Rifampicin is highly active against experimental tuberculosis in the mouse and has also established itself in clinical practice as a first-line tuberculostatic. It was therefore of interest to examine the activity of the drug in animals against another facultative intracellular microorganism, *Listeria monocytogenes*, and compare it with that of antibiotics recommended for the treatment of human listeriosis. In vitro, rifampicin was considerably more active against *Listeria* than tetracycline, chloramphenicol and sulphadiazine; the minimum inhibitory concentrations, however, were only slightly lower than those of ampicillin, penicillin G and gentamicin. These substances were tested in mice infected in various ways with *Listeria* and treated according to various dosage schedules. In all the experiments rifampicin proved very much superior to the other substances, being active in doses at least 30 times lower. After a single dose, the count of viable *Listeria* in the spleens of the treated animals was reduced more quickly by rifampicin than by ampicillin in a dose roughly 300 times greater. In animals given rifampicin in combination with ampicillin or tetracycline a synergistic effect was observed. The demonstrable superiority of rifampicin over ampicillin in animal experiments is presumably not due to the slight difference in their activity in vitro, but to the effect of rifampicin on intracellular micro-organisms. In this connection, however, the longer sojourn of rifampicin in the body of the mouse must also be taken into consideration.

Systematical Research of *Salmonellas* in Sewage and River Water in Strasbourg and Outskirts: Public Health Problem

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During the period from 16. 9. 1974 to 12. 11. 1975, in order to make a microbial study of sewage and of the river Ill in Strasbourg, we received each week 2 samples, taken at 41 different places, to trace *Salmonellas*. We treated 123 samples (each place is then represented three times at different seasons of the year) in which 75% contained *Salmonellas*; 209 strains were isolated – with 32 serotypes – from the groups B, C, D, E and G. We have to mention the great diversity of these serotypes; *Typhimurium* (13,5% of the 209 strains), Para B (10,5%) and Panama (10%) were the most frequently isolated. In many samples we often found 4 to 6 different serotypes. Finally we noted that the river Ill contains little *Salmonellas* when compared to sewage. Moreover the samples of sewage near hospitals and clinics did not contain more *Salmonellas* than others.

Description and Physiological Properties of an Autotrophic Hydrogen-Oxidizing *Spirillum*

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Two strains, SA 32 and SA 33, of a facultatively autotrophic hydrogen-oxidizing *Spirillum* were isolated from the water of a small eutrophic lake by the membrane filter method, using a mineral agar medium and an atmosphere of $H_2 + O_2 + CO_2$. They appear as typical, medium-sized spirilla, 0.6–0.8 μm in diameter, with bipolar lophotrichous flagellation. Spira has a wave length of 3–4 μm with clockwise orientation. Growth occurs either autotrophically in mineral medium under knallgas and carbon dioxide, or heterotrophically, with many organic acids or amino acids as sole C-source. Carbohydrates are not used as substrates, except for sugar acids such as gluconate and 2-keto-gluconate. Denitrification does not occur, but nitrates, as well as ammonium ions, urea and asparagine, are used as sole *N*-source. The G + C content of DNA is approximately 60–62%. Autotrophic growth is microaerophilic. Generation times of about 4.5 h were measured in the fermentor, with frequent adjustment of the oxygen partial pressure to cell concentration. With succinate as sole C-source, the generation time under air was about 2 h. Hydrogenase is localized in membranes and fails to reduce either NAD of NADP. It is inducible by H_2. Oxygen represses hydrogenase synthesis at concentrations higher than 1.8 mg/l, but has no inhibitory effect on the oxy-hydrogen reaction in vivo up to 11,3 mg/l. Autotrophically-grown cells are able to oxidize acetate, gluconate and succinate without a lag phase, but at a reduced rate compared to substrate-induced cells. Citrate is oxidized only after a lag phase.

Involvement of 4-Aminobutyrate Aminotransferase in Arginine Biosynthesis and Putrescine Catabolism of *Pseudomonas aeruginosa*

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4-Aminobutyrate aminotransferase (GABAT) from *Pseudomonas aeruginosa* was purified 64-fold to apparent electrophoresis homogeneity from cells grown with 4-aminobutyrate as the only source of carbon and nitrogen. Purified GABAT catalyzed the transamination of 4-aminobutyrate, N^2-acetyl-L-ornithine, L-ornithine, putrescine, L-lysine, and cadaverine with 2-oxoglutarate (listed in order to decreasing efficiency as substrates). The enzyme is induced in cells grown on 4-guanidinobutyrate, 4-aminobutyrate or putrescine as the only carbon and nitrogen source. Cells grown on arginine or on glutamate contained low levels of the enzyme. The regulation of the synthesis of GABAT as well as the growth properties of a mutant with an inactive N^2-acetyl-L-ornithine 5-aminotransferase suggest that GABAT functions in the biosynthesis of arginine by converting N^2-acetyl-L-ornithine 5-semialdehyde to N^2-acetyl-L-ornithine as well as in catabolic reactions during growth on putrescine or 4-guanidinobutyrate but not during growth on arginine.

Studies on the Mode of Action Phenylmercuric Borate on *Escherichia coli*

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The bactericidal activity of the disinfectant phenylmercuric borate (PHB: Phenylhydrargiriboras) was investigated on *Escherichia coli*. In the first step of this study, we attempted to localize PHB in cells exposed to it. The fractionation by differential centrifugation of a cell free extract of *E. coli* exposed to the drug gave about the same distribution of PHB and proteins in the different fractions. This result shows the poor specificity of PHB in regard to the numerous proteins of the cell, all of which appear as potential fixation sites. The fractionation by density gradient centrifugation permits the isolation of

cytoplasmic membrane vesicles whose content in PHB is noticeably higher. Thus, cytoplasmic membrane can be considered as a concentration site for PHB. The amount of PHB incorporated in cells is approximately proportional to the applied concentration. The incorporation kinetics show, in the first 2 min of exposure, a very rapid fixation, following which, the speed decrease rapidly. 80 to 100% of PHB incorporated in 30 min is already in the cells after the first 5 min. PHB binds directly to proteins or other SH- containing compounds without involvement of an active transport mechanism; moreover localization does not differ in cells exposed for a short time (5 min) or for a longer time (> 30 min). By comparing with an inorganic mercury, such as Hg $(NO_3)_2$, PHB appears to be more rapidly incorporated, but localization seems to be the same. The inhibitory effect of PHB on protein synthesis was demonstrated in an exponentially growing culture, whose L-leucine incorporation into cells was immediately blocked after addition of PHB.

Effect of Colimycin on the Decarboxylation of Pyruvate from Serine in *Escherichia coli* and *Citrobacter intermedius*

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The inhibition of pyruvate decarboxylase by colimycin in *E. coli* and *C. intermedius* has been assayed by enzymatic titration of pyruvate. The bacteria were maintained in undivided conditions since they were deprived of elements necessary for growth. The catabolism of serine passes through pyruvate when they receive the latter only in the form of either D, L or DL. In normal conditions one finds, for example, 0.17% (in *E. coli*) and 0.12% (in *C. intermedius*) of the initially given DL-serine as pyruvate in the medium after 4 h. After treatment with colimycin, pyruvate has a tendency to accumulate in the medium and the values are 1.47% for *E. coli* (about 9 times more) and 9.46% for *C. intermedius* (about 79 times more). However, between 2 and 4 h, the pyruvate in excess represents only 5 to 30% of the amount of serine degraded, no matter the type of serine provided. The decarboxylases producing pyruvate in the bacteria are only partially inhibited.

In conclusions, it seems that there is an inhibition of the pyruvate decarboxylase in *E. coli* and *C. intermedius* by colimycin, although the decarboxylation is not entirely inhibited. This suggests that not all decarboxylases are accessible to the antibiotic. (GOUDA, SCHORER and CHODAT, Archs Sci., Genève *18*, 591–598, 1965; MARTIN-HERNANDEZ, FUENTE-SANCHEZ and SANTOS-RUIZ, Rev. espan. Fisiol. *12*, 143–152, 1956.)

Antigens Extracted from Oral Microorganisms in Lymphocyte Stimulation

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Periodontal disease can be induced in a gnotobiotic rat model (Infect. Immun. *10*, 565, 1974). Rats monoassociated with *Actinomyces viscosus Ny 1* and fed with a high sucrose containing diet developed large amounts of dental plaque, gingival inflammation and severe bone loss. Antigen dependent lymphocyte reaction was studied in a miniculture lymphocyte stimulation assay. Lymphocytes were collected from either germ-free or conventional Sprague-Dawley rats by canulation of the thoracic duct. These cells were incubated with bacterial antigens for 24 h in serum-free RPMI 1640 medium. After cell harvesting ^{3}H-uridine incorporation was measured in a liquid scintillation counter. Lymphocytes from germ-free animals were activated by the extracellular heteroglycan of *A. viscosus*. No stimulation resulted when cells from conventional animals were used. Rantz-Randall extracts prepared from 10 different microorganisms were tested in the same culture system. An increased ^{3}H-uridine incorporation was measured only in cells from conventional animals incubated with the extract from *Pseudomonas aeruginosa* and *Proteus mirabilis*. This effect might be due to bacterial endotoxin. A comparison between the lymphocyte activation of germ-free animals and conventional animals suggests the presence of a suppressor factor in cultures with cells from conventional animals. The immune reaction of conventional animals as evaluated by this test system may be compared to that of human peripheral leucocytes obtained from healthy subjects.

Regulation of the Glyoxylate Cycle in *Acetobacter aceti*

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Growth of *Acetobacter aceti* LBG B 4114 on ethanol was inhibited for several hours by glyoxylate but not growth on pyruvate. During growth of the organism on pyruvate, no measureable activity of isocitrate lyase, the key enzyme of the glyoxylate cycle, could be detected. The glyoxylate cycle and especially the enzyme isocitrate lyase appeared therefore as a possible site for the growth inhibiting action of glyoxylate. Either the activity or the synthesis of the enzyme or both could be controlled by glyoxylate. Isocitrate lyase was strongly inhibited by succinate ($I_{0.5(V)} \leq 1.7$ mM), inhibited to a lesser extent by phosphoenolpyruvate (2.5 mM), malate (5.5 mM), oxaloacetate (8.7 mM), fumarate (18.2 mM) and not at all inhibited by glyoxylate. This is in contrast to observations with *Acinetobacter calcoaceticus* (KLEBER, Acta biol. med. germ. *34*, 723, 1975), where not only the other compounds but also glyoxylate were inhibitors of isocitrate lyase. The time course of growth and specific activity of isocitrate lyase was studied in shake flask experiments with and without glyoxylate in the growth medium. Without glyoxylate, there was a rapid rise of specific activity at the onset of growth, a maximum in the middle of the log phase (7 h) and a sharp decline towards the end of the log phase (10 h). With small amounts of glyoxylate in the growth medium (4 mM), there was practically no isocitrate lyase activity as long as growth inhibition by glyoxylate lasted (about 10 h). As soon as about one half of the glyoxylate had disappeared from the medium and growth was resumed, there was an increase in specific activity of the enzyme up to less than half the maximal value of growth without glyoxylate. To our knowledge, repression of isocitrate lyase by glyoxylate has not been observed before.

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