A CHEMICAL EXAMINATION OF SAMPLES OF WATER AND OF SHELLS FROM THE INLE VALLEY.

By Gilbert J. Fowler and M. A. Malandkar.

The chemical analyses, results of which are given in the attached tables, were undertaken after a discussion with Dr. N. Annandale concerning his investigations on the fauna of the Inlé Lake in the State of Yawnghwe on the Shan Plateau. It appeared of interest carefully to examine the saline constituents of the lake and its tributaries, and the composition of certain typical shells found in its waters, in order to discover whether any relation exists between the mineral matter present in the water and the characteristics of the lake fauna.

In particular it seemed important to determine the relation of magnesium to calcium, as an analysis by Mr. R. V. Briggs[1] of the surface water from the middle of the lake, corresponding to our samples Nos. 5 and 10, shows magnesium to be present in higher proportion than calcium, a circumstance which might be supposed to have some physiological effect on the calciferous glands of the pond snails. The present analyses fail to reveal so large a proportion of magnesium. In the majority of cases magnesium is present in relatively small amounts, and the highest proportion of MgO to CaO, in any analysis of the mineral residue left on evaporation of the water, is 1 : 2. The analysis of the shells failed to reveal more than traces of magnesium.

In Table I are given the main analyses of the water samples, in Table II the analyses of residues from selected samples of water, and in Table III, the analysis of sundry typical shells. Water samples Nos. 1 to 9 were examined about four months after collection. Water sample No. 10 was examined about one month after collection. It was thought that the determination of the hydrogen-ion concentration (P_H.) might reveal interesting differences between the samples. Colorimetric estimations with Phenol Red as indicator, showed no very striking contrasts. The other constituents were determined according to the methods given in *Standard Methods for the Examination of Water and Sewage* (American Public Health Association, 3rd Edition, 1917).

[1] *Records of the Indian Museum*, 1918, 14, 2.

The important observation was made that the residue on evaporation gave in all cases an alkaline reaction with phenol-phthalein. This at first was supposed to be due entirely to sodium carbonate, as the residue also gave a strong sodium flame, but careful examination showed that the presence of magnesium partly accounted for the alkalinity. Magnesium carbonate is appreciably soluble in water to a basic carbonate giving a strong phenol-phthalein reaction. According to Treadwell and Reuter[1] magnesium bicarbonate in solution is stable only in presence of a large excess of free dissolved carbon dioxide. The partial pressure necessary corresponds to between 2 and 4 per cent. carbon dioxide in the superincumbent atmosphere, (i.e. 20 mm. approx.) If the partial pressure sinks further, the solution loses all the free, and a portion of the half-bound carbon dioxide and a mixture of carbonate and bicarbonate results.

According to Dr. Annandale's description of the Inlé Lake[2] the amount of organic matter, and consequently of dissolved carbon dioxide and of ammonium salts, may vary considerably from time to time at any portion of the lake, owing to the formation and transport from one part of the lake to another of floating islands of vegetation. This might well account for varying proportions of magnesium being found at different times by different observers. It would seem worth while, therefore, as opportunity occurs, for further samples to be taken, under carefully observed conditions, and for rather larger volumes to be submitted to complete analysis, than were available in the present research.

The most interesting points revealed by the analysis of the shell samples, as recorded in Table III, are the practical absence of magnesium in all cases, and the variation in the percentage of silica. Before analysis, ordinary loosely adhering earthy matter was removed as far as possible, and the silica content of certain of the shells may be ascribed to a closely adhering thin layer of incrustation, due, according to Dr. Annandale, to algal activity.

[1] *Zeit. für Anorg. Chem.*, 1898, **17**, 202.
[2] *Records of the Indian Museum*, 1918, **14**.

TABLE

Results of analysis of samples of water from

Sample No.	P_H.	
	Before boiling.	After boiling.
1. Canal at Yawnghwe—water shallow and full of weeds such as *Potamogeton*, *Myriaphyllum*, *Ceratophyllum*, etc. Mollusca abundant *e.g. Taia naticoides*, *Hydrobioides nassa typica*, *Gyraulus velifer*, etc.	7·1	7·7
2. Hot spring at Nyaungwin—N.W. of Inlé Lake. Temp. of water on leaving earth 43°C. The water forming a small pool at the edge of which, further from the spring, the temp. was 40·1°. At this point a small ostracod was abundant at the bottom but it was absent at the hotter end of the pool. No other animal life was observed and water-weeds were absent.	6·7	7 7
3. Hotter spring a little N. of No. 2. The water issued at a temp. of 64°C and had a faint smell of H^2S. This was probably due to bubbles of gas issuing from the bottom of the little pools formed by the spring. There was no sign of any sulphurous deposit. No animal life observed.	6·7	7·3
4. Sample of water from warm spring about 1½ miles N. of Fort Steadman. Temp. of water 27°C at 7 a.m.: air temp. 17°C at same time. Water very clear. *Hydrobioides nassa avarix* abundant. Water forming a deep pool of considerable size.	6·9	7·5
5. Inlé lake: surface outside lake bungalow near middle of the lake, water very clear. *Taia intha* and *Hydrobioides nassa lacustris* abundant.	7·3	7·9
7. Marginal zone of Inlé Lake at Fort Stedman. Water turbid and blackish. *T. shanensis*, *Indoplanorbis exustus*, *Pachylabra maura*, etc. abundant.	6·9	7·6
8. Tai-o, stream towards the N. end of the Inlé valley. Most of the shells very thick, including a new species of *Taia* with a fairly prominent sculpture. Dead shells thickly incrusted with lime; living shells less deeply incrusted. People bathing in the water.	6·7	7·6
9. White Crow tank; a large artificial tank made about eight years ago by damming a stream in the N. part of the valley. Water clear. With dead leaves, *T. naticoides*, some with almost smooth shells, common.	6·7	7·7
10. The same as No. 5 but taken on the 25th January, 1923. Sent by Capt. W. R. Head.	7·4	8·0

Note.—All the water samples except No. 10 were collected March, 1922

I.

Inlé valley, Southern Shan States.

Parts residue per 100,000, Temp: 110°C.	The Methyl orange alkalinity in parts per 100,000 of $CaCO_3$.	Parts free CO_2 per 100,000.	Parts Chlorine per 100,000.	Remarks.
20 very little organic matter.	18·8 shows alkaline reaction with phenolphthalein.	Nil.	0·5	Sample clear and odourless sediment at the bottom which was found to be vegetable debris.
64 practically no organic matter.	46·0	8·0	1·5	Sample clear and odourless. practically no sediment.
78 practically no organic matter.	55·2	12·2	2·1	Same as No. 2.
40 practically no organic matter.	37·2	6·2	0·24	Do.
16 very little organic matter.	16·0	1·2	0·2	Same as No. 1.
60 some organic matter.	21·6	2·0	1·6	Sample not very clear. Odourless, much sediment at the bottom, a few bacteria in the sediment.
39 some organic matter.	32·8	3·8	0·8	Sample opalescent, very little sediment. A strong smell like sewage effluent but no action on lead acetate paper.
27 very little organic matter.	27·0	2·4	0·6	Clear and odourless. Very little sediment at the bottom.
14	14·8	Practically nil.	0·15	Same as No. 5.

and received 2nd May, 1922. Both samples of No. 6 arrived broken.

TABLE II.

Detailed analyses of water residues.

Parts per 100,000.

Sample No.	Residue	Silica	$CaCO_3$	$MgCO_3$	Water-soluble
5	15.5	tr.	7.1	2.3	6.1
10	13.7	0.4	6.8	4.0	*Nil.*
7	59.0	19.5	17.8	2.1	5.6
8	38.7	0.5	22.7	5.0	8.1
9	26.6	tr.	18.0	2.1	6.2

TABLE III.

Analyses of shells from Inlê Valley, S. Shan States and from Erh-hai, Yunnan.

——	Silica	Al_2O_3	CaO	MgO
1. Shell of *Acrostoma intermedium* showing calcareous incrustation; from shore of Tai-o stream No. 8	4.60	6.21	50.39	Trace
2. Shells of *Taia* sp. nov. . Tai-o stream towards the N. end of the valley. Sta. 8	4.10	1.11	53.05	,,
3. *Taia intha* Ann. Inlê Lake; surface outside lake bungalow near middle of the lake. Water very clear Sta. 5	0.22	0.73	53.80	,,
4. *T. shanensis* (Kob.) marginal zone of Inlê Lake at Fort Stedman. Water turbid and blackish. Sta. 7	0.12	trace	54.61	,,
5. *T. naticoides* (Theob.) White Crow tank; water clear with dead leaves. Sta. 9 ...	1.80	,,	53.84	,,
6. *Margarya melanoides* Nevill. Tali-fu, Erh-hai, Yunnan	0.20	*Nil*	54.51	,,
7. *Vivipara margariyoides* Ann. same locality ...	0.22	trace	54.90	,,

Figures represent percentage of constituents indicated, the calcium being present as carbonate.
No. 1 had a thick incrustation and the shell was analysed with it.
Nos. 2 and 5 had a slight incrustation and the shell was analysed with it.

BIOLOGICAL NOTE.

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In order to render Messrs. Fowler and Malandkar's chemical results available to zoologists it will be necessary for me to give further particulars about the molluscs and also to summarise their results from a malacological point of view. Very little work has been done hitherto on the comparative composition of the shells of different molluscs and none at all, so far as I know, on that of molluscan shells in relation to that of the water in which they live.

An interesting point in the chemical results is the comparatively large amount of silica apparently present in certain shells from a slow-running stream and an artificial pond and the smaller quantity certainly present in those living in a lake and in canals in the same district. It is further interesting to observe that its presence is not confined to one genus. In the Tai-o stream in the Inlé valley the shell of *Acrostoma intermedium* of the family Melaniidae appears to contain rather more silica (nearly 5 per cent) than that of a new species of *Taia* of the family Viviparidae from the same environment. The latter apparently contains a little over 4 per cent. The shell of another species of *Taia* (*T. naticoides*) from a large pond in the same valley apparently contains an appreciable amount (nearly 2 per cent) of silica, while those of *T. intha* and *T. shanensis* from the Inlé Lake contain less than a quarter per cent.

It is important to point out in this connection that conditions are abnormal both in the Tai-o stream and in the tank, in both of which a calcareous alga incrusts the shells of all the molluscs even while they are living. It is possible, therefore, that a large proportion of the silica in these shells is not proper to them but is really contained in the alga which covers their surface. Additional evidence for this view is to be found in the fact that the crust is thickest on the shell of *Acrostoma* and that this shell appears to contain the most silica. The layer of algal crust on the Viviparid shells examined, however, is so thin that it can hardly account for all the silica present.

The authors of the chemical part of this paper have examined not only shells from the Inlé valley but also those of Viviparidae from the edge of the great lake Erh-hai (Lake Tali) in Yunnan in Western

China. The results show the presence of silica in small amounts strictly comparable to those found in the shells of the same family from the Inlé Lake and much smaller than in those from the Tai-o stream and the White Crow tank.

My special object in asking Dr. Fowler to examine the samples of water and shells was to throw light on a problem which has been engaging much of my attention for some years. This problem can best be stated in terms of the phenomena with which it is concerned.

In the Inlé valley, which is situated on the limestone plateau of the Shan States at an altitude of about 3,000 feet, four species of the Viviparid genus *Taia*[1] occur living. This genus is remarkable for the tendency it exhibits to produce a highly sculptured shell with spiral ridges or series of projections. Three of the species have been found only in the valley and, indeed, only in the Inlé Lake, which still occupies a considerable part of it. The fourth species, which lives in swamps, canals and slow-running streams amidst dense aquatic vegetation, has a wide range in the Shan States, but is probably differentiated into different races in different valleys. This species is *Taia naticoides* (Theobald). It is remarkable for its extreme variability in shell sculpture. Some shells are nearly smooth, others, often from precisely the same environment, possess spiral sculpture of the characteristic kind in varying degrees. The three species from the lake are much more restricted both in geographical range and in their range of individual variability. *T. shanensis* (Kobelt) from water among the floating islands which surround the lake in a ring, has a strongly but irregularly sculptured shell and, though much less variable than *T. naticoides*, is by no means constant in shell-sculpture. *T. elitoralis* Annand, is a still more highly sculptured form and also more constant. It lives on the outer edge of the ring of floating islands in much less foul water ; while *T. intha* Annand. inhabits the clear water of the central region of the lake and is by far the most highly and regularly sculptured and also the most constant species of the three. It is, indeed, both the most extreme and the most constant living species known in the genus.

We know that similar phenomena have occurred at different geological periods and in different countries in the family Viviparidae, notably in the Plaistacian period of the tertiary[2] of Eastern Europe and the Levant and in tertiary and recent times in Yunnan.[3]

[1] For further particulars see Annandale, *Records of the Indian Museum*, XIV, pp. 101–182, (1918).

[2] See Neumayr, *Abh. k. k. geol. Reichanst.* VII, heft 3, pls. iv, V. vi, viii, ix (1875).

[3] See Neumayr in Béla Széchenyi's *Reise in Ostasien* II, p. 649, pl. iii (1888) and Mansuy, *Bull. Serv. Géol. Indochine* V, fasc. III, pls. i, ii (1918).

I have expressed the opinion elsewhere that such phenomena are probably induced in the first instance by some chemical action of the environment on the organism, but that they can have free play only in suitable conditions and when there is no bar in the path of eccentricity. It is of course impossible to say precisely when and where this influence was first exerted on the race, but I had hoped that an analysis of the water and of the shells themselves might cast some light on the conditions suitable for the development of specific differences. Interesting as are the results obtained by Dr. Fowler and his colleague they do not suggest any particular answer to the questions involved. It is evident that further investigations are necessary in more than one direction and that these investigations should be carried out in the habitat of the molluscs and not in a laboratory situated many hundred miles away. Other factors such as food and parasites would also have to be considered.

My thanks are nevertheless due to Dr. Fowler for undertaking this important enquiry. I would express the hope that it may at least lead to more extensive investigations of the kind.

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