

Chapter 11

Calcium

It is nearly 30 years since the last FAO/WHO recommendations on calcium intake were published in 1974 (1) and nearly 40 years since the experts' meeting in Rome (2) on which these recommendations were based. During this generation gap, a paradigm shift has occurred with respect to the involvement of calcium in the aetiology of osteoporosis. The previous reports were written against the background of the Albright paradigm (3), according to which osteomalacia and rickets were due to calcium deficiency, vitamin D deficiency, or both, whereas osteoporosis was attributed to failure of new bone formation secondary to negative nitrogen balance, osteoblast insufficiency, or both. The rediscovery of earlier information that calcium deficiency led to the development of osteoporosis (not rickets and osteomalacia) in experimental animals (4) resulted in a reexamination of osteoporosis in humans, notably in postmenopausal women. This reexamination yielded evidence in the late 1960s that menopausal bone loss was not due to a decrease in bone formation but rather to an increase in bone resorption (5-8), and this has had a profound effect on our understanding of other forms of osteoporosis. Although reduced bone formation may aggravate the bone loss process in elderly people (9) and probably plays a major role in corticosteroid osteoporosis (10) – and possibly in osteoporosis in men (11) – bone resorption is increasingly held responsible for osteoporosis in women and for the bone deficit associated with hip fractures in elderly people of both sexes (12). Because bone resorption is also the mechanism whereby calcium deficiency destroys bone, it is hardly surprising that the role of calcium in the pathogenesis of osteoporosis has received increasing attention and that recommended calcium intakes have risen steadily in the past 35 years from the nadir which followed the publication of the report from Rome in 1962 (13). The process has been accelerated by the growing realisation that insensible losses of calcium (via skin, hair, nails, etc.) need to be taken into account in the calculation of calcium requirement.

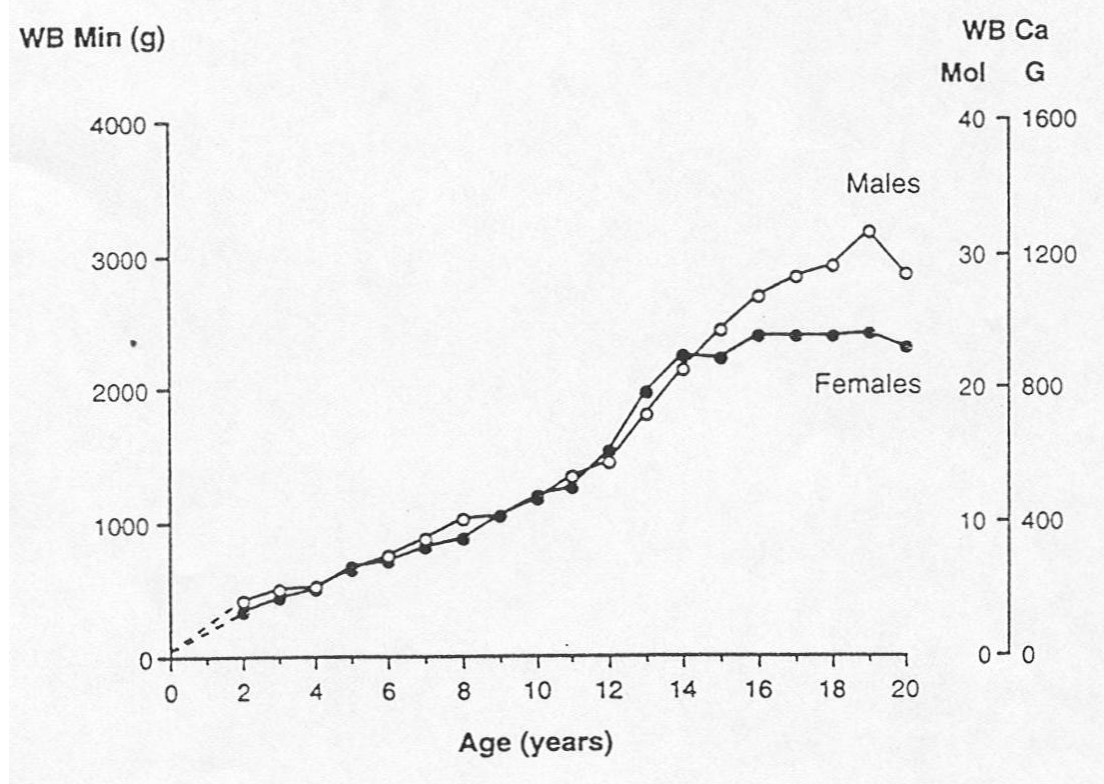
As the calcium allowances recommended for developed nations have been rising – and may still not have reached their peak – the gap between them and the actual calcium intakes in developing countries has widened. The concept that calcium requirement may itself vary from culture to culture for dietary, genetic, lifestyle, and geographical reasons is emerging. This report therefore seeks to make it clear that our main recommendations – like the latest recommendations from USA and Canada (14), Great Britain (15), the European Union (16), and Australia (17) – are largely based on data derived from the developed world and are not necessarily applicable to nations with different dietary cultures, different lifestyles, and different environments for which different calculations may be indicated.

Chemistry and distribution of calcium

Calcium is a divalent cation with an atomic weight of 40. In the elementary composition of the human body, it ranks fifth after oxygen, carbon, hydrogen, and nitrogen, and it makes up 1.9 percent of the body by weight (18). Carcass analyses show that it constitutes 0.1–0.2 percent of early foetal fat-free weight, rising to about 2 percent of adult fat-free weight. In absolute terms, this represents a rise from about 24 g (600 mmol) at birth to 1300 g (32.5 mol) at maturity, requiring an average daily positive calcium balance of 180 mg (4.5 mmol) during the 20 years of growth (*Figure 12*).

Figure 12

Whole-body bone mineral (WB Min) (left axis) and calcium (right axis) as a function of age as determined by total-body dual-energy X-ray absorptiometry



Note: Data supplied by Dr Zanchetta, IDIM, Buenos Aires, Argentina).

Ninety-nine percent of the body calcium is located in the skeleton. The remaining 1 percent is equally distributed between the teeth and soft tissues, with only 0.1 percent in the extracellular fluid (ECF). In the skeleton it constitutes 25 percent of the dry weight and 40 percent of the ash weight. The ECF contains ionised calcium at about 4.8 mg/100 ml (1.20 mmol/l) maintained by the parathyroid – vitamin D system as well as complexed calcium at about 1.6 mg/100 ml (0.4 mmol/l). In the plasma there is an additional protein-bound calcium fraction of 3.2 mg/100 ml (0.8 mmol/l). In the cellular compartment the total calcium concentration is comparable with that in the ECF, but the free calcium concentration is lower by several orders of magnitude (19).

Biological role of calcium

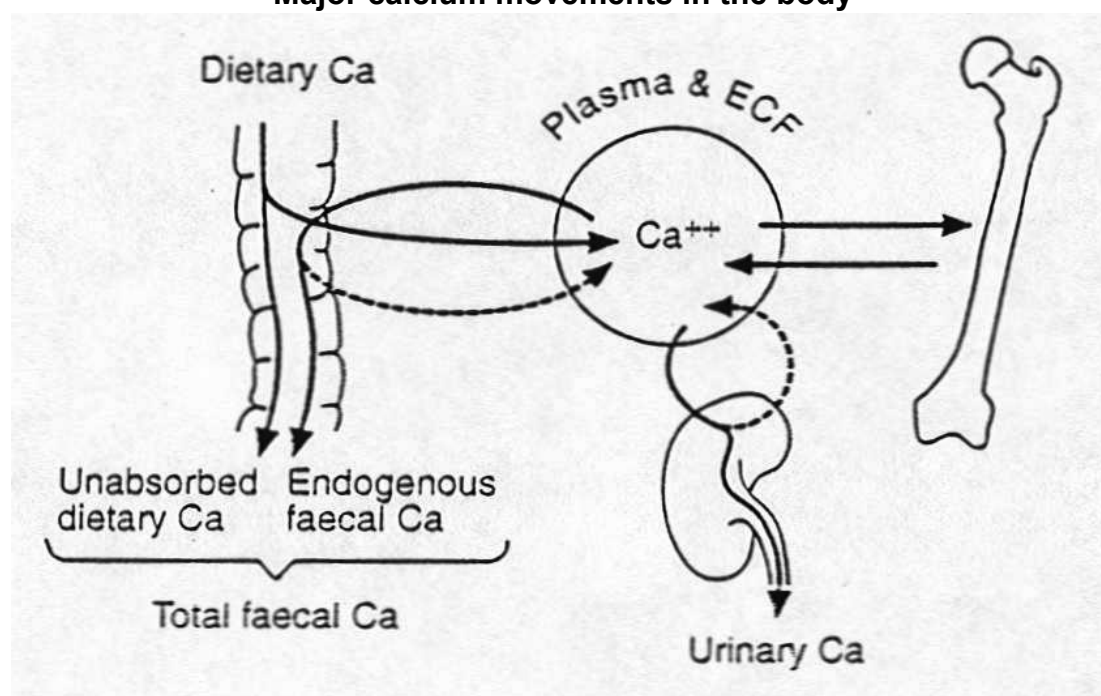
Calcium salts provide rigidity to the skeleton and calcium ions play a role in many if not most metabolic processes. In the primitive exoskeleton and in shells, rigidity is generally provided by calcium carbonate, but in the vertebrate skeleton it is provided by a form of calcium phosphate which approximates hydroxyapatite $[\text{Ca}_{10}(\text{OH})_2(\text{PO}_4)_6]$ and is embedded in collagen fibrils.

Bone mineral serves as the ultimate reservoir for the calcium circulating in the ECF. Calcium enters the ECF from the gut by absorption and from bone by resorption. Calcium leaves the ECF via the gastrointestinal tract, kidneys, and skin and enters into bone via bone formation (**Figure 13**). In addition, calcium fluxes occur across all cell membranes. Many neuromuscular and other cellular functions depend on the maintenance of the ionised calcium concentration in the ECF. Calcium fluxes are also important mediators of hormonal effects on target organs through several intracellular signalling pathways, such as the phosphoinositide and cyclic adenosine monophosphate systems. The cytoplasmic calcium concentration is kept

down by a series of calcium pumps, which concentrate calcium within the intracellular storage sites or extrude from the cells the calcium which flows in by diffusion. The physiology of calcium metabolism is primarily directed towards the maintenance of the concentration of ionised calcium in the ECF. This is protected and maintained by a feedback loop through calcium receptors in the parathyroid glands (20), which control the secretion of parathyroid hormone (see **Figure 10** of Chapter 8). This hormone increases the renal tubular reabsorption of calcium, promotes intestinal calcium absorption by stimulating the renal production of 1,25-dihydroxycoleciferol [$1,25(\text{OH})_2\text{D}$], and, if necessary, resorbs bone. However, the integrity of the system depends critically on vitamin D status; if there is a deficiency of vitamin D, the loss of its calcaemic action (21) leads to a decrease in the ionised calcium and secondary hyperparathyroidism and hypophosphataemia. This is why experimental vitamin D deficiency results in rickets and osteomalacia whereas calcium deficiency gives rise to osteoporosis (4,22).

Figure 13

Major calcium movements in the body



Determinants of calcium balance

Calcium intake

In a strictly operational sense, calcium balance is determined by the relationship between calcium intake and calcium absorption and excretion. A striking feature of the system is that relatively small changes in calcium absorption and excretion can neutralise a high intake or compensate for a low one. There is a wide variation in calcium intake among nations, generally following the animal protein intake and depending largely on dairy product consumption. The lowest calcium intakes are in developing countries, particularly in Asia, and the highest are in developed countries, particularly in USA, Canada and Europe (**Table 30**).

Table 30**Protein and calcium intakes in different regions of the world 1987–1989**

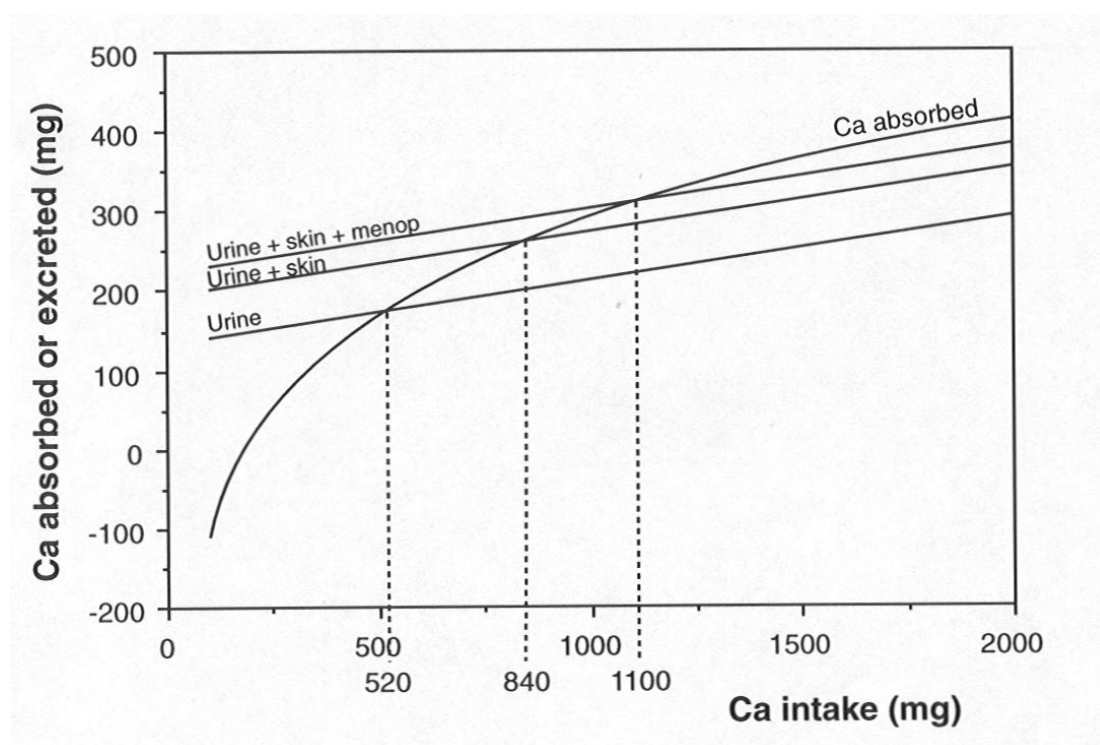
Region	Total	Protein (g)		Total	Calcium (mg)	
		Animal	Vegetable		Animal	Vegetable
USA and Canada	108.7	72.2	36.5	1031	717	314
Europe	102.0	59.6	42.4	896	684	212
Oceania	98.3	66.5	31.8	836	603	233
Other developed	91.1	47.3	43.8	565	314	251
USSR	106.2	56.1	50.1	751	567	184
All developed	103.0	60.1	42.9	850	617	233
Africa	54.1	10.6	43.5	368	108	260
Latin America	66.8	28.6	38.2	476	305	171
Near East	78.7	18.0	60.7	484	223	261
Far East	58.2	11.0	47.2	305	109	196
Other developing	55.8	22.7	33.1	432	140	292
All developing	59.9	13.3	46.6	344	138	206

Source: Adapted from FAO Yearbook, 1990 (23).

Calcium absorption

Ingested calcium mixes with digestive juice calcium in the proximal small intestine from where it is absorbed by a process, which has an active saturable component and a diffusion component (24-27). At low calcium intakes calcium is mainly absorbed by active (transcellular) transport, but at higher intakes an increasing proportion of calcium is absorbed by simple (paracellular) diffusion. The unabsorbed component appears in the faeces together with the unabsorbed component of digestive juice calcium known as endogenous faecal calcium. Thus, the faeces contain unabsorbed dietary calcium and unreabsorbed digestive juice calcium (**Figure 14**).

True absorbed calcium is the total calcium absorbed from the calcium pool in the intestines and therefore contains both dietary and digestive juice components. Net absorbed calcium is the difference between dietary calcium and faecal calcium and is numerically the same as true absorbed calcium minus endogenous faecal calcium. At zero calcium intake, all the faecal calcium is endogenous and represents the digestive juice calcium which has not been reabsorbed; net absorbed calcium at this intake is therefore negative to the extent of about 200 mg (5 mmol) (28,29). When the intake reaches about 200 mg (5 mmol), dietary and faecal calcium become equal and net absorbed calcium is zero. As calcium intake increases, net absorbed calcium also increases, steeply at first but then, as the active transport becomes saturated, more slowly until the slope of absorbed on ingested calcium approaches linearity with an ultimate gradient of about 5–10 percent (24,25,30,31). The relationship between intestinal calcium absorption and calcium intake, derived from 210 balance studies performed in 81 individuals collected from the literature (32-39), is shown in **Figure 14**.

Figure 14**Relationship between calcium absorption and calcium intake**

The relationships between calcium intake and calcium absorbed and excreted calcium calculated from 210 balance experiments in 81 subjects (32-39). Equilibrium is reached at an intake of 520 mg, which rises to 840 mg when skin losses of 60 mg are added and to 1100 mg when menopausal loss is included. The curvilinear relationship between intestinal calcium absorption and calcium intake can be made linear by using the logarithm of calcium intake to yield the equation: $C_{aa} = 174 \log_e C_{ai} - 909 \pm 71$ (SD) mg/day, where C_{ai} represents ingested calcium and C_{aa} net absorbed calcium. The relationship between urinary calcium excretion and calcium intake is given by the equation: $C_{au} = 0.078 C_{ai} + 137 \pm 11.2$ (SD) mg/day, where C_{au} is urinary calcium and C_{ai} calcium intake.

True absorption is an inverse function of calcium intake, falling from some 70 percent at very low intakes to about 35 percent at high intakes (**Figure 15**). Percent net absorption is negative at low intakes, becomes positive as intake increases, reaches a peak of about 30 percent at an intake of about 400 mg, and then falls off as the intake increases. The two lines converge as intake rises because the endogenous faecal component (which separates them) becomes proportionately smaller.

Many factors influence the availability of calcium for absorption and the absorptive mechanism itself. The former includes substances, which form insoluble complexes with calcium, such as the phosphate ion. The relatively high calcium-phosphate ratio of 2.2 in human milk compared with 0.77 in cow milk (18) may be a factor in the higher absorption of calcium from human milk than cow milk (see below).

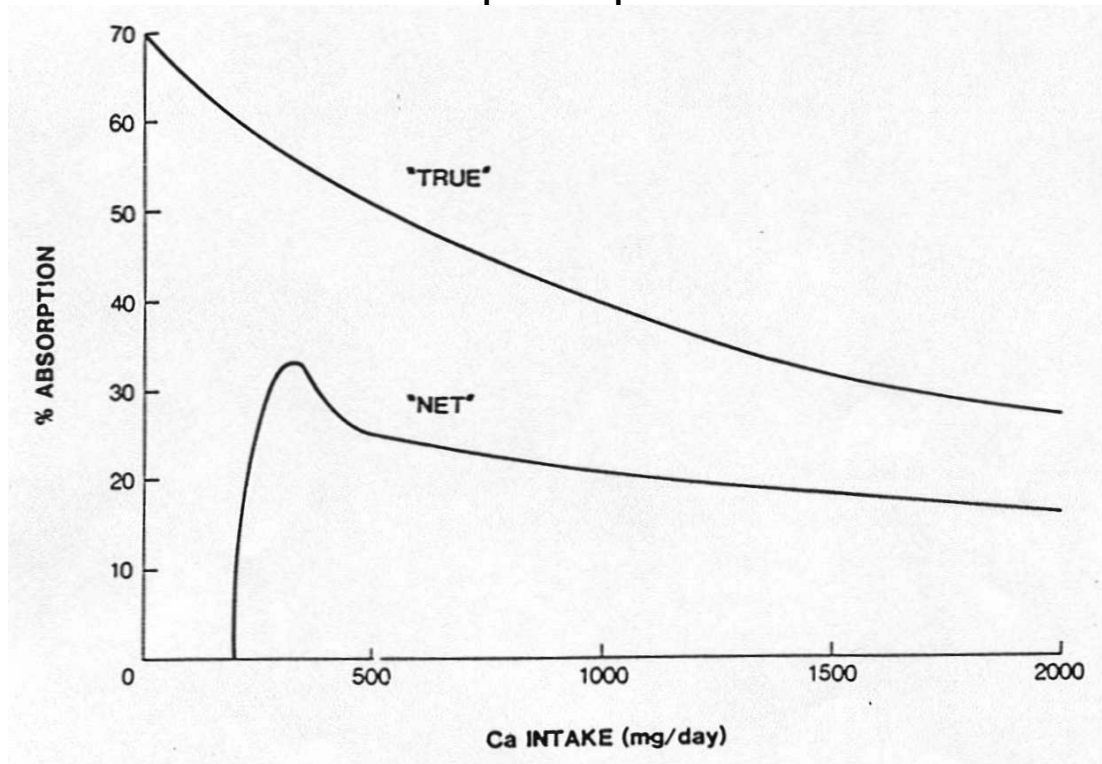
Intestinal calcium absorption is mainly controlled by the serum concentration of $1,25(\text{OH})_2\text{D}$ (see **Chapter 8**). The activity of the $1-\alpha$ -hydroxylase, which catalyses $1,25(\text{OH})_2\text{D}$ production from 25-hydroxycolecalciferol (25OHD) in the kidneys, is negatively related to the plasma calcium and phosphate concentrations and positively to plasma parathyroid hormone (21). Thus the inverse relationship between calcium intake and

fractional absorption described above is enhanced by the inverse relationship between dietary calcium and serum $1,25(\text{OH})_2\text{D}$ (21,40,41).

Phytates, present in the husks of many cereals as well as in nuts, seeds, and legumes, can form insoluble calcium phytate salts in the gastrointestinal tract. Excess oxalates can precipitate calcium in the bowel but are not an important factor in most diets.

Figure 15

True and net calcium absorption as percents of calcium intake



Note: The great differences between these functions at low calcium intakes and their progressive convergence as calcium intake increases.

Urinary calcium

Urinary calcium is the fraction of the filtered plasma water calcium, which is not reabsorbed in the renal tubules. At a normal glomerular filtration rate of 120 ml/min and ultrafiltrable calcium of 6.4 mg/100 ml (1.60 mmol/l), the filtered load of calcium is about 8 mg/min (0.20 mmol/min) or 11.6 g/day (290 mmol/day). Because the usual 24-hour calcium excretion in developed countries is about 160–200 mg (4–5 mmol), it follows that 98–99 percent of the filtered calcium is usually reabsorbed in the renal tubules. However, calcium excretion is extremely sensitive to changes in filtered load. A decrease in plasma water calcium of only 0.17 mg/100 ml (0.043 mmol/l), which is barely detectable, was sufficient to account for a decrease in urinary calcium of 63 mg (1.51 mmol) when 27 subjects changed from a normal- to a low-calcium diet (42). This very sensitive renal response to calcium deprivation combines with the inverse relationship between calcium intake and absorption to stabilise the plasma ionised calcium concentration and to preserve the equilibrium between calcium entering and leaving the ECF over a wide range of calcium intakes. However, there is always a significant obligatory loss of calcium in the urine (as there is in the faeces), even on a low calcium intake, simply because maintenance of the plasma ionised calcium and, therefore, of the filtered load, prevents total elimination of the calcium from the urine. The lower limit for urinary calcium in developed countries is about 140 mg (3.5 mmol) but

depends on protein and salt intakes. From this obligatory minimum, urinary calcium increases on intake with a slope of about 5–10 percent (30,31,43). In the graph derived from 210 balance studies referred to above (**Figure 14**), the relationship between urinary calcium excretion and calcium intake is represented by the line which intersects the absorbed calcium line at an intake of 520 mg.

Insensible losses

Urinary and endogenous faecal calcium are not the only forms of excreted calcium; losses through skin, hair, and nails need to be taken into account. These are not easily measured, but a combined balance and isotope procedure has yielded estimates of daily insensible calcium losses in the range of 40–80 mg (1–2 mmol), which are unrelated to calcium intake (44,45). The addition of a loss of 60 mg (1.5 mmol) as a constant to urinary calcium loss raises the dietary calcium at which absorbed and excreted calcium reach equilibrium from 520 to 840 mg (13 to 21 mmol) (**Figure 14**).

Calcium requirements and recommended intakes

Methodology

Although it is well established that calcium deficiency causes osteoporosis in experimental animals, the contribution that calcium deficiency makes to osteoporosis in humans is much more controversial, not least because of the great variation in calcium intakes across the world (**Table 30**), which does not appear to be associated with any corresponding variation in the prevalence of osteoporosis. This issue is dealt with at greater length below in the section on nutritional factors; in this section we will simply define what is meant by calcium requirement and how it may be calculated.

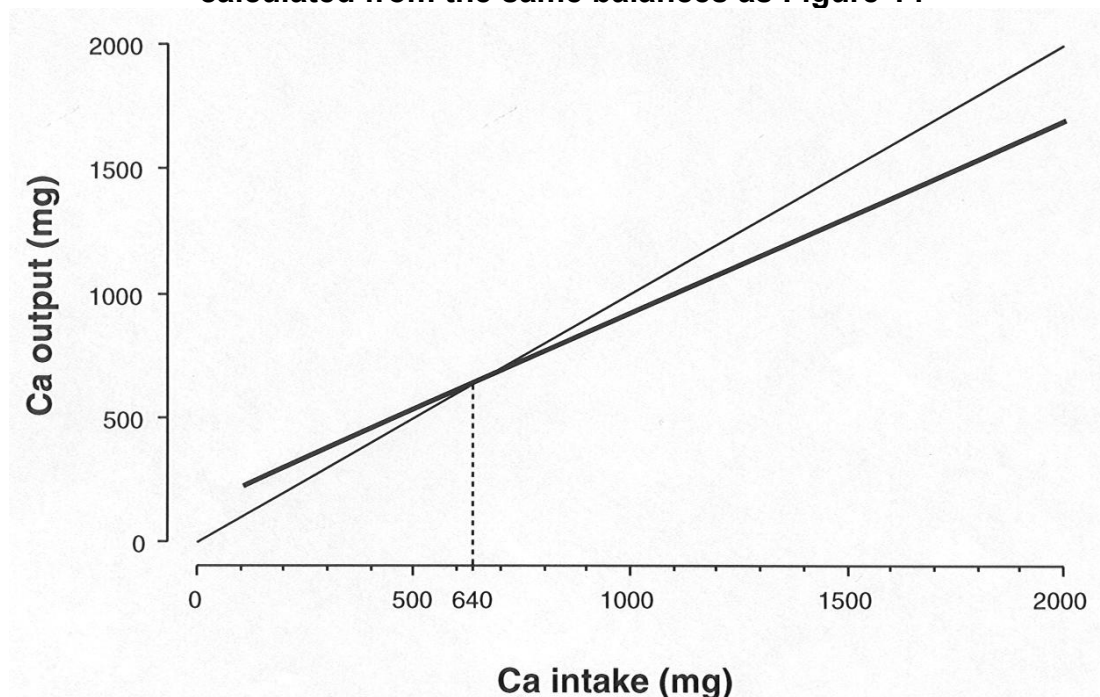
The calcium requirement of an adult is generally recognised to be the intake required to maintain calcium balance and therefore skeletal integrity. The mean calcium requirement of adults is therefore the mean intake at which intake and output are equal, which at present can only be determined by balance studies conducted with sufficient care and over a sufficiently long period to ensure reasonable accuracy and then corrected for insensible losses. The reputation of the balance technique has been harmed by a few studies with inadequate equilibration times and short collection periods, but this should not be allowed to detract from the value of the meticulous work of those who have collected faecal and urinary samples for weeks or months from subjects on well-defined diets. This meticulous work has produced valuable balance data, which are clearly valid; the mean duration of the balances in the 210 studies from eight publications used in this report was 90 days with a range of 6–480 days. (The four 6-day balances in the series used a non-absorbable marker and are therefore acceptable.)

The usual way of determining mean calcium requirement from balance studies has been by linear regression of calcium output (or calcium balance) on intake and calculation of the mean intake at which intake and output are equal (or balance is zero). This was probably first done in 1939 by Mitchell and Curzon (46), who arrived at a mean requirement of 9.8 mg/kg/day or about 640 mg (16 mmol) at a mean body weight of 65 kg. The same type of calculation was subsequently used by many other workers who arrived at requirements ranging from 200 mg/day (5 mmol/day) in male Peruvian prisoners (47) to 990 mg (24.75 mmol) in premenopausal women (48), but most values were about 600 mg (15 mmol) (31) without allowing for insensible losses. However, this type of simple linear regression yields a higher mean calcium requirement (640 mg in the same 210 balances) (**Figure 16a**) than the intercept of absorbed and excreted calcium (520 mg) (**Figure 14**) because it tends to underestimate the negative calcium balance at low intake and overestimate the positive balance at high intake. A better reflection of biological reality is obtained by deriving calcium

output from the functions given in the previous section and then regressing that output on calcium intake. This yields the result shown in **Figure 16b** where the negative balance is more severe at low intakes and less positive at high intakes than in the linear model and in which zero balance occurs at 520 mg as in **Figure 14**.

Figure 16a

**Calcium output as a linear (A) function of calcium intake
calculated from the same balances as Figure 14**



Note: The regression line crosses the line of equality at an intake of 640 mg.
The equation is: $Ca_o = 0.779 Ca_i + 142$ mg where Ca_o is calcium output.

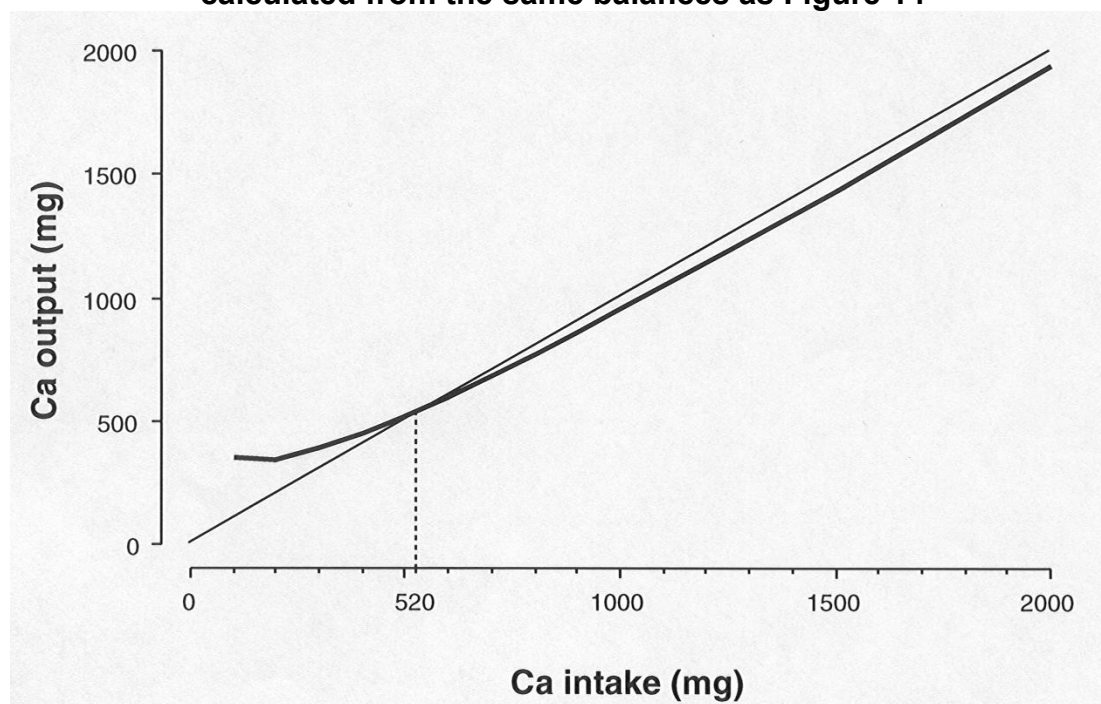
An alternative way of calculating calcium requirement is to determine the intake at which the mean maximum positive balance occurs. This has been done with a two-component, split, linear regression model in which calcium balance is regressed on intake to determine the threshold intake above which no further increase in calcium retention occurs (49). This may well be an appropriate way of calculating the calcium requirement of children and adolescents (and perhaps pregnant and lactating women) who need to be in positive calcium balance and in whom the difference between calcium intake and output is therefore relatively large and measurable by the balance technique. However, in normal adults the difference between calcium intake and output at high calcium intakes represents a very small difference between two large numbers, and this calculation therefore carries too great an error to calculate their requirement.

We are inclined to think that the most satisfactory way of calculating calcium requirement from current data is as the intake at which excreted calcium equals net absorbed calcium, which has the advantage of permitting separate analysis of the effects of changes in calcium absorption and excretion. This intercept has been shown in **Figure 14** to occur at an intake of about 520 mg, but when insensible losses of calcium of 60 mg (1.5 mmol) (44,45) are taken into account, the intercept rises to 840 mg, which we believe is as close as it is possible to get at present to the calcium requirement of adults on Western-style diets. The addition to this excretion line of an additional obligatory urinary calcium of 30 mg (0.75 mmol) at menopause (50) raises the amount to about 1100 mg, which we suggest is the mean calcium requirement of postmenopausal women (see below). However, this type of

calculation cannot easily be applied to other high-risk populations (such as children) because there are not sufficient published data from these groups to permit a similar analysis of the relationship among calcium intake, absorption, and excretion. An alternative is to estimate how much calcium each population group needs to absorb to meet obligatory calcium losses and desirable calcium retention and then to calculate the intake required to provide this rate of calcium absorption. This is what has been done in the following section.

Figure 16b

**Calcium output as a non-linear function of calcium intake
calculated from the same balances as Figure 14**



Note: The regression line crosses the line of equality at an intake of 520 mg.
The equation is: $Ca_o = Ca_i - 174 \text{ Log}_e Ca_i - 909 + 0.078 Ca_u + 137 \text{ mg}$.

Populations at risk

It is clear from **Figure 12** that a positive calcium balance (i.e., net calcium retention) is required throughout growth, particularly during the first 2 years of life and during puberty and adolescence. These age groups therefore constitute populations at risk for calcium deficiency, as are pregnant women (especially in the last trimester), lactating women, postmenopausal women, and, possibly, elderly men. Our calculations for these groups, ultimately derived from Western European and North American data, are given below.

Recommendations by group

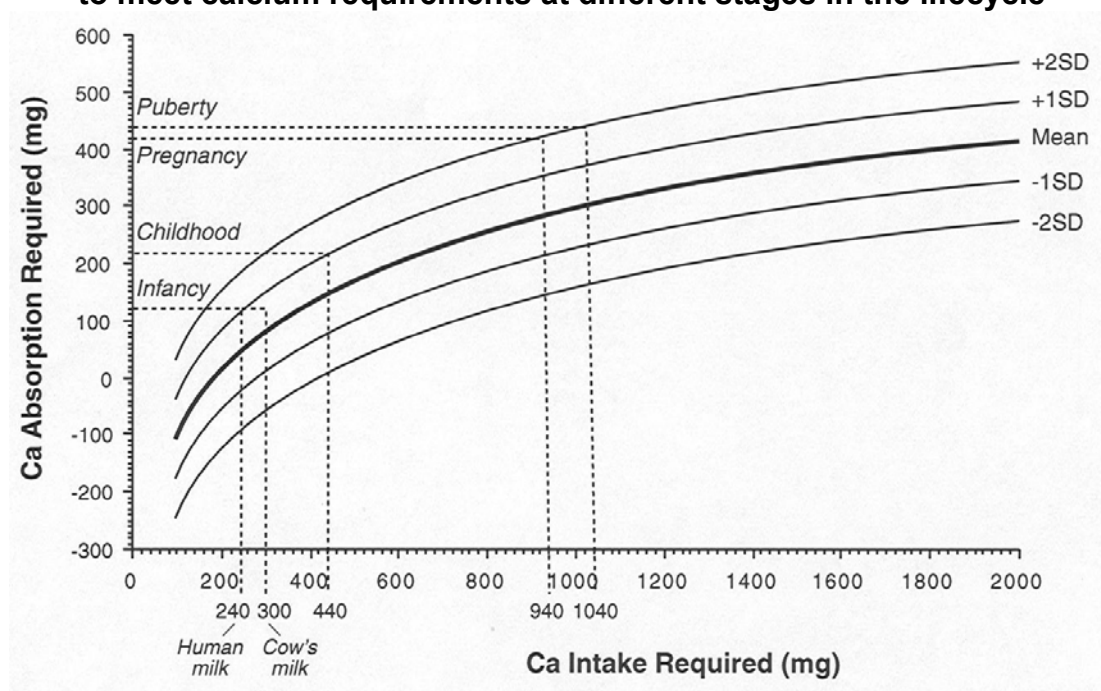
Infancy

In the first 2 years of life, the daily calcium increment in the skeleton is about 100 mg (2.5 mmol) (51). The urinary calcium of infants is about 10 mg/day (0.25 mmol/day) and is virtually independent of intake (52-56) and insensible losses are perhaps the same. Therefore, infants need to absorb some 120 mg (3 mmol) of calcium daily to allow for normal growth. What this represents in dietary terms can be calculated from calcium absorption studies in newborn infants (52-56), which suggest that the absorption of calcium from cow milk by

infants is about 0.5 SD above the normal adult slope and from human milk is more than 1 SD above the normal adult slope. If this information is correct, different recommendations need to be made for infants depending on milk source. With human milk, an absorption of 120 mg (3 mmol) of calcium requires a mean intake of 240 mg (6 mmol) (**Figure 17**) and a recommended intake of say 300 mg (7.5 mmol), which is close to the amount provided in the average daily milk production of 750 ml. With cow milk, calcium intake needs to be about 300 mg (7.5 mmol) to meet the requirement (**Figure 17**) and the allowance should be 400 mg (10 mmol) (**Table 31**).

Figure 17

Calcium intakes required to provide the absorbed calcium necessary to meet calcium requirements at different stages in the lifecycle



Note: The solid lines represent the mean and range of calcium absorption as a function of calcium intake derived from the equation in **Figure 14**. The interrupted lines represent the estimated calcium requirements based on Western European and North American data.

Childhood

The accumulation of whole-body calcium with skeletal growth is illustrated in **Figure 12**. It rises from about 120 g (3 mol) at age 2 years to 400 g (10 mol) at age 9 years. These values can be converted into a daily rate of calcium accumulation from ages 2 to 9 of about 120 mg (3 mmol), which is very similar to the amount calculated by Leitch and Aitken (57) from growth analyses. Although urinary calcium must rise with the growth-related rise in glomerular filtration rate, a reasonable estimate of the mean value from ages 2 to 9 might be 60 mg (1.5 mmol) (58). When this is added to a daily skeletal increment of 120 mg (3 mmol) and a dermal loss of perhaps 40 mg (1.0 mmol), the average daily net absorbed calcium needs to be 220 mg (5.5 mmol) during this period. If the net absorption of calcium by children is 1 SD above that of adults, the average daily requirement during this period is about 440 mg (11 mmol) (**Figure 17**) and the average recommended intake is 600 mg (15 mmol) – somewhat lower in the earlier years and somewhat higher in the later years (**Table 31**).

Table 31**Recommended calcium allowances based on Western European,
American and Canadian data**

Group	Recommended intake mg/day
Infants and children	
0–6 months	
Human milk	300
Cow milk	400
7–12 months	400
1–3 years	500
4–6 years	600
7–9 years	700
Adolescents, 10–18 years	1300 ^a
Adults	
Females	
19 years to menopause	1000
Postmenopause	1300
Males	
19–65 years	1000
65 +	1300
Pregnancy (last trimester)	1200
Lactation	1000

^a Particularly during the growth spurt.

Puberty and adolescence

As can be seen in **Figure 12**, a striking increase in the rate of skeletal calcium accretion occurs at puberty – from about ages 10 to 17 years. The peak rate of calcium retention in this period is 300–400 mg (7.5–10 mmol) daily (57); it occurs earlier in girls but continues longer in boys. For a target value of 300 mg (7.5 mmol) for the skeleton, 100 mg (2.5 mmol) for urinary calcium (58), and insensible losses of 40 mg (1.0 mmol), the net absorbed calcium during at least part of this period needs to be 440 mg (11 mmol) daily. Even with assuming high calcium absorption (+2 SD), this requires an intake of 1040 mg (26.0 mmol) daily (**Figure 17**) and an allowance of 1300 mg (32.5 mmol) during the peak growth phase (**Table 31**). It is difficult to justify any difference between the allowances for boys and girls because, as mentioned above, although the growth spurt starts earlier in girls, it continues longer in boys. This recommended intake (which is close to that derived differently by Matkovic and Heaney [49,58]) is not achieved by many adolescents even in developed countries (59–61), but the effects of this shortfall on their growth and bone status are unknown.

Adults

As indicated earlier and for the reasons given, we accept that the mean apparent calcium requirement of adults in developed countries is about 520 mg (13 mmol) but that this is increased by insensible losses to some 840 mg (21 mmol) (**Figure 14**). This reasoning forms the basis of our recommended intake for adults of 1000 mg (**Table 31**).

Menopause

The most important single cause of osteoporosis – at least in developed countries – is probably menopause, which is accompanied by an unequivocal and sustained rise in obligatory urinary calcium of about 30 mg (0.75 mmol) daily (50,62,63). Because calcium absorption certainly does not increase at this time – and probably decreases (64,65) – this extra urinary calcium represents a negative calcium balance which is compatible with the average bone loss of about 0.5–1.0 percent per year after menopause. There is a consensus that these events are associated with an increase in bone resorption but controversy continues about whether this is the primary event, the response to an increased calcium demand, or both. The results of calcium trials are clearly relevant. Before 1997, there had been 20 prospective trials of calcium supplementation in 857 postmenopausal women and 625 control subjects; these trials showed highly significant suppression of bone loss by calcium (65). Another meta-analysis covering similar numbers showed that calcium supplementation significantly enhanced the effect of oestrogen on bone (66). It is therefore logical to recommend sufficient additional calcium after the menopause to cover at least the extra obligatory loss of calcium in the urine. The additional dietary calcium needed to meet an increased urinary loss of 30 mg (0.75 mmol) is 260 mg/day (6.5 mmol/day) (**Figure 14**), which raises the daily requirement from 840 mg (21 mmol) to 1100 mg (27.5 mmol) and the recommended intake from 1000 to 1300 mg/day (25 to 32.5 mmol/day), which is a little higher than that recommended by the United States and Canada (14) (**Table 31** and **32**).

Table 32

Current calcium intake recommendations (mg/day)

	Australia 1991 Recommended Dietary Intake	United Kingdom 1991 Reference Nutrient Intake	European Union 1993 Population Reference Intake	United States and Canada 1997 Adequate Intake
Pregnancy (last trimester)	1100	700	700	1000–1300
Lactation	1200	1250	1200	1000–1300
Infancy	300 (human milk) 500 (cow milk)	525	400	210–270
Childhood	530–800	350–550	400–550	500–800
Puberty and adolescence				
Boys	1000–1200	1000	1000	1300
Girls	800–1000	800	800	1300
Maturity				
Males	800	700	700	1000
Females	800	700	700	1000
Later life				
Males >65 years	800	700	700	1200
Postmenopausal women	1000	700	700	1200

Ageing

Not enough is known about bone and calcium metabolism during ageing to enable calculation of the calcium requirements of older men and women with any confidence. Calcium absorption tends to decrease with age in both sexes (67-69) but whereas the evidence that calcium requirement goes up at the menopause is strong, corresponding evidence about ageing men is less convincing (32,36). Nonetheless, as a precaution we propose an extra allowance of 300 mg/day (7.5 mmol/day) for men over 65 to bring them into line with postmenopausal women (**Table 31**).

Pregnancy

The calcium content of the newborn infant is about 24 g (600 mmol). Most of this calcium is laid down in the last trimester of pregnancy, during which the foetus retains about 240 mg (6 mmol) of calcium daily (51). There is some evidence that pregnancy is associated with an increase in calcium absorption [associated with a rise in the plasma $1,25(\text{OH})_2\text{D}$ level] (70-72). For a maternal urinary calcium of 120 mg (3 mmol) and a maternal skin loss of 60 mg (1.5 mmol), the absorbed calcium should be 420 mg (10.5 mmol) daily. Even at optimal calcium absorption, the corresponding calcium intake would need to be 940 mg (23.5 mmol) (**Figure 17**) and the recommended allowance would need to be 1200 mg (30 mmol) (**Table 31**), which is similar to that proposed by the United States and Canada (14) (**Table 32**).

Lactation

The calcium content of human milk is about 36 mg per 100 ml (9 mmol/l) (18). A lactating woman produces about 750 ml of milk daily, which represents about 280 mg (7.0 mmol) of calcium. For a maternal urinary calcium of 100 mg/day (2.5 mmol/day) and maternal skin loss of 60 mg/day (1.5 mmol/day), the required absorption is 440 mg/day (11 mmol/day) – the same as at puberty. If calcium absorption efficiency is maximal (i.e., 2 SD above the normal mean) – possibly because of the effect of prolactin on the production of $1,25(\text{OH})_2\text{D}$ (72) – the requirement would be about 1040 mg (26.0 mmol) and the recommended intake would be about 1300 mg (32.5 mmol). However, although it is known that bone is lost during lactation and restored after weaning (73,74), early reports that this bone loss could be prevented by calcium supplementation (75) have not been confirmed in controlled studies (76-78). The prevailing view now is that calcium absorption does not increase and may decrease during lactation. It is increasingly thought that lactational bone loss is not a nutritional problem but may be due to parathyroid hormone-related peptide secreted by the breast (79) and therefore beyond the control of dietary calcium. In view of this uncertainty, we do not at present recommend any extra calcium allowance during lactation; any risk to adolescent mothers is covered by our general recommendation of 1300 mg for adolescents.

Upper limits

Because of the inverse relationship between fractional calcium absorption and calcium intake (**Figure 15**), a calcium supplement of 1000 mg (2.5 mmol) added to a Western-style diet only increases urinary calcium by about 60 mg (1.5 mmol). Urinary calcium also rises very slowly with intake (slope of 5–10 percent) and the risk of kidney stones from dietary hypercalciuria must therefore be negligible. In fact, it has been suggested that dietary calcium may protect against renal calculi because it binds dietary oxalate and reduces oxalate excretion (80,81). Toxic effects of a high calcium intake have only been described when the calcium is given as the carbonate in very high doses; this toxicity is caused as much by the alkali as by the calcium and is due to precipitation of calcium salts in renal tissue (milk-alkali syndrome) (82). However, in practice we recommend an upper limit on calcium intake of 3 g (75 mmol).

Comparisons with other recommendations

Our recommendations in **Table 31** can be compared with the current recommendations of Australia, the United Kingdom, the European Union, and the United States and Canada in **Table 32**. Our recommendations for adults are very close to those of the United States and Canada but higher than those of the United Kingdom and Australia, which do not take into account insensible losses, and higher than those of the European Union, which assumed 30 percent absorption of dietary calcium. The British and European values make no allowance for ageing or menopause. Recommendations for other high-risk groups are very similar in all five sets of recommendations except for the rather low allowance for infants by the United States and Canada. Nonetheless, and despite this broad measure of agreement, we have some misgivings about the application of these recommendations, all of which rely ultimately on data from developed nations, to developing countries where other dietary constituents – such as animal protein and sodium – and environmental factors may be very different. We shall therefore in the next sections briefly review current knowledge about the prevalence of osteoporosis across racial national boundaries and its relevance to calcium requirement.

Ethnic and environmental variations in the prevalence of osteoporosis

Intakes of calcium have been known for many years to vary greatly from one country to another, as is clearly shown in FAO food balance sheets (**Table 30**). Until fairly recently, it was widely assumed that low calcium intakes had no injurious consequences. This view of the global situation underlay the very conservative adequate calcium intakes recommended by WHO/FAO in 1962 (2). At that time, osteoporosis was still regarded as a bone matrix disorder and the possibility that it could be caused by calcium deficiency was barely considered. The paradigm has changed since then. Calcium deficiency is taken more seriously than it was and the apparent discrepancy between calcium intake and bone status across the world has attracted more attention. In general, recent investigations have sought for evidence of low bone density and high fracture incidence in countries where calcium intake is low; rickets has not been looked for, but the low calcium rickets recently reported from Nigeria (83) will no doubt attract attention.

This issue can be considered at several levels. The first level is genetic: Is there a genetic (ethnic) difference in the prevalence of osteoporosis between racial groups within a given society? The second level might be termed environmental-cultural (e.g., dietary): Is there a difference in the prevalence of osteoporosis between national groups of similar ethnic composition? The third level is environmental-geographical (e.g., latitude, affluence, and lifestyle): Is there a difference in the prevalence of osteoporosis between countries regardless of ethnic composition? At each of these levels, the prevalence of osteoporosis can in theory be determined in at least two ways – from the distribution of bone density within the population and from the prevalence of fractures, notably hip fractures. In practice, hip fracture data (or mortality from falls for elderly people which has been used as a surrogate [84]) are more readily available than bone densitometry.

Ethnicity

Comparisons between racial groups within countries suggest substantial racial differences in the prevalence of osteoporosis. This was probably first noted by Trotter (85) when she showed that bone density (weight/volume) was significantly higher in skeletons from black than from Caucasian subjects in the United States. It was later shown that hip fracture rates were lower in blacks than Caucasians in South Africa (86) and the United States (87). These observations have been repeatedly confirmed (88,89) without being fully explained but appear to be genetic in origin because the difference in bone status between blacks and Caucasians in the United States is already apparent in childhood (90) and cannot be explained by differences in body size (91). The difference in fracture rates between blacks and Caucasians cannot be

explained by differences in hip axis length (91); it seems to be largely or wholly due to real differences in bone density. Comparisons between Caucasians and Samoans in New Zealand (92) have also shown the latter to have the higher bone densities whereas the lower bone densities of Asians than Caucasians in New Zealand are largely accounted for by differences in body size (92). In the United States, fracture rates are lower among Japanese than among Caucasians but may be accounted for by their shorter hip axis length (93) and their lower incidence of falls (94). Bone density is generally lower in Asians than Caucasians within the United States (95) but this again is largely accounted for by differences in body size (96). There are also lower hip fracture rates for Hispanics, Chinese, Japanese, and Koreans than Caucasians in the United States (97,98). The conclusion must be that there are probably genetic factors influencing the prevalence of osteoporosis and fractures, but it is impossible to exclude the role of differences in diet and lifestyle between ethnic communities within a country.

Geography

There are wide geographical variations in hip fracture incidence, which cannot be accounted for by ethnicity. In the United States, the age-adjusted incidence of hip fracture in Caucasian women aged 65 and over varied with geography but was high everywhere – ranging from 700 to 1000 per 100 000 per year (99). Within Europe, the age-adjusted hip fracture rates ranged from 280 to 730 per 100 000 women in one study (100) and from 419 to 545 per 100 000 in another study (97) in which the comparable rates were 52.9 in Chile, 94.0 in Venezuela, and 247 in Hong Kong. In another study (101) age-adjusted hip fracture rates in women in 12 European countries ranged from 46 per 100 000 per year in Poland to 504 per 100 000 in Sweden, with a marked gradient from south to north and from poor to rich. In Chinese populations, the hip fracture rate is much lower in Beijing (87–97 per 100 000) than in Hong Kong (181–353 per 100 000) (102), where the standard of living is higher. Thus there are marked geographic variations in hip fracture rates within the same ethnic groups.

Ethnicity, environment, and lifestyle

The conclusion from the above is that there are probably ethnic differences in hip fracture rates within countries but also environmental differences within the same ethnic group which may complicate the story. For international comparisons on a larger scale, it is impossible to separate genetic from environmental factors, but certain patterns emerge which are likely to have biological meaning. The most striking of these is the positive correlation between hip fracture rates and standard of living noted by Hegsted when he observed that osteoporosis was largely a disease of affluent Western cultures (103). He based this conclusion on a previously published review of hip fracture rates in 10 countries (104), which strongly suggested a correlation between hip fracture rate and affluence. Another review of 19 regions and racial groups (105) confirmed this by showing a gradient of age- and sex-adjusted hip fracture rates from 31 per 100 000 in South African Bantu to 968 per 100 000 in Norway. In the analysis of hip fracture rates in Beijing and Hong Kong referred to above (102), it was noted that the rates in both cities were much lower than in the United States. Many other publications point to the same conclusion – that hip fracture prevalence (and by implication osteoporosis) is related to affluence and, consequently, to animal protein intake, as Hegsted pointed out, but also and paradoxically to calcium intake.

The calcium paradox

The paradox that hip fracture rates are higher in developed nations where calcium intake is high than in developing nations where calcium intake is low clearly calls for an explanation. Hegsted (103) was probably the first to note the close relation between calcium and protein intakes across the world (which is also true within nations [63]) and to hint at but dismiss the

possibility that the adverse effect of protein might outweigh the positive effect of calcium on calcium balance. Only recently has fracture risk been shown to be a function of protein intake in American women (106). There is also suggestive evidence that hip fracture rates (as judged by mortality from falls in elderly people across the world) are a function of protein intake, national income, and latitude (107). The latter is particularly interesting in view of the strong evidence of vitamin D deficiency in hip fracture patients in the developed world (108-114) and the successful prevention of such fractures with small doses of vitamin D and calcium (115,116) (see **Chapter 8**). It is therefore possible that hip fracture rates may be related to protein intake, vitamin D status, or both and that either of these factors could explain the calcium paradox. We shall therefore consider how these and other nutrients (notably sodium) affect calcium requirement.

Nutritional factors affecting calcium requirements

The calculations of calcium requirements proposed above were based on data from developed countries (notably the United States and Norway) and can only be applied with any confidence to nations and populations with similar dietary cultures. Other dietary cultures may entail different calcium requirements and call for different recommendations. In particular, the removal or addition of any nutrient that affects calcium absorption or excretion must have an effect on calcium requirement. Two such nutrients are sodium and animal protein, both of which increase urinary calcium and must be presumed therefore to increase calcium requirement. A third candidate is vitamin D because of its role in calcium homeostasis and calcium absorption.

Sodium

It has been known at least since 1961 that urinary calcium is related to urinary sodium (117) and that sodium administration raises calcium excretion, presumably because sodium competes with calcium for reabsorption in the renal tubules. Regarding the quantitative relationships between the renal handling of sodium and calcium, the filtered load of sodium is about 100 times that of calcium (in molar terms) but the clearance of these two elements is similar at about 1 ml/min, which yields about 99 percent reabsorption and 1 percent excretion for both (118). However, these are approximations, which conceal the close dependence of urinary sodium on sodium intake and the weaker dependence of urinary calcium on calcium intake. It is an empirical fact that urinary sodium and calcium are significantly related in normal and hypercalciuric subjects on freely chosen diets (119-122). The slope of urinary calcium on sodium varies in published work from about 0.6 percent to 1.2 percent (in molar terms); a representative figure is about 1 percent – that is, 100 mmol of sodium (2.3 g) takes out about 1 mmol (40 mg) of calcium (63,120). The biological significance of this relationship is supported by the accelerated osteoporosis induced by feeding salt to rats on low-calcium diets (123) and the effects of salt administration and salt restriction on markers of bone resorption in postmenopausal women (124,125). Because salt restriction lowers urinary calcium, it is likely also to lower calcium requirement and, conversely, salt feeding is likely to increase calcium requirement. This is illustrated in **Figure 18**, which shows that lowering sodium intake by 100 mmol (2.3 g) from, for example, 150 to 50 mmol (3.45 to 1.15 g), reduces the theoretical calcium requirement from 840 mg (21 mmol) to 600 mg (15 mmol). However, the implications of this on calcium requirement across the world cannot be computed because information about sodium intakes is available from very few countries (126).

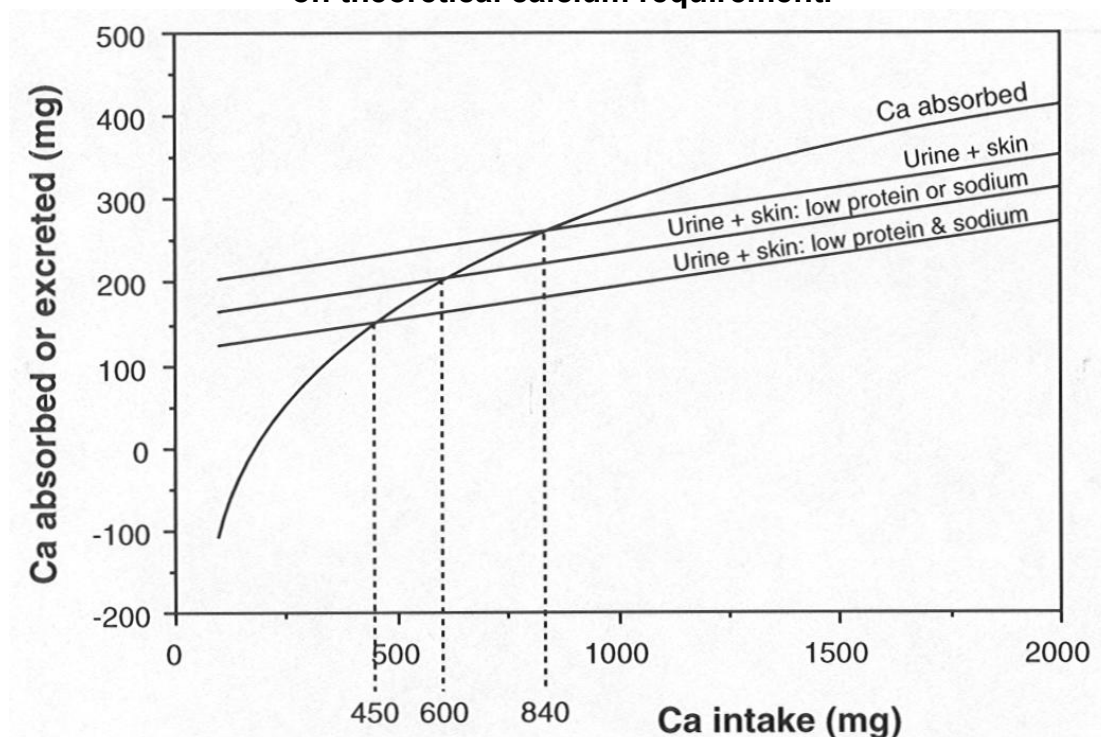
Protein

The positive effect of dietary protein – particularly animal protein – on urinary calcium has also been known at least since the 1960s (127-129). One study found that 0.85 mg of calcium

was lost for each gram of protein in the diet (130). A meta-analysis of 16 studies in 154 adult humans on protein intakes up to 200 g found that 1.2 mg of calcium was lost in the urine for every 1g rise in dietary protein (131). A small but more focussed study showed a rise of 40 mg in urinary calcium when dietary animal protein was raised from 40 to 80 g (i.e., within the physiological range) (132). This ratio of urinary calcium to dietary protein ratio (1mg to 1g) is a representative value, which we have adopted. This means that a 40g reduction in animal protein intake from 60 to 20 g (or from the developed to the developing world [Table 30]) would reduce calcium requirement by the same amount as a 2.3g reduction in dietary sodium, i.e. from 840 to 600 mg. (Figure 18).

Figure 18

**The effect of varying protein or sodium intake
on theoretical calcium requirement.**



Note: In a western-style diet, absorbed calcium matches urinary and skin calcium at an intake of 840 mg as in Figure 14. Reducing animal protein intakes by 40 g reduces the intercept value and requirement to 600 mg. Reducing both sodium and protein reduces the intercept value to 450 mg.

How animal protein exerts its effect on calcium excretion is not fully understood. A rise in glomerular filtration rate in response to protein has been suggested as one factor (128) but this is unlikely to be important in the steady state. The major mechanisms are thought to be the effect of the acid load contained in animal proteins and the complexing of calcium in the renal tubules by sulphate and phosphate ions released by protein metabolism (133,134). Urinary calcium is significantly related to urinary phosphate (as well as to urinary sodium), particularly in subjects on restricted calcium intakes or in the fasting state, and most of the phosphorus in the urine of people on Western-style diets comes from animal protein in the diet (63). Similar considerations apply to urinary sulphate but it is probably less important than the phosphate ion because the association constant for calcium sulphate is lower than that for calcium phosphate (135). The empirical observation that each 1 g of protein results in 1 mg of calcium in the urine agrees very well with the phosphorus content of animal protein (about 1 percent by weight) and the observed relationship between calcium and phosphate in the urine (63).

Vitamin D

One of the first observations made on vitamin D after it had been identified in 1918 (136) was that it promoted calcium absorption (137). It is now well established that vitamin D (synthesised in the skin under the influence of sunlight) is converted to 25OHD in the liver and then to 1,25(OH)₂D in the kidneys and that the latter metabolite controls calcium absorption (21) (see **Chapter 8**). However, plasma 25OHD closely reflects vitamin D nutritional status and because it is the substrate for the renal enzyme which produces 1,25(OH)₂D, it could have an indirect effect on calcium absorption. The plasma level of 1,25-(OH)₂D is principally regulated through increased gene expression of the 1- α -hydroxylase (CYP1 α) and not by increased 25OHD levels. This has been seen consistently in animal studies, and the high calcium absorption (138) and high plasma 1,25-(OH)₂D (139) observed in Gambian mothers is consistent with this type of adaptation. However, increasing latitude may compromise vitamin D synthesis to the degree that 25OHD levels are no longer sufficient to sustain adequate 1,25-(OH)₂D levels and efficient intestinal calcium absorption, although this theory remains unproved. Regardless of the mechanism of compromised vitamin D homeostasis, the differences in calcium absorption efficiency have a major effect on theoretical calcium requirement, as illustrated in **Figure 18**, which shows that an increase in calcium absorption of as little as 10 percent reduces the intercept of excreted and absorbed calcium (and therefore calcium requirement) from 840 to 680 mg. (The figure also shows the great increase in calcium requirement that must result from any impairment of calcium absorption.)

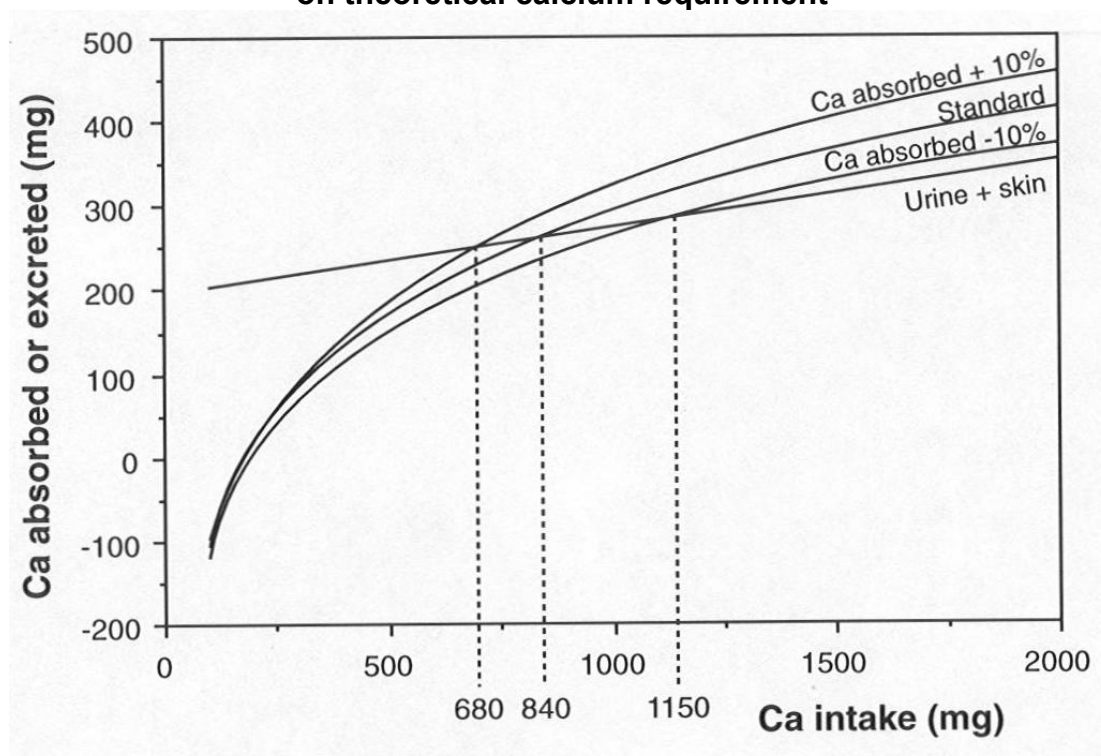
Implications

The major reduction in theoretical calcium requirement which follows animal protein restriction has led us to attempt to show in **Table 33** how the calcium allowances recommended in **Table 31** could be modified to apply to nations where the animal protein intake per capita is around 20–40 g rather than around the 60–80 g in developed countries. These hypothetical allowances take into account the need to protect children, in whom skeletal needs are much more important determinants of calcium requirement than are urinary losses and in whom calcium supplementation had a beneficial effect in the Gambia (140). However, adjustment for animal protein intake has a major effect on the recommended calcium allowances for adults as the table shows. It also brings the allowances nearer to what the actual calcium intakes are in many parts of the world.

If sodium intakes were also lower in developing than developed nations or urinary sodium were reduced for other reasons such as increased sweat losses, the calcium requirement might be even lower, for example, 450 mg (**Figure 18**). This would be reduced still further by any increase in calcium absorption, whether resulting from better vitamin D status because of increased sunlight exposure or for other reasons, as illustrated in **Figure 19**. Because the increase in calcium absorption in the Gambia is much more than 10 percent (138), this is likely to have a major – although not at present calculable – effect on calcium requirement there. However, the adjusted bone mineral density in Gambian women is reported to be some 20 percent lower in the spine (but not in the forearm) than in British women (141), which emphasises the need for more data from developing countries.

Figure 19

**The effect of varying calcium absorptive efficiency
on theoretical calcium requirement**



Note: At normal calcium absorption, the intercept of urinary plus skin calcium meets absorbed calcium at an intake of 840 mg as in **Figure 14**. A 10 percent reduction in calcium absorption raises the intercept and requirement to 1150 mg and a 10 percent increase in calcium absorption reduces it to 680 mg.

Table 33

Theoretical calcium allowances based on an animal protein intake of 20–40 g

Group	Recommended intake mg/day
Infants and children	
0–6 months	
Human milk	300
Cow milk	400
7–12 months	450
1–3 years	500
4–6 years	550
7–9 years	700
Adolescents, 10–18 years	1000 ^a
Adults	
Females	
19 years to menopause	750
Postmenopause	800
Males	
19–65 years	750
65 +	800
Pregnancy (last trimester)	800
Lactation	750

^a Particularly during the growth spurt.

Conclusions

Calcium is an essential nutrient that plays a vital role in neuromuscular function, many enzyme-mediated processes, blood clotting, and providing rigidity to the skeleton by virtue of its phosphate salts. Its non-structural roles require the strict maintenance of ionised calcium concentration in tissue fluids at the expense of the skeleton if necessary and it is therefore the skeleton which is at risk if the supply of calcium falls short of requirement.

Calcium requirements are essentially determined by the relationship between absorptive efficiency and excretory rate – excretion being through the bowel, kidneys, skin, hair, and nails. In adults, the rate of calcium absorption from the gastrointestinal tract needs to match the rate of all losses from the body if the skeleton is to be preserved; in children and adolescents, an extra input is needed to cover the requirements of skeletal growth.

Compared with that of other minerals, calcium economy is relatively inefficient. On most intakes, only about 25–30 percent of dietary calcium is effectively absorbed and obligatory calcium losses are relatively large. Absorbed calcium has to match these obligatory losses and the dietary intake has to be large enough to ensure this rate of absorption if skeletal damage is to be avoided. The system is subject to considerable inter-individual variation in both calcium absorption and excretion for reasons that are not fully understood but which include vitamin D status, sodium and protein intake, age, and menopausal status in women. Although it needs to be emphasised that calcium deficiency and negative calcium balance must sooner or later lead to osteoporosis, this does not mean that all osteoporosis can be attributed to calcium deficiency. On the contrary, there may be more osteoporosis in the world from other causes. Nonetheless, it would probably be generally agreed that any form of osteoporosis must inevitably be aggravated by negative external calcium balance. Such negative balance – even for short periods – is prejudicial because it takes so much longer to rebuild bone than to destroy it. Bone that is lost, even during short periods of calcium deficiency, is only slowly replaced when adequate amounts of calcium become available.

In seeking to define advisable calcium intakes on the basis of physiologic studies and clinical observations, nutrition authorities have to rely largely on data from developed nations living at relatively high latitudes. Although it is now possible to formulate recommendations that are appropriate to different stages in the life cycle of the populations of these nations, extrapolation from these figures to other cultures and nutritional environments can only be tentative and must rely on what is known of nutritional and environmental effects on calcium absorption and excretion. Nonetheless, we have made an attempt in this direction, knowing that our speculative calculations may be incorrect because of other variables not yet identified.

No reference has been made in this account to the possible beneficial effects of calcium in the prevention or treatment of pre-eclampsia (142), colon cancer (143), or hypertension (144) and no attempt has been made to use these conditions as endpoints on which to base calcium intakes. In each of the above conditions, epidemiologic data suggested an association with calcium intake, and experimentation with increased calcium intakes has now been tried. In each case the results have been disappointing, inconclusive, or negative (145-147) and have stirred controversy (148-150). Because there is no clear consensus about optimal calcium intake for prevention or treatment of these conditions and also no clear mechanistic ideas on how dietary calcium intakes affect them, it is not possible to allow for the effect of health outcomes in these areas on our calcium recommendations. However, although the anecdotal information and positive effects of calcium observed in these three conditions cannot influence our recommendations, they do suggest that generous calcium allowances may confer other benefits besides protecting the skeleton. Similarly, no reference has been made to the effects of physical activity, alcohol, smoking, or other known risk

factors on bone status because the effects of these variables on calcium requirement are beyond the realm of simple calculation.

Future research

Future research should:

- recognise that there is an overwhelming need for more studies of calcium metabolism in developing countries;
- investigate further the cultural, geographical, and genetic bases for differences in calcium intakes in different groups in developing nations;
- establish the validity of different recommended calcium intakes based on animal protein and sodium intakes;
- clarify the role of dietary calcium in pre-eclampsia, colon cancer, and hypertension; and
- investigate the relationship of latitude, sun exposure, and synthesis of vitamin D with intestinal calcium absorption in different geographical locations.

REFERENCES

1. **FAO.** 1974. Handbook on Human Nutritional Requirements. Rome, FAO.
2. **FAO/WHO Expert Group.** 1962. Calcium Requirements. Rome, FAO.
3. **Albright, F. & Reifenstein, E.C.** 1948. The Parathyroid Glands and Metabolic Bone Disease. Baltimore: Williams & Wilkins
4. **Nordin, B.E.C.** 1960. Osteomalacia, osteoporosis and calcium deficiency. *Clin. Orthop.*, 17: 235-258.
5. **Young, M.M. & Nordin, B.E.C.** 1967. Effects of natural and artificial menopause on plasma and urinary calcium and phosphorus. *Lancet*, 2: 118-120.
6. **Stepan, J.J., Posphichal, J., Presl, J. & Pacovsky, V.** 1987. Bone loss and biochemical indices of bone remodeling in surgically induced postmenopausal women. *Bone* 8: 279-284.
7. **Kelly, P.J., Pocock, N.A., Sambrook, P.N. & Eisman, J.A.** 1989. Age and menopause-related changes in indices of bone turnover. *J. Clin. Endocrinol. Metab.*, 69: 1160-1165.
8. **Christiansen, C., Christensen, M.S., Larsen, N-E. & Transbøl, I.B.** 1982. Pathophysiological mechanisms of estrogen effect on bone metabolism. Dose-response relationships in early postmenopausal women. *J. Clin. Endocrinol. Metab.*, 55: 1124-1130.
9. **Parfitt, A.M.** 1990. Osteomalacia and related disorders. In: *Metabolic Bone Disease and Clinically Related Disorders*. Second Edition. Avioli, L.V., Krane, S.M., eds. p. 329-396. Philadelphia: W.B. Saunders,
10. **Need, A.G.** Corticosteroid hormones. In: *Metabolic Bone and Stone Disease*. Third Edition. Nordin, B.E.C., Need, A.G., Morris, H.A., eds. p.70-78. Edinburgh: Churchill Livingstone.
11. **Horowitz, M.** 1993. Osteoporosis in men. In: *Metabolic Bone and Stone Disease*. Third Edition. Nordin, B.E.C., Need, A.G., Morris, H.A., eds. Edinburgh: Churchill Livingstone,
12. **Lips, P., Netelenbos, J.C. & Jongen, M.J.M.** 1982. Histomorphometric profile and vitamin D status in patients with femoral neck fracture. *Metab. Bone Dis. Relat. Res.*, 4: 85-93.
13. **Truswell, S.** 1983. Recommended dietary intakes around the world. Report by Committee 1/5 of the International Union of Nutritional Sciences. *Nutr. Abstracts Revs.*, 53: 939-1119.
14. **Food and Nutrition Board, Institute of Medicine.** 1997. *Dietary reference intakes for calcium, phosphorus, magnesium, vitamin D, and fluoride*. Washington DC: National Academy Press.
15. **Department of Health.** 1991. Dietary Reference Values for Food Energy and Nutrients for the United Kingdom. *Report of the Panel on Dietary Reference Values of the Committee on Medical Aspects of Food Policy*. London: HMSO.
16. **Directorate-General Industry.** 1993. *Reports of the Scientific Committee for Food* (Thirty-first series). Nutrient and energy intakes for the European Community. Luxembourg: Office for Official Publications of the European Communities.
17. **National Health and Medical Research Council.** 1991. *Recommended Dietary Intakes for use in Australia*. Canberra: Commonwealth of Australia.
18. **Nordin, B.E.C.** 1976. Nutritional considerations. In: *Calcium, Phosphate and Magnesium Metabolism*. Nordin, B.E.C., ed. p. 1-35. Edinburgh: Churchill Livingstone.

19. **Robertson, W.G. & Marshall, R.W.** 1981. Ionised calcium in body fluids. *Crit. Revs. Clin. Lab. Sci.*, 15: 85-125.
20. **Brown, E.M. & Hebert, S.C.** 1997. Calcium-receptor-regulated parathyroid and renal function. *Bone*, 20: 303-309.
21. **Jones, G., Strugnell, S.A. & DeLuca, H.F.** 1998. Current understanding of the molecular actions of vitamin D. *Physiol. Revs.*, 78: 1193-1231.
22. **Wu, D.D., Boyd, R.D., Fix, T.J. & Burr, D.B.** 1990. Regional patterns of bone loss and altered bone remodeling in response to calcium deprivation in laboratory rabbits. *Calcif. Tissue Int.*, 47: 18-23.
23. **Food and Agriculture Organization of the United Nations.** 1991. Production Yearbook Vol. 44, 1990. Rome, FAO.
24. **Ireland, P. & Fordtran, J.S.** 1973. Effect of dietary calcium and age on jejunal calcium absorption in Humans studied by intestinal perfusion. *J. Clin. Investig.*, 52: 2672-81.
25. **Heaney, R.P., Saville, P.D. & Recker, R.R.** 1975. Calcium absorption as a function of calcium intake. *J. Lab. Clin. Med.*, 85: 881-890.
26. **Wilkinson, R.** 1976. Absorption of calcium, phosphorus and magnesium. *Calcium, Phosphate and Magnesium Metabolism*. Nordin, B.E.C. ed. p. 36-112. Edinburgh: Churchill Livingstone.
27. **Marshall, D.H.** 1976. Calcium and phosphate kinetics *Calcium, Phosphate and Magnesium Metabolism*. Nordin, B.E.C. ed. p. 257-297. Edinburgh: Churchill Livingstone.
28. **Heaney, R.P. & Skillman, T.G.** 1964. Secretion and excretion of calcium by the Human gastrointestinal tract. *J. Lab. Clin. Med.*, 64: 29-41.
29. **Nordin, B.E.C., Horsman, A. & Aaron, J.** 1976. *Diagnostic procedures*. Calcium, Phosphate and Magnesium Metabolism. Nordin, B.E.C. ed. p. 469-524. Edinburgh: Churchill Livingstone.
30. **Marshall, D.H., Nordin, B.E.C. & Speed, R.** 1976. Calcium, phosphorus and magnesium requirement. *Proc. Nutr. Soc.*, 35: 163-173.
31. **Nordin, B.E.C. & Marshall, D.H.** 1988. Dietary requirements for calcium. In: *Calcium in Human Biology*. Nordin, B.E.C., ed. p. 447-471. Berlin: Springer-Verlag.
32. **Bogdonoff, M.D., Shock, N.W. & Nichols, M.P.** Calcium, phosphorus, nitrogen, and potassium balance studies in the aged male. *J. Gerontol.* 1953;8:272-288.
33. **Clarkson, E.M., Durrant, C. & Phillips, M.E., Gower, P.E., Jewkes, R.F., De Wardener, H.E.** 1970. The effect of a high intake of calcium and phosphate in normal subjects and patients with chronic renal failure. *Clin. Sci.*, 39: 693-704.
34. **Johnston, F.A., McMillan, T.J. & Derby Falconer, G.** 1952. Calcium retained by young women before and after adding spinach to the diet. *J. Am. Diet. Assoc.*, 28: 933-938.
35. **Malm, O.J.** 1958. Calcium requirement and adaptation in adult men. *Scand. J. Clin. Lab. Investig.*, 10(Suppl 36):1-289.
36. **Owen, E.C., Irving, J.T. & Lyall, A.** 1940. The calcium requirements of older male subjects with special reference to the genesis of senile osteoporosis. *Acta Medica. Scand.*, 103: 235-250.
37. **Steggerda, F.R. & Mitchell, H.H.** 1939. The calcium requirement of adult man and the utilisation of the calcium in milk and in calcium gluconate. *J. Nutr.*, 17: 253-262.
38. **Steggerda, F.R. & Mitchell, H.H.** Further experiments on the calcium requirement of adult man and the utilisation of the calcium in milk. *J. Nutr.*, 21: 577-588.

39. **Steggerda, F.R. & Mitchell, H.H.** 1946. Variability in the calcium metabolism and calcium requirements of adult Human subjects. *J. Nutr.*, 31: 407-422.
40. **Gallagher, J.C., Riggs, B.L. & Eisman, J.** 1979. Intestinal calcium absorption and serum vitamin D metabolites in normal subjects and osteoporotic patients. *J. Clin. Investig.*, 64: 729-736.
41. **Wishart, J.M., Horowitz, M., Need, A.G., Scopacasa, F., Morris, H.A., Clifton, P.M. & Nordin, B.E.C.** 1997. Relations between calcium intake, calcitriol, polymorphisms of the vitamin D receptor gene, and calcium absorption in premenopausal women. *Am. J. Clin. Nutr.*, 65: 798-802.
42. **MacFadyen, I.J., Nordin, B.E.C., Smith, D.A., Wayne, D.J. & Rae, S.L.** 1965. Effect of variation in dietary calcium on plasma concentration and urinary excretion of calcium. *Br. Med. J.*, 1: 161-164.
43. **Heaney, R.P., Recker, R.R. & Ryan, R.A.** 1999. Urinary calcium in perimenopausal women: normative values. *Osteoporos. Int.*, 9: 13-18.
44. **Charles, P., Taagehøj, F., Jensen, L., Mosekilde, L. & Hansen, H.H.** 1983. Calcium metabolism evaluated by Ca^{45} kinetics: estimation of dermal calcium loss. *Clin. Sci.*, 65: 415-422.
45. **Hasling, C., Charles, P., Taagehøj, J. & Mosekilde, L.** 1990. Calcium metabolism in postmenopausal osteoporosis: the influence of dietary calcium and net absorbed calcium. *J. Bone Miner. Res.*, 5: 939-946.
46. **Mitchell, H.H. & Curzon, E.G.** 1939. The dietary requirements of calcium and its significance. Actualites Scientifique et Industrielles No. 771. p.36-101. Paris: Hermann.
47. **Hegsted, J.M., Moscoso, I. & Collazos, C.H.C.** 1952. Study of minimum calcium requirements by adult men. *J. Nutr.*, 46:181-201.
48. **Heaney, R.P., Recker, R.R. & Saville, P.D.** 1978. Menopausal changes in calcium balance performance. *J. Lab. Clin. Med.*, 92: 953-963.
49. **Matkovic, V. & Heaney, R.P.** 1992. Calcium balance during Human growth: evidence for threshold behavior. *Am. J. Clin. Nutr.*, 55: 992-996.
50. **Nordin, B.E.C., Need, A.G., Morris, H.A. & Horowitz, M.** 1999. Biochemical variables in pre- and postmenopausal women: reconciling the calcium and estrogen hypotheses. *Osteoporos. Int.*, 9: 351-357.
51. **American Academy of Pediatrics Committee on Nutrition.** 1978. Calcium requirements in infancy and childhood. *Pediatrics*, 62: 826-832.
52. **Williams, M.L., Rose, C.S., Morrow, G., Sloan, S.E. & Barness, L.A.** 1970. Calcium and fat absorption in neonatal period. *Am. J. Clin. Nutr.*, 23: 1322-1330.
53. **Hanna, F.M., Navarrete, D.A. & Hsu, F.A.** 1970. Calcium-fatty acid absorption in term infants fed Human milk and prepared formulas simulating Human milk. *Pediatrics*, 45: 216-224.
54. **Widdowson, E.M.** 1965. Absorption and excretion of fat, nitrogen, and minerals from "filled" milks by babies one week old. *Lancet*, 2: 1099-1105.
55. **Shaw, J.C.L.** 1976. Evidence for defective skeletal mineralisation in low birthweight infants: the absorption of calcium and fat. *Pediatrics*, 57: 16-25.
56. **Widdowson, E.M., McCance, R.A., Harrison, G.E. & Sutton, A.** 1963. Effect of giving phosphate supplements to breast-fed babies on absorption and excretion of calcium, strontium, magnesium and phosphorus. *Lancet*, 2:1250-51.
57. **Leitch, I. & Aitken, F.C.** 1959. The estimation of calcium requirements: a re-examination. *Nutr. Abstracts Revs.*, 29: 393-411.

58. **Matkovic, V.** 1991. Calcium metabolism and calcium requirements during skeletal modeling and consolidation of bone mass. *Am. J. Clin. Nutr.*, 54: 45S-260S.
59. **Abrams, S.A. & Stuff, J.E.** 1994. Calcium metabolism in girls: current dietary intakes lead to low rates of calcium absorption and retention during puberty. *Am. J. Clin. Nutr.*, 60: 739-743.
60. **Truswell, A.S. & Darnton-Hill, I.** 1981. Food habits of adolescents. *Nutr. Revs.* 39: 73-88.
61. **Marino, D.D. & King, J.C.** 1980. Nutritional concerns during adolescence. *Pediatr. Clin. N. Am. J.*, 27: 125-139.
62. **Prince, R.L., Dick, I. & Devine, A.** 1995. The effects of menopause and age in calcitropic hormones: a cross-sectional study of 655 healthy women aged 35 to 90. *J. Bone Miner. Res.*, 10: 835-842.
63. **Nordin, B.E.C. & Polley, K.J.** 1987. Metabolic consequences of the menopause. A cross-sectional, longitudinal, and intervention study on 557 normal postmenopausal women. *Calcif. Tissue Int.*, 41: S1-S60.
64. **Heaney, R.P., Recker, R.R., Stegman, M.R. & Moy, A.J.** 1989. Calcium absorption in women: relationships to calcium intake, estrogen status, and age. *J. Bone Miner. Res.*, 4: 469-475.
65. **Nordin, B.E.C.** 1997. Calcium and osteoporosis. *Nutrition*, 13: 664-686.
66. **Nieves, J.W., Komar, L., Cosman, F. & Lindsay, R.** 1998. Calcium potentiates the effect of estrogen and calcitonin on bone mass: review and analysis. *Am. J. Clin. Nutr.*, 67: 18-24.
67. **Morris, H.A., Need, A.G., Horowitz, M., O'Loughlin, P.D. & Nordin, B.E.C.** 1991. Calcium absorption in normal and osteoporotic postmenopausal women. *Calcif. Tissue Int.*, 49: 240-243.
68. **Ebeling, P.R., Yergey, A.L. & Vleira, N.E. et al.** 1994. Influence of age on effects of endogenous 1,25-dihydroxyvitamin D on calcium absorption in normal women. *Calcif. Tissue Int.*, 55: 330-334.
69. **Need, A.G., Morris, H.A., Horowitz, M., Scopacasa, F. & Nordin, B.E.C.** 1998. Nordin. Intestinal calcium absorption in men with spinal osteoporosis. *Clin. Endocrinol.*, 48: 163-168.
70. **Heaney, R.P. & Skillman, T.G.** 1971. Calcium metabolism in normal Human pregnancy. *J. Clin. Endocrinol. Metab.*, 33: 661-670.
71. **Kent, G.N., Price, R.I. & Gutteridge, D.H.** 1991. The efficiency of intestinal calcium absorption is increased in late pregnancy but not in established lactation. *Calcif. Tissue Int.*, 48: 293-295.
72. **Kumar, R., Cohen, W.R., Silva, P. & Epstein, F.H.** 1979. Elevated 1,25-dihydroxyvitamin D plasma levels in normal Human pregnancy and lactation. *J. Clin. Investig.*, 63: 342-344.
73. **Kent G.N., Price, R.I. & Gutteridge, D.H.** 1990. Human lactation: forearm trabecular bone loss, increased bone turnover, and renal conservation of calcium and inorganic phosphate with recovery of bone mass following weaning. *J. Bone Miner. Res.*, 5: 361-369.
74. **López, J.M., González, G., Reyes, V., Campino, C. & Díaz, S.** 1996. Bone turnover and density in healthy women during breastfeeding and after weaning. *Osteoporos. Int.*, 6: 153-159.
75. **Chan, G.M., McMurphy, M., Westover, K., Engelbert-Fenton, K & Thomas, M.R.** 1987. Effects of increased dietary calcium intake upon the calcium and status of lactating adolescent and adult women. *Am. J. Clin. Nutr.*, 46: 319-323.

76. **Prentice, A., Jarjou, L.M.A. & Cole, T.J.** 1995. Calcium requirements of lactating Gambian mothers: effects of a calcium supplement on breast-milk calcium concentration, maternal bone mineral content and urinary calcium excretion. *Am. J. Clin. Nutr.*, 62: 58-67.
77. **Kalkwarf, H.J., Specker, B.L., Bianchi, D.C., Ranz, J. & Ho, M.** 1997. The effect of calcium supplementation on bone density during lactation and after weaning. *N. Engl. J. Med.*, 337: 523-528.
78. **Allen, L.H.** 1998. Women's dietary calcium requirements are not increased by pregnancy or lactation. *Am. J. Clin. Nutr.*, 67: 591-592.
79. **Sowers, M.F., Hollis, B.W. & Shapiro, B.** 1996. Elevated parathyroid hormone-related peptide associated with lactation and bone density loss. *JAMA*, 276: 549-554.
80. **Curhan, G.C., Willett, W.C., Speizer, F.E., Spiegelman, D. & Stampfer, M.J.** 1997. Comparison of dietary calcium with supplemental calcium and other nutrients as factors affecting the risk for kidney stones in women. *Ann. Internal Med.*, 126: 497-504.
81. **Curhan, G.C., Willett, W.C., Rimm, E.B. & Stampfer, M.J.** 1993. A prospective study of dietary calcium and other nutrients and the risk of symptomatic kidney stones. *N. Engl. J. Med.*, 328: 833-838.
82. **Burnett, C.H., Commons, R.R., Albright, F. & Howard, J.E.** 1949. Hypercalcaemia without hypercalciuria or hypophosphatemia, calcinosis and renal insufficiency. A syndrome following prolonged intake of milk and alkali. *N. Engl. J. Med.*, 240: 787-794.
83. **Thacher, T.D., Fischer, P.R. & Pettifor, J.M.** 1999. A comparison of calcium, vitamin D, or both for nutritional rickets in Nigerian children. *N. Engl. J. Med.*, 341: 563-568.
84. **Eddy, T.P.** 1972. Deaths from domestic falls and fractures. *Br. J. Prev. Soc. Med.*, 26: 173-179.
85. **Trotter, M., Broman, G.E. & Peterson, R.R.** 1960. Densities of bones of white and Negro skeletons. *J. Bone Joint Surg.* 1960;42-A:50-59.
86. **Solomon, L.** 1968. Osteoporosis and fracture of the femoral neck in the South African Bantu. *J. Bone Joint Surg.*, 50-B-2:2-13.
87. **Bollet, A.J., Engh, G. & Parson, W.** 1965. Sex and race incidence of hip fractures. *Arch. Internal Med.*, 116:191-194.
88. **Cohn, S.H., Abesamis, C., Yasumara, S., Aloia, J.F., Zanzi, I. & Ellis, K.J.** 1977. Comparative skeletal mass and radial bone mineral content in black and white women. *Metabolism*, 26: 171-178.
89. **DeSimone, D.P., Stevens, J., Edwards, J., Shary, J., Gordon, L. & Bell, N.H.** 1989. Influence of body habitus and race on bone mineral density of the midradius, hip, and spine in aging women. *J. Bone Miner. Res.*, 5: 827-830.
90. **Bell, N.H., Shary, J., Stevens, J., Garza, M., Gordon, L. & Edwards, J.** 1991. Demonstration that bone mass is greater in black than in white children. *J. Bone Miner. Res.*, 6: 719-723.
91. **Nelson, D.A., Jacobsen, G., Barondess, D.A. & Parfitt, A.M.** 1995. Ethnic differences in regional bone density, hip axis length, and lifestyle variables among healthy black and white men. *J. Bone Miner. Res.*, 10: 782-787.
92. **Cund, T., Cornish, J., Evans, M.C., Gamble, G., Stapleton, J. & Reid, I.R.** 1995. Sources of interracial variation in bone mineral density. *J. Bone Miner. Res.*, 10: 368-373.
93. **Cummings, S.R., Cauley, J.A., Palermo, L., Ross, P.D., Wasnich, R.D., Black, D. & Faulkner, K.G.** 1994. Racial differences in hip axis lengths might explain racial differences in rates of hip fracture. *Osteoporos. Int.*, 4: 226-229.

94. **Davis, J.W., Ross, P.D., Nevitt, M.C. & Wasnich, R.D.** 1997. Incidence rates of falls among Japanese men and women living in Hawaii. *J. Clin. Epidemiol.*, 50: 589-594.
95. **Yano, K., Wasnich, R.D., Vogel, J.M. & Heilbrun, L.K.** 1984. Bone mineral measurements among middle-aged and elderly Japanese residents in Hawaii. *Am. J. Epidemiol.*, 119: 751-764.
96. **Ross, P.D., He, Y-F. & Yates, A.J.** 1996. Body size accounts for most differences in bone density between Asian and caucasian women. *Calcif. Tissue Int.*, 59: 339-343.
97. **Silverman, S.L. & Madison, R.E.** 1988. Decreased incidence of hip fracture in Hispanics, Asians, and blacks: California hospital discharge data. *Am. J. Public Health*, 78: 1482-1483.
98. **Lauderdale, D.S., Jacobsen, S.J., Furner, S.E., Levy, P.S., Brody, J.A. & Goldberg, J.** 1997. Hip fracture incidence among elderly Asian-American populations. *Am. J. Epidemiol.*, 146: 502-509.
99. **Villa, M.L. & Nelson, L.** 1996. Race, ethnicity and osteoporosis. In: Osteoporosis. Marcus, R., Feldman, D., Kelsey, J., eds. p. 435-447. San Diego: Academic Press.
100. **Bacon, W.E., Maggi, S. & Looker, A.** 1996. International comparison of hip fracture rates in 1988-89. *Osteoporos. Int.*, 6: 69-75.
101. **Johnell, A., Gullberg, B., Allander, E. & Kanis, J.A.** 1992. The apparent incidence of hip fracture in Europe: A study of national register sources. *Osteoporos. Int.*, 2: 298-302.
102. **Xu, L., Lu, A., Zhao, X., Chen, X. & Cummings, S.R.** 1996. Very low rates of hip fracture in Beijing, People's Republic of China: the Beijing Osteoporosis Project. *Am. J. Epidemiol.*, 144: 901-907.
103. **Hegsted, D.M.** 1986. Calcium and osteoporosis. *J. Nutr.*, 116: 2316-2319.
104. **Gallagher, J.C., Melton, L.J., Riggs, B.L. & Bergstrath, E.** 1980. Epidemiology of fractures of the proximal femur in Rochester, Minnesota. *Clin Orthop.*, 150: 163-171.
105. **Maggi, S., Kelsey, J.L., Litvak, J. & Heyse, S.P.** 1991. Incidence of hip fractures in the elderly. A cross-national analysis. *Osteoporos. Int.*, 1: 232-241.
106. **Feskanich, D., Willett, W.C., Stampfer, M.J. & Colditz G.A.** 1996. Protein consumption and bone fractures in women. *Am. J. Epidemiol.*, 143: 472-479.
107. **Nordin, B.E.C.** 1997. Calcium in health and disease. Food, Nutrition and Agriculture, 20: 13-24.
108. **Aaron, J.E., Gallagher, J.C., Anderson, J., Stasiak, L., Longton, E.B. & Nordin, B.E.C.** 1974. Frequency of osteomalacia and osteoporosis in fractures of the proximal femur. *Lancet*, 2: 229-233.
109. **Aaron, J.E., Gallagher, J.C. & Nordin, B.E.C.** 1974. Seasonal variation of histological osteomalacia in femoral neck fractures. *Lancet*, 2: 84-85.
110. **Baker, M.R., McDonnell, H., Peacock, M. & Nordin, B.E.C.** 1979. Plasma 25-hydroxy vitamin D concentrations in patients with fractures of the femoral neck. *Br. Med. J.*, 1: 589.
111. **Morris, H.A., Morrison, G.W., Burr, M., Thomas, D.W. & Nordin, B.E.C.** 1984. Vitamin D and femoral neck fractures in elderly South Australian women. *Med. J. Aust.*, 140: 519-521.
112. **Von Knorring, J., Slatis, P., Weber, T.H. & Helenius, T.** 1982. Serum levels of 25-hydroxy vitamin D, 24,25-dihydroxy vitamin D and parathyroid hormone in patients with femoral neck fracture in southern Finland. *Clin. Endocrinol.*, 17: 189-194.
113. **Pun, K.K., Wong, F.H. & Wang, C.** 1990. Vitamin D status among patients with fractured neck of femur in Hong Kong. *Bone*, 11: 365-368.

114. **Lund, B., Sorenson, O.H. & Christensen, A.B.** 1975. 25-hydroxycholecalciferol and fractures of the proximal femur. *Lancet*, 2: 300-302.
115. **Chapuy, M.C., Arlot M.E. & Duboeuf, F.** 1992. Vitamin D₃ and calcium to prevent hip fractures in elderly women. *N. Engl. J. Med.*, 327: 1637-1642.
116. **Boland, R.** 1986. Role of vitamin D in skeletal muscle function. *Endocr. Revs.*, 7: 434-448.
117. **Walser, M.** 1961. Calcium clearance as a function of sodium clearance in the dog. *Am. J. Physiol.*, 200: 769-773.
118. **Nordin, B.E. & Need, A.G.** 1994. The effect of sodium on calcium requirement. *Advances in Nutritional Research. Volume 9 Nutrition and Osteoporosis.* Draper, H.H. ed. p.209-230. New York: Plenum Press.
119. **Goulding, A. & Lim, P.E.** 1983. Effects of varying dietary salt intake on the fasting excretion of sodium, calcium and hydroxyproline in young women. *NZ Med. J.*, 96: 853-854.
120. **Sabto, J., Powell, M.J., Breidahi, M.J. & Gurr, F.W.** 1984. Influence of urinary sodium on calcium excretion in normal individuals. *Med. J. Aust.*, 140: 354-356.
121. **Kleeman, C.R., Bohannon, J., Bernstein, D., Ling, S. & Maxwell, M.H.** 1964. Effect of variations in sodium intake on calcium excretion in normal Humans. *Proc. Soc. Exp. Bio. (NY)*, 115: 29-32.
122. **Epstein, F.H.** 1968. Calcium and the kidney. *Am. J. Med.*, 45: 700-714.
123. **Goulding, A. & Campbell, D.** 1983. Dietary NaCl loads promote calciuria and bone loss in adult oophorectomized rats consuming a low calcium diet. *J. Nutr.*, 113: 1409-1414.
124. **McParland, B.E., Goulding, A. & Campbell, A.J.** 1989. Dietary salt affects biochemical markers of resorption and formation of bone in elderly women. *Br. Med. J.*, 299: 834-835.
125. **Need, A.G., Morris, H.A., Cleghorn, D.B., DeNichilo, D., Horowitz, M. & Nordin, B.E.C.** 1991. Effect of salt restriction on urine hydroxyproline excretion in postmenopausal women. *Arch. Internal Med.*, 151: 757-759.
126. **Elliott, P., Stamler, J., Nichols, R., Dyer, A.R., Stamler, R., Kesteloot, H. & Marmot, M.** 1996. Intersalt revisited: further analyses of 24 hour sodium excretion and blood pressure within and across populations. *Br. Med. J.*, 312: 1249-1253.
127. **Hegsted, M. & Linkswiler, H.M.** 1981. Long-term effects of level of protein intake on calcium metabolism in young adult women. *J. Nutr.*, 111: 244-251.
128. **Margen, S., Chu, J-Y., Kaufmann, N.A. & Callow, D.H.** 1974. Studies in calcium metabolism. I. The calciuretic effect of dietary protein. *Am. J. Clin. Nutr.*, 27:584-589.
129. **Linkswiler, H.M., Zemel, M.B., Hegsted, M. & Schuette, S.** 1981. Protein-induced hypercalciuria. *Federation Proc.*, 40: 2429-2433.
130. **Heaney, R.P.** 1993. Protein intake and the calcium economy. *J. Am. Diet. Assoc.*, 93: 1259-1260.
131. **Kerstetter, J.E. & Allen, L.H.** 1989. Dietary protein increases urinary calcium. *J. Nutr.*, 120: 134-136.
132. **Nordin, B.E.C., Morris, H.A., Need, A.G. & Horowitz M.** 1993. Dietary calcium and osteoporosis. *Second WHO Symposium on Health Issues for the 21st Century: Nutrition and Quality of Life.* Kobe.
133. **Schuette, S.A., Zemel, M.B. & Linkswiler, H.M.** 1980. Studies on the mechanism of protein-induced hypercalciuria in older men and women. *J. Nutr.*, 110: 305-315.

134. **Schuette, S.A., Hegsted, M., Zemel, M.B. & Linkswiler, H.M.** 1981. Renal acid, urinary cyclic AMP, and hydroxyproline excretion as affected by level of protein, sulfur amino acid, and phosphorus intake. *J. Nutr.*, 111: 2106-2116.
135. **Need, A.G., Horowitz, M. & Nordin, B.E.C.** 1998. Is the effect of dietary protein on urine calcium due to its phosphate content? *Bone*, 23(Suppl):SA344.
136. **Mellanby, E.** 1918. The part played by an "accessory factor" in the production of experimental rickets. A further demonstration of the part played by accessory food factors in the aetiology of rickets. *J. Physiol.*, 52: 11-53.
137. **Telfer, S.V.** 1926. Studies in calcium and phosphorus metabolism. *Q. J. Med.*, 20:1-6.
138. **Fairweather-Tait, S., Prentice, A., Heumann, K.G. et al.** 1995. Effect of calcium supplements and stage of lactation on the calcium absorption efficiency of lactating women accustomed to low calcium intakes. *Am. J. Clin. Nutr.*, 62:1188-1192.
139. **Prentice, A., Jarjou, L.M.A., Stirling, D.M., Buffenstein, R. & Fairweather-Tait, S.** 1998. Biochemical markers of calcium and bone metabolism during 18 months of lactation in Gambian women accustomed to a low calcium intake and in those consuming a calcium supplement. *J. Clin. Endocrinol. Metab.*, 83: 1059-1066.
140. **Dibba, B., Prentice, A., Ceesay, M., Stirling, D.M., Cole, T.J. & Poskitt, E.M.E.** 2000. Effect of calcium supplementation on bone mineral accretion in Gambian children accustomed to a low calcium diet. *Am. J. Clin. Nutr.*, 71: 544-549.
141. **Aspray, T.J., Prentice, A., Cole, T.J., Saw, Y., Reeve, J. & Francis, R.M.** 1996. Low bone mineral content is common but osteoporotic fractures are rare in elderly rural Gambian women. *J. Bone Miner. Res.*, 11: 1019-1025.
142. **Bucher, H.C., Guyatt, G.H. & Cook, R.J.** 1996. Effect of calcium supplementation on pregnancy-induced hypertension and preeclampsia. *JAMA*, 275:1113-17.
143. **Garland, C.F., Garland, F.C. & Gorham, E.D.** 1991. Can colon cancer incidence and death rates be reduced with calcium and vitamin D? *Am. J. Clin. Nutr.*, 54(Suppl):193s-201s.
144. **McCarron, D.A.** 1997. Role of adequate dietary calcium intake in the prevention and management of salt-sensitive hypertension. *Am. J. Clin. Nutr.*, 65(Suppl): 712s-716s.
145. **Joffe, G.M., Esterlitz, J.R., Levine, R.J., Clemens, J.D., Ewell, M.G., Siba, I.B.M. & Catalano, P.M.** 1998. The relationship between abnormal glucose tolerance and hypertensive disorders of pregnancy in healthy nulliparous women. Calcium for Preeclampsia Prevention (CPEP) Study Group. *Am. J. Obstet. Gynecol.*, 179:1032-1037.
146. **Martinez, M.E. & Willett, W.C.** 1998. Calcium, vitamin D, and colorectal cancer: a review of the epidemiologic evidence. *Cancer Epidemiol. Biomarkers Prev.*, 7: 163-168.
147. **Resnick, L.M.** 1999. The role of dietary calcium in hypertension: a hierarchical overview. *Am. J. Hypertens.*, 12: 99-112.
148. **DerSimonian, R. & Levine, R.J.** 1999. Resolving discrepancies between a meta-analysis and a subsequent large controlled trial. *JAMA*, 282: 664-670.
149. **Mobarhan, S.** 1999. Calcium and the colon: recent findings. *Nutr. Revs.*, 57: 124-126.
150. **McCarron, D.A. & Reusser, M.E.** Finding consensus in the dietary calcium-blood pressure debate. *J. Am. Coll. Nutr.*, 18(Suppl): 398S-405S.