using a toxin-immuno sorbent column; conjugates of these antibodies and horse radish peroxidase were used for the assay.

The ELISA developed showed no cross reactivity with two other enterotoxins of *S. aureus* tested i.e. enterotoxins A and C. Since antibodies from sheep were used the cross reactivity with protein A, a cell wall component of *S. aureus* having a high affinity for the Fc-part of immunoglobulins, was very low i.e. less than 1:2000 compared with enterotoxin B. Apart from application of the assay to detect the presence of toxin in raw materials and products, the ELISA test was used to analyse heat denaturation of the toxin. The observed decimal reduction time of immunoreactive toxin at 90°C, using the ELISA, was about 90 min. This is in agreement with the high degree of heat resistance as reported from bioassay measurements (Schantz et al., 1965).

Our results indicate that the ELISA-technique is very appropriate to analyse routinely minute amounts of toxic contaminants in foods.

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Mixed substrates and mixed culture

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The last few years have seen a growing interest in the growth of microorganisms on mixed substrates is only observed when either one or both substrates are in excess, and that simultaneous ecology of microorganisms. In the first place, it has been shown that the biphasic utilization of two substrates is only observed when either one or both substrates are in excess, and that simultaneous utilization under limitation of growth by the two substrates is the rule rather than the exception. Secondly, it is now realized that not only in the natural environment but also in many waste water treatment plants, growth of microorganisms is often simultaneously limited by more than one (organic) nutrient. Thirdly, in industrial production processes the nutrients seldom contain only one carbon and energy source, but mostly a mixture of organic substrates. Fourthly, it has now become apparent that many recalcitrant molecules can only be metabolized by microorganisms when a second "auxiliary" substrate is available. Finally, the interest in mixed substrates has increased since these, in many cases, form the basis for the coexistence of microorganisms in mixed cultures. In themselves, mixed cultures have drawn much attention from both basic and applied science due to their importance in ecological systems, in waste water purification plants and because of a renewed interest in the use of mixed cultures for production processes.

Biphasic growth of pure cultures on mixtures of two substrates is a well-known phenomenon, an example of which is the diauxic growth of *E. coli* on a mixture of glucose and lactose. Silver and Mateles (1969) were the first to show that growth under dual limitation by a mixture of two carbon and energy sources was possible: In a glucose- and lactose-limited chemostat, *E. coli* utilized these compounds simultaneously at lower dilution rates. Above a certain dilution rate, *E. coli* gradually switched to the exclusive use of glucose. In a variety of experiments by several authors, it has now been shown that simultaneous utilization is possible for varying mixtures of acetate-formate; acetate-oxalate; oxalate-formate (*Pseudomonas oxalaticus*, Dijkhuizen and Harder, 1979a, b); of acetate-thiosulfate, fructose-thiosulfate, glycolate-thiosulfate (*Thiobacillus* A2, Gottschal et al., 1979; Gottschal and Kuenen, 1980b); of mannitol-methanol (*Paracoccus denitrificans*, Van Versveld et al., 1979) and glucose-arginine (coryneform bacterium, Law and Button, 1977).

During mixed substrate utilization, enzymes required for the metabolism of the two substrates are often proportionally adapted to the required turnover rates of the two substrates. Sometimes growth on mixed substrates is more efficient (in terms of yield per mole substrates utilized) than on the single substrates. (Gottschal and Kuenen, 1980b; Van Versveld et al., 1979). In the natural environment, a mixture of many substrates is often present at growth-limiting concentrations and it might therefore be expected that during evolution microorganisms have been under a constant pressure to develop and improve their ability to utilize more than one substrate simultaneously. Such a strategy might have led to development of metabolically highly versatile organisms. An alternative strategy may have been for a pronounced specialization towards the ability to utilize one or few substrates. One such example is the occurrence, among the colorless sulfur bacteria, of specialist *Thiobacillus* species, the obligate chemolitho(auto)trophs, and of versatile *Thiobacillus* species, the facultative chemolithotrophs or "mixotrophs". In the natural environment these organisms can be shown to coexist and thus must have different ecological niches. In mixed batch cultures, in the presence of excess substrates, the versatile thiobacilli always grow more slowly than specialist chemolithotrophs or specialist heterotrophs. It has now been shown in our laboratory and also by Smith and Kelly (1979) that, during growth limitation of chemostat cultures of these organisms by mixtures of acetate-thiosulfate, glycolate-thiosulfate or glucose-thiosulfate, the versatile thiobacilli can dominate or even sometimes outcompete the specialist-chemolithotrophs and heterotrophs. Using this principle, it has now been possible for the first time to reproducibly enrich in continuous culture for facultatively chemolithotrophic *Thiobacillus* species from different fresh water samples (Gottschal and Kuenen, 1980a).

Although in both the model experiments, with only two or three different species, and the chemostat enrichment cultures the versatile organisms may become dominant, the coexistence of two species is often observed. This is in accordance with mathematical models which have been constructed by several authors including some from our laboratory. These models predict that the number of species that can coexist must always be lower or equal to the number of growth-limiting substrates that are available (Taylor and Williams, 1975; Yoon et al., 1977; J. C. Gottschal and T. F. Thingstad, submitted). This holds under the condition that no other interactions occur. However, the coexistence of more species than predicted from the mathematical model is often observed. This is undoubtedly due to interactions other than pure competition (Frederickson, 1977). In the experiments described (Gottschal et al., 1979; Gottschal and Kuenen, 1980a), we observed that, for example, vitamins and metabolic products such as glycolate were excreted by one organism and utilized by another. Of course, many other versions of interactions would be possible. An interesting point which emerges from the same mathematical modelling is that the yield of the different organisms on the various substrates may play an important role in the outcome of competition. This is in contrast to the competition of microorganisms for one substrate where, at least in theory, only $\mu_{\max}$ and K_s for the substrate play a role in the competition for the growth-limiting substrate.

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