92. THE ABSORPTION AND EXCRETION OF 'MINOR' ELEMENTS BY MAN

1. SILVER, GOLD, LITHIUM, BORON AND VANADIUM

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(*Received 23 June 1941*)

OUR knowledge of the biological principles underlying the excretion of minerals, whether physiological, pharmacological or pathological, is surprisingly meagre. Even the route by which many of them leave the human body is not yet known, much less the rate at which they do so. Within recent years iron has been shown to be excreted much less readily than was previously supposed [McCance & Widdowson, 1938], and not by the large intestine [Nicolayson, 1935; Welch *et al.* 1936], but there are many problems underlying the excretion of calcium and other elements in the same periodic group [McCance & Widdowson, 1939, 1, 2]. The introduction of radioactive elements into the physiologist's armamentarium has stimulated interest in this subject and provided some data of great value. The use of these 'marked' elements, however, is still in its infancy, and has by no means displaced older methods. For some purposes it never can. Accordingly, as opportunities presented themselves of studying the excretion of various metals, it was natural to do so, and the analytical technique, together with some of the results, form the substance of this paper.

Subjects and methods

The subjects of these experiments have been patients, and normal persons. The former were used for the studies of Ag and Au. The latter had submitted themselves to prolonged balance experiments in order to solve other problems in mineral metabolism, and it was while they were thus engaged that their excretion of V, Li and B was investigated. The technique of these balance experiments has been so fully described by McCance & Widdowson [1941] that nothing further need be said.

The minerals were introduced into the body in various ways which will be described in the appropriate sections.

The analyses were done spectrochemically. A Hilger medium quartz spectrograph (E. 3) was used with Ilford Auto-Filter (ortho) and Rapid Process Panchromatic plates. The electrodes, the purest obtainable, were supplied by Messrs Adam Hilger Ltd., with an analyst's report of their impurities. The light source was an oxy-coal gas flame, a low voltage D.C. arc or a high voltage A.C. spark. Ag was estimated by a modification of the Ramage flame method, full details of which have been published [Kent, 1940]. The filter paper standards contained 100, 10, 5, 2, 1, 0·5 and 0·1 μg. of Ag. The other elements were determined by comparing their lines with those of an internal reference element. Mg or Fe was generally selected for this purpose, and if so its concentration in the analytical material was estimated chemically. Bi and Mo were used as the reference elements for Sn (see Table 1). They were added in equal quantities both to the

'unknown' and to the 'standard' solutions, neither of which contained any until this was done.

For the spark, a pair of graphite electrodes (6·5 mm. diameter) was used (Hilger lab. no. 12313), the upper of which was bored longitudinally with a 1 mm. diameter hole. One end of this electrode was pointed, while the other end was fitted with a rubber teat, by means of which the liquid for analysis was drawn up into the electrode. The lower electrode had a flat end, and the spark gap was about 2 mm. wide. The electrodes were connected through a condenser (0·005 μ farad) to the secondary circuit of a transformer (generating 15,000 V.), the primary circuit of which was connected through a resistance to the A.C. mains. The spark was about 25 cm. from the slit of the spectrograph, and the exposure was usually 20 sec. For the arc a pair of pure copper electrodes was used, of which the upper (5 mm. diameter, Hilger lab. no. 11184) was pointed, the lower (7 mm. diameter, Hilger lab. no. 10300) flattened. A small quantity of residue from the evaporated solutions (*vide infra*) was placed on the lower electrode. The arc was struck by bringing the electrodes together, and then separating them until the current was approximately 1·8 A. This current was maintained for 60 sec. The arc was connected in series with the D.C. mains and with a resistance which cut down the potential to about 60 V.

The intensity of the lines in the spectra was measured by a microphotometer which was constructed in the Department of Physical Chemistry, Cambridge, where all the spectrographic work was carried out. A framework held the plate vertically, and permitted a coarse up-and-down motion and a fine sideways motion. Light from a 12 V., 60 W., lamp was focussed on the plate by means of two lenses, and an image of the line was focussed by a third lens on the slit of a photoelectric cell. The latter was connected to a non-recording mirror galvanometer. The most sensitive line of the element to be estimated was usually chosen for microphotometric measurements. The choice of internal reference element was governed by the distribution of lines in the spectrum as it was essential to have the line of the reference element fairly close to the line of the element to be estimated. In Table 1 are listed the elements estimated and the method of excitation, the internal reference element and the pairs of comparison lines used for photometric determinations.

Table 1. *Summary of quantitative spectrographic methods*

Element estimated	Process	Internal standard	Comparison pairs of lines: Element estimated	Comparison pairs of lines: Standard
Lithium	Arc	Magnesium	Li 3232	Mg 2803
Boron	Spark	Iron	B 2496·7	Fe 2493
Vanadium	Spark	Magnesium	V 3102	Mg 3097
		Iron	V 3102	Fe 2783
Silver	Ramage flame	—	Ag 3280	—
			Ag 3383	—
Gold	Arc	Magnesium	Au 2676	Mg 2795
Manganese*	Spark	Iron	Mn 2606	Fe 2607
Nickel*	Spark	Iron	Ni 2394	Fe 2388
		,,	Ni 2394	Fe 2384
Cobalt*	Spark	Iron	Co 2378	Fe 2379
		,,	Co 2388·9	Fe 2388·6
Tin*	Arc	Bismuth	Sn 3034	Bi 2997
		Molybdenum	Sn 3034	Mo 2911

* The absorption and excretion of these elements will be discussed in a subsequent paper.

When the density of a line was being measured with the microphotometer, the plate was moved very slowly with the fine screw so that the line passed completely across the slit of the photoelectric cell. The spot light from the galvanometer was watched and the maximum deflexion noted. A reading was also recorded for the lighter ground adjacent to each line measured. The intensity of the line of the internal reference element was measured before passing on to another spectrogram on the same plate.

Food, urine and faeces were prepared for chemical analysis as described by McCance & Widdowson [1941], and were supplied in duplicate for spectrochemical analysis in the form of a dilute HCl extract of the ash. The ash extracts of the foods and urines were prepared for analysis by the spark method by making the concentration of the Fe in them approximately equal to that in the ash extracts of the faeces. For this purpose 0·8 ml. of an Fe solution, containing 0·5 mg. of Fe/ml., was added to 9·2 ml. of the ash extracts of the food, and 1 ml. of the same Fe solution to 9 ml. of the ash extracts of the urines. Unless this had been done, erroneously high results were obtained for Mn both in the foods and urines, and as a precaution, therefore, the Fe concentration was similarly adjusted when other elements were being estimated. In the arc method the Fe concentration was adjusted as outlined above, the Bi and Mo were added if Sn was being determined, and the solutions were evaporated to dryness. Two exposures were made of each ash extract or of the dry residue prepared from it. Hence, quadruplicate spectrograms were taken of the food, urine and faeces in each experimental period.

For each element to be estimated a series of standard solutions was prepared containing varying quantities of that element, but the same quantities of the internal reference element. The concentrations of the reference elements in these serial standards were 0·05 mg. of Bi and of Mo/ml., 0·4 mg. of Mg/ml., and 0·05 mg. of Fe/ml. To each was also added the same quantity of a 'background' solution containing Na, K and Ca, the purpose of which was to give these solutions a composition similar to that of the unknowns. From the microphotometric readings of these standard solutions a series of ratios, Intensity of the element to be estimated/Intensity of reference element, was obtained. When the logarithm of these ratios was plotted on squared paper against the logarithm of the percentage of the element to be estimated, straight lines or smooth curves were obtained. From these 'working curves' the log of the percentage of the unknown in any ash extract could be obtained by working out the log of the intensity ratio from the microphotometric results, and reading off the answer from the graph.

Results

Silver

The study was made on a woman who was suffering from generalized argyria caused by washing out her nose for many years with an organic silver preparation. She was nearly, but not quite as pigmented as the case shown in Harker & Hunter's [1935] coloured plate. Her Ag metabolism was followed for three analytical periods, each of 7 days in length. The food, urine and faeces were prepared for analysis as described by McCance & Widdowson [1941]. The results of the balances are given in Table 2. It will be noted that the patient was excreting very little of the silver with which she appeared to be so saturated, and this result is in keeping with the clinical observations, all of which go to show that the pigmentation, once acquired, does not fade rapidly, if at all. The small negative balances which are shown may not even be physiological in the true

Table 2. *The intake by mouth and the excretion of silver by a patient with argyria*

Week	Food mg./week	Urine mg./week	Faeces mg./week	Total excretion mg./week	Balance mg./week
1	0·05	0	1·3	1·3	−1·25
2	0	0	1·5	1·5	−1·5
3	0·7	0	2·3	2·3	−1·6

sense of the term, for the mucous membranes of the patient's mouth and nose were deeply pigmented, and the proctoscope revealed similar, although less deep, staining in the visible parts of the rectum and lower bowel. The whole alimentary canal, therefore, may have been lined by cells containing Ag. Desquamation of some of these cells may have been the cause of the negative balances.

Gold

Gold is given therapeutically by injection, and attempts have been made from time to time to discover the organs in which it is deposited [Gerlach, 1935; Koppenhöfer, 1936; Leulier & Béruard, 1938; Leulier & Payre-Ficot, 1934, 2; Lumière & Julliard, 1930], and the route by which it is excreted [Arloing *et al.* 1932; Leulier *et al.* 1937; Leulier & Béruard, 1938; Leulier & Payre-Ficot, 1934, 1]. Most of this work appears to have been done in France and Germany. Both men and laboratory animals have been investigated. All species do not necessarily react in quite the same way [Leulier & Payre-Ficot, 1934, 2], but it has been established that the drug may cause nephritis, that it is excreted very slowly, and that the more soluble compounds, such as Solganol B and Myochrysine, are excreted predominantly by the kidney, whereas the more insoluble, such as Lipaurol and Myoral, in so far as they are excreted at all, are passed out mainly by the gut [Leulier & Payre-Ficot, 1934, 1].

The present work was carried out on a woman who was being treated for rheumatism with injections of Solganol B oleosum, an oily suspension of aurothioglucose which is said to be water-soluble and to contain 50% of Au [Leulier & Payre-Ficot, 1934, 1; Gerlach, 1935]. There were two analytical periods, each of 7 days in length, and in each the patient received 50 mg. of gold by intramuscular injection. 500 mg. of Au had already been given before the study began. The results, which are shown in Table 3, corroborate those of the French authors.

Table 3. *Excretion of gold after intramuscular injection*

Week	Food mg./week	Urine mg./week	Faeces mg./week	Total excretion mg./week	% of the total excreted in the urine
1	0	5·2	1·5	6·7	78
2	0	7·1	2·0	9·1	78

Some [Arloing *et al.* 1932; Leulier & Payre-Ficot, 1934, 1, 2; Leulier *et al.* 1937] have found a rather higher proportion of these soluble salts excreted in the urine, but the present partition is of very much the usual order. It is evident, however, that some Au is genuinely excreted by the gut. How it reaches the intestine is unknown, and seems to be a matter for investigation.

Lithium

Practically no papers on the absorption and excretion of lithium salts have been found since Good's [1903] article on the pharmacology of this metal. Good studied the oral and subcutaneous administration of toxic doses of LiCl to cats

and dogs, and found that, however given, they produced vomiting, enteritis and death. In spite of these signs of gastro-intestinal excretion, and the finding of lithium in the saliva and gastric juices, the metal was eliminated mainly, although incompletely, by the kidney. This process did not cause nephritis. Table 4 shows the lithium balances of two men and one woman before, during

Table 4. *Lithium balances before, during and after ingesting soluble lithium salts*

Subject	Week	Intake by mouth mg./week	Lithium in urine mg./week	'Total' lithium excreted (urine + faeces) mg./week	Lithium balance mg./week	Lithium recovered mg.
N. K.	1	11·1	4·7	9·7	+ 1·4	
(Male)	2	6·3 + 250	218	229	+27	236
	3	7·9	15	21	−13	
E. W.	1	17	4·3	17	± 0	
(Female)	2	17 + 250	218	233	+34	258
	3	16	46	58	−42	
R. M.	1	18	4·5	19	− 1	
(Male)	2	16 + 250	195	208	+58	247
	3	17	60	72	−55	

and after a time in which 250 mg. of LiCl were taken by mouth. The salt was given after meals, over a period of 3 days, during the second week of a 3-week metabolism experiment. N. K. took the Li in the early part of the week, the others in the last 3 days, and this explains why E. W. and R. M. excreted more of the administered Li in the third week. It is evident that soluble Li salts added to a diet are readily absorbed and quantitatively excreted in the urine. In N. K.'s second week there were 5 mg. more lithium in the faeces than there were in the solid food. This must have come from the medicinal 250 mg., but apart from this trifling exception, none of the administered Li seemed to reach the faeces.

The present results, therefore, differ somewhat from those of Good, but the experimental conditions were totally different. In the light of modern knowledge it is easy to explain the excretion by the gut of small amounts of any metal which can replace Na in the extracellular fluids, and of quite large amounts if the metal in its passage to the lumen of the gut produces a gastroenteritis and diarrhoea.

If now the first week of each person's experiment is inspected, it will be seen that the urine contained only 25–50 % of the total amount excreted. This suggests a very poor absorption of Li from natural diets. Table 5 shows the metabolic data in more detail, and also gives three other 'balances'. The brown

Table 5. *The intake and excretion of lithium when 40–50 % of the total calories in the diets were supplied by white bread, or by brown*

Subject	Diet	Week	Food mg./week	Urine mg./week	Faeces mg./week	Li absorbed (food − faeces) mg./week
N. K.	White bread	1	11·1	4·7	5·0	6·1
R. M.	,,	1	12·5	4·6	6·6	5·9
	,,	2	11·4	4·2	6·8	4·6
	,,	3	12·4	6·5	7·3	5·1
E. W.	Brown bread	1	16·9	4·3	12·4	4·5
R. M.	,,	1	17·7	4·5	14·0	3·7

bread diets contained more Li than the white because the 92% flour had much more in it than the 69%. The figures obtained by analysis were 0·42 and 0·17 mg./100 g. respectively. It is, however, strange that the Li was absorbed so badly from both diets, and particularly so from the brown bread. Potassium was absorbed much more freely [McCance & Widdowson, 1941]. Admittedly, calcium and magnesium were not well absorbed from the 92% flour, but this was found to be due to their precipitation as phytates. The poor absorption of Li cannot be explained in this way, for lithium phytate is soluble. There was, moreover, very little phytic acid in the white flour diets, the lithium in which was also relatively poorly absorbed. It is possible that the Li in the branny parts of wheat is in very close association with the cellulose particles and never really goes into solution in the intestine. Li has physiological properties in plants which suggest that this may be so, for it has been found that Li does not return from the senescent wheat leaf as potassium does, but remains with the calcium *in situ* [Mason & Maskell, 1931; Kent, 1941]. Its manner of fixation in these leaves is not yet known. It is evident, however, that for some reason the Li in natural foods may be very much less freely absorbed than soluble Li salts, even if the latter have been taken with food.

Boron

Plants were first recognized to contain B in 1857 [Brenchley, 1927] and this element is now accepted as a constituent of all vegetable tissues. Consequently, it is an inevitable component of our daily diet, and, apart from this physiological aspect of the matter, boric acid has had such a checkered career as a food preservative that some experiments have been carried out to determine the fate of the B which is naturally present in foods, and of boric acid when taken by mouth. Something is known of this latter. Röst [1905] and Wiley [1907] both studied the excretion of boric acid after doses of 3 g. and upwards had been given, and concluded that 82–100% was quickly excreted in the urine. The faeces were never an important avenue of excretion, and although small amounts were found in the sweat, this also was quantitatively unimportant. Oddly enough, neither author considered the B naturally present in food, but this was probably due to their technical limitations. The upper half of Table 6 shows the results which were obtained when two normal women each took (in all) 352 mg. of B as boric acid during the 3rd, 4th and 5th days of a 28-day metabolism experiment. 40% of the calories of their diets were derived from white flour. It will be seen that 93–94% of the B was recovered during the first week, which demonstrates its rapid absorption and metabolism, and confirms the results of Röst [1905], Wiley [1907] and Presnell & Brill [1937]. It will also be noted from the last 2 weeks of the metabolism experiment that the B in natural foods was metabolized in very much the same way. The lower half of Table 6 shows the B balances for the same two women over a period of 21 days when 40% of their calories were derived from flour of 92% extraction. 100 g. of the white flour contained 0·045 mg. of B. 100 g. of the 92% flour contained 0·16 mg., a figure which agrees quite well with those of Bertrand & De Wall [1936] for whole wheat and cereals generally. It may seem strange, therefore, that the intakes were not higher during the brown bread than they were during the white bread experiments. The explanation is that even the 92% flour accounted for only 4·0 mg., or about 5% of the B in the week's food. The greater part of the B in these diets seems to have been introduced by the large amounts of fruit, particularly plums and greengages, which were being consumed [Allen & Tankard, 1904]. It is not thought that any of the foods had had any boric acid added to

Table 6. *Boron balances on white bread diets after ingesting boric acid*

Subject	Week	Intake by mouth mg./week	Boron in urine mg./week	Total boron excreted (urine + faeces) mg./week	Boron balance mg./week	Boron recovered
		Boron balances on white bread diets.				
E. W.	1	89 + 352	408	417	+24	351
	2	81	100	104	−23	
	3	64	53	57	+ 7	
	4	88	86	92	− 4	
A. M.	1	113 + 352	435	445	+20	332
	2	142	123	129	+13	
	3	111	100	104	+ 7	
	4	138	124	135	+ 3	
		Boron balances on brown bread diets.				
E. W.		75	62	68	+ 7	
A. M.		96	100	109	−13	

them. These results on the whole are in keeping with those of Löhnis [1936] and of Bertrand & De Wall [1936], who stated that cereals contained very much less B than fruits and vegetables.

Vanadium

Vanadium has been employed in the therapy of syphilis. Proescher *et al.* [1917] investigated some of its actions and reported that the hexa- were less toxic than the tetra-vanadates, but that there was not a large margin between the therapeutic and the toxic doses. They also reported that the drug was not 'cumulative'. When 12·5 mg. of V_2O_5 were taken by mouth daily for 12 days, 87·6 % of it was recovered in the faeces, and 12·4 % in the urine. Thus all the administered V was recovered, but no attempt was made to find out how much of the 87 % represented unabsorbed V, and how much re-excreted.

In the present experiments two normal men, whose metabolism was being studied for three successive weeks, were given six intravenous injections of sodium tetravanadate dissolved in normal saline during the second week; 3–5 min. were taken over each injection. The results are shown in Table 7. It

Table 7. *The excretion of vanadium after intravenous injections of Na tetravanadate*

		Intake		Excretion			
Subject	Week	Food mg./week	Injection mg./week	Urine mg./week	Faeces mg./week	Total mg./week	Vanadium recovered
N. K.	1	0	18	10·4	1·4	11·8	16·8 mg. ≡ 93 % of the dose
	2	0	0	5·0	0·0	5·0	
C. B.	1	0	24	13·2	1·3	14·5	21·2 mg. ≡ 89 % of the dose
	2	0	0	6·8	0·0	6·8	

will be seen that most of the V was excreted during the week in which it was given, and that very little remained in the body by the end of the subsequent week. During the week of injections some 10 % of the excreted V was found in the faeces, but it is clear that V which has once been absorbed is mainly eliminated by the kidney. The greater part of the oral dose found by Proescher *et al.* [1917] in the faeces should therefore be regarded as unabsorbed V and this element can be said to be one of those which is not readily absorbed when taken by mouth as the vanadate but, once absorbed, is excreted almost entirely by the kidney.

SUMMARY

Balance experiments have shown that:—

1. A patient with severe generalized argyria excreted less than 2 mg. of silver per week, all of it in the faeces.

2. A patient who had received 550 mg. of gold intramuscularly, excreted 6·7 and 9·1 mg. in two successive weeks, 78% of it in the urine.

3. Soluble lithium salts taken orally by normal persons were rapidly and completely excreted in the urine, but the Li in natural foods was poorly absorbed. Only 25% of the Li in a brown bread diet was excreted in the urine.

4. Boron taken by mouth as boric acid was rapidly excreted in the urine. B in foods was much more readily absorbed than Li, 92% of it appearing in the urine.

5. 81% of the vanadium, which had been administered intravenously as sodium tetravanadate to normal persons in six daily doses, was excreted in the urine by the 7th day after the last injection. 9% was excreted in the faeces in the same time.

The authors are grateful to Prof. Norrish for placing a spectrograph at their disposal and to Dr W. C. Price for some very valuable technical advice. They have also to thank Drs J. H. Sheldon and F. B. Parsons for allowing their patients to be investigated. The work could never have been undertaken, without the help and co-operation of the subjects, and also of Miss B. Alington and Miss E. M. Widdowson. The greater part of the expenses were covered by a grant made by the Medical Research Council.

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