

- Harbour Laboratory, Cold Spring Harbour, New York, 1976, p. 587.
6. Shepherd, N. S., Churchward, G., and Bremer, H., *J. Bacteriol.*, 1980, **141**, 1098.
 7. Leduc, E., Hoekstra, M. and Spiegelman, G. B., *Can. J. Microbiol.*, 1982, **28**, 1280.
 8. Patel, I. R. and Rao, K. K., *Arch. Microbiol.*, 1983 (in press).
 9. Adams, M. H., *Bacteriophages*, Interscience Publishers Inc. New York, 1959, p. 450.
 10. Dulbecco, R., *J. Bacteriol.*, 1952, **63**, 209.
 11. Hutchinson, C. A. III, Sinsheimer, R. L., *J. Mol. Biol.*, 1966, **18**, 429.

COMMENTS ON ESTIMATION OF SULPHATE REDUCING BACTERIA IN SEAWATER

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THE pollution of natural waters eventually leads to the multiplication of sulphate-reducing bacteria. These anaerobes thriving under rapidly reduced oxygen tension and Eh values have many undesirable consequences. One of the major problems has been an accelerated metallic corrosion of the water-front structures in industrially growing coastal towns¹.

The identification and enumeration of sulphate-reducing bacteria are of considerable interest to both pollution and corrosion scientists. One of the methods², commonly used for enumerating the sulphate reducers and to measure hydrogen sulphide generated by these anaerobes has been inadequate. A relationship between the amount of H₂S formed and the population of sulphate reducers quantified in the water sample has been found to be indifferent.

The American Standard Test Method (ASTM)² is based upon the iodometric determination of the hydrogen sulphide produced by sulphate-reducing bacteria in a suitable culture medium. Chaudhuri³ found that the formula recommended by ASTM for calculating H₂S value was "incorrect" and stated that "such values of H₂S will still be valid provided they are multiplied ten times". The sulphate-reducing bacteria according to this method are considered to be absent if the calculated value of H₂S is less than 50 ppm or the same amount as in control bottle. They are considered as present if the value exceeds 50 ppm and further, the

growth is termed as heavy if the H₂S value is as high as 350 ppm.

In our studies related to the monitoring of sulphate reducers in coastal waters, the use of ASTM method giving H₂S value as well as the extent of anaerobes in the water samples was employed. After repeated samplings monitored for one year, it was realised that the H₂S values obtained were very low even after multiplying the values by a factor of ten as suggested by Chaudhuri³. After a short incubation period, the culture bottles in reality showed a copious amount of black to gray flocculent precipitate of iron sulphide and heavy cloudiness of the medium accompanied by a very strong smell of H₂S. Higher values of H₂S were also reckoned by the intense blackening of wet lead acetate paper. The bacterial count executed simultaneously also gave very high values. In this study H₂S was estimated according to ASTM method and the sulphate-reducing bacteria were enumerated by adopting the 'shake tube' technique⁴.

Table 1 gives the values of bacterial count and the amount of H₂S generated for various volumes of seawater samples incubated. Hydrogen sulphide values corrected according to Chaudhuri's suggestion are also mentioned. It is seen that even at a corrected concentration value of 37.3 ppm of H₂S, a bacterial count is 1.2×10^4 and at a concentration of 46.2 ppm, the count is as high as 13×10^4 . Postgate⁵ considered that a count of bacteria between 10^4 and 10^6 was adequate to cause serious pollution. Therefore, ASTM grading suggesting the absence of the organisms below 50 ppm of H₂S does not seem to be correct. Sasaki *et al*⁶ observed that a concentration of 40 ppm of H₂S rendered the seawater corrosive. In any case, the H₂S values obtained by the ASTM method are very low.

Table 1 Relationship between the number of sulphate reducers and the amount of H₂S formed in seawater samples

Seawater sample (ml)	H ₂ S (ppm) ASTM method	H ₂ S (ppm) after correction	Bacterial count
0.5	2.31	23.1	4×10^2
1.0	3.73	37.3	1.2×10^4
5.0	4.62	46.2	3×10^4
10.0	5.84	58.4	13×10^4
Control	0.37	—	Nil

A communication received from ASTM conveys that "method D 993-58 part 31 (1977) will be replaced by a completely new method" and that "the new method will not give the user any indication on the amount of

H₂S that is formed or being formed". It is evident therefore that since the bacterial presence or absence in the sample is related to the amount of H₂S generated, the method will also cease to be of any assistance in estimating the sulphate-reducing bacteria in water samples.

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1. De, C. P., Deshmukh, M. B. and Rodrigues, P. V., *Proc. 5th Int. Congress on Marine Corrosion and Fouling*, Barcelona, 1980, 527.
2. ASTM, D, 993-58 in *The 1977 Annual Book of ASTM Standards*, Part 31, American Society for Testing and Materials, 1977, 843.
3. Chaudhuri, J. C., *Curr. Sci.*, 1967, 36, 430.
4. Mara, D. D. and Williams, D. J. A., In *Biodeterioration investigation techniques*, (ed.) A. Harry Walters, Applied Science Publishers, London, 1977, 115.
5. Postgate, J. R., *Bacteriol. Rev.*, 1965, 29, 425.
6. Sasaki, H., Nakahara, T., Kanda, Y., Osato, K. and Togano, H., *Boshoku Gijutsu*, 1977, 26, 229.

NOTE ON MORE ADDITIONS TO THE NATURAL ENEMY COMPLEX OF *SPODOPTERA LITURA* F. AND *MYZUS PERSICAE* SULZ. ON TOBACCO IN ANDHRA PRADESH.

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A SURVEY of natural enemies of *Spodoptera litura* F. and *Myzus persicae* Sulz. the two important pests of tobacco was carried out in the tobacco growing areas of Andhra Pradesh. Information on the indigenous natural enemies is essential for augmentation of biological control either by the promising indigenous ones or the exotic natural enemies imported for the purpose. Earlier workers^{1,2} reported the natural enemies of *S. litura* and *M. persicae* from this area. In the present investigations some more parasites and predators are recorded on these two important pests. They are as follows.

<i>Name of the parasite/ predator</i>	<i>Stage of the host</i>
I. <i>S. litura</i> F.	
A. <i>parasites</i>	
i) <i>Zele chlorophthalma</i> Nees (Hymenoptera—Braconidae)	Larval
ii) <i>Brachymeria lasus</i> Walker (Hymenoptera—Chalcididae)	Pupal
iii) <i>Lesiochalcidia? erythropoda</i> Cameron (Hymenoptera—Chalcididae)	Pupal
iv) <i>Chelonus carbonator</i> Marshall (Hymenoptera—Braconidae)	Egg-larval
B. <i>Predators</i>	
i) <i>Rhinocoris squalis</i> (Disk.) (Heteroptera—Reduviidae)	Predaceous on larval stage
ii) <i>Chrysopa crassinervis</i> Esben Peterson (Neuroptera—Chrysopidae)	Predatory on eggs and larvae.
II. <i>M. persicae</i> Sulz.	
A. <i>Parasites</i>	
i) <i>Aphelinus</i> sp. (Hymenoptera—Aphelinidae)	
ii) <i>Aphidencyrthus aphidivorus</i> Mayr (Hymenoptera—Encyrtidae)	
iii) <i>Liocyrtus aphidivorus</i> Shafee (Hymenoptera—Encyrtidae)	
B. <i>Predator</i>	
i) <i>Anisochrysa boninensis</i> Okamoto. (Neuroptera—Chrysopidae)	Predaceous on nymphs and adults

Zele chlorophthalma was earlier recorded by Kamal³ in Egypt on *Prodenia litura*. But this is the first record in India on this pest. Narendran and Joseph⁴ compared the parasitisation of *Brachymeria lasus* Walker on *Plusia peponis* and *S. litura* under laboratory conditions. Whereas the present record happens to be the first in India under natural field conditions. *Lesiochalcidia? erythropoda*, *Anisochrysa boninensis* and *Tetrastichus* sp also happen to be first time records on the respective hosts.

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1. Joshi, B. G., Sitaramaiah, S., Satyanarayana, S. V. V. and Ramaprasad, G., *Sci. Cult.* 1979, 45, 51.