

## 68 (2591)

## The source of energy of the sulphur bacteria.

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Bacteria which deposit sulphur, either within or outside the cell, are supposed to derive this substance from the oxidation of hydrogen sulphide (Winogradsky,<sup>1</sup> Buder<sup>2</sup>). This hydrogen sulphide is supposedly the source of energy of these autotrophs.

In an investigation of the external and internal milieu of the sulphur bacteria (to be published elsewhere) the author has investigated mass cultures of the following nine genera:

Thiorhodaceae: Thiospirillum, Chromatium, Rhabdochromatium, Amoebobacter. Thiopedia, Thiopolycoccus, Lamprocystis.

Thioleucaceae: Beggiatoa, Thiothrix.

Among other points it was noted that the organisms occurred exclusively in alkaline waters (pH 7.6-8.6) either fresh water, seawater or brine.

As H<sub>2</sub>S is a weak acid it will undergo hydrolytic dissociation in aqueous medium, and this dissociation will progress with a decrease in [H<sup>+</sup>]. We will have:

$$k_1 [\text{H}_2\text{S}] = [\text{H}^+] [\text{HS}^-] \quad (1)$$

in which  $k_1 = .91 \times 10^{-7}$  at 18° C.

$$k_2 [\text{HS}^-] = [\text{H}^+] [\text{S}^-] \quad (2)$$

in which  $k_2$  is very small, probably about  $10^{-15}$ . We have also

$$[\text{B}^+] + [\text{H}^+] = [\text{OH}^-] + [\text{HS}^-] + 2[\text{S}^-] \quad (3)$$

in which B<sup>+</sup> represents total metal ion.

$$k_w = [\text{H}^+] [\text{OH}^-] \quad (4)$$

From these four equations [H<sub>2</sub>S] can be calculated

$$[\text{H}_2\text{S}] = \frac{([\text{B}^+] + [\text{H}^+]) [\text{H}^+]^2 - k_w [\text{H}^+]}{k_1 [\text{H}^+] + 2k_1 k_2} \quad (5)$$

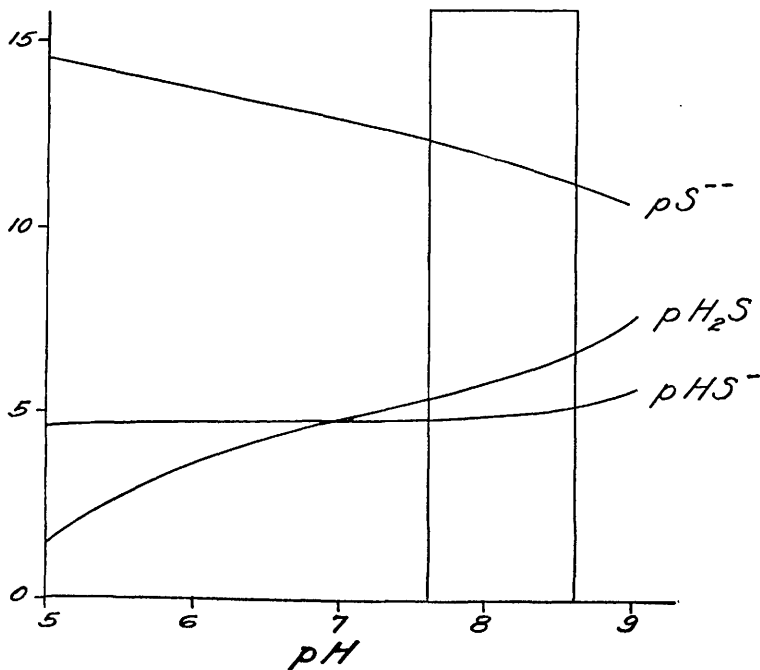
<sup>1</sup> Winogradsky, S., *Bot. Ztg.*, 1887, xlv, 488.

<sup>2</sup> Buder, J., *Jahrb. Wiss. Bot.*, 1922, vii, 231.

Assuming the metal ions to be present in a low concentration, let us say  $10^{-5}$ , the other ionic concentrations can be plotted

pH	pH <sub>2</sub> S	pHS <sup>-</sup>	pS <sup>-2</sup>
5	1.66	4.79	14.70
6	3.92	4.96	13.96
7	4.96	5.00	13.00
8	6.00	5.04	12.04
9	7.66	5.70	11.70

A graphical representation is given in the accompanying diagram.



As can be seen from the graph there is from 50 to 100 times more hydrosulphide ion than undissociated hydrogen sulphide in the natural milieu of the sulphur bacteria. There is, therefore, no *a priori* evidence that H<sub>2</sub>S *per se* is the source of energy of the S bacteria. Keil has failed to replace the H<sub>2</sub>S by sulphides, in the case of *Beggiatoa*. The author of this paper found .01 percent CaS decidedly beneficial to the growth of *Lamprocystis*. The amount of H<sub>2</sub>S in equilibrium with the metal ion is, as we have seen, dependent on the pH. It is apparent from Keil's data that most of the H<sub>2</sub>S in his solutions was decidedly in excess of the

metal ion. This gives more evidence that  $H_2S$  *per se* is inassimilable, perhaps even toxic to the bacteria.

Arranging the sulphide and hydrosulphide ion and  $H_2S$  according to their free energy levels in relation to liquid sulphur and sulphate ion, it will be seen that  $H_2S$  aq. has a lower energy level than the sulphur. It is therefore unfit to serve as a source of energy for the sulphur bacteria unless a compensating oxidation of the hydrogen would occur. As the oxygen tension in the black mud (the natural habitat of the bacteria) is extremely low, it seems more natural to assume that some other substance, like the  $HS^-$  or the  $S^{--}$  ion will be the source of energy. The low concentration of the latter ion in natural waters, however, points to the importance of the  $HS^-$  ion as the source of energy. Dehydrogenation of this ion in the absence of oxygen would imply the presence of a hydrogen acceptor in the same sense as used by Hopkins in his glutathione theory. A mechanism of sulphur formation on the basis of this theory will be developed elsewhere.

$S^{--}$	
	20000 calories
$HS^-$	
	3000
S	
	6500
$H_2S$ aq.	
	170000
$SO_4^{--}$	