comprehensive and accurate genealogical classification of reptiles. The principles upon which the present classification is founded are discussed and an account of the early evolutionary history of the Reptilia is included. A diagram showing the classification of the reptiles and amphibians follows this chapter.

The classification of the reptiles is outlined in synoptic form in Chapter VII. Five subclasses, Anapsida, Synapsida, Synaptosauria, Parapsida and Diapsida are listed and a brief definition of the characters of each is given. Nineteen orders and their various suborders are listed and defined briefly.

Five chapters following the synoptic classification deal with the five reptilian subclasses. In each of these the various orders and suborders, intermediate groups and families are listed and defined. One or two apparently inadvertent omissions in this treatment, such as the coelosaurid, megalosaurid and deinodont theropods, are unfortunate.

While there may not be unanimous agreement regarding the status of some groups in the classification, such as the Parapsida possibly, there can be no doubt that this is the most adequate, well-defined classification of the Reptilia that has been proposed, possibly the most natural one. It is largely in agreement with that published by Broom in 1924.

The book is illustrated by nearly two hundred text figures, partly drawings by the author and partly derived from other sources, for which credit is always given. The figures are well chosen and illustrate very well the characters discussed. Complete cross-references in the text bind text and figures together in a way that increases the value of both.

The editorial work upon the volume consists of more than the usual editorial preparation and oversight of the text and illustrations. Numerous comments in brackets by the editor call attention to different viewpoints by various authors, especially to work done since Williston's death in 1918. This applies particularly to notice of recent work by Broom, Watson and other specialists on the earlier reptiles. These comments serve partially to bridge over the gap between the preparation of the manuscript by Williston in 1918 and earlier and its publication in 1925. An alphabetical finding index of genera would have been a useful addition.

The small and convenient size of the book (three hundred pages, octavo), its attractive appearance and its reasonable price ought to insure widespread use of the volume aside from the value of its contents. It is really surprising to find the wealth of useful, accurate, thoroughly digested information this book contains in so small a space. *"The Osteology of the Reptiles"* is the final work of a distinguished morphologist and paleontologist. It is based upon many years of thorough, patient work upon the earlier reptiles by a brilliant mind. It includes also the mature results of the author's studies of the later reptiles. The information is clearly expressed and compact and is readily available to inexperienced students and more mature research workers alike.

The book fills a place long empty in scientific literature and is a credit to every one concerned with its appearance, first its author, the late Professor Samuel Wendell Williston, and also to its sponsors, Professor T. H. Barbour and the Harvard University Press, and its editor, Professor William King Gregory and his associates.

CHARLES C. MOOK

AMERICAN MUSEUM
OF NATURAL HISTORY

SPECIAL ARTICLES

THE PRESENCE OF SULPHATE REDUCING BACTERIA IN OIL FIELD WATERS

Statement of the problem: The reduction of sulphates in sea water and the waters of certain springs through the agency of living microscopic organisms has been well established by the researches of Sir John Murray and many other observers whose work will be cited in a more detailed paper to be published later. The following equations are types of those which have been postulated by these observers to explain the results of these reactions of metabolism.

1a. $CaSO_4 + CH_4 \rightarrow CaS + CO_2 + 2H_2O$.
1b. $CaS + CO_2 + H_2O \rightarrow CaCO_3 + H_2S$.
2. $Na_2SO_4 + CH_4 \rightarrow Na_2S + CO_2 + 2H_2O$.
2a. $Na_2S + CO_2 + H_2O \rightarrow Na_2CO_2 + H_2S$.
3. $CaSO_4 + 2C \rightarrow CaS + 2CO_2$.
4. $MgSO_4 + 2C \rightarrow MgS + 2CO_2$.
5. $Na_2SO_4 + 2C \rightarrow Na_2S + 2CO_2$.

Within recent years geological studies principally in the United States have disclosed evidence of the reduction of sulphates with the concomitant development of hydrogen sulphide and of carbonates in the deeply buried waters of oil fields.[1] Somewhat analogous conditions have been postulated by Siebenthal[2] in the zinc districts of Missouri, Oklahoma and Kansas.

Although most of the equations above listed were originally written to express reactions recognized as due to living organisms, geologists concerned with the reduction of sulphates in connection with oil have

[1] G. Sherburne Rogers, "The Sunset-Midway Oil Field, California," Professional Paper 117, U. S. Geological Survey, 1919.

[2] C. E. Siebenthal, "Origin of the Lead and Zinc deposits of the Joplin region," Bull. 606, U. S. Geological Survey, 1915.

assumed, not unnaturally, that sulphate reducing organisms could not live under the conditions existing in the oil sands. They have postulated therefore that reaction of the types cited could proceed without the aid of living organisms.

In view of the great geologic importance of the natural reduction of sulphates, it has seemed desirable to critically examine the validity of the two postulates:

I. That sulphates may be reduced under conditions existing in nature by reactions like those cited above, *without the aid of living organisms.*

II. That living sulphate-reducing organisms do not and can not exist in the waters found in oil sands.

The preliminary results of this critique will now be summarized, the full details being reserved for a later paper.

I

Sulphate Reduction by Dead Organic Material

While it is well known that some of the reactions cited above can take place in the absence of organisms at *high temperatures,* no experimental evidence has thus far been recorded, so far as known to the writers, to show that they can proceed at the moderate temperatures that characterize oil-field waters. On the other hand, thermo-chemical considerations seem to be opposed to their progress at ordinary temperatures.

The validity of equation No. 1, for the reduction of $CaSO_4$ by methane at ordinary temperatures, is brought into serious question by the work of Marino and Danesi,[3] who succeeded in obtaining this reduction only at high temperatures close to the explosion point for methane. At these temperatures some dissociation to CaS and CaS_2O_3 took place.

The reduction of $CaSO_4$ by carbon, as expressed by equation 3, has been investigated by Hofman and Mostowitsch.[4] Chemically pure calcium sulphate and sugar charcoal were used and the reduction carried on in a neutral atmosphere of nitrogen at atmospheric pressures. It was found that reduction began at 700° C. and was practically complete at 1000° C.

As pointed out by Palmer[5] the reaction of equation 5 is the one involved in the LeBlanc process of soda manufacture, in which the mixture of sodium sulphate and charcoal is heated for a considerable period at 1000° C. The reaction is strongly endothermic.

With regard to the reduction of ferrous sulphate by organic matter, Allen, Crenshaw and Johnson[6] conclude: "Some experiments have been tried in this laboratory in the hope of 'reducing' ferrous sulphate with organic matter, but the results have not been promising. The action of starch and glucose in aqueous solutions at 300° C. was either slight or nil. On the other hand, the possibilities of hydrogen sulphide are suggestive."

Some simple experiments were conducted by Bastin and Merritt to test further the possibilities of reduction of sulphates at ordinary temperatures by coal, crude petroleum and oil shales. Calcium sulphate and cupric sulphates singly or together were the sulphates chosen. The use of cupric sulphate, it should be noted, insured aseptic conditions and eliminated any possibility of bacterial action. Furthermore, any development of hydrogen sulphide or other soluble sulphide would immediately have manifested itself by the precipitation of cupric sulphide. Hydrogen sulphide was further tested for with lead acetate, and carbonates were tested for by acidulation with hydrochloric acid. Most of the tests were continued over periods of from 60 to 170 days. *In no instance was there any evidence of reduction of sulphates or development of carbonates.*

From the data given above, it must be concluded that no valid evidence has as yet been obtained of the reduction of sulphates by dead organic material at ordinary temperatures. It is of course entirely possible that such evidence may be obtained in the future.

II

Presence of Sulphate-reducing Bacteria in Oil-Field Waters

In 1919, G. Sherburne Rogers published the results of his very detailed chemical and geologic study of ground water conditions in the Sunset-Midway Oil Field, California. In this report he showed that whereas the surface and near-surface waters of the region are rich in sulphates, the waters associated with the oil are free or nearly free from sulphate but carry carbonate instead. He attributed the paucity or absence of sulphates to the reducing action of the petroleum, the sulphates originally present being reduced with the development of hydrogen sulphide or other sulphides. He concluded that the reduction took place at temperatures little above ordinary and through the agency of the dead organic materials of the oil.

The field and laboratory data presented by Rogers

[3] L. Marino and D. Danesi, *Gazz. chim. Ital.,* Vol. 43, pp. 423–434.

[4] H. O. Hofman and W. Mostowitsch, "The reduction of calcium sulphate by carbon monoxide and carbon, and the oxidation of calcium sulphide," *Trans. Amer. Inst. Min. Eng.,* Vol. XLI, 1910, pp. 763–785.

[5] Chase Palmer, "California oil field waters," *Econ. Geol.,* XIX, 1924, p. 628.

[6] Allen, Crenshaw and Johnson, "The mineral sulphides of iron," *Amer. Journ. Sci.,* XXXIII, 1912, p. 171.

are very convincing, but are in conflict with the experimental results that have been summarized in Section I above. It seemed desirable, therefore, to test the waters from certain oil fields to determine whether the tacit assumption of the absence in them of sulphate-reducing bacteria was correct. Unfortunately these tests could not be carried out in the Sunset-Midway Field, but the Illinois State Geological Survey offered facilities for collecting water samples from oil-bearing horizons in southeastern and southwestern Illinois. Preliminary tests on oil-field waters from Clark County were successful in showing some sulphate-reducing bacteria and were followed by the development of favorable culture mediae and by further tests of a considerable number of samples. These further tests showed such bacteria to be abundant in a large majority of the samples collected.

Samples were collected from 19 wells in southeastern Illinois and 6 in southwestern Illinois. The wells selected were most of them yielding waters low in sulphates and containing hydrogen sulphide. All the waters either have been or will be submitted to chemical analysis and the results will be reported in the final paper. All thus far analyzed are dominantly sodium chloride brines with minor amounts of K, Ca, Mg, HCO_3 and SO_4 radicles. They resemble normal sea-water in composition but are lower in sulphate content. They resemble also the salt waters associated with the oil in the Sunset-Midway Field, California.

The oil-producing horizons from which the samples were obtained ranged from Pennsylvanian to Ordovician in age and lay at depths of from 450 to 1,700 feet.

Mr. Gail Moulton and his assistant, Mr. C. R. Clark, collected many of the samples and supplied all the data concerning their field occurrence. The collections were made in bottles previously sterilized in the bacteriological laboratory, and all the precautions usual in taking samples for bacterial analysis were observed. The samples were the mixtures of salt water and oil that constitute the regular yield of the wells, the oil being much subordinate to the water. The samples were taken direct from the pumping jacks of wells that had been pumping continuously for days or weeks and the sample clearly came from the producing oil horizon with little possibility of contamination from sources higher up. Any organisms present in these salt waters were tolerant of (if not dependent on) the presence of petroleum.

The samples were submitted to bacteriological study by Frank E. Greer, in the bacteriological laboratory of the University of Chicago, within a few days of their collection.

Two culture mediae in eight phases were used as follows:

Medium 1.1—	K_2HPO_4	0.5 g.
	Asparagin	1.0
	Magnesium sulphate	2.5
	Sodium lactate	5.0
	Mohr's salt (Ferrous ammonium sulphate)	Trace.
	Distilled water	1 liter.

" 1.2—Same as 1.1 with 30 g. NaCl.
" 1.3—Same as 1.1 with 30 g. agar.
" 1.4—Same as 1.2 with 30 g. agar.

Medium 2.1—	K_2HPO_4	0.5 g.
	Ammonium sulphate	2.0
	Sodium sulphate	2.0
	Iron lactate	5.0
	Distilled water	1 liter.

" 2.2—Same as 2.1 with 30 g. NaCl.
" 2.3—Same as 2.1 with 30 g. agar.
" 2.4—Same as 2.2 with 30 g. agar.

These mediae were tubed in large test-tubes and sterilized in autoclave at 15 lbs. pressure for 20 minutes. All mediae heated in Arnold before use to expel air from liquid mediae and to melt solid mediae. All were cooled to about 45°–50° C. before inoculating. Each tube was inoculated with 10 c.c. of water sample. Duplicates of each type of medium were inoculated. All liquid mediae were sealed with sterile white vaseline. Incubation at 37° C.

The iron compounds in the mediae served as indicators. By the bacterial reduction of the sulphates H_2S was generated which reacted with the iron salts to form ferrous sulphate, FeS. In the liquid mediae this appeared as a black precipitate often clinging to filamentous bacterial growths. In the solid media it colored the sperical colonies black.

All water samples carrying enough H_2S so that they might blacken the mediae by direct inorganic precipitation on inoculation were placed in a sterile vacuum dessicator and the H_2S removed by evacuation before the inoculations were made.

Of the nineteen waters collected from the southeastern Illinois fields, all but two showed sulphate-reducing bacteria. One of those giving negative results was a water free from H_2S. Eleven showed black precipitate or numerous black colonies at the end of the second or third day after inoculation when the first observations were made. After from six to ten days, the precipitates in the liquid mediae were profuse and the solid mediae were filled or nearly filled with a black growth of coalescing colonies.

Of the six waters collected from the Waterloo

Field, in southwestern Illinois, every one showed, on the third day after inoculation, either a black precipitate or black colonies in at least one of the five culture mediae used. By the tenth day, five out of the six waters showed results with three or more mediae, many of the solid mediae being nearly or completely converted into a dense black mass of coalescing colonies.

Conclusions. No evidence has, to our knowledge, been adduced as yet for the reduction of sulphates with the formation of sulphides through the agency of *dead organic* matter at ordinary temperatures. At high temperatures such reductions do take place. Reduction of sulphates with the development of hydrogen sulphide or other sulphides has long been known to take place in the slimes of the sea-bottom through the agency of anaerobic bacteria and also in sewerage and about the orifices of certain springs.

From the work here reported it is evident that anaerobic bacteria of the sulphate-reducing type are present in abundance in some of the waters associated with oil in productive fields and it is very probable that they are largely responsible for the low sulphate content of these waters. The hydrogen sulphide so abundantly associated with most of the waters tested is probably in part at least a product of bacterial reduction of sulphates. It probably influences in important measure the quality of the oil since it constitutes a potential source of sulphur.

Bacteria other than the sulphate-reducing type are present in some of the waters, but their nature and functions have not been studied.

The oil-field waters in which the sulphate-reducing bacteria occur are similar in general composition to sea-water, and by some geologists are regarded as ancient sea waters buried within the rocks when the latter were laid down on the sea bottom. It would seem probable that any waters entombed in the rocks as long ago as Pennsylvanian times must have undergone repeated changes in concentration and at least minor changes in composition by contact with other waters and with the rocks. Whether the bacteria found in these waters to-day are lineal descendants of forms living on the sea-bottom at the time the sediments were laid down or have been introduced later by ground waters descending from the surface to the oil-bearing horizons is an interesting question that it may never be possible to answer.

Edson S. Bastin

With the cooperation of

Frank E. Greer
C. A. Merritt
Gail Moulton

The University of Chicago

TENNESSEE ACADEMY OF SCIENCE

The fourteenth annual meeting of the Tennessee Academy of Science was held at Vanderbilt University, Nashville, on November 27.

The report of the secretary showed a large increase in membership. The academy took the necessary steps to apply for affiliation with the American Association for the Advancement of Science.

Officers elected for the ensuing year: *President,* A. F. Ganier, Nashville; *vice-president,* C. A. Mooers, University of Tennessee, Knoxville; *editor,* G. R. Mayfield, Vanderbilt University, Nashville; *secretary-treasurer,* J. T. McGill, Vanderbilt University.

The program of papers was as follows: Chemistry in Its Every-day Uses, J. M. Breckenridge; A Theory of the Source of the Sun's Heat, Beecher Moore; Sanitary Conditions in Tennessee, with Special Reference to Water Supplies, Howard R. Fullerton; Reasons for Belief in the Theory of Evolution, L. C. Glenn; annual address of the president, Possibilities for a Biological Station at Reelfoot Lake, Scott C. Lyon; Observations of the Lunar Crater Eratofphenes, Latimer J. Wilson; Practice Effects on Serial Responses and their Physical Bases, Joseph Peterson; Facts which Support the Electron Theory of the Structure of Matter, Louis J. Bircher; The Great Smoky Mountain National Park Project, Henry E. Colton; The Rainbow Natural Bridge, Utah, Hugh D. Miser; A New Method of Determining Water Pollution, William Litterer.

The following resolution was passed unanimously:

Resolved, That the Tennessee Academy of Science, having as its principal objects scientific research and the diffusion of knowledge concerning the various departments of science, is directly interested in the teaching of science in the public schools and colleges of Tennessee, and this society deplores and regards as contrary to the basic principles of freedom and the best interests of citizenship, any attempt to hamper by statute the pursuit of truth and the discovery and teaching of the facts of nature. We therefore desire to record our belief that the Act of the Tennessee Legislature (House Bill No. 185, approved by the governor March 21, 1925) is in effect an unfortunate limitation of the intellectual freedom of teachers of science in our public schools; that it marks a backward step in our educational program; that it takes away important privileges of study and instruction heretofore available to students, especially those in our higher institutions. In justice to our people and for the welfare of the state, the Tennessee Academy of Science hereby earnestly recommends the repeal of the act above mentioned at the next session of the State Legislature.

Roscoe Nunn,
Secretary-Treasurer