Short Communication

Pak. j. sci. ind. res., vol. 32, no. 2, February 1989

CULTIVATION OF SYMBIOTIC MICRO-ORGANISMS UNSUCCESSFUL AS DUE TO PEROXIDES FORMED BY SMEARED TISSUE

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(Received January 3, 1989)

Tissues contain fats and when smeared give rise to peroxides. These are antibacterial and also injurious to healthy body cells. Tissues containing symbiotic germs, when smeared, likewise produce antibacterial peroxides and prevent isolation of germs. But leaving a piece of intestine of insects, in the midst of smears of tissue with germs, serves as reducing centres and counteracts the action of peroxides. Tissue smears with pieces of intestines left in the culture plates enable successful isolation of symbiotic germs. The role of peroxides in human pathology has been properly expounded in Bradford Research Institute in California.

Keywords: Germs, Antibacterial, Peroxides.

Buchner [1] has published a classic on symbiosis mostly between insects and micro-organisms like yeasts and bacteria. That symbiosis is a widely spread phenomenon in nature appears as the most impressive feature of this work. However, the role of symbiosis is poorly indicated since the isolation of symbiotes had not been carried out except in a couple of solitary cases. What however has greatly hampered the isolation of the symbiotes is the fact that cellular degradation products or cellular debris have been mistaken for real germs, while these were overlooked as components of normal tissues. Nevertheless in some insects, like species of Lecanium, which is a scale insect or coccid, the symbiote is an yeast-like microorganism. Even Prof. P. Linder of Berlin, who was a specialist of yeasts and an author of a handbook on that subject, could not isolate the symbiote of a Lecanium. I had the honour of knowing him personally and I can report what had been his actual experience. Now the lac insect, again a Coccid, also contains symbiotic yeasts. I tried to grow them but at first with negative results. Believing that the symbiotic germ would prefer an extract of lac insects, since it grew within their bodies, a special media was prepared. Even with such an additive to the culture media attempts to grow the symbiotic germ were negative.

Consulting literature I came across the work of Meyer [2]. He found bacteria in the nephreadia of some land molluses. Trying to isolate them he mostly got negative results. He writes (on p. 72) that "it was expected that the smearing of crushed concretions, teeming with bacteria, on suitable medium would produce a confluent growth. Instead however sterile plates or a few scattered colonies have been obtained."

Now the germs which were isolated as exception belonged to the common soil bacterium *Pseudomonas fluorescens*. That it should not grow well from tissue containing the same could not be explained. Here was a parallel case to what was observed while isolating the symbiotic yeast of the lac insect.

After much persistent efforts, I did succeed in isolating the symbiotic yeast of the lac insect and also *P. fluorescens* from *Cyclostoma elegans* and two other molluscs. What enabled me to succeed can be stated what has been mentioned before [3]. On p. 172 I wrote "When a tissue infected with a germ is teased out and exposed to air it acquires bactericidal properties speicfic to that germ. I have come to imagine that the bacteria produce fatty acids and they give rise to specific peroxides which are highly bactericidal. The one trick I resort to in culturing symbiotic germs was to leave a bit of intestine as a centre where reduction is assured and peroxide formation avoided. The lac yeasts are embeded in fatty tissue so that the insect fat itself can be a potent source of producing peroxides or toxins. It may be realized that the symbiotic germ does not grow at random within the host and is kept within control. What then is the mechanism by which the germ is kept in check by the host."

Realizing that a tissue can produce a metabolite which is antibiotic, along with a colleague, Dr. Bakshi, I prepared liver extracts. What was water soluble enabled bacteria to grow well but the fraction which was also alcoholsoluble was antibiotic (Mahdihassan and Bakshi, [4]. Further it was found that Stein [5] published an illuminating article on antibacterial substances produced by tissues. And we have to assure that peroxides, acting upon tissue metabolites,

these would function as antibiotic substances. My interest arose again when I got a copy of the publication, through my son,Dr. Mohsin Ali Hassan, "Oxidology" by Drs. R.W. Bradford, H.W. Allen and M.L. Culbert [6]. They have shown how peroxides can explain many a degenerative disease. What I had also found before was that liver extract can contain antibacterial metabolites. Such a substance is not reported in the above work, "oxidology". But it does appear that no one before had emphasized the wide occurance of peroxides causing degeneration of tissues.

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