forearm, and spine, on diets averaging about 17.5 mmol/day. Bone acquisition was positively correlated with physical activity and with calcium intake, and negatively correlated with protein intake and age. Regressing rate of skeletal gain on age, these investigators found X-axis intercepts (zero gain) for the various bony sites to be at about age 29. The single most powerful factor influencing this skeletal consolidation was the calcium:protein ratio of the diet.

In a cross-sectional study Hirota et al (65) found a positive association between calcium intake and bone mass in Japanese college women aged 19–25, as did Fehily et al (41) in young Irish women. Prentice et al (104), in another cross-sectional study, found gain in bone mass until sometime in the fourth decade in Gambian women and noted in passing that, though rates of gain seemed slower, they remained positive in British women, as well, until sometime in the fourth decade.

On the other hand, Bonjour and colleagues (15), in cross-sectional studies of Swiss children, found substantial slowing of skeletal acquisition by age 18 and suggested that there was no further accumulation after that age. However, these authors did not specifically evaluate women in the third decade. It may also be that, with higher national calcium intakes, Swiss adolescents reach their genetic maximum earlier than do North American youngsters.

Thus, from both longitudinal and cross-sectional data, most studies show continuing skeletal consolidation throughout most of the third decade, dietary intake permitting. These studies also suggest that current estimates for the RDA may be low. Note in this regard that the RDA for children and adolescents is explicitly based on an assumption that calcium absorption efficiency at these ages is at least 50% (111). Recent studies of absorption by Miller et al (91, 92) using double tracer, stable isotope methods, showed that, in the ages concerned, absorption fraction averages only about 35%, or nearly one-third lower than the RDA Committee assumed.

A further reason for a higher than expected requirement during growth is seen in the observation of high urinary calcium loss, particularly in adolescents, noted first by Matkovic et al (84), and then found in the balances assembled by Matkovic & Heaney (85). During growth, urine calcium is a function mainly of body size, and during adolescence, particularly, it is quite independent of calcium intake. While a high urinary calcium loss might be taken as evidence of calcium sufficiency in an adult, that is not a tenable conclusion in a growing adolescent. Yet in both studies (84, 85) urinary calcium values in adolescent girls were high even on very restricted intakes—so restricted that no skeletal acquisition was possible. Hence this loss through the kidneys must be considered obligatory, and coupled with the lower than presumed absorption efficiency, explains why the threshold intake may be higher than estimated in the current RDAs.

Current nutritional thinking about this issue presumes that the body would conserve calcium during times of need—such as during the rapid growth of adolescence. Thus, the finding of values as low as those of adults for absorption and excretion at this age comes as a surprise. That reaction may reflect some degree of nutritional provincialism. Eaton & Konner (35) and Eaton & Nelson (36) point out that the calcium nutrient density of the diets of both the chimpanzee and contemporary hunter-gatherers is in the range of 1.75–2.0 mmol of calcium/100 kCal, two to four times what typical Western diets provide. They suggest that human physiology has adapted to such calcium-rich diets, and that the time that has elapsed from the agricultural revolution to the present has been insufficient to have allowed substantial evolutionary resetting of the basic physiological mechanisms involved.

Secondary Prevention: The Conservation of Acquired Bone Mass

Because of critical physiological differences at various life-stages, conservation of acquired bone mass is discussed under three temporal headings: premenopause, early postmenopause, and late postmenopause. While these three terms explicitly refer to women, available evidence suggests that the situation for aging men is similar to that for late postmenopausal women.

PREMENOPAUSE Most authors have suggested that bone mass declines slowly with age after the adult peak is reached at age 30–35, and cross-sectional studies have tended to be consistent with that presumption. However, most such studies have inappropriately applied linear models across significant watershed events (e.g. menopause), and cross-sectional studies are also prone to the confounding of cohort effects (109). When they measured it directly, Mazess & Barden (87) found no measurable loss with age at any bony site in premenopausal women, and Recker et al, in a longitudinal study of over 70 late premenopausal women, found no measurable loss over periods up to three years in duration (110). These two observations suggest that ambient dietary calcium intakes (averaging above 700 mg for the Recker study) were sufficient to prevent calcium deficiency-related bone loss in premenopausal women.

Only one study suggests that augmented calcium intake may be beneficial in premenopausal women. Baran et al (5) found no loss in women supplemented with two additional servings of dairy products daily, but substantial loss in unsupplemented control subjects. However, the difference was detectable only at the third year study point, at which time there had been substantial losses of sampling units; and so it is hard to know how much of the apparent effect was a consequence of these losses.

This period of life has received much less attention than other phases of a

woman's life, but the available data are consistent with a conclusion that intakes in the range of the current RDA are adequate to prevent intake-related bone loss in healthy premenopausal women.

EARLY POSTMENOPAUSE Typical women lose roughly 15% of their bone mass following menopause. The loss is well described as an exponential function with a rate constant of about $-0.25\ yr^{-1}$ (55). Thus, most of the loss will occur within the first 5 yr after menopause, and by 15 yr postmenopause, the estrogen withdrawal loss merges into the slow, age-related loss of senescence, seen in both men and women. The early postmenopausal change in the skeleton is clearly due to withdrawal of gonadal hormones, and is preventable indefinitely by estrogen replacement therapy. An apparently very similar loss occurs in castrated males.

The mechanism of this shift is uncertain but has been plausibly explained as a resetting of the putative bone density "mechanostat," the bony apparatus that adjusts bone density through a canonical feedback loop that responds to sensed mechanical loading. Ostensibly, the setpoint is lowered in the presence of gonadal hormones, and accordingly bone density increases to reduce the load-induced strains. The shift from high estrogen to low estrogen levels at menopause thus produces a down-sizing of bones exactly analogous to what would be produced by decreased exercise.

Virtually all published studies show little or no effect of dietary calcium during this five-year period following menopause (39, 71, 114–116, 121). Before the centrality of the load-density feedback system and its sensitivity to gonadal hormones were appreciated, this failure of calcium was over-interpreted to mean that calcium intake had no role in age-related bone loss at any age. It is now clear that the situation is rather different.

During the first few years after menopause, so much calcium is made available from downward revision of bone density that in effect there may be no external calcium requirement whatsoever. Only with intakes high enough to suppress remodeling would an effect of calcium intake be expected. Elders and co-workers (38), in fact, show exactly such an effect in controlled trials with intakes of 29, 54, and 79 mmol/day. And, while the highest intakes in early postmenopausal women delayed menopausal loss, they did not prevent it.

The early postmenopausal years are, of course, the time when bone loss is the most rapid and the value of successfully intervening is most evident, which may explain why so many previous studies of calcium effect chose to address the early postmenopausal period (39, 71, 114–116, 121). But loss at that life stage is best prevented by estrogen. This topic has been extensively explored elsewhere (55) and is elucidated clearly in the study by Dawson-Hughes et al (31), in which, with the same investigational design, the same measurement

methods, and the same calcium sources, a modest calcium supplement abolished age-related bone loss in women six or more years postmenopausal, but was quite without effect in women within 0–5 years following menopause. This point is also illustrated by Prince et al (106), who studied women an average of 5–6 years after menopause. Their subjects thus straddle the age-dividing line used by Dawson-Hughes et al (31) and would be predicted to exhibit only a partial response to increased calcium intake. Prince et al, in fact, found a significant reduction in rate of loss in the calcium-supplemented group, but the effect was less than produced by estrogen, precisely as predicted for a group this close to menopause.

LATE POSTMENOPAUSE/SENESCENCE As just noted, women more than five years postmenopausal, and middle-aged and older men generally, show decreased bone loss with age if they have high calcium intakes (28, 56). The double-blind, placebo-controlled study by Dawson-Hughes et al, (31) already cited, makes that point clear, as do other studies, which, though mainly observational, nevertheless all come to the same conclusion. The Dawson-Hughes study concentrated on women with calcium intakes below 16 mmol/day, and it is sometimes interpreted as showing a benefit of calcium supplements only in those women with intakes below 10 mmol/day, and even that the threshold intake itself must be below 22 mmol/day. Both interpretations go well beyond the data. What the study clearly shows is that women with low intakes lose bone, that calcium supplements reduce or prevent that loss, and that the effect is not seen in women less than five years postmenopausal but becomes quite clear at six or more years after menopause. The study was not designed to provide more quantitative information. In fact, the data show a clear, dose-related effect, with women below 10 mmol/day losing bone most rapidly, women supplemented with calcium losing not at all, and placebo-treated women with intakes between 10 and 16 mmol/day, losing at a rate intermediate between the two. While the loss in the latter group was not significantly different from the loss in women with the lowest intake, this is nearly always true for adjacent groups in any continuous variable. Again, as Beaton pointed out for nutrients generally (11), there must be a range of intake if one is to find a statistically significant relationship.

Similarly, the absence of loss in this study in the supplemented women, whose intakes averaged about 22 mmol/day, is interpreted as indicating that 22 mmol is sufficient. This also would overinterpret the data. Calcium supplements (or any remodeling suppressor, for that matter) induce a remodeling transient that interferes with the ability to measure new steady state rates. Only changes beginning after one year or more of therapy can safely be used to infer the new steady state. The Dawson-Hughes study was not designed to address such questions.

That the effective response threshold may well be higher than the levels achieved by Dawson-Hughes is indicated both by the older metabolic balance studies of Heaney et al (61) and the more recent, very similar metabolic studies of Charles et al (23, 50), both of which point to equilibrium intakes well in excess of 25 mmol/day. Reid et al (112) in a design similar to that of the Dawson-Hughes study, but starting with women with higher basal intakes and supplementing with 25 mmol/day, rather than 12.5 mmol, found results essentially similar to those of Dawson-Hughes et al (31). Furthermore, their data indicated that the effect continued into the second year of treatment and thus showed that the much higher calcium intake had induced a new steady state—in other words, that women with higher basal intakes were nevertheless responsive to additional calcium. Finally, in older adults, and even in those with prior hip fracture, calcium supplementation has been shown to slow or prevent further bone loss (22a, 24).

Calcium Intake and Risk of Fracture

For reasons of feasibility, most studies of nutrition and bone health have used bone mass (or change in bone mass over time) as a surrogate for bone strength, which is the ultimate focus of our interest. And, while there is an undoubted relation between the two, it is important to bear in mind that low trauma fractures and their prevention must ultimately be the focus of interest.

The study of Matkovic and colleagues (86) was perhaps the first to demonstrate anti-fracture efficacy for high calcium intakes. However, the authors found protection only for hip fracture, not for the equally common wrist fracture; and because it was both observational and cross-sectional, the study did not control for many possibly confounding variables. However, Holbrook et al, in a 14-year longitudinal study, also found that hip fracture rates were lower in individuals ingesting higher calcium diets (67). Three other epidemiological studies have been reported (27, 77, 130), one showing that high calcium intakes protect both men and women, one showing protection for men only, and one showing protection for neither.

In perhaps the largest and best controlled study using fracture itself as the endpoint, Chapuy and colleagues have shown substantial reductions in fracture rate in a large group of institutionalized elderly given supplements of both calcium and vitamin D (22a). Protection became apparent after 6–12 months of treatment, and by 18 months, fractures were reduced by more than 30% in the treated individuals. Heikinheimo et al (64), using vitamin D alone in a randomized controlled trial, also found a substantial reduction in all fractures in the elderly.

Three studies in elderly individuals have reported decreased hip fracture risk in patients receiving thiazide diuretics, an effect presumably due to thiazide-induced reduction of urinary calcium losses (42, 76, 107). Nordin

and colleagues have reported a tendency to a renal calcium leak in postmenopausal women (98), and, as has already been noted, renal conservation of calcium is an important determinant of the intake requirement. Hence these results complement the data from direct studies of calcium intake. Finally, while fractures in the elderly are the main concern, it is worth noting that Chan and colleagues, in a long-running series of studies, have shown fracture rates and bone density in children inversely proportional to calcium intake (20–22). So, at all life-stages, other things being equal, the higher the calcium intake, the stronger the skeleton.

Calcium as a Component of Treatment of Established Osteoporosis

The goals of treatment of osteoporosis, in addition to symptom control and rehabilitation, include arrest of further bone loss and, where possible, restoration of lost bone mass. Estrogen, the bisphosphonates, and calcitonin act mainly to arrest bone loss, although they often produce a small increase in mass that is probably at least in part a remodeling transient. Fluoride and PTH, both still experimental, increase trabecular bone density more dramatically, sometimes restoring it to young adult values.

However, for any of these modalities to produce these effects, calcium intakes must be sufficient to prevent further bone loss and/or to support the laying down of new bone, without taking calcium from other regions of the skeleton. Both PTH and fluoride will increase spine trabecular density irrespective of intake, and both agents are thus capable of producing an intra-body shift of bone from appendicular cortical sites to axial, trabecular sites. This seems to be a part of the reason why, in some studies, appendicular bone mass has actually declined while spine density was improving and why peripheral fracture rates increased under these therapies. In this connection, Dure-Smith and colleagues have demonstrated a striking degree of bone hunger in patients responding to fluoride treatment (34).

Hence, calcium supplementation, usually beyond what can feasibly be provided by diet alone, constitutes an essential component of virtually every therapeutic regimen for this disorder. Given the relatively poor absorption efficiency found in the elderly, intakes of at least 40–60 mmol/day seem necessary. Adequate attention needs to be paid, as well, to vitamin D status (see below).

Nutrient-Nutrient Interactions

FIBER The term *fiber* refers to a varied group of plant polymers that are not hydrolyzed by human digestive enzymes and therefore pass intact through the principal absorptive region of the intestine. These polymers frequently contain

multiple, negatively charged groups capable of complexing with cations. In theory, therefore, they might interfere with the availability of co-ingested calcium, at least until the intestinal contents reach the colon where bacterial hydrolysis of the polymer chains may free bound cations. Barger-Lux et al (8), in examining the time course of tracer absorption from labeled calcium intakes, noted that 95% of the calcium that will be absorbed from a mixed food meal is absorbed by 4 to 5 hr after ingestion, and that the remaining 5% is slowly absorbed over the next 20 hr. This performance is consistent with the notion of release of bound calcium after entry into the colon. At the same time it also indicates that not much calcium is made available that way in the typical adult human.

Contemporary diets typically contain from 5 to 15 g fiber. Current recommendations are for intakes in the range of 15–20 g/day. This increase, if realized, might be expected to interfere to some extent with calcium absorption. While such interference can be demonstrated in various animal models, it has been harder to show in humans. When reviewing various aspects of calcium nutrition in the elderly in 1982, Heaney et al (58) concluded that only large increases in fiber intake were likely to produce much effect, and an expert panel, in reviewing the topic more recently (101), came to much the same conclusion.

Knox et al (73) found that increasing the fiber content of a test meal from 0.5 to 10.5 g (using raw wheat bran as the fiber source), resulted in a decrease in apparent calcium absorption (measured by whole body tracer retention) from about 25–26% for the low fiber meal to about 19–20% for the high fiber meal. Weaver et al (129) found that 40 g of wheat bran cereal (containing 12 g of bran) reduced calcium absorption from co-ingested milk by about one third—from a value of 37.5% to 25.8%. On the other hand, the same paper reported higher absorption for the calcium contained in whole wheat bread than for milk ingested at a comparable calcium load. The same authors (62, 63) also report good absorbability for calcium from kale and from low phytate soybeans, both of which contain significant amounts of fiber. Consistent with this finding are the data of Wisker et al (131), who found that a large addition (15 g/day) of a low phytate barley fiber concentrate did not interfere with either calcium absorption or calcium balance in normal adults. Apparently, the various forms of fiber contained in different food sources do not interfere with calcium absorption to an equivalent extent.

It may be concluded that at least certain fiber sources, such as wheat bran, do interfere with absorption of co-ingested calcium, though the effect over an intake range typically encountered in adult diets is not likely to be very great. The experiments of Knox et al (73) and of Weaver et al (129) are concordant and can be summarized to predict a decrease in absorbability of about 20–30% upon going from an intake of 0–1 g bran fiber per day to roughly 30 g per

day, values that span the plausible range of fiber intakes for most adults. Thus, an adult with a basal intake of 10 g per day, who chooses to double that intake, might therefore expect a 6–10% decrease in food calcium availability. This is not negligible, particularly if calcium intake is already low. Still, it could be easily compensated for by relatively small increases in dietary calcium intake. Finally, it must be stressed that these calculations apply only to wheat bran fiber. At least some other plant fibers interfere not at all.

PHOSPHORUS Phosphorus is as important for bone health as is calcium. As phosphate, it makes up roughly half the weight of bone mineral and hence must be present in adequate quantities in the diet both to mineralize and to maintain the skeleton. Phosphorus is generally present in relatively adequate quantities in the US diet, and most of the recent concern of the nutrition community has centered on the question of whether its presence in the diet might be excessive, with consequent harm (25).

Increased phosphorus intake transiently depresses ionized calcium and thereby leads to increased secretion of PTH, which could clearly influence bone. However, the effect wanes in a few days (118), unless calcium intake is also suppressed, in which case PTH levels remain high (17, 18). Under steady state conditions, an increased phosphorus intake reduces urinary calcium loss and increases digestive juice secretion of calcium (25). The two effects are approximately equal in magnitude; hence total body calcium balance tends not to be affected.

Calvo et al (18) placed young adults on high phosphorus, low calcium diets, and noted elevations of iPTH and urinary hydroxyproline lasting for at least four weeks. However, Barger-Lux & Heaney (6), found identical changes in both variables when *both* calcium and phosphorus intake were lowered. Thus, it seems unlikely that the effects noted by Calvo et al are due specifically to high phosphorus intakes, as these authors suggested.

Increased phosphorus intake also powerfully suppresses renal synthesis of calcitriol, which could lead to decreased calcium absorption (103). At the same time, as already noted, increased phosphorus intake suppresses urinary calcium loss and is used for that purpose in patients with renal stone disease. The effect appears to be direct, since it can readily be demonstrated by adjusting phosphate loads in patients on total parenteral nutrition (133).

Although low phosphorus intakes are less common in the United States, low intakes may limit the body's utilization of calcium for building and remodeling bone. According to NHANES II (19), only about 10 to 15% of women aged 65 to 74 have phosphorus intakes less than two-thirds the RDA. The proportion is probably higher for the old elderly. Low phosphorus intakes lead to excessive urinary calcium loss and hence could aggravate the effect of low calcium intakes. Conversely, large calcium supplements will lower

absorbed phosphorus and, in individuals who already have low phosphorus intakes, this effect could produce phosphate deficiency. Individually, these interactions are generally well studied, but taken together in the very old elderly, who may have other problems (such as declining renal function, which will affect ECF phosphorus levels), they present complexities that have not been adequately explored.

PROTEIN AND SODIUM Dietary excesses of both protein and sodium increase urinary calcium loss and hence interfere with calcium conservation in response to a restricted calcium intake. Protein is a powerful determinant of urine calcium, partly, at least, because of the associated increase in acid load (1, 79, 132). At least three different groups of investigators have found that a doubling of protein intake results in roughly a 50% increase in urine calcium loss (59). Hence, protein intake is an important determinant of calcium requirement. This is shown clearly, for example, by the finding cited above that the calcium:protein ratio of the diet was the most important measured determinant of the rate of bone acquisition in third-decade women (108).

While excess protein intake (relative to calcium) is more of a problem in the US than is protein insufficiency, the relationship is biphasic, and low intake is also important for bone health, especially in the elderly. Bone mineral density has been reported to be positively correlated with protein intake in elderly Caucasian and Asian women (75, 125), and protein intake was inversely correlated with rates of bone loss (46) in older women. Many other variables are involved in these studies, and by themselves they are only suggestive. However, in view of the clear benefit from protein supplements at the time of repair of hip fracture (10, 32, 69), it seems reasonably clear that in some patients, particularly the very old elderly, insufficient protein intake may contribute importantly to the osteoporosis problem.

Urine calcium rises by 1–2 mmol for every 100 mmol increment of ingested sodium (97). The sodium effect appears more marked at low calcium intakes and may be less important at intakes above 25 mmol/day. It is as if a large renal sodium load interferes with ability to reabsorb calcium from the tubular lumen under conditions when calcium is being conserved, but has less effect when excess calcium is being excreted.

VITAMIN D

That vitamin D is important for absorption of calcium from the diet has long been recognized. Its role lies in facilitating active transport, in part by inducing the formation of calcium-binding protein in intestinal mucosal cells. This function is particularly important for adaptation to low intakes. However, passive transport occurs by other means, not as well elucidated, which are

not dependent upon vitamin D. The proportion of absorption by the two mechanisms varies with intake and is not well characterized; at high calcium intakes (above 60–75 mmol/day) absorption fraction approaches that observed in anephric individuals (~10–15% of intake). Under these circumstances it is likely that active transport contributes relatively little to the total absorbed load. Nevertheless, it is generally recognized that vitamin D status can influence absorptive performance and hence effective calcium requirement.

A principal storage form of the vitamin is 25-hydroxyvitamin D [25(OH)D], and its plasma level is the best clinical indicator of vitamin D status. Although orders of magnitude less potent than calcitriol in promoting active transport, 25(OH)D may possess physiological functions in its own right (12). Vitamin D status commonly deteriorates in the elderly, whose plasma 25(OH)D levels are generally lower than in young adults (44, 88). These elderly persons, without histological or biochemical evidence of osteomalacia, nevertheless exhibit high PTH levels and low absorptive performance, both of which change when they are given physiological amounts of supplemental vitamin D (30, 52, 74). Low dosage vitamin D supplementation of ostensibly healthy postmenopausal women significantly slows wintertime bone loss and reduces the annual parathyroid-mediated activation of the bone remodeling system that occurs in winter through late spring (30). The rate of age-related loss of bone has been found to be inversely correlated to dietary vitamin D (81). Heikinheimo et al (64) in a 4-yr, randomized, controlled trial found a substantial reduction in all fractures in elderly Finns given a single injection of 150,000–300,000 IU vitamin D in the fall of each year. And, in a 3-yr randomized controlled trial, combined supplementation with calcium and vitamin D in 3,270 institutionalized elderly significantly reduced both bone loss and fracture rate after the first year of treatment (22a). The latter study is particularly noteworthy because it concentrated on the most vulnerable population, used the ultimate endpoint (fracture), and employed a strong design (randomized controlled trial).

The foregoing studies, and many others as well, lead inexorably to the conclusion that vitamin D insufficiency is prevalent in the middle-aged and elderly of Europe and North America. In virtually none of these studies was frank osteomalacia a significant feature of the problem. Hence this criterion for true vitamin D deficiency may well be much too strict to be nutritionally useful.

Low 25(OH)D levels in the elderly are partly due to decreased solar exposure, partly to decreased efficiency of skin vitamin D synthesis, and partly to decreased intake of milk, the principal dietary source of the vitamin in the US. Moreover, the elderly exhibit other abnormalities of the vitamin D endocrine system that may further impair their ability to adapt to reduced calcium intake. These include decreased responsiveness of the renal 1-α-

hydroxylase to parathyroid hormone (120) and decreased mucosal responsiveness to calcitriol (45). For all these reasons there is a growing body of opinion that the requirement for vitamin D rises with age (48, 52, 100, 124).

How much of the effect of vitamin D in the above studies is due to facilitating gut adaptation to marginal calcium intakes and how much may represent an extra-intestinal effect of the vitamin in its own right is not clear. Calcitriol receptors are widely distributed in many tissues, and calcitriol enhances PTH-mediated bone resorption, exhibits autocrine action in cell differentiation and in the immune response, and inhibits parathyroid hormone synthesis by direct action on the parathyroid gland. Furthermore, calcitriol elicits a prompt and sizable increase in osteoblast synthesis of osteocalcin (105). Nevertheless, patients with vitamin D–dependent rickets Type II, who lack functional calcitriol receptors, show essentially complete remission of their pathophysiological processes with intravenous calcium infusions alone (14). Furthermore, while subtle impairment of immune function can be demonstrated in nutritional vitamin D deficiency, the defects appear to be sufficiently mild to be of little or no clinical consequence. Hence the issue remains quite unclear.

Nevertheless, whether solely through an effect on calcium absorption, or through other mechanisms as well, a growing body of data strongly suggests that relative vitamin D deficiency plays a role in several components of the osteoporosis syndrome.

VITAMIN K

Vitamin K was reviewed in 1984 (99) and its role in the synthesis of bone proteins in 1988 (105). Thus the current review focuses mainly on developments since 1988.

Vitamin K is necessary for the gamma-carboxylation of glutamic acid residues in a large number of proteins, and the vitamin K-dependent carboxylase is widely distributed in many tissues. At least seven vitamin K dependent proteins are involved in one way or another in blood coagulation. The gamma-carboxyglutamic acid residues in the peptide chain bind calcium, either free or on the surface layers of crystals, and are thought to function in varying ways—from inhibiting mineralization (as in urine) (95) to serving as osteoclast chemotactic signals (51). Vitamin K deficiency classically produces bleeding disorders, but the liver, where the clotting factors are produced, is highly efficient in extracting vitamin K from the circulation, and gamma-carboxylation declines substantially in other tissues before the deficiency is serious enough to result in bleeding disorders. Thus the bleeding tendencies

that have been the hallmark of vitamin K deficiency may, in fact, be the last manifestation of deficiency. If so, the other clinical manifestations of vitamin K deficiency remain uncertain.

Two vitamin K-dependent proteins are of special interest in the context of this review: osteocalcin [bone Gla protein (BGP)] and a kidney Gla protein (nephrocalcin) (94), both of which are dependent upon vitamin K for their synthesis. BGP is the principal noncollagenous protein of bone. It is synthesized and gamma-carboxylated by osteoblasts as they synthesize bone matrix. Roughly 30% of the synthesized BGP is not incorporated into matrix but is released instead into the circulation, where, like alkaline phosphatase, it can be measured and used as an indicator of bone formation. In vitamin K deficiency, as would occur with coumarin anticoagulants, serum BGP levels decline, and the degree of carboxylation of the circulating BGP protein falls dramatically. Further, binding to hydroxyapatite of the BGP synthesized under these conditions to hydroxyapatite falls precipitously soon after starting anticoagulant therapy. It would seem, therefore, that vitamin K deficiency would have detectable skeletal effects. The problem is that they have been very hard to find. Rats reared and sustained to adult life under near total suppression of BGP gamma-carboxylation show only minor skeletal abnormalities (105). Hauschka et al have suggested that BGP bound to hydroxyapatite is chemotactic for osteoclasts, and that the absence of such binding might impede remodeling (51)—an effect that may not be apparent in the rat, which does not remodel bone the way larger animals do.

Various vitamin K-related abnormalities have been described in association with osteoporosis, but their significance remains unclear. Circulating vitamin K and menaquinone levels are low in hip fracture patients (66). BGP is under-carboxylated in osteoporotics, and this defect responds to physiological doses of vitamin K (102). Finally, Vermeer and colleagues have reported that urine calcium is high in osteoporotics and falls in response to physiological doses of vitamin K (72, 128). In the same subjects, urine hydroxyproline, an indicator of bone resorption, was also high and fell after vitamin K treatment. The urinary effects could plausibly be explained as a defect first in a calcium transport protein, with resulting renal leak of calcium, and a consequent PTH-mediated increase in bone resorption (reflected in the increased hydroxyproline excretion).

Whether or not vitamin K is important for bone health, vitamin K levels are indicators of general nutritional status; the observation of low vitamin K levels in osteoporotics, especially in those with hip fracture, may simply reflect the often poor nutrition of these individuals (47, 113). Clearly, however, much about vitamin K and bone health remains an enigma, and more work must be done before the picture will become clear.

TRACE MINERALS

Several trace minerals, notably zinc, manganese, and copper, are essential metallic cofactors for enzymes involved in synthesis of various bone matrix constituents. In growing animals, diets deficient in these elements produce definite skeletal abnormalities (29, 89). Additionally, zinc deficiency is well known to produce growth retardation and other abnormalities in humans. But it is not known with certainty whether significant deficiencies of these elements occur in previously healthy adults, or at least, if they do, whether such deficiencies contribute to the osteoporosis problem. Copper deficiency is said to be associated with osteoporotic lesions in sheep, cattle, and rats (29, 122). Copper has not been much studied in connection with human osteoporosis, but in one study serum copper levels were negatively correlated with lumbar spine BMD, even after adjusting for body weight and dietary calcium intake (68). Further, in one four-way, randomized intervention trial, copper, as a part of a trace mineral cocktail including also zinc and manganese, slowed bone mineral loss in postmenopausal women, when given either with or without supplemental calcium (123). Most of the effect in this study was shown to be due to the calcium supplement, but there appeared to be a small additional benefit from the extra trace minerals. Since both osteoporotic and age-related bone loss are multifactorial, and since there was no way to select subjects for inclusion on the basis of presumed trace mineral need, the findings of this study, while inconclusive, have to be considered strongly suggestive.

NUTRITION AND HIP FRACTURE

The impact of nutrition on the problem of hip fracture is twofold: in predisposing to fracture and in recovery from and repair of the injury. Fractures in the very old elderly, and particularly hip fractures, are concentrated in institutionalized persons with multiple disabilities (119). The osteoporotic elderly generally have depleted lean body mass and fat mass (113), as well as low circulating values for several key nutritional indicator variables, from serum albumin to ferritin and vitamin A (32). Survival two years after injury is four times higher in patients with serum albumin values above 3.5 g/dl than in patients with values below 3.0 g (16).

Additionally, patients with hip fracture often have low calcium intakes, and dietary calcium earlier in life has been inversely associated with hip fracture risk. Moreover, it is likely that there are other intrinsic abnormalities in the bone at the femoral neck (40). Thus hip fracture occurs in multiply compromised individuals, and the prospect of successfully intervening to reduce risk of fracture has proved daunting.

However, one aspect of the problem may be more amenable to control. The

relative malnutrition of patients suffering hip fracture and coming to hospital for repair may contribute significantly to the often unsatisfactory outcomes for this common fracture (15–20% excess mortality; 50% institutionalization of the survivors). If so, appropriate nutritional treatment could improve outcomes. Exactly such improvement was found by Delmi et al (32) in a randomized trial of a protein-based nutrient supplement given to patients newly hospitalized for hip fracture. Only 26% of unsupplemented individuals had outcomes classified as good at six months after injury, whereas nearly 60% of supplemented individuals had good outcomes. The investigators noted that the hospital diets offered the unsupplemented individuals were nutritionally adequate, but were frequently unconsumed, whereas the investigators saw to the ingestion of the supplement. Subsequent work from this same group both confirmed these dramatic benefits and narrowed the effect of the supplement to its protein content. This is not an isolated observation; others had earlier found qualitatively similar benefit from nutritional supplementation in these patients (10, 69). Thus the consistency of these findings presents a challenge to the medical profession to apply these basic nutritional principles in the management of their patients.

Literature Cited

1. Adams, N. D., Gray, R. W., Lemann, J. Jr. 1979. The calciuria of increased fixed acid production in humans: evidence against a role for parathyroid hormone and 1,25(OH)2-vitamin D. *Calcif. Tissue Int.* 28:233–38
2. Arnaud, C. D., Sanchez, S. D. 1990. The role of calcium in osteoporosis. *Annu. Rev. Nutr.* 10:397–414
3. Avioli, L. V. 1984. Calcium and osteoporosis. *Annu. Rev. Nutr.* 4:471–91
4. Avioli, L. V., Heaney, R. P. 1991. Calcium intake and bone health (editorial). *Calcif. Tissue Int.* 48:221–23
5. Baran, D., Sorensen, A., Grimes, J., Lew, R., Karellas, A., et al. 1990. Dietary modification with dairy products for preventing vertebral bone loss in premenopausal women: a three-year prospectve study. *J. Clin. Endocrinol. Metab.* 70:264–70
6. Barger-Lux, M. J., Heaney, R. P. 1993. Metabolic characteristics of premenopausal women after calcium restriction or calcium supplementation. *J. Clin. Endocrinol. Metab.* 76:103–7
7. Barger-Lux, M. J., Heaney, R. P., Packard, P. T., Lappe, J. M., Recker, R. R. 1992. Nutritional correlates of low calcium intake. *Clinics Appl. Nutr.* 2:39–44
8. Barger-Lux, M. J., Heaney, R. P., Recker, R. R. 1989. Time course of calcium absorption in humans: evidence for a colonic component. *Calcif. Tissue Int.* 44:308–11
9. Barrett-Connor, E. 1989. The RDA for calcium in the elderly: too little, too late (editorial). *Calcif. Tissue Int.* 44:303–7
10. Bastow, M. D., Rawlings, J., Allison, S. P. 1983. Benefits of supplementary tube feeding after fractured neck of femur. *Br. Med. J.* 287:1589–92
11. Beaton, G. H. 1986. Toward harmonization of dietary, biochemical, and clinical assessments: the meanings of nutritional status and requirements. *Nutr. Rev.* 44:349–58
12. Bell, N. H., Epstein, S., Shary, J., Greene, V., Oexmann, M. J., Shaw, S. 1988. Evidence of a probable role for 25-hydroxyvitamin D in the regulation of calcium metabolism. *J. Bone Miner. Res.* 3:489–95
13. Bergman, E. A., Boyungs, J. C., Erickson, M. L. 1990. Comparison of a food frequency questionnaire and a 3-day diet record. *J. Am. Diet. Assoc.* 90:1431–33

14. Bliziotes, M., Yergey, A. L., Nanes, M. S., Muenzer, J., Begley, M. G., et al. 1988. Absent intestinal response to calciferols in hereditary resistance to 1,25-dihydroxyvitamin D; documentation and effective therapy with high dose intravenous infusions. *J. Clin. Endocrinol. Metab.* 66:294–300
15. Bonjour, J.-P., Theintz, G., Buchs, B., Slosman, D., Rizzoli, R. 1991. Critical years and stages of puberty for spinal and femoral bone mass accumulation during adolescence. *J. Clin. Endocrinol. Metab.* 73:555–63
16. Bruyere, A., Rapin, C. H., Dirren, H. 1984. Nutritional blood values in patients with femoral neck fracture. A comparative study. In *Nutrition, Immunity and Illness in the Elderly*, ed. R. K. Chandra, pp. 242–46. London: Pergamon
17. Calvo, M. S., Kumar, R., Heath, H. III. 1988. Elevated secretion and action of serum PTH in young adults consuming high phosphorus, low calcium diets assembled from common foods. *J. Clin. Endocrinol. Metab.* 66:823–29
18. Calvo, M. S., Kumar, R., Heath, H. III. 1990. Persistently elevated PTH secretion and action in young women after four weeks of ingesting high phosphorus, low calcium diets. *J. Clin. Endocrinol. Metab.* 70:1334–40
19. Carroll, M. D., Abraham, S., Dresser, C. M. 1983. Dietary intake source data: US, 1976–80. *Vital & Health Statistics, Ser. 11-NO. 231, DHHS. Publ. No. (PHS) 83-PHS, March.* Washington, DC: Gov. Print. Off.
20. Chan, G. M. 1991. Dietary calcium and bone mineral status of children and adolescents. *Am. J. Dis. Child.* 145:631–34
21. Chan, G. M., Hess, M., Hollis, J., Book, L. S. 1984. Bone mineral status in childhood accidental fractures. *Am. J. Dis. Child.* 138:569–70
22. Chan, G. M., McMurry, M., Westover, K., Engelbert-Fenton, K., Thomas, M. R. 1987. Effects of increased dietary calcium intake upon the calcium and bone mineral status of lactating adolescent and adult women. *Am. J. Clin. Nutr.* 46:319–23
22a. Chapuy, M. C., Arlot, M. E., Duboeuf, F., Brun, J., Crouzet, B., et al. 1992. Vitamin D3 and calcium to prevent hip fractures in elderly women. *New Engl. J. Med.* 327:1637–42
23. Charles, P. 1989. Metabolic bone disease evaluated by a combined calcium balance and tracer kinetic study. *Dan. Med. Bull.* 36:463–79
24. Chevalley, T., Rizzoli, R., Nydegger, V., Slosman, D. O., Rapin, C.-H., et al. 1992. Effects of calcium supplement on femoral bone mineral density and vertebral fracture rate in vitamin D replete elderly patients with and without a recent hip fracture. *J. Bone Miner. Res.* 7:S322. (Abstr.)
25. Chinn, H. I. 1981. Effects of dietary factors on skeletal integrity in adults: calcium, phosphorus, vitamin D, and protein. Report prepared for the Bureau of Foods, Food and Drug Admin. (FDA), Sept. Life Sciences Research Office, FASEB, Bethesda, Md.
26. Consensus Development Conference: prophylaxis and treatment of osteoporosis. 1991. *Am. J. Med.* 90:107–10
27. Cooper, C., Barker, D. J. P., Wickham, C. 1988. Physical activity, muscle strength, and calcium intake in fracture of the proximal femur in Britain. *Br. Med. J.* 297:1443–46
28. Cumming, R. G. 1990. Calcium intake and bone mass: a quantitative review of the evidence. *Calcif. Tissue Int.* 47:194–201
29. Davis, G. K., Mertz, W. 1987. Copper. See Ref. 89, pp. 301–64
30. Dawson-Hughes, B., Dallal, G. E., Krall, E. A., Harris, S., Sokoll, L. J., Falconer, G. 1991. Effect of vitamin D supplementation on wintertime and overall bone loss in healthy postmenopausal women. *Ann. Int. Med.* 115:505–12
31. Dawson-Hughes, B., Dallal, G. E., Krall, E. A., Sadowski, L., Sahyoun, N., Tannenbaum, S. 1990. A controlled trial of the effect of calcium supplementation on bone density in postmenopausal women. *New Engl. J. Med.* 323:878–83
32. Delmi, M., Rapin, C.-H., Bengoa, J.-M., Delmas, P. D., Vasey, H., Bonjour, J.-P. 1990. Dietary supplementation in elderly patients with fractured neck of the femur. *Lancet* 335: 1013–16
33. Dodds, R. A., Emery, R. J. H., Klenerman, L. 1990. Selective depression of metabolic activities in cortical osteoblasts at the site of femoral neck fractures. *Bone* 11:157–61
34. Dure-Smith, B. A., Farley, S. M., Linkhart, S. G., Baylink, D. J. 1992. Evidence that increased bone formation leads to calcium deficiency and peripheral bone loss in fluoride treated patients: correction with 1,25 vitamin D. *J. Bone Miner. Res.* 7:S188 (Abstr.)
35. Eaton, S. B., Konner, M. 1985. Paleolithic nutrition. A consideration of

its nature and current implications. *New Engl. J. Med.* 312:283–89
36. Eaton, S. B., Nelson, D. A. 1991. Calcium in evolutionary perspective. *Am. J. Clin. Nutr.* 54:S281–87
37. Elders, P. J. M., Netelenbos, J. C., Lips, P., Khoe, E., van Ginkel, F. C., et al. 1989. Perimenopausal bone mass and risk factors. *Bone Miner.* 7:289–99
38. Elders, P. J. M., Netelenbos, J. C., Lips, P., van Ginkel, F. C., Khoe, E., et al. 1991. Calcium supplementation reduces vertebral bone loss in perimenopausal women: a controlled trial in 248 women between 46 and 55. *J. Clin. Endocrinol. Metab.* 73: 533–40
39. Ettinger, B., Genant, H. K., Cann, C. E. 1987. Postmenopausal bone loss is prevented by treatment with low-dosage estrogen with calcium. *Ann. Intern. Med.* 106:40–45
40. Eventov, I., Frisch, B., Cohen, Z., Hammel, I. 1991. Osteopenia, hematopoiesis and osseous remodeling in iliac crest and femoral neck biopsies: a prospective study of 102 cases of femoral neck fractures. *Bone* 12:1–6
41. Fehily, A. M., Coles, R. J., Evans, W. D., Elwood, P. C. 1992. Factors affecting bone density in young adults. *Am. J. Clin. Nutr.* 56:579–86
42. Felson, D. T., Sloutskis, D., Anderson, J. J., Anthony, J. M., Kiel, D. P. 1991. Thiazide diuretics and the risk of hip fracture. Results from the Framingham Study. *J. Am. Med. Assoc.* 265:370–73
43. Forbes, R. M., Weingartner, K. E., Parker, H. M., Bell, R. M., Erdman, J. Jr. 1979. Bioavailability to rats of zinc, magnesium and calcium in: casein-, egg- and soy protein-containing diets. *J. Nutr.* 109:1652–60
44. Francis, R. M., Peacock, M., Storer, J. H., Davies, A. E. J., Brown, W. B., Nordin, B. E. C. 1983. Calcium malabsorption in the elderly: the effect of treatment with oral 25-hydroxy-vitamin D3. *Eur. J. Clin. Invest.* 13:391–96
45. Francis, R. M., Peacock, M., Taylor, G. A., Storer, J. H., Nordin, B. E. C. 1984. Calcium malabsorption in elderly women with vertebral fractures: evidence for resistance to the action of vitamin D metabolites on the bowel. *Clin. Sci.* 66:103–7
46. Freudenheim, J. L., Johnson, N. E., Smith, E. L. 1986. Relationships between usual nutrient intake and bone-mineral content of women 35–65 years of age: longitudinal and cross-sectional analysis. *Am. J. Clin. Nutr.* 44:863–76
47. Geinoz, G., Rapin, C. H., Rizzoli, R., Kraemer, R., Buchs, B., et al. 1993. Relationship between bone mineral density and dietary intakes in elderly. *Osteoporosis Int.* In press
48. Gloth, F. M. III, Tobin, J. D., Sherman, S. S., Hollis, B. W. 1991. Is the recommended daily allowance for vitamin D too low for the homebound elderly? *J. Am. Geriatr. Soc.* 39:137–41
49. Hansen, M. A., Overgaard, K., Riis, B. J., Christiansen, C. 1991. Potential risk factors for development of postmenopausal osteoporosis—examined over a 12-year period. *Osteoporosis Int.* 1:95–102
50. Hasling, C., Charles, P., Jensen, F. T., Mosekilde, L. 1990. Calcium metabolism in postmenopausal osteoporosis: the influence of dietary calcium and net absorbed calcium. *J. Bone Miner. Res.* 5:939–46
51. Hauschka, P. V., Lian, J. B., Cole, D. E. C., Gundberg, C. M. 1989. Osteocalcin and matrix gla protein: vitamin K-dependent proteins in bone. *Physiol. Rev.* 69:990–1047
52. Heaney, R. P. 1986. Calcium, bone health, and osteoporosis. In *Bone and Mineral Research, Annual IV*, ed. W. A. Peck, pp. 255–301. Amsterdam: Elsevier Sci.
53. Heaney, R. P. 1989. Osteoporotic fracture space: an hypothesis. *Bone Miner.* 6:1–13
54. Heaney, R. P. 1989. Nutritional factors in bone health in elderly subjects: methodological and contextual problems. *Am. J. Clin. Nutr.* 50:S1181–89
55. Heaney, R. P. 1990. Estrogen-calcium interactions in the postmenopause: a quantitative description. *Bone Miner.* 11:67–84
56. Heaney, R. P. 1992. Calcium intake and bone health in an aging population. *Clin. Appl. Nutr.* 2:10–29
57. Heaney, R. P., Davies, K. M., Recker, R. R., Packard, P. T. 1990. Long-term consistency of nutrient intakes in humans. *J. Nutr.* 120:869–75
58. Heaney, R. P., Gallagher, J. C., Johnston, C. C. Jr., Neer, R., Parfitt, A. M., Whedon, G. D. 1982. Calcium nutrition and bone health in the elderly. *Am. J. Clin. Nutr.* 36:986–1013
59. Heaney, R. P., Recker, R. R. 1982. Effects of nitrogen, phosphorus, and caffeine on calcium balance in women. *J. Lab. Clin. Med.* 99:46–55
60. Heaney, R. P., Recker, R. R. 1986. Distribution of calcium absorption in

middle-aged women. *Am. J. Clin. Nutr.* 43:299–305
61. Heaney, R. P., Recker, R. R., Saville, P. D. 1978. Menopausal changes in calcium balance performance. *J. Lab. Clin. Med.* 92:953–63
62. Heaney, R. P., Weaver, C. M. 1990. Calcium absorption from kale. *Am. J. Clin. Nutr.* 51:656–57
63. Heaney, R. P., Weaver, C. M., Fitzsimmons, M. L. 1991. Soybean phytate content: effect on calcium absorption. *Am. J. Clin. Nutr.* 53:741–44
64. Heikinheimo, R. J., Inkovaara, J. A., Harju, E. J., Haavisto, M. V., Kaarela, R. H., et al. 1992. Annual injection of vitamin D and fractures of aged bones. *Calcif. Tissue Int.* 51:105–10
65. Hirota, T., Nara, M., Ohguri, M., Manago, E., Hirota, K. 1992. Effect of diet and lifestyle on bone mass in Asian young women. *Am. J. Clin. Nutr.* 55:1168–73
66. Hodges, S. J., Pilkington, M. J., Stamp, T. C. B., Catterall, A., Shearer, M. J., et al. 1991. Depressed levels of circulating menaquinones in patients with osteoporotic fractures of the spine and femoral neck. *Bone* 12:387–89
67. Holbrook, T. L., Barrett-Connor, E., Wingard, D. L. 1988. Dietary calcium and risk of hip fracture: 14-year prospective population study. *Lancet* 2: 1046–49
68. Howard, G., Andon, M., Bracker, M., Saltman, P., Strause, L. 1992. Low serum copper, a risk factor additional to low dietary calcium in postmenopausal bone loss. *J. Trace Elements Exp. Med.* 5:23–31
69. Jensen, J. E., Jensen, T. G., Smith, T. K., Johnson, D. A., Dudrick, S. J. 1982. Nutrition in orthopaedic surgery. *J. Bone Joint Surg.* 64A:1263–72
70. Johnston, C. C. Jr., Miller, J. Z., Slemenda, C. W., Reister, T. K., Hui, S., et al. 1992. Calcium supplementation and increases in bone mineral density in children. *New Engl. J. Med.* 327:82–87
71. Kanis, J. A., Passmore, R. 1989. Calcium supplementation of the diet—I and II. *Br. Med. J.* 298:137–40; 205–8
72. Knapen, M. H. J., Hamulyak, K., Vermeer, C. 1989. The effect of vitamin K supplementation on circulating osteocalcin (bone gla protein) and urinary calcium excretion. *Ann. Int. Med.* 111:1001–5
73. Knox, T. A., Kassarjian, Z., Dawson-Hughes, B., Golner, B. B., Dallal, G. E., et al. 1991. Calcium absorption in elderly subjects on high- and low-fiber diets: effect of gastric acidity. *Am. J. Clin. Nutr.* 53:1480–86
74. Krall, E. A., Sahyoun, N., Tannenbaum, S., Dallal, G. E., Dawson-Hughes, B. 1989. Effect of vitamin D intake on seasonal variations in parathyroid hormome secretion in postmenopausal women. *New Engl. J. Med.* 321:1777–83
75. Lacey, J. M., Anderson, J. J. B. 1991. Correlates of cortical bone mass among premenopausal and postmenopausal Japanese women. *J. Bone Miner. Res.* 6:651–59
76. LaCroix, A. Z., Wienpahl, J., White, L. R., Wallace, R. B., Scherr, P. A., et al. 1990. Thiazide diuretic agents and the incidence of hip fracture. *New Engl. J. Med.* 322:286–90
77. Lau, E., Donnan, S., Barker, D. J. P., Cooper, C. 1988. Physical activity and calcium intake in fracture of the proximal femur in Hong Kong. *Br. Med. J.* 297:1441–43
78. Leighton, G., McKinlay, P. L. 1930. Milk consumption and the growth of schoolchildren. Dept. of Health for Scotland H.M.S.O.
79. Lemann, J. Jr., Gray, R. W., Maierhofer, W. J., Cheung, H. S. 1986. The importance of renal net acid excretion as a determinant of fasting urinary calcium excretion. *Kidney Int.* 29:743–46
80. Lloyd, T., Andon, M. B., Rollings, N., Demers, L. M., Eggli, D. F., et al. 1992. The effect of calcium supplementation on total body bone mineral density in adolescent females. *J. Bone Miner. Res.* 7:S136 (Abstr.)
81. Lukert, B., Higgins, J., Stoskopf, M. 1992. Menopausal bone loss is partially regulated by dietary intake of vitamin D. *Calcif. Tissue Int.* 51:173–79
82. Marcus, R. 1987. Calcium intake and skeletal integrity: is there a critical relationship? *J. Nutr.* 117:631–35
83. Matkovic, V. 1991. Calcium metabolism and calcium requirements during skeletal remodeling and consolidation of bone mass. *Am. J. Clin. Nutr.* 54:S245–60
84. Matkovic, V., Fontana, D., Tominac, C., Goel, P., Chesnut, C. H. III. 1990. Factors that influence peak bone mass formation: A study of calcium balance and the inheritance of bone mass in adolescent females. *Am. J. Clin. Nutr.* 52:878–88
85. Matkovic, V., Heaney, R. P. 1992. Calcium balance during human growth. Evidence for threshold behavior. *Am. J. Clin. Nutr.* 55:992–96

86. Matkovic, V., Kostial, K., Simonovic, I., Buzina, R., Brodarec, A., Nordin, B. E. C. 1979. Bone status and fracture rates in two regions of Yugoslavia. *Am. J. Clin. Nutr.* 32:540–49
87. Mazess, R. B., Barden, H. S. 1991. Bone density in premenopausal women: effects of age, dietary intake, physical activity, smoking, and birth-control pills. *Am. J. Clin. Nutr.* 53:132–42
88. McKenna, J. M., Freaney, R., Meade, A., Muldowney, F. P. 1985. Hypovitaminosis D and elevated serum alkaline phosphatase in elderly Irish people. *Am. J. Clin. Nutr.* 41:101–9
89. Mertz, W., ed. 1987. *Trace Elements in Human and Animal Nutrition,* Vol. 1. San Diego: Academic. 5th ed.
90. Deleted in proof
91. Miller, J. Z., Smith, D. L., Flora, L., Peacock, M., Johnston, C. C. Jr. 1989. Calcium absorption in children estimated from single and double stable calcium isotope techniques. *Clin. Chem. Acta* 183:107–13
92. Miller, J. Z., Smith, D. L., Flora, L., Slemenda, C., Jiang, X., Johnston, C. C. Jr. 1988. Calcium absorption from calcium carbonate and a new form of calcium (CCM) in healthy male and female adolescents. *Am. J. Clin. Nutr.* 48:1291–94
93. Musgrave, K. O., Giambalvo, L., Leclerc, H. L., Cook, R. A., Rosen, C. J. 1989. Validation of a quantitative food frequency questionnaire for rapid assessment of dietary calcium intake. *J. Am. Diet. Assoc.* 89:1484–88
94. Nakagawa, Y., Abram, V., Kezdy, F. J., Kaiser, E. T., Coe, F. L. 1983. Purification and characterization of the principal inhibitor of calcium monohydrate crystal growth in human urines. *J. Biol. Chem.* 258:12594–12600
95. Nakagawa, Y., Ahmed, M. A., Hall, S. L., Deganello, S., Coe, F. L. 1987. Isolation from human calcium oxalate renal stones of nephrocalcin, a glycoprotein inhibitor of calcium oxalate crystal growth. *J. Clin. Invest.* 79: 1782–87
96. Nordin, B. E. C., Heaney, R. P. 1990. Calcium supplementation of the diet: justified by present evidence. *Br. Med. J.* 300:1056–60
97. Nordin, B. E. C., Need, A. G., Morris, H. A., Horowitz, M. 1991. Sodium, calcium, and osteoporosis. In *Nutritional Aspects of Osteoporosis, Serono Symp. Publ.* ed. P. Burckhardt, R. P. Heaney, 85:279–95. New York: Raven
98. Nordin, B. E. C., Need, A. G., Morris, H. A., Horowitz, M., Robertson, W. G. 1991. Evidence for a renal calcium leak in postmenopausal women. *J. Clin. Endocrinol. Metab.* 72:401–7
99. Olson, R. E. 1984. The function and metabolism of vitamin K. *Annu. Rev. Nutr.* 4:281–337
100. Parfitt, A. M., Gallagher, J. C., Heaney, R. P., Johnston, C. C. Jr., Neer, R., Whedon, G. E. 1982. Vitamin D and bone health in the elderly. *Am. J. Clin. Nutr.* 36:1014–31
101. Pilch, S. M. 1987. Physiological effects and health consequences of dietary fiber. Prepared for Center for Food Safety Appl. Nutr. FDA, Dept. Health Hum. Serv., Washington, DC. LSRO, FASEB
102. Plantalech, L. C., Chapuy, M. C., Guillaumont, M., Chapuy, P., Leclerq, M., Delmas, P. D. 1990. Impaired carboxylation of serum osteocalcin in elderly women: Effect of vitamin K1. In *Osteoporosis 1990,* ed. C. Christiansen, K. Overgaard, pp. 345–47. Copenhagen: Osteopress
103. Portale, A. A., Halloran, B. P., Morris, R. C. Jr. 1989. Physiologic regulation of the serum concentration of 1,25-dihydroxyvitamin D by phosphorus in normal man. *J. Clin. Invest.* 83:1494–99
104. Prentice, A., Shaw, J., Laskey, M. A., Cole, T. J., Fraser, D. R. 1991. Bone mineral content of British and rural Gambian women aged 18–80 + years. *Bone Miner* 12:201–14
105. Price, P. A. 1988. Role of vitamin-K-dependent proteins in bone metabolism. *Annu. Rev. Nutr.* 8:565–83
106. Prince, R. L., Smith, M., Dick, I.M., Price, R. I., Webb, P. G., et al. 1991. Prevention of postmenopausal osteoporosis. *New Engl. J. Med.* 325:1189–95
107. Ray, W. A., Griffin, M. R., Downey, W., Melton, L. J. III. 1989. Long-term use of thiazide diuretics and risk of hip fracture. *Lancet* 1:687–90
108. Recker, R. R., Davies, K. M., Hinders, S. M., Heaney, R. P., Stegman, M. R., Kimmel, D. B. 1992. Bone gain in young adult women. *J. Am. Med. Assoc.* 268:2403–8
109. Recker, R. R., Heaney, R. P. 1990. Age-related bone loss (editorial). *J. Bone Miner. Res.* 4:795–96
110. Recker, R. R., Lappe, J. M., Davies, K. M., Kimmel, D. B. 1992. Change in bone mass immediately before menopause. *J. Bone Miner. Res.* 7:857–62
111. *Recommended Dietary Allowances.* 1989. Washington, DC: Natl. Acad. Press. 10th ed.
112. Reid, I. R., Ames, R. W., Evans, M.

C., Gamble, G. D., Sharpe, S. J. 1993. Effect of calcium supplementation on bone loss in late postmenopausal women. *New Engl. J. Med.* 328: In press
113. Rico, H., Revilla, M., Villa, L. F., Hernandez, E. R., Fernandez, J. P. 1992. Crush fracture syndrome in senile osteoporosis: a nutritional consequence. *J. Bone Miner. Res.* 7:317–19
114. Riggs, B. L., Wahner, H. W., Melton, L. J. III, Richelson, L. S., Judd, H. L., O'Fallon, W. M. 1987. Dietary calcium intake and rates of bone loss in women. *J. Clin. Invest.* 80:979–82
115. Riis, B. J., Nilas, L., Christiansen, C. 1987. Does calcium potentiate the effect of estrogen therapy on postmenopausal bone loss? *Bone Miner.* 2:1–9
116. Riis, B. J., Thomsen, K., Christiansen, C. 1987. Does calcium supplementation prevent postmenopausal bone loss? *New Engl. J. Med.* 316:173–77
117. Robinovitch, S. N., Hayes, W. C., McMahon, T. A. 1991. Prediction of femoral impact forces in falls on the hip. *J. Biomech. Eng.* 113:366–74
118. Silverberg, S. J., Shane, E., Clemens, T. L., Dempster, D. W., Segre, G. V., et al. 1986. The effect of oral phosphate administration on major indices of skeletal metabolism in normal subjects. *J. Bone Miner. Res.* 1:383–88
119. Simonen, O. 1991. Incidence of femoral neck fractures: senile osteoporosis in Finland in the years 1970–1985. *Calcif. Tissue Int.* 49:S8–S10
120. Slovik, D. M., Adams, J. S., Neer, R. M., Holick, M. F., Potts, J. T. Jr. 1981. Deficient production of 1,25-dihydroxyvitamin D in elderly osteoporotic patients. *New Engl. J. Med.* 305:372–74
121. Stevenson, J. C., Whitehead, M. I., Padwick, M., Endacott, J. A., Sutton, C., et al. 1988. Dietary intake of calcium and postmenopausal bone loss. *Br. Med. J.* 297:15–17
122. Strain, J. J. 1988. A reassessment of diet and osteoporosis—possible role for copper. *Med. Hypotheses* 27:333–38
123. Strause, L., Saltman, P., Smith, K., Andon, M. 1991. The role of trace elements in bone metabolism. See Ref. 97, pp. 223–33
124. Suter, P. M., Russell, R. M. 1987. Vitamin requirements of the elderly. *Am. J. Clin. Nutr.* 45:501–12
125. Tylavsky, F. A., Anderson, J. J. B. 1988. Dietary factors in bone health of elderly lactoovovegetarian and omnivorous women. *Am. J. Clin. Nutr.* 48:842–49
126. van Beresteijn, E. C. H., van't Hof, M. A., Schaafsma, G., de Waard, H., Duursma, S. A. 1990. Habitual dietary calcium intake and cortical bone loss in perimenopausal women: a longitudinal study. *Calcif. Tissue Int.* 47:338–44
127. van Beresteijn, E. C. H., van't Hof, M. A., de Waard, H., Raymakers, J. A., Duursma, S. A. 1990. Relation of axial bone mass to habitual calcium intake and to cortical bone loss in healthy early postmenopausal women. *Bone* 11:7–13
128. Vermeer, C., Hamulyak, K. 1991. Pathophysiology of vitamin K-deficiency and oral anticoagulants. *Thromb. Haemostas.* 66:153–59
129. Weaver, C. M., Heaney, R. P., Martin, B. R., Fitzsimmons, M. L. 1991. Human calcium absorption from whole wheat products. *J. Nutr.* 121:1769–75
130. Wickham, C. A. C., Walsh, K., Cooper, C., Barker, D. J. P., Margetts, B. M., et al. 1989. Dietary calcium, physical activity, and risk of hip fracture: a prospective study. *Br. Med. J.* 299:889–92
131. Wisker, E., Nagel, R., Tanudjaja, T. K., Feldheim, W. 1991. Calcium, magnesium, zinc, and iron balances in young women: effects of a low-phytate barley-fiber concentrate. *Am. J. Clin. Nutr.* 54:553–59
132. Wood, R. J., Sitrin, M. D., Rosenberg, I. H. 1984. Calciuria in total parenteral nutrition: effects of amino acids and glucose in rats. *Am. J. Clin. Nutr.* 40:101–6
133. Wood, R. J., Sitrin, M. D., Rosenberg, I. H. 1988. Effect of phosphorus on endogenous calcium losses during total parenteral nutrition. *Am. J. Clin. Nutr.* 48:632–36