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Intracellular Magnesium Crystallized in the Arteries and Myocardium: New Chemical–Physical Hypothesis

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Detailed monitoring of Mg in the body should reveal five interacting chemical pools: pool 1 with the free, ionic, and most mobile Mg in chemical equilibrium with the others; pool 2 with the Mg salts precipitated mainly in the arteries and myocardium; pool 3 and 4, two different reservoirs of Mg; pool 5 with the physiologically bound Mg. The pool size needs to be determined exactly. The Mg content of the mitochondria in the arteries and myocardium is less than the content of K and greater than the content of Ca. These elements are in close equilibrium with each other and with PO_4^{3-}. Only within certain limits can the body adjust itself to the interdependent changes of these ions; it is, therefore, important that their combined presence, rather than Mg alone, be quantitatively determined.

When the solubility products of $Mg_3(PO_4)_2.8H_2O$ or of $MgKPO_4$ are reached, the two salts precipitate inside the cells, seeding further irreversible crystallization of Ca–Mg–K. TriMg phosphate nucleates triCa phosphate and the two cause the sarcoma to be filled with tightly packed crystals. On the inside surface of the membrane electromagnetic interactions exist with the polar salts in solution, which form a kind of fluid layer adherent to the membrane. Crystallization of $MgKPO_4$ occurs solely at this interface, where the K_{sp} is reached. The salt becomes polarly coupled with the membranes and, together, they favor successive epitaxial crystallization. After several decades the entire arterial wall is "calcified" and arteriosclerosis is in a well advanced stage; myocardial scars and other cardiovascular lesions should originate in a similar way.

Introduction

Magnesium is highly reactive; it is the only technological metal displacing hydrogen from boiling water (Appendix 1). Since its presence is widespread, it is difficult to accept some of the reports on its non-pathological deficiency in the body (Seelig, 1980). Quite the contrary, the results of Lee & Britton (1980), and of Lee, Britton & Rowland (1980) on chickens, prove that dietary Mg can be toxic.

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Mg follows Ca, K, and Na in terms of abundance in the body. Its concentration inside the cells, however, is second only to K and amounts to 10 times more than the extracellular concentration (Aikawa, 1976). The normal serum or plasma Mg is 2·5 mg/dl (0·885 mmol/l) (Flink, 1976); such an amount varies little with age and sex. The average of the 21 values in infancy listed by Seelig (1980) in her Table A-2B is 2·09 mg/dl; while Keating *et al.* (1969) found a Mg increase of only 4·5% in passing from the average 20 year old to the 70 year old.

It has been suggested that multiple Ca–Mg mixed apatites are the chemical and physical cause of atherosclerosis, of calcification and of calcinosis in the arteries (Boldrini, 1981). This is supported by the chemical-physical laws governing crystallization of the multiple mixed apatites, by their solubility products and by the pathological findings that Mg increases with the degree of atherosclerosis (Buck, 1951; Crawford & Crawford, 1967; Yu, 1973). These findings indicate that biological Mg is much more important than the essential elements: Co, Cr, Cu, Fe, etc.

"There is a need for serious investigation of the problem (of Mg) by chemists to help in the formulation of hypotheses and guide to further research" (Crawford & Crawford, 1967). This note aims at offering such a hypothesis, limited to the chemistry and physics of Mg not to its physiology and pathology, for which there are four recent books: Aikawa, Cantin & Seelig, Seelig, and Wacker.

Pools of Biological Magnesium†

About 3000 papers in 50 years show that the biology of Mg cannot be studied in a satisfactory way only by distinguishing Mg as ultrafiltrable or tightly bound, neither by the various other classifications (Aikawa, 1976; Nordin, 1976). These all fail to pinpoint which form of Mg is responsible for the arterial deposits, how much of it is involved, where in the cellular tissue it is precipitated and why it is there.

A better understanding is achieved by subdividing Mg into five pools (Fig. 1).

Pool 1 is the input pool; all Mg of the body at one time or another passes through it, irrespective of its final destination. This pool contains the most soluble, mobile, and dissociated Mg, partially surrounded by water of hydration. The content of this pool is in irreversible chemical equilibrium

† Definition of pool from the New Collegiate: "The whole quantity of a particular material present in the body and available for function or the satisfying of metabolic demands". In the paper we speak of biochemical pools or of chemical ones; these pools are hypothetical as far as their size is concerned, which has to be determined.

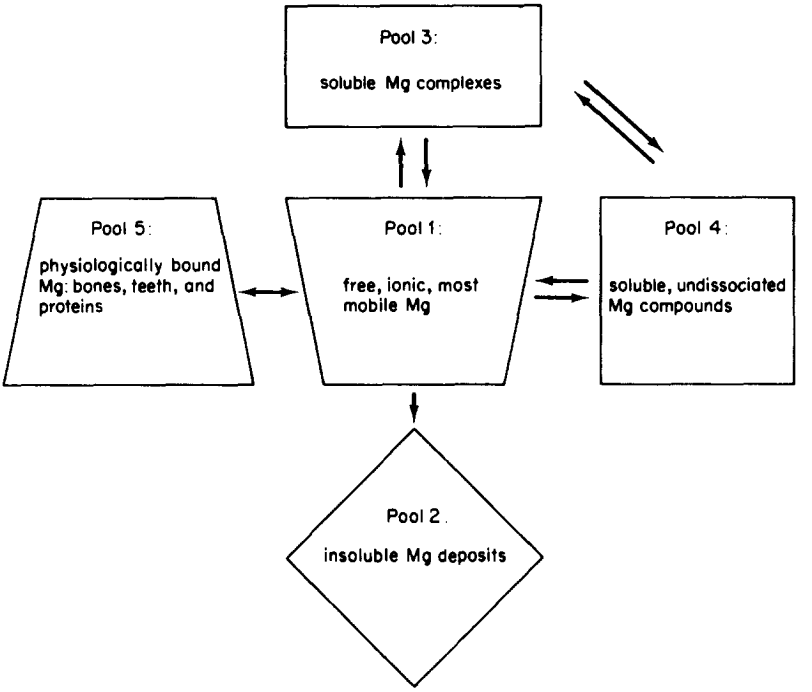

FIG. 1. The five Mg pools of the body; *pool 1* is the pool where the free, ionic and most mobile Mg is located; in *pool 2* is the crystalline Mg precipitated mainly in the arteries and myocardium; *pool 3* has the Mg complexes; *pool 4* is constituted by the undissociated Mg salts; *pool 5* is the pool with the physiologically bound Mg of bones and proteins. Pool 1 is in chemical reversible equilibrium with pools 3 and 4, while it is in irreversible equilibrium with pool 2. The double headed arrow between pools 1 and 5 indicates the physiological equilibrium existing between the two pools. Pools 3 and 4 communicate with each other.

with the content of pool 2, it is in reversible equilibrium with pools 3 and 4, but it is only in physiological equilibrium with pool 5 (Boldrini, 1982). Mg of pool 1 may be directly converted into any other combination of chemical importance.

Pool 2 contains the insoluble Mg deposits described by Buck (1951); Crawford & Crawford (1967) and Yu (1973). Unwanted Mg is crystallized in the arteries and myocardium; this crystallization is quite different from the "mineralization" of bones. Among other things, it is a chemical phenomenon and as such may be studied strictly according to the chemical laws. The physical association of these crystallized salts with the cell membranes will be considered in this note, as well as the chemical-physical aspects of it. Introduction of pool 2 should help physiologists to understand

why plasma Mg may not directly relate to the Mg salts already deposited in arteries, kidneys, myocardium, spleen, etc. The latter deposits are permanent, the former is constantly renewed.

Pools 3 and 4 are two transition pools, both operating as chemical-biological reservoirs. Pool 3 contains the complexes with amino acids, citric acid, urea, adenosine phosphate, other polyphosphates, metal salts, etc. Pool 4 includes the soluble but undissociated Mg salts (Boldrini, 1982). Mg does not precipitate directly from these pools, unless it is first ionized and then transferred into pool 1. Pools 3 and 4, however, communicate with each other (Fig. 1) by allowing some of their components to pass in both directions when variations of their chemical environment occur, e.g. the undissociated citrates in the presence of excess of citric acid behave more like complexes and partially move from pool 4 into pool 3. The presence of other substances, the pH, the temperature, and the disease conditions also influence the equilibrium between pool 3 and 4. The chemical distinction between pool 3 and 4 allows a full monitoring of the metal, although physiologically pools 3 and 4 are practically identical.

Pool 5 is the physiological output pool; it contains the biologically and physiologically bound Mg of bone, teeth, protein, etc. From this pool Mg is not directly available for the chemical reactions described in this note, nor can it be mobilized by chemical reactions only. It is, of course, recognized that Mg is physiologically released from pool 5 into pool 1. This note, however, considers Mg only at its arrival in pool 1 and is not concerned with the modality of its transfer. It will not be speculated whether or not pool 5 has any relation with the others, apart from pool 1.

The distribution of Mg among the various pools is calculated in Table 1(b) and 1(c) from Table 1(a). The size of pool 1 has been deduced from the suggestion of many investigators that less than 1% of mobile Mg is in a very mobile, ionic condition, as well as by taking into account the rate of exchange of intra- and extra-cellular Mg. Table 1(b) represents only the ideal situation, when pool 2 is empty. This changes as soon as Mg starts to supersaturate the arteries and myocardium; this might even happen in infants (Monif & Savory, 1972). Thus, in Table 1(c) a comparison is made between ideal and possible Mg distributions. The third and fourth columns have been computed assuming that pools 1, 3, and 4 do not change throughout one's life. Another assumption is that a total of 1 g of Mg may accumulate in the arteries and myocardium until we reach 50 years; then, from 50 years onwards Mg may crystallize in pool 2 at an average rate of about 1 mg/day.

Several circumstances may influence the equilibria among the pools. In uremia conditions, pool 3 can be from 20 to 40% bigger than the estimate

TABLE 1

The overall distribution of 24·32 g *of* Mg (1 mole) *in human body of* 70 kg

(a) Percentage of the distribution, according to Aikawa (1976):

60% in bones;	10% in soft tissues
29% in muscles;	1% extracellular

(b) Proposed percentage among the various pools, taking into account that a third of the non-bones Mg is protein bound. This distribution is the ideal one only, whereby pool 2 contains no precipitate at all.

73% (pool 5)	60%	in bones	25%	in muscles, soft tissues, intracellular, (pools 3 and 4)
	13%	protein bound; 9·7 muscles, 3 soft tissues, 0·3 extracellular	2%	in muscles, soft tissues, intracellular, (pool 1)

(c) Comparison between ideal Mg distribution in the body and possible distribution at 50 and at 75 years. If no Mg were accumulated in pool 2 before 50 years of age, but the total Mg of the body increased 4·5 times with age (Keating *et al.*, 1969), then at 75 we would have 10·12 g of Mg in pool 2.

Pools	Ideal Mg distribution (g)	Possible distribution at 50 years (g)	Possible distribution at 75 years (g)
5	17·75	16·75	8·63
3–4	6·08	6·08	6·08
2	0·00	0·001–1·00	9·12–10·12
1	0·49	0·49	0·49

of Table 1(c), because of the extra urea complexes (Jackson & Meier, 1968; Wallach *et al.*, 1966). As well, when soluble undissociated Mg salts are injected (e.g. Mg gluconate) they go mainly into pool 4; on the contrary, if $MgSO_4$ is injected, it goes into pool 1. One of the latter cases is presented by Monif & Savory (1972).

Insolubility of Biological Magnesium

While the similarity between Mg and Ca is well acknowledged, the resemblance between Mg and K is much less documented. Some of the mixed solid compounds given by K and Mg are shown in Table 2; their formation indicates the existence of affinity between the two cations. This is confirmed by the solubility properties exhibited in the body fluids. Though Mg salts are in general less soluble than those of K and more soluble than those of Ca, Table 3 reports some Ca, Mg, and K salts found in food and water which are exceptions to the rule. For instance, the solubility of $MgCl_2$

TABLE 2

Some of the mixed salts formed between potassium and magnesium (Handbook of Chemistry and Physics). *Their formation indicates chemical affinity between the two cations*

$KMgF_3$	$KHCO_3.MgCO_3.4H_2O$
K_2MgF_4	$K_2CrO_4.MgCrO_4.2H_2O$
$KCl.MgCl_2$	$K_2SeO_4.MgSeO_4.6H_2O$
$KCl.MgSO_4.3H_2O$	$K_2SO_4.MGSO_4.4H_2O$
$K_2SO_4.2MgSO_4$	$K_2SO_4.MgSO_4.6H_2O$

lies between that of the corresponding Ca and K salts, but $CaCl_2$ is the most soluble salt, with KCl being the least soluble. It can also be seen that $MgSO_4$ is the most soluble of the three analogues, while MgO is the least soluble of all. For these and other reasons, as is seen by considering the K_{sp}'s listed in Table 4, it is important to deal with the insolubility of biological Mg. All the Ca–Mg salts of Table 4 are involved in the precipitates of the arteries; the Mg salts, however, are more important in the intracellular precipitation considered in this note.

The insolubility of the three salts studied by Taylor, Frazier & Gurney (1963) will be considered further here, because of their peculiarity. $Mg_3(PO_4)_2$ has been described in the anhydrous† (Marshall, 1976) and in the hydrated form (Taylor *et al.*, 1963). The latter belongs to a series of natural compounds of general formula $X_3(YO_4)_2$, where Y = P or As, and X = Mg, Fe, Co, Ni. The fact that $Mg_3(PO_4)_2.8H_2O$ is a natural mineral indicates its thermodynamic stability; as well, it suggests the possibility of isomorphism within the series and polymorphism for the individual compound. Both these characteristics follow from the crystal structure of the material (Wyckoff, 1965). The solubility of both Mg phosphates (Table 4) is very similar; both of them precipitate in the body at the same time. The inorganic materials with a K_{sp} around 10^{-25} or below are so insoluble that their precipitates are irreversible. Such is $Mg_3(PO_4)_2$.

$MgKPO_4.6H_2O$ has the marked tendency to crystallize together with similar salts in solution (Pascal, 1958). It exhibits the same general characteristics given above for triMg phosphate, with the peculiarity of also being piezoelectric. For this reason, even though its insolubility is not as great as that of triMg phosphate, its importance is apparent at the membrane

† Seelig (1980) reports $Mg_3(PO_4)_2$ in a composite diagram, but never refers to it.

TABLE 3

A comparison of the solubility (g/100 cm^3) *of* Ca, Mg *and* K *salts which are in the waters and in food* (Handbook of Chemistry and Physics)

Salts	Ca (g)		Mg (g)		K (g)	
Chloride	75·5 (20°C)	159 (100°C)	54·25 (20°C)	72·7 (100°C)	37·7 (20°C)	56·7 (100°C)
Sulfate	0·209 (30°C)	0·1619 (100°C)	26 (0°C)	73·8 (100°C)	12 (25°C)	24·1 (100°C)
Oxide	0·131 (10°C)	0·07 (80°C)	62×10^{-5} (?)	86×10^{-4} (30°C)	very soluble	very soluble
Stearate	0·004 (15°C)	0·89 (80°C)	0·003 (15°C)	0·008 (50°C)	soluble	soluble
d-tartrate†	0·0266 (0°C)	0·0689 (37·5°C)	0·8 (18°C)	1·44 (90°C)	150 (14°C)	278 (100°C)

† $Ca = 4H_2O$; $Mg = 5H_2O$; $K = \frac{1}{2}H_2O$.

TABLE 4

The solubility product constants (K_{sp}) *calculated for some of the most insoluble simple* Mg *and* Ca *salts, those most likely to seed crystallization of the* Ca–Mg *mixed apatites in the arteries and in myocardium* (†from Taylor *et al.*, 1963; ‡from Marshall, 1976; §from Boldrini, 1982). *The temperature is indicated*

Salts	K_{sp}
$Mg_3(PO_4)_2.8H_2O$	$6{\cdot}3\times10^{-26}$ (25°C)†
$Mg_3(PO_4)_2$	$1{\cdot}25\times10^{-25}$‡
$Ca_3(PO_4)_2$	$1{\cdot}1\times10^{-21}$ (c.w.)§
$MgNH_4PO_4.6H_2O$	$7{\cdot}1\times10^{-14}$ (25°C)†
$MgNH_4PO_4$	3×10^{-13} (c.w.)§
$Mg(OH)_2$	$6{\cdot}7\times10^{-12}$ (c.w.)§
CaF_2	9×10^{-12} (c.w.)§
$MgKPO_4.6H_2O$	$2{\cdot}4\times10^{-11}$ (25°C)†

c.w. = cold water.

interface where ferroelectricity becomes a remarkable phenomenon of the arteries (Boldrini, 1980b). The same can be said of $MgNH_4PO_4$, which is isomorphous with the K equivalent. Precipitation of the K^+ or of the NH_4^+ salts on the membranes will depend entirely upon the local concentration of each ion.

In summary, even though the Mg salts have the reputation of being soluble, those of Table 4 belong to the most insoluble compounds. Unfortunately, up to now none of them has received the attention they deserve for being part of the intracellular materials. Their high insolubility cannot be altered from the chemical or pharmacological point of view and for these reasons the chemical equilibria where the compounds are formed are worthy of more detailed consideration. In studies with humans, Heaton, Hodgkinson & Rose (1964) showed that increasing dietary phosphates decreased Mg absorption. This further demonstrates formation of triMg phosphate and its insolubility already noticeable in the gastrointestinal tract.

Equilibria with the Orthophosphates

Some of the ions taking part in the biological equilibria of the body are:

$$Cl^-, SO_4^{2-}, HCO_3^-, PO_4^{3-}, F^-, OH^-, HPO_4^{2-}, \text{etc.} \quad (1)$$

$$Na^+, K^+, Ca^{2+}, Mg^{2+}, NH_4^+, Fe^{2+}, \text{etc.} \quad (2)$$

Each of these ions interacts with the others producing new chemical reactions, changes in the concentrations of the individual elements in solution, recycling back into the body of some of them, etc. Sometimes these interactions enhance the solubility of the dissolved electrolites because of complex formation (pool 3). Generally speaking, only the dissociated salts of pool 1 precipitate into pool 2, when their concentrations reach the solubility product. Such a precipitation has many origins, but it always obeys the laws of chemical equilibria, e.g. the various phosphates of plasma may come from inside or outside the body; in plasma, however, they transform easily into the tribasic phosphates (Appendix 2 of Boldrini, 1982). Such a transformation has consistently been overlooked, with the result that many exogeneous phosphates which are fed, go to increase the unwanted pool 2.

While the ions of (1) and (2) participate in the physiological activity of the arterial cells, only a few have the ability to precipitate with Mg in the intracellular fluids, namely:

$$Ca^{2+}, Mg^{2+}, K^+, NH_4^+, \text{and } PO_4^{3-}. \quad (3)$$

These are the ions which originate the nucleating reactions; the first of them is:

$$3A^2 + 2PO_4^{3-} \rightleftarrows A_3(PO_4)_2; \text{ with } A = \text{Mg or Ca}. \quad (4)$$

The ions in (4), although they are not the only ones of importance, constitute the basic chemical equilibrium of Mg and Ca in the body.

An example of close interaction between dietary Ca, Mg, and PO_4, and their equilibria in plasma is given by Berlyne, Bedrak & Yagil (1973). They monitored people who had been drinking from 7 to 20 l/day of special waters, not usually drunk anywhere in the world (with concentrations of dissolved solids higher than 2000 ppm). Their findings are tabulated in Table 5 where it can be seen that Grofit (1970) has by far the lowest Ca–Mg molarity, but at the same time also the highest PO_4^{3-} concentration. In contrast, Grofit (1971) has the highest Ca–Mg molarity, with the lowest PO_4^{3-} content. Further comparison between Grofit (1971) and (1970) is also informative. In the 1971 case, the K_{sp} of triMg phosphate is less than that of 1970, in spite of the fact that the Mg concentration found is exactly the average of the experimental values.

An important point, then, is to consider the ions of equilibrium (4) together and not each one separately, Grofit (1970) and (1971) (Table 5) once again indicate the close relationship between Ca, Mg, and PO_4^{3-}. In the last two columns of Table 5 we see that triCa phosphate never reaches

TABLE 5

Molar concentrations of Ca^{2+}, Mg^{2+} *and* PO_4^{3-} *in plasma of people usually drinking extremely hard mineral waters in quantities of* 7 to 20 l/day (*from* Table 2 *of* Berlyne *et al.*, 1973). *The first column gives the locality of the water supply and the year of the experiment. For each element, in addition to the molar concentration, its ratio with respect to* PO_4^{3-} *is given. The last two columns report the calculated solubility product constants for the concentrations of this table. It can be seen that* triCa *phosphate never reaches the solubility product and, therefore, can never precipitate in such a condition. The opposite is true for the* Mg *compound. For the reasons explained in the text, only* 1% *of each ion has been used in the calculations of the* K_{sp}'s *The last column has the four larger values greater than the* K_{sp} *of the compound, which then will precipitate*

Original of water	Ca/PO_4 molar ratio	Ca (mmol/ml)	Mg/PO_4 molar ratio	Mg (mmol/ml)	PO_4/PO_4 molar ratio	PO_4 (mmol/ml)	$Ca_3(PO_4)_2$ K_{sp}	$Mg_3(PO_4)_2.8H_2O$ K_{sp}
Sa'ad (1970)	7·6	$2{\cdot}5\times10^{-3}$	2·2	$0{\cdot}72\times10^{-3}$	1·0	$0{\cdot}32\times10^{-3}$	$1{\cdot}6\times10^{-24}$	$4{\cdot}0\times10^{-26}$
Ein Gedi (1971)	7·2	$2{\cdot}5\times10^{-3}$	2·6	$0{\cdot}89\times10^{-3}$	1·0	$0{\cdot}34\times10^{-3}$	$1{\cdot}8\times10^{-24}$	$8{\cdot}3\times10^{-26}$
Grofit (1970)	4·3	$2{\cdot}1\times10^{-3}$	1·7	$0{\cdot}83\times10^{-3}$	1·0	$0{\cdot}49\times10^{-3}$	$2{\cdot}1\times10^{-24}$	$13{\cdot}7\times10^{-26}$
Grofit (1971)	7·9	$2{\cdot}4\times10^{-3}$	2·8	$0{\cdot}84\times10^{-3}$	1·0	$0{\cdot}30\times10^{-3}$	$1{\cdot}3\times10^{-24}$	$5{\cdot}5\times10^{-26}$
Yotvatah (1970)	5·8	$2{\cdot}3\times10^{-3}$	2·1	$0{\cdot}83\times10^{-3}$	1·0	$0{\cdot}40\times10^{-3}$	$1{\cdot}8\times10^{-24}$	$8{\cdot}8\times10^{-26}$
Keturah (1971)	6·9	$2{\cdot}4\times10^{-3}$	2·5	$0{\cdot}86\times10^{-3}$	1·0	$0{\cdot}34\times10^{-3}$	$1{\cdot}6\times10^{-24}$	$7{\cdot}3\times10^{-26}$

its solubility product and thus does not precipitate in the intracellular fluids, while the last column indicates that four times out of six the K_{sp} of triMg phosphate is reached and this is the compound which has the highest probability of crystallizing in the arteries. Macroscopic errors have been introduced into the literature simply by starting from the erroneous hypothesis that triCa phosphate is the most insoluble compound of the body.

Last two columns of Table 5 were calculated on the assumption that half of pool 1 for the 3 ions in equilibrium (4) participate (about 1% of their total). The results of Berlyne *et al.* (1973) suggest that such an assumption is close to reality, as confirmed by different examples from Acharya & Payne (1965). In Table 6 we can see that the three children studied have

TABLE 6

Inorganic ions and urea in three children's plasma (1, 2, 3) *immediately after birth*

(a) Molar concentrations and ratios calculated from results of Acharya & Payne (1965)

	1		2		3	
	Ca/PO_4 molar ratio	Ca (mmol/ml)	Mg/PO_4 molar ratio	Mg (mmol/ml)	PO/PO_4 molar ratio	PO (mmol/ml)
K^+	26	$6{\cdot}20\times10^{-3}$	25	$5{\cdot}50\times10^{-3}$	20	$6{\cdot}10\times10^{-3}$
Ca^{2+}	10	$2{\cdot}32\times10^{-3}$	11	$2{\cdot}40\times10^{-3}$	10	$2{\cdot}27\times10^{-3}$
Mg^{2+}	3	$0{\cdot}63\times10^{-3}$	3	$0{\cdot}60\times10^{-3}$	4	$0{\cdot}84\times10^{-3}$
PO_4^{2+}	1	$0{\cdot}24\times10^{-3}$	1	$0{\cdot}22\times10^{-3}$	1	$0{\cdot}22\times10^{-3}$
Urea	226	$53{\cdot}70\times10^{-3}$	300	$66{\cdot}60\times10^{-3}$	562	$124{\cdot}0\times10^{-3}$

(b) The solubility product constants for the two pure apatites, in the Acharya cases (first two lines) and in the case of doubling of Ca or of Mg (last two lines)

	1	2	3
$K_{sp}: Ca_3(PO_4)_2$	$7{\cdot}4\times10^{-25}$	$7{\cdot}3\times10^{-25}$	$5{\cdot}9\times10^{-25}$
$K_{sp}: Mg_3(PO_4)_2$	$1{\cdot}5\times10^{-26}$	$1{\cdot}2\times10^{-26}$	$3{\cdot}0\times10^{-26}$
$K_{sp}: Ca_3(PO_4)_2$	$5{\cdot}8\times10^{-24}$	$5{\cdot}4\times10^{-24}$	$5{\cdot}5\times10^{-24}$
$K_{sp}: Mg_3(PO_4)_2$	$1{\cdot}2\times10^{-25}$	$0{\cdot}9\times10^{-25}$	$2{\cdot}3\times10^{-25}$

a plasma which does not reach the solubility product of the pure Ca and Mg apatites. The situation is not changed if we double the effective Ca concentration (Table 6(b)); on the other hand, doubling the Mg or the PO_4 concentration leads to precipitation of triMg apatite. This means that the newborn infant can tolerate a sudden rise in Ca content, but not of Mg or of PO_4, without incurring the danger of starting the intracellular buildup of pool 2. Of the three ions of (4), Mg^{2+} is the closest to saturation.

Nucleating Agents

In the myocardium, where the concentration of Mg is 2·5 times that of bones, there are no other Mg or Ca salts less soluble than $Mg_3(PO_4)_2$, the nucleolus (nucleus of a nucleus) of crystallization. $Mg_3(PO_4)_2$ is also the least soluble of the intracellular fluids of the arteries, where Mg concentration is 10 times greater than the extracellular one. In its turn, triMg phosphate seeds crystallization of triCa phosphate, and the two nucleate the other Ca–Mg–K salts, when the latter reach their saturation values (Table 7). Two of the additional seeds of intracellular crystallization are reported in the last two lines of Table 7.

TABLE 7

The ideal Ca/P, Ca/Mg *and* P/Mg *ratios of the crystalline deposits of the extracellular fluids (top two lines) and in the intracellular fluids of the arteries (bottom two lines). The middle line represents the possible precipitation in the cell membranes*

	Ca/P molar	Ca/Mg molar	P/Mg molar
$[Ca_3(PO_4)_2]_3.MgNH_4PO_4$	1·41/1	9/1	7/1
$[Ca_3(PO_4)_2]_3.CaF_2.MgNH_4PO_4$	1·7/1	10/1	6/1
$[Ca_3(PO_4)_2]_3.MgNH_4PO_4.MgKPO_4$	1·1/1	4·5/1	4/1
$[Mg_3(PO_4)_2]_3.[Ca_3(PO_4)_2]_3$	0·75/1	1/1	1·3/1
$[Mg_3(PO_4)_2]_3.[Ca_3(PO_4)_2]_3.CaF_2$	0·84/1	1·1/1	1·3/1

The molar ratios: Ca/P, Ca/Mg, P/Mg = 0·8, 1·0, 1·3, respectively, reported in the literature for the intracellular deposits represent the crystal as a whole, rather than the nucleus, which constitutes only $\frac{1}{10}$ or $\frac{1}{15}$ of the whole crystal. Table 7 gives the other ratios of intracellular crystallization and of the crystallization inside the membranes.

The chemical laws of extracellular crystallization are simpler than those just discussed (Boldrini, 1982). Pure Ca apatite is the nucleating agent there, seeding crystallization of the other Ca–Mg salts. Their molar ratios are those of the first two lines of Table 7, which are close to those of line 2–3 of Table 8a of Boldrini (1982). Extracellular crystallization is that of grade 1 and 3 of atherosclerosis. By comparing the last line of Table 7 with the first line of Table 8a of Boldrini (1982), we deduce that cases described as being of 0 grade atherosclerosis look like seeding stages of arteriosclerosis.

A special situation arises at the cell membranes. Since Ca–Mg salts are electrolytes, they can filter through the membranes in either direction; however, because of the opposite electrical sign of the two membrane surfaces, at their interfaces two divergent configurations appear. The polar salts of the $MgXPO_4$ type ($X = K$ or NH_4), are attracted and polarized by the membranes, but their concentration is higher and layer oriented on the inside of the membranes. The K_{sp} of the $MgXPO_4$ salts is then reached differently from those of the ions in equilibrium (4). Due to the electromagnetic nature of the phenomenon, crystallization on the inside of the membranes is a very selective process: only polar crystals are deposited in the successive slow phases of crystallization. Their probable molar ratios are those of the middle line of Table 7.

There are, then, particular seeds of crystallization, according to where the phenomenon takes place in the cells. Both the intracellular and the inside-the-membrane crystallization are extremely slow processes (of the order of decades), because the intracellular Ca–Mg–K equilibria are not easily influenced by dietary manipulations. The K content is also not homogeneous inside the cells, being higher in the vicinity of the membranes. For these reasons, intracellular crystallization is not comparable with that occurring in other parts of the body: gallbladder, kidneys, etc. What is most important is that intracellular crystallization (especially the inside-the-membrane process) constitutes the chemical and physical origin of arteriosclerosis.

One of the many examples of misinterpretation of Ca for Mg is documented in Fig. 9–12 of Heggtveit *et al.* (1964) showing "progressive filling of mitochondria with electron dense granules, presumably calcium". In that Fig. 9 the crystals shown actually have 'clear centers' of pure triMg phosphate; in Fig. 10 the proportions of Mg are probably still 35%, while in Figs 11 and 12 the internal Mg salts have been totally covered by triCa phosphate, as well as by other mixtures of Ca–Mg–K salts. The same Figs 9 and 10 clearly show that the Mg salt rather than the Ca salt is the nucleolus of crystallization; this can also be deduced by the authors' statement: "the dense granules are, at first, arranged as spheres". What Figs 11 and 12 show most clearly is that, unlike the inside of the crystals of Figs 9 and 10, their outside is made up of polycrystalline multicomponent crystal phases.

Seelig (1980) describes the risks of phosphate therapy. Unknowingly, she also gives the risks of Mg therapy (pp. 351–352). The patient who "had died of a massive infection the day after the phosphate infusion" would have met the same fate with an equimolar Mg infusion. In both cases $Mg_3(PO_4)_2$ is the nucleating agent.

The Danger of Soft Waters

Epidemiological evidence has been presented relating death from cardiovascular disease with the softness of the water supply in various countries (Seelig, 1980). No agreement, however, is reached in explaining why the two phenomena are so correlated. The problem is being studied from too narrow a point of view, often with insufficient, and sometimes with incorrect, data. Exclusive attention is paid to the contribution of water, neglecting other concurrently related factors such as the effect of Mg from the solid food supply. Table 5 shows that even by drinking from 7 to 20 l/day of the hardest waters, Ca cannot be made to precipitate while Mg can.

The most important consideration is the simultaneous equilibrium of Ca, Mg, K and PO_4, which determines whether or not water causes Mg crystallization in pool 2. With this equilibrium we may explain why "Cow's milk has Mg in reasonable amount, but the high phosphate and Ca content adversely affect the Mg utilization" (Flink, 1976). Mg in any insoluble form cannot be utilized by the body, a fact that holds for dietary as well as for intracellular Mg. This explanation is important in the connection between Mg in water and cardiovascular disease; yet the literature does not report chemical and physical comparisons of the ions of (3) and, therefore, cannot offer a unique cause–effect relationship. Indeed, investigators do not report the chemical methods used in determining Mg any longer, or whether or not they have individually analyzed the crystalline precipitates of the arteries separated from their tissue (Crawford & Crawford, 1967). The same holds for many analyses of water.

Few people are aware that the artificial softening of waters frequently decreases the Ca/Mg ratio, as a consequence of the fact that ion exchange methods proportionally eliminate 50% less Mg than Ca (Weber, 1972). A water with an initial Ca/Mg ratio of 10/1 in two exchanges would reduce it to 2·5/1. At the same time there would be a parallel increase of K, to partially compensate for the disappearance of the divalent cations. The overall result is that in soft waters the solubility product of $MgKPO_4$ is reached more frequently than in hard waters. This has never been pointed out before and is commented upon in the next sections.

Bearing in mind that some Mg salts are much more insoluble than $CaCO_3$, we may explain why they are also more corrosive: "Excessively soft waters are almost always corrosive whatever their pH value, because they do not contain enough $CaCO_3$ to deposit a protective film. Wherever the hardness of a water is less than 30 ppm (equiv. $CaCO_3$) the addition of lime is probably advisable. The effluents from base-exchange softening plants are 'dead-soft' and, even with a pH value above 7·0 they tend to be corrosive,

particularly if even a small amount of CO_2 is present" (Twort, 1963). My explanation is somewhat different. Both the above corrosive effects and the scars produced by the Mg salts in the myocardium have the same origin: the extreme insolubility of the Mg salts. These salts are not likely to be easily separated from their original matrix. Due to the high energy released during their precipitation, they tend to combine directly with the matrix in a chemical-physical way.

Referring to the municipal water supplies, it makes little sense to arbitrarily state: "The material (Mg) is not harmful to health but the U.S. Public Health Service recommends an upper limit of 125 p.p.m." (Twort, 1963). Nor does the opposite suggestions of indiscriminate dietary supplements of Mg make sense, on the wrong presumption of dietary deficiencies (Seelig, 1980). Until and unless soft waters are analyzed correctly for their Ca, Mg, K, and PO_4 contents, how can we definitely attribute to them negative effects (Marier, 1979)? Furthermore, the consequences of drinking soft water can be determined only after several decades. In conjunction with this, accurate chemical analyses of the crystalline precipitates in the arteries and myocardium are required (Heinrich, 1981).

Magnesium in the Myocardium

The rate of entry of Mg into the intracellular pools is about 1% of the rate of its removal from the extracellular pools by all routes (Wallach *et al.*, 1966). This indicates that intracellular Mg is the most constant and the least influenced by dietary manipulations or deprivation. The constancy of Mg is especially true in the myocardium, where cells accumulate proportionally more Mg in response to stimuli that cause cellular hypertrophy (such as mechanical constriction of the ascending aorta). Under such conditions there is also an increase in sequestered myofibrillar Mg (Seelig, 1980).

Heart mitochondria accumulate large amounts of Mg and of orthophosphates by transport across the inner membrane (Brierly, Bachman & Green, 1963). Because of the high mitochondrial and myofibrillar contribution, myocardium shows the highest Mg concentration of the body: 14 times the normal extracellular distribution (Table 8), supplied mainly by pools 1 and 3. Myocardium, then, has the potential of contributing heavily to pool 2. The biological importance of such contribution is that it seeds precipitation of Ca as well as of Mg.

The chemical-physical principles discussed previously concerning Mg–Ca crystallization are proven by many electron microscope pictures of myocardial cells. Unfortunately, the majority of the crystalline precipitates present in those pictures have generally been indicated as "granules", in connection

TABLE 8

Estimated content of Mg *in selected tissues* (Seelig, 1980)

Tissue	Mg (mg/dl)	(mmol/dl)
Myocardium	30·1	1·265
Spleen	21·7	0·895
Liver	21·4	0·880
Kidney	16·6	0·680
Skeletal muscle	14·4	0·590
Bone	12·1	0·495

with their spherical shape. In a few cases they have been suggested as probable Ca salts. Such a vague interpretation, which started over 50 years ago, lacks chemical credibility. No one has indicated which anions are present in those granules; above all, no one explained how Ca could precipitate in a solution which has 10 times more Mg and 80 times more K. Finally, no one indicates how and when Mg comes out of the cells in the presumed deficiency cases.

"In the face of this multiplicity of theories, the depletion of the intracellular ions, Mg^{2+} and PO_4^{3+}, representing early and consistent changes in the development of myocardial necrosis produced by two unrelated procedures, emerges as a finding which deserves closer scrutiny in view of the known significance of these two ions in vital enzymatic and energy processes" (Lehr *et al.*, 1966).

"The earliest descernible submicroscopical changes in myocardial cells consisted of swelling and degeneration of sarcomes and enlargement of the sarcotubular reticulum. These alterations preceded disintegration of myofibrils and were associated with significant deviations from the norm of the myocardial electrolyte content, consisting of a decrease in Mg and inorganic phosphorus and an increase in Na, whereas K remained initially remarkably constant, and Ca stayed within the physiological range throughout" (Lehr *et al.*, 1969).

"Mg^{2+} is unlikely to be the ion accumulated into the dense bodies (granules), since mitochondrial Mg^{2+} decreases (with the autolysis) rather than increases at the time the dense bodies appear. On the other hand, the data suggest that the dense bodies may contain a significant amount of Ca" (Jennings *et al.*, 1970).

The above quotations have in common some lack of understanding of how and why crystallization occurs in myocardium, and what its consequences are on the depleted fluids of the cells. It is also amazing that apparently

no investigator has ever analyzed the intracellular precipitates either. As a matter of hard fact, some common phosphates have been mislabeled, molarities are sometimes incorrectly calculated, and a few investigators did not realize that the extremely insoluble Ca–Mg phosphates do not dissolve at all in the common organic solvents and, consequently, have not been accounted for in their analyses.† As a direct consequence of these oversights, many found decreased amounts of intracellular Mg and phosphates, where in fact, together with the crystalline precipitates, there are more Mg and phosphates than usual. The mere presence of cellular crystals indicates highly supersaturated and dynamic conditions.

Under the name of electron dense bodies or granules, spherulitic crystallization is consistently shown in the electron micrographs of the last 20 years. Spherulites are formed in the myocardium so frequently only because of intrinsic nucleation. They grow then by successive layer deposition any time the K_{sp}'s of some of the electrolytes are reached. TriMg phosphate is one, if not the only one, of the seeds of crystallization in myocardium. This follows from the usually high Mg presence in it, frequently supplemented by exogenous Na_2HPO_4 or NaH_2PO_4. The latter two salts are highly soluble and penetrate the mitochondrial membranes more easily and quickly than NaCl. The two Na salts are then transformed into the tribasic phosphate, which participates in equilibrium (4).

The consequences in the myocardium of seeded crystallization are the depletion of the intracellular solutions of part of their content: namely, Mg^{2+}, K^+ or PO_4^{3-}. The mother liquids in equilibrium with their solids are less saturated than the initial supersaturated solutions. The insoluble precipitates seen in the myocardium are the initial cause of the necrotic processes, because their presence hampers the normal functioning of the cells. Degeneration and necrosis of myocardial tissue is always associated with substantial precipitation in the cytoplasm or on the cellular membranes. It is, then, the excessive presence of Mg^{2+}, PO_4^{3-} and of the other important cellular ions that starts the physical process of crystallization. The chemical and physical mechanisms of intracellular necrosis are rather similar to those already described by Boldrini (1980a,b) for C–C–2W. The biological origin

† Referee one is of the opinion that there is no convincing evidence on the existence of pool 2, "and no evidence at all that its size increases with age as indicated in Table 1(c)". Quite the contrary, such evidence exists: Buck (1951); Crawford & Crawford (1967); Yu (1973); plus about 100 EM photographs like those of Jennings *et al.* (1970). (A Table of them had been prepared, but that alone would have doubled the bibliography.) Unfortunately, many of the investigators who pretended to quantify Mg did not know analytical chemistry to do it correctly; e.g. they ignore that Mg salts are volatile when using the dry ashing method. The use of alcohol to pretend dissolution of the Ca–Mg phosphates is another error, etc.

of the new precipitates is, of course, quite different from that of C–C–2W. Intracellular Ca–Mg precipitation is much slower than the extracellular one, because of the lower variability of the intracellular concentrations of ions. The best example of how far intracellular precipitation may go is given by Figs. 2–5 of Jennings *et al.* (1970), where the end result is the complete obliteration of the cell.

Ferroelectricity in the Myocardium

The conditions at the membrane interface of the myocardium are somewhat different than those inside the mitochondria. $MgKPO_4$ is a polar salt and already when in solution it is electrically coupled with the membrane. At the interface the concentrations are not homogeneous with the rest of the intracellular fluids, because of the existence of a bipolar layer of oriented molecules. When the solubility product of $MgKPO_4$ is reached, the salt is deposited epitaxially on the membrane surface; it precipitates there, followed only by other polar materials. Such crystallization has been seen less frequently than the other of triMg phosphate and is more a physical than a chemical event.

The same kind of electromagnetic coupling with the membrane, as well as similar consequences from such an incrustation are explained elsewhere for C–C–2W (Boldrini, 1980*b*). Successive deposition on the membranes hampers their physiological activity, independently from the internal crystallization in the mitochondria. It is this second kind of epitaxial crystallization that leads to arteriosclerosis in the long run. Of all calcification processes, this is the slowest, the more permanent and widespread. This is confirmed by Heggtveit, Herman & Mishra (1964). These authors stated: "Calcification commenced with the deposition of particulate electron dense material on the cristae and sarcomas... the cristae are distorted, some cristae adjacent to the vacuoles are highly dense and may represent the earliest deposition of Ca".

When the first $MgKPO_4$ is deposited, the primary cause of its retention there is the stress-induced polarization of the membrane. $MgKPO_4$ possesses a permanent dipole moment which may be reversed by the field produced by the radial expansion/contraction of the myocardium. The initial $MgKPO_4$ increases the polarization of the myocardium, favoring accumulation of additional oriented material. $MgKPO_4$ constitutes then a polar continuum with the myocardium, which becomes ferroelectric storing energy in its walls under the form of residual polarization (Boldrini, 1982).

Conclusions

The biology and analytical chemistry of Mg have been constantly obscured by its companionship with Ca. Many intracellular crystalline precipitates of Mg have been incorrectly tagged as being Ca salts and, therefore, Mg has not been accounted for in many published data. Paradoxically, in an age when parts per billion of certain elements are detected, we loose track of 10% of Mg in some Ca salts! Such a situation is analogous to that of cholestanol present in cholesterol (Boldrini, 1980*a*).

It follows that not all cases called Mg deficiencies are real deficiencies; some of them are hypermagnesemia instead, while others are pathological conditions. One of the latter is that of Clark & Carré (1967). "The definition of Mg deficiency is one of tremendous practical difficulty . . . the diagnosis of Mg deficiency is almost impossible (Pors Nielsen, 1974). "Mg is widely distributed in both plant and animal foods and it has generally been believed that usual intakes are adequate" (Hathaway, 1962). "Mg like Na, is a component of the body that can be rather rigidly conserved in the presence of a specific (dietary) deficiency" (Barnes, Cape & Harrison, 1958).

A non-thorough investigator accepting the precipitates of Heggtveit *et al.* (1964), shown in their Figs 9–12, as Ca would not test analytically for Mg in those crystals and would then be faced with the apparent disappearance of Mg from the cells. The investigator would then think that he is in the presence of Mg deficiency when in reality the situation at the start was exactly the opposite: Mg crystallized after the solubility product of triMg phosphate is reached (which indicates Mg supersaturation or hypermagnesemia).

The false belief that the only deposition of Mg was the physiological process of "mineralization" (Mg deposition in bones) has been another reason why non-physiological crystallization in the arteries and myocardium has to date been passed. However, the laws of chemistry and physics, as

TABLE 9

Effect of magnesium deprivation on Mg *and* Ca *content of rat viscera* (Tufts & Greenberg, 1936)

	Control animals		Deficient animals	
	Range of Mg (mg/dl)	Ca (mg/dl)	Mg (mg/dl)	Ca (mg/dl)
Heart	22·6–29.1	3·6–6·1	21·2–26·5	6·1–14·7
Muscle	24·6–36·2	4·8–7·9	15·2–32·0	8·5–15·2
Kidney	18·0–25·3	6·9–12·4	16·9–25·3	48·5–23·2

well as many electron microscope pictures, clearly indicate that Mg is indeed irreversibly deposited in the arteries and myocardium. These salts of Mg make up pool 2, which may be negligible till "middle age", but becomes the biggest pool in advanced age (Table 1(c)).

Pool 2 is the mitochondrial pool, because Mg obliterates the mitochondrial structure (Cantin & Seelig, 1980; Di Giorgio, 1962). Pool 2 grows

TABLE 10

Biological significance of the new theory

The five Mg pools of the body are: *pool 1* which includes the free, ionic and most mobile Mg; *pool 2* includes the crystalline Mg precipitated mainly in the arteries and myocardium; *pool 3* includes the Mg complexes; *pool 4* the undissociated Mg salts; *pool 5* the physiologically bound Mg of bones and proteins (Fig. 1).

The size of the five pools must be quantitatively determined by way of accurate analyses, accounting for the insoluble Mg salts precipitated and for the physiological variations of those in solution. Special attention should be focused on the identification of the very beginning of crystallization in the arteries and myocardium.

When saturation is reached in the intracellular fluids, triMg phosphate nucleates triCa phosphate and the other Mg–Ca–K salts. The unwanted insoluble salts build up in the cells and can hinder their physiological functions.

The combined presence of Mg, Ca, K and PO_4^{3-} must be monitored in the intracellular fluids, and not Mg alone, as has been done up to now, since the body normally adjusts itself to intervariations of each of these ions.

The equilibrium of Mg^{2+}, Ca^{2+}, K^+, and PO_4^{3-} is constantly maintained by the blood supply; however, new PO_4^{3-} is also continuously formed as a result of the transformation of the other phosphates of serum.

Simultaneous feeding of Mg, Ca, K and PO_4^{3-} is neither useful nor healthy. The ions already combine during digestion and are poorly absorbed; the part of them which passes into blood displaces equilibrium (4) toward precipitation.

There is a polar interaction between $MgKPO_4$ and the internal surface of the membranes of the myocardium and arterial cells. The salt crystallizes on the surface of the membranes becoming electrically coupled with them, with the consequent scarring of the myocardium and calcification of the media of the arteries (arteriosclerosis).

Direct injection of the most ionic Mg salts does not seem safe under any circumstances, since it saturates pool 1 and might induce instant precipitation. On the other hand, injection of soluble, undissociated salts (like Mg gluconate) increases pool 4 instead: one of the reservoirs of Mg in the body.

In healthy persons Mg deficiency (inadequacy or shortage of a substance necessary to health) is not as frequent as some authors believe, since Mg is chemically very active. Kidneys recycle Mg with very high efficiency.

Hypermagnesia is frequently and unnecessarily reached in healthy bodies, with the consequences of accumulation of insoluble Mg in pool 2. Some of the unexplained incidences of field rickets and other related leg and skeletal problems may be due to excessive dietary Mg intake.

During the later decades of life, it is estimated that the Mg crystallized into the arteries and myocardium might reach 1 mg/day, causing pool 2 to become larger and larger, at the expenses of pool 3 and 5.

A more specific role for Mg in muscles must be ascertained, independently from the role of Ca.

with the increase of calcification of the arteries and is a new parameter to be considered in cardiovascular disease. Mg role in the body has to be reconsidered in view of the damage it can possibly do to the arteries and myocardium (Ciaccio, 1973).

The concentration of Mg should be considered together with that of Ca, K and PO_4, because of the chemical equilibrium existing among them. Jackson & Meier (1968) showed that because of this relationship the majority of patients affected by arteriosclerosis have the highest Mg content. Mg deprivation alone does not then alter the Mg in the heart (Table 9), nor that of the intracellular fluids. A more clear distinction between hypo- and hypermagnesemia, both in the diet and in plasma is required; only in this way, may we hope to answer the question of Aikawa (1976): "What is the role of Mg at this interface of chemistry and physics to molecular biology?"

In Mg deposition an important role is that played by the polarity of the membrane and of the Mg salts. There is an electrical coupling between the inside surface of the membranes and the polar crystallographic direction of $MgKPO_4$: the seed of nucleation. As a result of this electromagnetic coupling, even the inside part of the membrane becomes ferroelectric, in a way similar to that already described (Boldrini, 1980*b*).

The implications of the new theory presented here are summarized in Table 10.

REFERENCES

ACHARYA, P. T. & PAYNE, W. (1965). *Arch. Dis. Childhood* **40,** 430.

AIKAWA, J. K. (1976). Biochemistry and physiology of Mg. In: *Trace Elements in Human Health and Disease.* Vol. 2. p. 47. London: Academic Press.

AIKAWA, J. K. (1980). *Magnesium.* Cleveland, Ohio: CRC Press.

BARNES, B. A., COPE, O. & HARRISON, T. (1958). *J. Clin. Invest.* **37,** 430.

BERLYNE, G. M., BEDRAK, E. & YAGIL, R. (1973). *Q. J. Med.* **42,** 793.

BOLDRINI, P. (1980*a*). *Physiol. Chem. Phys.* **12,** 153.

BOLDRINI, P. (1980*b*). *J. theor. Biol.* **87,** 263.

BOLDRINI, P. (1982). *J. theor. Biol.* **95,** 325.

BRIERLY, G., MURER, E., BACHMANN, E. & GREEN, D. (1963). *J. biol. Chem.* **238,** 3482.

BRISCOE, A. M. & RAGAN, C. (1966). *Am. J. Clin. Nutrition* **19,** 296.

BUCK, R. C. (1951). *Arch. Pathol.* **51,** 319.

CANTIN, M. & SEELIG, M. S. (eds) (1980). *Magnesium in Health and Disease.* Utrecht: Spectrum.

CIACCIO, L. L. (1973). *Water and Water Pollution.* Vol. 3. New York: Marcel Dekker.

CLARK, P. C. & CARRÉ, I. J. (1967). *J. Pediatrica* **70,** 806.

CRAWFORD, T. & CRAWFORD, M. (1967). *Lancet* **1,** 229.

DI GIORGIO J. (1962). *Biochem. J.* **82,** 184.

FLINK, E. B. (1976). Mg deficiency and Mg toxicity in man. In: *Trace Elements in Human Health and Disease.* Vol. 2. London: Academic Press.

GROSS, W. H. (1949). *The Story of Magnesium.* Cleveland: American Society for Metals.

HATHAWAY, M. L. (1962). *Magnesium in human nutrition.* U.S. Dept. of Agriculture.

HEATON, F. W., HODGKINSON, A. & ROSE, G. A. (1964). *Clin. Sci.* **27,** 31.
HEINRICH, K. F. (1981). *Electron Beam X-ray Microanalysis.* New York: Van Nostrand.
HEGGTVEIT, H. A., HERMAN, L. & MISHRA, R. K. (1964). *Am. J. pathol.* **45,** 757.
HERDSON, P. B., KALTENBACH, J. P. & JENNINGS, R. B. (1969). *Am. J. Pathol.* **57,** 539.
JACKSON, C. E. & MEIER, D. W. (1968). *Ann. Internal Med.* **69,** 743.
JENNINGS, R. B., BAUM, J. H. & HERDSON, P. B. (1965). *Arch. Pathol.* **79,** 135.
JENNINGS, R. B., MOORE, C. B., SHEN, A. C. & HERDSON, P. B. (1970). *Proc. Soc. Exp. Biol. Med.* **135,** 515.
KEATING, F. R., JONES, J. D., ELVEBACK, L. R. & RANDALL, R. V. (1969). *J. Lab. Clin. Med.* **73,** 825.
LEE, S. R. & BRITTON, W. M. (1980*a*). *Poultry Sci.* **59,** 1989.
LEE, S. R., BRITTON, W. M. & ROWLAND, G. N. (1980*b*). *Poultry Sci.* **59,** 2403.
LEHR, D. (1969). *Ann. N.Y. Acad. Sci.* **156,** 344.
MARSHALL, R. W. (1976). Plasma fractions. In: *Calcium, Phosphate and Magnesium Metabolism.* Edinburgh: Churchill Livingstone.
MARIER, J. R. (1979). *Water Hardness, Human Health and the Importance of Magnesium.* Ottawa: NRCC.
MONIF, G. R. G. & SAVORY, T. (1972). *J. Am. Med. Ass.* **219,** 1469.
NORDIN, B. E. C. (ed.) (1976). *Calcium, Phosphate and Magnesium Metabolism.* Edinburgh: Churchill Livingstone.
PASCAL, P. (1958). *Nouveau Traité de Chimie Minérale.* Vol. 4. Paris: Masson.
PORS NIELSEN, S. (1974). *Interrelations of Magnesium and Calcium Metabolism.* Copenhagen: Munksgaard.
PRASAD, A. S. (ed.) (1976). *Trace Elements in Human Health and Disease.* Vol. 2. London: Academic Press.
SEELIG, M. S. (1980). *Magnesium Deficiency in the Pathogenesis of Disease.* New York: Plenum Press.
TAYLOR, A. W., FRAZIER, A. W. & GURNEY, E. L. (1963). *Trans. Faraday Soc.* **59,** 1580, 1585.
TUFTS, E. V. & GREENBERG, D. M. (1936). *Proc. Soc. Exp. Biol. Med.* **34,** 292.
TWORT, A. C. (1963). *A Textbook of Water Supply.* London: Edward Arnold.
WACKER, W. E. (1980). *Magnesium and Man.* Cambridge, Massachusetts: Harvard University Press.
WALLACH, S., RIZEK, J. E., DIMICH, A., PRASAD, N. & SILER, W. (1966). *J. Clin. Endocrinol. Metabol.* **26,** 1069.
WEBER, W. J. (1972). *Physico-chemical Processes.* New York: Wiley. p. 277.
WYCKOFF, R. W. G. (1965). *Crystal Structures.* 2nd ed. Vol. 3. New York: Interscience.
YU, S. Y. (1973). In: *Arterial Mesenchyme and Arteriosclerosis* (W. D. Wagner & T. B. Clarkson eds), New York: Plenum Press.

APPENDIX 1

Magnesium in Our Everyday Life

Lee *et al.* (1980*b*) suggest that chick feed may be adequate in Mg because of soil richness; Gross (1949) shows how widespread the use of Mg is in technology: "The world may be said to be in a light metal age, and Mg, the lightest metal suitable for structural use, is rapidly becoming indispensable in our everyday life. Household appliances and furnishings employ light weight to advantage in easing the burden of the housewife and modernizing the home".

"It is extremely difficult to form a clear picture of the syndrome of Mg deficiency owing to the confusion which exists between hypomagnesemia and depletion of body stores... In man, hypomagnesemia per se does not appear to cause any disability and should perhaps be regarded as an adaptation to inadequate intake rather than a sign of true Mg deficiency" (Nordin, 1976). "In spite of the probability of diets being low in Mg, under certain circumstances, Mg deficiency does not occur in human beings with healthy kidneys.... Retention of Mg by the kidney occurs rapidly in response to a restriction in the dietary intake. This is why it is so difficult to produce Mg depletion in the adult without some source of abnormal loss from the body" (Aikawa, 1976). "Establishing a diagnosis of Mg deficit is still difficult. Hypomagnesemia (more than 2 standard deviations below the mean) is not conclusive evidence per se, and a big deficit can occur without hypomagnesemia. Erythrocyte Mg concentration has been used by man, but red cells are not quickly responsive to a deficit. Total external balance studies are the most convincing evidence, but such a study is often not feasible" (Flink, 1976).

Prasad (1976) said that "... for some trace elements, i.e. Ni, Si, Sn and V there is little likelihood of spontaneous deficiency in man due to inadequate dietary intake. Stringent measures are required to induce deficiencies of these elements in experimental animals." This is very true of Mg, if one makes the analyses correctly.

APPENDIX 2

Magnesium Intoxication

"Mg intoxication occurs primarily in three settings: treatment of eclampsia with attendant neonatal depression of vital functions, accidental or intentional poisoning, and moderately to severe renal failure. When Mg-containing medications are taken by a patient with renal failure, intoxication often results... Fatal intoxication occurred in infants given $MgSO_4$ enemas" (Flink, 1976). Some of the past experiments where Mg has been directly administered n large quantities to humans can be challenged on ethical, as well as on scientific grounds. The heuristic value of the experiments is not in doubt, but rather their interpretation.

Chicks fed 1% Mg in their diet usually develop skeletal abnormalities: bone lesions, bowing of the metatarsus, decrease in per cent of the tibia ash, leg weakness, shortening of the tibia, etc. (Lee *et al.*, 1980*a*, *b*). These and other effects of excessive Mg intake may be studied in the way one wants, but their conclusions are univocal: they represent abnormal

situations and quick events leading to extracellular crystallization. This deposition into pool 2 is also preventing the normal physiological activity of the cells, as described elsewhere in this paper, with few secondary details of its own. We have to conclude that $MgSO_4$ or $MgCl_2$, the two most soluble Mg salts, cannot be injected in living organisms having normal Ca^{2+} and PO_4^{3-} levels; such direct supplementation induces hypocalcemia, because more Ca is precipitated in equilibrium (4) by the extra Mg. This is shown by the results of many autopsies.

The skeletal abnormalities, therefore, are a combined consequence of reduced availability of Ca and of increased calcification in the soft tissues. The collateral effects of Mg deposition are dramatic on the growth of the chicks as well; their weight is reduced by as much as 3·4 times when compared with the controls (Lee *et al.*, 1980a, b). "As a result of poor growth, tissues of these chicks are smaller and may tend to confound analyses or inspections of lesions found, especially in soft tissues" (Lee *et al.* 1980*b*). Hypermagnesemia may then be a danger for healthy individuals.