LV. THE BIOCHEMISTRY OF SILICIC ACID
VI. THE SOLUTION AND EXCRETION OF SILICA

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In papers II and III of this series [1933, 1, 2] it was shown that the excretion of silicic acid in the urine could be influenced by the administration of silica. The urinary level was largely dependent on the amount of silica which was orally ingested, either as a constituent of the food, or as added mineral material. Both oral and parenteral administrations lead to prompt elimination in the urine. Large amounts of silicic acid, given by continuous intravenous injection, were quickly excreted by the kidney. No marked increase in the silica content of the blood was observed, even when the concentration in the urine was enormously increased.

Evidence was obtained that solution of silica dust may take place in the lung [King & Dolan, 1934]. Increased urinary excretion of silica in animals was obtained following administration by intratracheal insufflation, and in humans following dust exposure. This silicic acid arising from the solution of particles in the lung was presumably carried by the blood to the kidney where it was excreted. Some evidence was obtained that the urinary silica was at a higher level amongst gold miners than in a non-mining population, but the urinary silica values were found to be so dependent on the nature of the diet that no conclusions could be drawn in single cases [see also Goldwater, 1936]. Bloomfield *et al.* [1935], however, claimed to show a relation between the silica dust exposure of men in the anthracite coal industry and the amount of silica in the urine. The urinary excretion of abnormal amounts of silica cannot be taken as evidence of exposure to dust, but only as indicating an excessive intake of silica, either through ingestion or through inhalation.

In the present study an attempt is made to correlate the solution of various forms of silica in body fluids [King & McGeorge, 1938] with the urinary excretion of silicic acid following administration of the different forms of silica by the oral, intraperitoneal and in some cases the intratracheal route.

Experimental

The mineral materials used for administration were those described in the previous communication. Their mode of preparation, size distribution and analyses are given there. A brief statement of the chemical nature of some of the specimens is contained in Table I (see also Table I of the preceding paper).

All the experiments were conducted on cats. The use of a carnivorous animal has the advantage that the normal daily urinary excretion of silica is very low. Any increase caused through the administration of silica will consequently be much more easily recognized than would be the case in a herbivorous animal, such as the rabbit, where the concentration of silica is high in the urine. The cats were maintained on a diet of cooked meat, fish, milk and water. During an experiment each animal was kept in a metabolism cage equipped to collect the faeces and urine separately. The cages were constructed of galvanized metal and heavy wire screening. Metal surfaces were coated with baked white enamel. The urine was collected in bakelite-coated vessels.

Table I. *Nature of mineral specimens*

Quartz—free crystalline silica
Flint—free crystalline silica
Felspar—anhydrous silicate of potassium, sodium and aluminium
Sericite—hydrous silicate of potassium and aluminium
Fuller's earth—a hydrated silicate of aluminium
Kaolin—silicate of aluminium
Talc—a hydrous silicate of magnesium
Steatite—a variety of talc
Stone dust (for underground dusting)—5·4% free silica, 6·3% felspar
Colliery stone dust (1)—55% SiO_2, 21% free silica, 5·7% felspar
Colliery stone dust (2)—64% SiO_2, 39·7% free silica, 13% felspar

(The silicate was present chiefly as kaolin in these stone dusts. There was practically no carbonate.)

Determination of silica in urine (method of King & Dolan [1934])

The urine is simultaneously freed of most of its pigment, any protein and of its phosphate (which interferes with the determination of silica) by precipitation with basic ferric acetate. The silica is determined colorimetrically in the filtrate as silicomolybdic acid reduced to a blue complex.

5 ml. of urine diluted with 5 ml. of water are treated with 20 ml. of ferric chloride[1] and 20 ml. of sodium acetate solution in a 250 ml. conical flask. The mixture is brought quickly to the boil, and filtered hot through a fine paper. The filtrate must be tested for iron and phosphate (see the original method, King & Stantial [1933]), and if either be found the precipitation should be repeated using larger amounts of ferric chloride and sodium acetate solutions.

20 ml. of the filtrate are transferred to a 25 ml. volumetric flask. 2 ml. of molybdate solution and 0·5 ml. of aminonaphtholsulphonic acid are added and water to 25 ml. At the same time standards are prepared from 2, 5 and 10 ml. of standard solution, with molybdate, sulphonic acid and water to 25 ml. The coloured solutions are mixed and the test compared against the nearest standard after 10 min. In the case of urines suspected of containing a large amount of silicic acid it is advisable to prepare an additional test from 10 ml. of filtrate.

Normal silica excretion

The daily excretions of a series of cats are shown in Table II. The normal daily output of silica was determined on all animals used. The figures shown are

Table II. *Urinary excretion on low silica diet*

Silica excreted		No. of cat 1	2	3	4	5	6	7	8
1st day	mg./100 ml.	1·6	1·4	1·5	1·6	1·2	0·8	0·8	0·9
	mg./day	1·8	2·2	1·7	1·8	1·9	1·7	2·0	0·9
2nd day	mg./100 ml.	1·6	1·2	1·7	1·8	1·2	0·9	0·6	0·9
	mg./day	1·7	1·4	2·2	2·3	1·7	1·9	0·9	1·4
3rd day	mg./100 ml.	0·9	1·3	0·8	0·8	1·1	0·6	0·7	0·9
	mg./day	1·2	1·7	1·3	1·4	1·6	1·2	2·4	1·2
4th day	mg./100 ml.	0·8	1·4	0·8	0·7	0·9	0·6	0·5	1·0
	mg./day	0·9	1·5	0·7	0·6	1·6	2·3	1·0	0·9
6th day	mg./100 ml.	1·1	1·1	1·0	0·9	0·8	0·6	0·6	0·8
	mg./day	1·5	1·2	1·4	1·4	1·7	1·0	1·9	1·0
7th day	mg./100 ml.	0·8	1·4	0·8	1·0	0·5	0·5	0·6	0·8
	mg./day	1·5	1·3	1·8	1·9	1·9	1·1	1·2	0·6

[1] For preparation of solutions see previous communication.

representative of the normal daily variations in silica output which are encountered with cats kept on a constant diet.

Oral administration of silica

The cats were subjected to light anaesthesia with ether, and a stomach tube was passed. 5 g. of the siliceous material were suspended in 20 ml. of milk and injected into the stomach by means of a glass syringe. An additional 10 ml. of milk were then used to wash out the syringe and tube. The animals were replaced in the metabolism cages and the daily specimens of urine taken for analysis. Two cats were usually treated on the same day with different silica or silicate samples, and the experiments repeated until at least four cats had been used for each mineral sample.

The results are given as the mg. of SiO_2 excreted in the urine in 24 hr. following administration—and in 48 hr., 72 hr. etc.—until the excretion had been followed for at least 7 days. Before this time the daily output had invariably returned to the range of the normal figures. The difference between the figures for the excretion at two time intervals (e.g. 24 and 48 hr.) represents, of course, the amount excreted during the intervening 24 hr. period.

Four representative experiments are illustrated in Table III, where the daily silica excretion is shown in four cats for each form of silica administered. The variation in the silicic acid output from animal to animal is here illustrated for

Table III. *Excretion of silica in cats' urine following oral administration of siliceous materials*

	mg. SiO_2 excreted in					
Material administered	24 hr.	48 hr.	72 hr.	96 hr.	120 hr.	144 hr.
None	1·5	3·4	5·2	6·9	8·3	9·4
	1·9	3·9	6·6	9·0	10·9	13·0
	1·8	3·8	5·6	7·4	9·0	10·6
	1·2	2·9	3·3	4·6	6·2	8·8
Average excretion	1·6	3·5	5·2	7·0	8·6	10·4
Freshly precipitated gelatinous silicic acid	25	32	35	37	39	40
	26	34	38	39	40	44
(≡5 g. SiO_2)	23	38	43	46	48	49
Average excretion	25	35	39	41	42	44
Quartz (5 g.)	6·9	19·5	20·6	22·1	24·0	25·6
	5·6	15·3	18·2	19·7	20·6	22·3
	4·6	12·5	15·9	17·5	20·8	24·1
	7·7	10·3	14·5	15·2	17·8	20·4
Average excretion	6·2	14·4	17·5	18·6	20·8	23·3
Kaolin (5 g.)	2·0	3·7	5·4	7·2	8·4	9·1
	2·0	3·4	5·2	7·1	8·1	9·1
	3·3	5·1	6·2	7·4	7·4	8·6
	1·7	3·4	4·0	4·7	5·4	6·4
Average excretion	2·2	3·9	5·2	6·6	7·6	8·4
Steatite (5 g.)	1·5	3·0	4·4	6·0	6·9	7·9
	1·1	2·1	6·2	7·8	9·2	10·5
	2·5	3·7	4·9	5·8	7·1	8·3
	3·2	4·0	5·0	6·0	7·0	8·2
Average excretion	2·1	3·2	5·1	6·4	7·5	8·7

several levels of silica excretion. The average excretions of silicic acid for all the silica and silicate preparations, samples of powdered minerals and air-borne dusts are given in Table IV.

Table IV. *Average urinary excretions of silica after oral administration of siliceous preparations and dusts*

	mg. SiO_2 excreted in				
	24 hr.	48 hr.	72 hr.	96 hr.	120 hr.
Amorphous silica:					
Silicic acid (fresh)	24·2	35·3	39·6	42·0	43·3
Silicic acid (moist B.D.H.)	11·7	20·6	23·9	27·2	29·0
Precipitated silica (dry)	5·8	8·8	11·2	13·7	16·4
Kieselguhr (acid-washed)	2·2	4·8	6·3	7·7	8·8
Crystalline silica:					
Quartz (Porcupine)	5·7	7·8	9·9	11·5	12·6
Quartz (air-sedimented—very fine)	6·2	14·4	17·5	18·6	20·8
Quartz, pure (rock crystal)	4·4	7·3	11·4	14·5	17·1
Flint	4·8	6·9	8·4	10·6	12·4
Amorphous silicates (synthetic):					
Calcium silicate (B.D.H.)	12·6	24·5	29·6	34·1	37·2
Iron silicate (B.D.H.)	18·4	22·5	24·4	27·1	28·7
Magnesium trisilicate [Mutch, 1936]	4·2	12·3	22·1	28·0	34·1
Natural silicates:					
Felspar	6·9	10·4	13·4	15·7	17·7
Fuller's earth	15·0	20·7	23·0	25·2	27·0
Kaolin	2·2	3·9	5·2	6·6	7·6
Scotch Whinstone	17·7	24·0	25·6	27·6	29·7
Steatite	2·1	3·2	5·1	6·4	7·5
Talc	4·1	6·3	7·6	8·8	9·2
Air-borne dusts:					
Mine dust (South Africa)	5·0	10·1	13·8	15·8	17·3
Pennant Rock	3·6	5·5	8·0	9·4	11·1
Somerset Greys	6·8	11·9	16·7	18·8	20·3
Stone dust (used for underground stone dusting)	3·6	5·5	7·0	8·7	9·0
Colliery stone dust (1)	2·6	3·8	5·0	6·1	6·9
Colliery stone dust (2)	2·6	5·1	6·8	8·1	11·5

Intraperitoneal administration

The animals were anaesthetized with chloralose and a retention catheter was inserted in the bladder. 2 g. of the siliceous material were suspended in 20 ml. of sterile saline and injected intraperitoneally. The samples or urine were taken for

Table V. *Excretion of silica in the urine after intraperitoneal administration of siliceous dusts*

	Av. excretion before administration	Excretion of silica (mg. SiO_2) (hourly)						
		1	2	3	4	5	6	7
Amorphous silica	0·09	0·39	1·18	1·03	0·56	0·67	0·63	—
Quartz (very fine)	0·07	0·63	0·74	0·38	0·17	0·12	0·11	—
Flint (air-sedimented)	0·05	0·12	0·35	0·21	0·12	0·15	0·17	0·17
Flint	0·08	0·28	0·18	0·11	0·13	0·07	0·10	0·08
Felspar	0·08	0·15	0·13	0·04	0·03	0·09	—	—
Sericite	0·09	0·15	0·20	0·08	0·11	0·07	0·11	0·06
Whinstone	0·08	0·15	0·09	0·17	0·15	0·18	0·06	0·09
Fuller's earth	0·08	0·26	0·19	0·18	0·05	0·04	0·07	—
Daily:								
Quartz	1·6	4·3	2·7	2·8	2·2	2·9	2·3	—
Flint (air-sedimented)	1·2	4·6	4·1	4·1	2·0	1·6	1·9	—
Flint	0·9	3·5	1·9	1·1	1·1	1·0	—	—
Felspar	0·9	1·1	1·9	1·1	0·8	1·0	—	—
Whinstone	1·5	1·6	1·4	0·5	1·0	1·1	1·1	—
Fuller's earth	2·0	2·9	2·9	1·6	0·8	1·5	—	—

analysis at hourly intervals. 10 ml. of 5 % "gluco-saline" were given by stomach tube once every 2 hr. The cats were replaced in the metabolism cages after being kept under the anaesthetic for 8 hr., and the daily excretion of silica followed thereafter until it had returned to normal. Results of several experiments conducted in this way are given in Table V.

Inhalation of silicic acid

A fog of silicic acid solution was produced by means of an ordinary atomizer blowing into a short wide tube (1 × 6 in.). 2 % sodium metasilicate, carefully neutralized with conc. HCl to avoid precipitation of silicic acid, was used. The cat was anaesthetized with nembutal, a catheter passed and a stomach tube which had a large ($\frac{3}{4}$ in.) dilatation 5 in. from its tip by means of which the oesophagus was occluded and swallowing prevented. Hourly feedings of milk or 5 % "gluco-saline" were given through the stomach tube. The nose of the cat was connected, by means of a thin rubber mask, to the tube in which the fog of silicic acid was being produced. The urine collecting tubes were changed hourly, and samples analysed for silica. A marked increase in the silicic acid of the urine was obtained which persisted for several days after the experiment was concluded. The results are given in Table VI.

Table VI. *Silica excretion during inhalation of silicic acid fog*

	Before	1	2	3	4	5	6
Hourly excretion during exposure to silicic acid:							
mg. SiO_2/100 ml.	0·6	11·7	17·7	22·0	31·6	38·2	22·0
mg. SiO_2/hr.	0·1	0·7	1·1	1·1	1·3	1·2	1·1
Daily excretion following exposure:							
mg. SiO_2/100 ml.	1·1	17·0	5·8	4·5	4·1	3·1	2·0
mg. SiO_2/24 hr.	1·3	11·9	2·9	3·6	4·8	2·5	1·6

Inhalation of amorphous silica

Well-washed freshly precipitated silicic acid from neutralized sodium metasilicate was air-dried for 2 days and finely ground in an agate mortar. The coarse particles were removed by means of a 200-mesh sieve. 5 g. of this powder were placed in a sintered glass funnel attached to the bottom of a glass column (2 × 30 in.). The large T-tube was attached to the top of the column. The cat was anaesthetized with nembutal, the stomach tube passed and its nose connected to the side-arm of the T-tube by means of the rubber mask. A slow current of air was blown through the sintered glass funnel at such a rate that a visible cloud of dust rose to about $\frac{3}{4}$ the height of the glass column. The concentration of dust at the level of the cat's nostrils corresponded to about 15,000 particles per ml. The majority of the particles were under 1 μ in size. The animal was allowed to breathe in this atmosphere for 6 hr. Samples of urine taken during the exposure showed only small increases in silica content, but the accumulated 24 hr. sample of urine, and those on subsequent days, showed a big increase over the normal daily output. The results are shown in Table VII.

Table VII. *Silica excretion following inhalation of amorphous silica*

	Daily excretion following exposure					
	Before	1	2	3	4 and 5	6
mg. SiO_2/100 ml.	0·9	4·0	3·2	4·6	2·0	1·7
mg. SiO_2/24 hr.	1·7	7·7	6·5	6·2	3·1	3·9

Inhalation of powdered minerals

Several experiments were conducted with the mineral dusts, using the type of experiment described in the preceding section. Only very minor increases in the urinary excretion of silica were obtained. Most of the specimens used gave either no increase at all, or slightly raised figures were obtained which could not be considered significant. The finely powdered pure quartz (rock crystal) gave greater urinary silica values than the other specimens, but even in this case the figures were not sufficiently increased for any importance to be attached to them. It was considered probable that in a short experiment of this sort the amount of fine dust finding its way past the ciliated epithelium of the bronchial tree into the lung was too small for detectable amounts of silicic acid to be formed. This impression was strengthened by the finding that the lungs of one of the animals submitted to quartz dusting for 8 hr. showed on analysis for total silica only 0·158 % SiO_2, which is within the normal range of silica content.

Discussion

The silicic acid excreted in the urine is probably mainly derived from the silica of the food. This is almost certainly true for humans and animals who are not habitually exposed to inhalation of large amounts of siliceous dust, and even in these cases it is possible that the bulk of the silica excreted in the urine originates in the alimentary tract—either as swallowed mineral matter or as a siliceous constituent of the diet. The hulls and bran of grain—and for this reason whole grain preparations, and vegetable foods in general—contain variable and in some cases large amounts of silica. In paper II of this series it was shown that the high urinary silica excretion, which normally takes place in a herbivorous animal, could be reduced at will to the low level of excretion pertaining in humans and carnivorous animals by altering the diet from whole grain, hay and vegetables, to bolted flour (white bread) and tomato juice. It seemed clear, therefore, that the silica of vegetable constitution was acted on by the fluids of the gastro-intestinal tract with the production of silicic acid.

In the present study the urinary excretion of silicic acid has been observed following the oral administration of several forms of silica. The synthetic amorphous varieties—both pure silica and silicates—gave greatly increased outputs of silicic acid in the urine. This finding is not surprising since precipitated silica, unless thoroughly dehydrated, is fairly soluble in alkaline fluids. The synthetic silicates are probably broken down by the hydrochloric acid of the stomach to free silica which should be readily attacked by the alkaline fluids of the small intestine.

The natural mineral forms of silica exhibit some interesting properties. In most instances the enhanced values of urinary silica excretion are related to the administration of free forms of crystalline silica or to dusts containing a high proportion of free silica, and these findings run parallel in a general way with the solution figures in ascitic fluid shown in the previous communication. But there are some contrasts. The complex natural silicates (felspar and those contained in the Scotch Whinstone), which are normally subject in the geological processes of weathering to a breakdown to simpler products, are apparently attacked to an appreciable extent. It is probable that there is some decomposition of these silicates by the hydrochloric acid of the gastric juice [cf. Whitehouse, 1937], followed by a partial solution of some of the products by the alkaline fluid of the

duodenum. Likewise, the simpler, but hydrated silicate represented in fuller's earth (a hydrated aluminium silicate) appears to be attacked. The simple silicates steatite, talc and kaolin which are natural breakdown products of the complex silicates, appear to suffer little or no solution.

The results of intraperitoneal administration correspond fairly well with the solution figures in body fluids *in vitro*. There is no complication of the solution process through a preliminary treatment with hydrochloric acid, as in the case of oral administration where the siliceous material has first to pass through the stomach. The dusts are subjected to a simple leaching process by the mildly alkaline fluid bathing the lung and peritoneum, and solution of a small proportion of the dust seems to have taken place in some cases.

The excretion of extra silica was only transitory, lasting in the most marked case for only 3 or 4 days before the normal level of excretion was restored. Only a very small proportion of the total silica injected was subsequently recovered in the urine, before the extra silica excreted became too small to represent a significant increase over the normal output.

Whether the extra silica in the urine is to be regarded as derived from the very finest of the particles present in the dust administered, or whether it represents material leached from the surface of all the particles is not clear. The transitory nature of the increase is understandable on either explanation. But if the process be thought of as a simple, though slow, solution of all the particles, then some additional explanation is necessary. In the early stages there may be an interference with the circulation of the tissue fluids by inflammatory reactions, the precipitation of fibrin etc., and later an exclusion (e.g. by phagocytosis or a walling-off of the injected mass by tissue proliferation) of the particles from the active process of continuous leaching by the body fluids.

In all these experiments massive doses of the siliceous materials were given. The results, therefore, must be considered as being associated with exposure of the animals to abnormal conditions. These are only exaggerations, however, of normal conditions of exposure, either by ingestion or inhalation of mineral material. The same processes which influence in a greater or lesser degree the disposal of small amounts of siliceous material in the normal animal, are here concerned with excessive amounts, but in which is probably not a qualitatively but only a quantitatively different fashion. It seems plain that siliceous mineral material suffers some solution in the body, and that the free forms of silica are attacked more readily *in vivo* as well as *in vitro*.

Summary

Oral administration of siliceous materials leads in some cases to the excretion of extra silicic acid in the urine. Different mineral substances influence the excretion to different degrees. Free silica is attacked to a variable extent depending on its physical and chemical condition. Several silicates are apparently unattacked and lead to no increase of urinary silica. Some complex silicates appear to suffer partial decomposition by the hydrochloric acid of the stomach, with partial solution of some of the products in the intestine and an increase of silica output in the urine. Intraperitoneally administered silica may lead to small increases in urinary silica excretion. The increases were most marked with the free forms of silica. Inhalation experiments with mineral dusts gave inconclusive results. Inhalation of amorphous silica led to increased elimination of silicic acid in the urine.

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