I find that there is no blank colour. Furthermore, at the lower 1-naphthol reagent levels the pink arginine colour is always more intense than the yellow colour given by OHQ for the same concentration of arginine, and a two-to threefold increase in the sensitivity of the original reaction may readily be achieved.

The monosubstituted guanidine compounds, methyl guanidine, 1-guanidino acetate, 1-guanidino butyrate and octopine (N-[1-carboxyethyl]arginine) are known to show a similar colour reaction to arginine 1,2. The molar chromogenicity of these and some other compounds is indicated in Table II. Not all monosubstituted guanidine derivatives are 'Sakaguchi-reactive', e.g. canavanine yields no coloured product. Under optimum reaction conditions, methyl guanidine is the most chromogenic of the Saka-GUCHI-reactive compounds. It is of interest that the Sakaguchi products of human and bovine albumin have equal molecular extinction per guanidine group and each is equal to that produced from 1-guanidino acetate (Table II). Trace amounts of copper have no effect on the SAKAGUCHI reaction of albumins. Furthermore, mild alkaline hydrolysis (2N KOH, 1 h, 20°C) of albumin has no appreciable enhancing effect on the subsequent

SAKAGUCHI colour yield, so that the reaction may yet have analytical applications in the chemistry of macromolecules that contain monosubstituted guanidines 10.

Zusammenfassung. Es werden neue Beobachtungen über die Zweckmässigkeit der Sakaguchi-Farbreaktion für Arginin mitgeteilt: Aminosäuren, die die Farbreaktion gewöhnlich hindern, wirken in niedriger Quantität (0,05 bis $0.2~\mu M$) verstärkend. Histidin ergibt eine positive Sakaguchi-Reaktion.

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10 These observations were made at the Regional Urological Centre, Liverpool, England, during the course of a study of blood serum amidine transferases, full details of which will be published elsewhere. The work was supported by a grant from the Department of Surgery, University of Liverpool.

Evidence for an Intracytoplasmic Membrane in the Core of Spores of Bacillus popilliae

Fine structural studies reveal bacterial spores to be cells of singular complexity. Particularly striking is the elaborate layering in the integument. Few published electron micrographs, however, show much structural detail in the so-called 'core', the protoplast of bacterial spores.

Experiments on the fine structure of *Bacillus popilliae* have recently yielded a surprisingly detailed spore core, prominent in which is a membranous element described herewith.

Spores of Bacillus popilliae, harvested from the hemolymph of infected larvae of Japanese beetles, were fixed

¹ C. L. Hannay, J. biophys. biochem. Cytol. 9, 285 (1961).

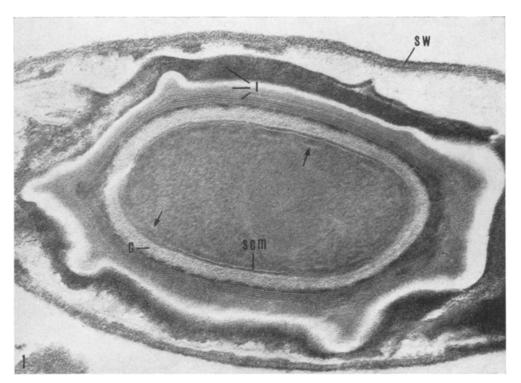
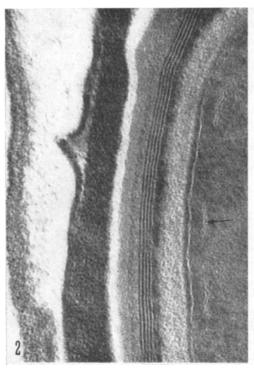


Fig. 1. Longitudinal section of a spore of Bacillus popilliae. Note the sporangial wall (SW) and the various layers of the spore wall (I) peripheral to the cortex (C). The core is bounded by the spore core membrane (SCM) parallel to which runs interruptedly another membrane (arrows) of comparable dimensions. ×93,500.



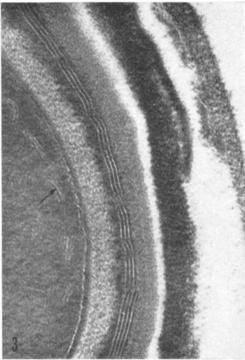


Fig. 2. Enlargement of a small section of the spore shown in Figure 1. Note the prominent segments of intracytoplasmic membrane. \times 170,000.

Fig. 3. Section of another spore showing the intracytoplasmic membrane. \times 187,500.

at 24°C for 3 h in 2% (w/v) KMnO₄ and 2% (w/v) OsO_4 in veronal acetate buffer, pH 6.1, bathed in 0.5% (w/v) uranyl acetate in veronal acetate buffer for 2 h, and embedded in Araldite (Cargille Laboratories Inc., New York). Sections were cut with a diamond knife on a Porter-Blum microtome and viewed in an RCA electron microscope (EMU 3G) with an accelerating voltage of 50 kV.

Figure 1 pictures a spore cut longitudinally. Inside the complex integument, consisting of the sporangial wall together with the various spore coats, and the cortical region, is the core, bounded by a 'unit' membrane² that lies just beneath the inner layer of the cortex. This spore core membrane encloses a cytoplasm that is packed with electron-dense particles corresponding in size to ribosomes. Centrally located is a large, ovoid body that probably contains the nucleoplasm. Just inside the spore core membrane, and coursing interruptedly but roughly parallel to it, is another membrane-like structure the dimensions of which are similar to those of the spore core membrane; but continuity of the two has not been observed. Details of this structure are shown in Figures 2 and 3.

The functional significance of this intracytoplasmic membrane is not known. These membranous wisps may be

intrasporal mesosomes, the existence of which Fitz-James has shown in developing forespores of *Clostridium pectinovorum* but which, to our knowledge, have not been observed before in the core of mature spores of *Bacillus* species.

Zusammenfassung. Die Arbeit bringt den elektronenmikroskopischen Beweis für das Bestehen einer intracytoplasmatischen Membran im Cytoplasma ('core') der Sporen von Bacillus popilliae.

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Department of Microbiology, Baylor University College of Medicine, Houston (Texas, USA), August 6, 1965.

- ² J. D. Robertson, Biochem. Soc. Symp. 16, 3 (1959).
- ³ P. C. Fitz-James, J. Bacteriol. 84, 104 (1962).
- ⁴ This investigation was supported by a research contract from the Northern Utilization Research and Development Division, US Department of Agriculture, Peoria (Illinois).

Fate of Injected 4-Iodoantipyrine (131I) in Rats

Radioiodinated 4-iodoantipyrine (RIAP) was used early in the study of the total body water compartment 1,2. Recently an increasing use for it was found in the measurement of cerebral 3,4 and coronary blood flow 5,6. Straub et al. 7 have pointed out that the RIAP is converted rapidly to a more diffusible compound: radioiodide, which invalidates the use of this compound in total body water studies.

In this experimental work, the distribution and elimination of the RIAP and its metabolic products have been studied on the wider basis of total body distribution and more time of observation.

Material and methods. 7 groups of 5 adult Wistar rats each were injected through the tail vein with 300 μ C of RIAP (specific activity 480 μ C/mg and radiochemical purity 99.0%). Then the animals were sacrificed at different intervals. The radioactivity of the different organs was measured with a scintillation counter and the values,