METHUEN'S MONOGRAPHS ON BIOCHEMICAL SUBJECTS

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CALCIUM METABOLISM

TO JANET

Preface

When professor of physiology in the University of Cape Town, I gave advanced lectures on calcium metabolism and they form the basis of this monograph. As would be expected in such a large field, much has had to be curtailed and much omitted, but the important highlights, as I see them, have been stressed and an attempt has been made to portray as all round a picture as possible. References have been given to important works in specialized aspects for further reading.

In discussing calcium, it is obvious that phosphorus comes often into the picture, since the two elements exist in combination in bone and have a close relationship in blood; problems of phosphorus metabolism have therefore been mentioned where these have impinged on those of calcium. No attempt has been made to deal with the clinical aspects of calcium metabolism, save when these illustrate physiological principles, as in rickets and diseases of the parathyroid glands. For information on the clinical side, the reader should consult works by authors such as Albright.

A well-known biochemist once told me that he found mineral metabolism most difficult to follow. Seeing that his research field was in the porphyrins, I felt a certain mutual sympathy. I hope he, and others who feel like him, will find, if they read the following pages, that the problems, though manifest, are not incomprehensible.

J. T. IRVING

Johannesburg
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Contents

CHAPTER I

Introduction

CALCIUM is the most important inorganic element in the body, and occurs in it in the highest amount. It is somewhat surprising that in human diets, at any rate, the supply is so exiguous. The highest Ca content of foods is of course in bone, and it is a regular agricultural practice to feed farm animals on bone-meal. This dietary practice has never appealed to civilized man, who has to rely on dairy products and certain green vegetables for his supply of Ca. In spite of this small distribution in the diet, Ca deficiency is not common, and the reason for this will be explained in more detail later.

The form in which Ca occurs in foods has not been much investigated. In milk it is probably associated in part with the protein fractions, and in vegetables it must in part be in a simple soluble form since some of it is easily leached out by water during cooking.

In the intestine an acid reaction is necessary for the solution and absorption of Ca. Thus $Ca_3(PO_4)_2$, insoluble at neutral reaction, is readily available for utilization in the body. However, in human studies there is little description of Ca deficiency due to achlorhydria, though this has been caused experimentally. The degree of absorption of Ca is difficult to determine, owing to the concurrent processes of secretion of digestive juices containing Ca, and the possible active excretion of Ca in addition. Taking the total balance of intake and output into account, one finds a very low figure of retention in humans, but a considerably higher one in animals like rats.

The absorption of Ca is governed by a number of factors, the chief of which is vitamin D. It seems probable that the degree of absorption is also controlled by the needs of the moment, especially during the growing period, though this has not yet been proved.

After absorption, Ca can exist in at least three forms in the body—Ca ions in solution, Ca bound to organic compounds, and Ca present in a crystal lattice as in bone. A close equilibrium exists between these three forms, governed by the endocrine glands and the dietary status, and obeying the rules of physical chemistry as closely as most biological reactions.

The excretion of Ca is controlled by factors still incompletely understood. The faecal Ca in man consists largely of unabsorbed dietary Ca, together with an unknown amount of secreted and actively excreted Ca. The kidney excretes Ca continually, even during starvation, but this may be due to the fact that the kidney tubule can deal with only certain types of Ca compounds. The urinary production of Ca is controlled by several endocrine glands.

The time-honoured method of investigating Ca metabolism is by the metabolic balance experiment. This involves an accurate knowledge of Ca intake and excretion and is tedious and often unsatisfactory to carry out. Errors can easily creep in and the full co-operation of the subject is essential. When errors do occur, they usually cause a fictitiously high positive balance of the element under consideration. As a result many writers have condemned balance methods, but unfortunately they have substituted nothing better in their place. The more recent use of radioactive isotopes has given us another tool which can be used freely in animal experiments, but which has naturally been sparingly used with human subjects.

Tetany was first described by Steinheim over 100 years ago and Gley [1] in 1893 recognized the role of the parathyroids as regulators of Ca metabolism. In 1918 Osborne and Mendel [2], working with rats, found that while the body could adapt to deprivation of most mineral elements, it was unable to adapt, either by substituting other elements or conserving what it had, to absolute Ca deficiency. The question of dietary Ca requirements was first put on a quantitative basis by Sherman in 1920 [3], and while his premisses were not in all cases correct, he started an aspect of the problem which is not yet solved. H. H. Mitchell and his school have thoroughly probed the question of utilization and retention of Ca and the effects of the body stores upon these [4].

The role of Ca in bone structure has been investigated with success by biologists working in collaboration with crystallographers, especially by Hodge and his colleagues and by Dallemagne. During the last two decades our knowledge of the physiology and biochemistry of bone formation has greatly advanced; the metabolic changes in calcification appear to follow the carbohydrate phosphorylytic cycle found in other parts of the body [5]. Much remains to be sorted out, especially the function of phosphatase. Bone formation has also been studied from the point of view of both endocrine and dietary influences [6, 7].

The teeth, until recently the Cinderellas of the calcified tissues, are now receiving their due share of attention, largely in an attempt to solve the caries problem. Apart from this, much fundamental work has appeared from Schour and his colleagues in Chicago, from the Bethesda group, and from other workers such as Bevelander, Armstrong and Sognnaes.

These aspects will be expanded in the following pages. And further it will be seen that Ca, although an apparently inert mineral element, undergoes some surprising adventures between its advent in the mouth and its final excretion.

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CHAPTER II

Content and Availability of Calcium in Foods

THE Ca content of common foodstuffs has now been widely determined and many tables giving the composition of foods have been published. The best source of Ca in human diets is milk or milk products excluding butter. Cows' milk contains about 0·12 per cent. of Ca and goats' milk has slightly more. Skimmed milk, liquid or dried, is just as good a source of Ca as whole milk. Cheddar cheese contains about 800 mg. Ca per 100 g. and thus is a very good source of Ca, but acid types of cottage cheese have only about 1/9th of the Ca content of Cheddar cheese. Human milk contains far less Ca than cows' milk. Leitch [1] has surveyed the literature on this and reported figures ranging from 0·013 to 0·04 per cent., the Ca content being to some extent modified by the diet of the mother. It is an interesting coincidence, as pointed out by Bunge [2], that the Ca content of the milk has a relationship with the rate of growth of the young, as these figures show:

Species	*Days to double birth weight*	*Ca % in mother's milk*
Man	180	0·02
Cow	47	0·12
Dog	7	0·32

The other main dietary source of Ca is green vegetables, and especially the outer green leaves, which may contain as much as twenty to thirty times as much Ca as the inner ones [3]. The methods of growing vegetables can influence their composition, artificial fertilizers tending to lower the Ca and raise the K content [4], the Ca possibly being replaced by some other element.

McCance and his colleagues [5] have analysed a number of vegetable foodstuffs. The most interesting conclusions are those relating to the effects of methods of preparation and cooking on the final Ca content of the food. They investigated the effects of steaming, boiling, baking, frying and roasting on the common constituents of vegetables. The only procedure affecting Ca is boiling. Boiling scarlet runner beans for 40 minutes and carrots for 120 minutes caused a loss of between 12 and 20 per cent. of the Ca in the cooking water. This was much less than the loss of, for example, Cl and K, of which over 60 per cent. were dissolved in the case of beans. Addition of alkali to the water made no difference to the loss. A more recent American study [6] showed that cooking cabbage in water resulted in a loss of over 20 per cent. of Ca by leaching, but with pressure cooking only 9 per cent. was lost. Averaging the results from eleven vegetables investigated, the same was found, nearly 25 per cent. of Ca being lost if the vegetable was covered with cooking water, and much less when the water was considerably reduced. It must be admitted that the Ca content of many of these vegetables is so low that these losses make little difference to the daily dietary intake. The outer leaves of cabbage contain 0·429 per cent., but carrots have only 0·045 per cent. and beans 0·055 per cent.

Several foods now have Ca added to them, the commonest being bread. In England, chalk is added to the flour to overcome the action of phytic acid, and in the U.S.A. dried milk and improvers are added, considerably increasing the Ca content. Prouty and Cathcart [7] found that the Ca content of fresh bread in the U.S. might vary from 0·065 per cent. to 0·141 per cent., six slices of bread giving 30 per cent. of the daily requirement, supposing all the Ca to be available.

One can summarize by saying that the bulk of our Ca intake comes from milk or milk products, taken either as such or in various cooked foods. The Ca from vegetable sources provides only a small fraction of the total daily intake.

Availability of Ca in foodstuffs. A knowledge of this is more important biologically than that of the Ca content in total amount, since it may be that the Ca in the given foodstuff is not in a condition to be nutritionally useful. This is especially true in the case of spinach and other chenopodiaceae, the Ca of which is in the form of oxalate and quite unavailable for use in the body. Spinach contains so much oxalate that it may also precipitate and render useless Ca in other constituents of the diet. In fact, Sherman [8] says that 'spinach was a mistaken choice for popularization as a typical green-leaf vegetable'. It will take some time before the feats of the seaman associated with this vegetable are forgotten.

The dipyridyl method can be used for testing the availability of iron, but no such procedure is available for Ca and rather tedious metabolic balance experiments have to be employed. Many experiments have been done on animals, especially calves and rats. Milk Ca is taken as the reference standard and the availability of other forms of Ca compared with it. Almost all inorganic forms of Ca seem to be available to some degree, such as carbonate, sulphate, phosphate and even silicate [9]. Ca lactate is also a good form of Ca and supported all biological functions in rats, except growth, as effectively as the Ca of milk [10].

As already stated, the Ca of spinach is not available, and blanching removed very little of it, the Ca being quite insoluble and not extractable. The Ca of cauliflower and broccoli is not as available as that of milk [11], nor is that of carrots, fresh lettuce or beans. Cooking of carrots or beans did not alter the availability [12].

These animal experiments have given useful information, but owing to species differences cannot necessarily be directly transferred to man. In general it has been found that Ca from inorganic sources is used as effectively by humans as that from milk, and the same applies to Ca gluconate or lactate [13, 14]. Aykroyd and Krishnan [15] gave Ca lactate in a large-scale experiment to school children in India and reported a significant improvement in weight and growth. The availability of inorganic salts appears to depend on their solubility. Steggerda and Mitchell [16] found that the availability of Ca to man from dried-milk solids or homogenized milk was the same as that from whole milk. Cheese Ca was as good as that from milk [17].

As far as vegetable Ca is concerned, as a rule this Ca is not so available as milk Ca. Breiter *et al.* [18] found that the utilization of carrot Ca was on an average 13·4 per cent., which is lower than the usual figure for milk (about 20 per cent.). The figures they give also illustrate the drawbacks of relying on vegetables as the main source of Ca. To obtain 202 mg. of Ca, 700 g. of carrots had to be eaten, this giving only about one-quarter of the daily requirement. On the other hand, lettuce Ca was even better than milk Ca when fed in amounts giving the same Ca intake, but here again the Ca content is so low (33 mg. per 100 g.) that, like most vegetables, lettuce could only be used as a subsidiary source of dietary Ca.

Pasteurization of milk. A great deal of controversy has raged over this process, and unfortunately considerations other than scientific ones have at times been used as arguments. As far as Ca is concerned, its availability is not very much affected. Kramer *et al.* [19] found in children and adults that the Ca of raw milk was better retained than that of pasteurized milk. Ellis and Mitchell [20] put rats on to diets of suboptimal Ca content and found that 98 per cent. of the Ca of raw milk was retained, and 92 per cent. of that of pasteurized milk. On the other hand, Henry and Kon [21] and Auchinachie [22] carried out simultaneously and independently balance experiments on rats similar to those of Ellis and Mitchell and found no differences in the availability of Ca from these two sources. Experiments undertaken by Crichton and Biggar [23] showed no differences whatever in the nutritional status of calves fed on either type of milk. Steggerda and Mitchell found in man [24] that heating milk to 160° F. for 30 minutes did not affect the utilization of the Ca. It seems therefore unlikely that pasteurization materially affects the nutritional value of Ca in milk, and if any slight change occurs, this is more than counterbalanced by the general improvement in the milk.

Water as a source of Ca. Hardness in water is expressed in a variety of ways, usually as parts of $CaCO_3$ per 100,000 or 1,000,000. This assessment makes no distinction between the various salts which can cause hardness, and may be bicarbonates, sulphates or chlorides of Ca, Mg and other metals. For this reason it is very difficult to assess water as a source of Ca. Soft water with a hardness of 15 parts of $CaCO_3$ per million is obviously a poor source of Ca since it contains only 0·6 mg. per 100 ml. (if the hardness is all Ca), and a daily intake of, say, 4 pints would give only 14 mg. Fairly hard waters would give on this basis a daily intake of about 200 mg. [25]. Really hard waters such as are found in the Kenhardt district of South Africa [26], with a total hardness of 1,000 parts of $CaCO_3$ per million, would give 900 mg. per day if 4 pints were consumed. Walker [27] has calculated from figures available in South Africa that the Ca contributed by the drinking water is negligible in the case of one-third of the population; that half of the population get about 60 mg. daily from water; and that with less than one-sixth of the population the Ca ingested from the drinking water is from 100 to 300 mg. per day. Except in the case of the latter group, the Ca from drinking water makes an insignificant contribution to the daily Ca requirement, and in the case of the last group it is not known if the hardness is all due to Ca or how available this Ca is. It thus appears wise to disregard water as a source of Ca till more knowledge is forthcoming. Hutchison [28] in his well-known textbook considers water to be negligible in this respect.

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