DEVELOPMENT OF SELECTIVE BIOSENSORS EMPLOYING IMMOBILISED MICROALGAE

In recent years, biosensors are discussed as an alternative technology in the analysis of environmental perturbations. Regarding the potential of algal diversity in biotechnology, this study aimed at utilising the advantages of biosensors incorporating microalgae.

As a requirement for the long-term use of microalgae in biosensor systems, a novel procedure for the stable immobilisation of microalgal cultures was developed. The method is based on the separation of the immobilised cells from a continuous flow of culture medium by a semi-permeable membrane and provided a sufficient physiological stability of microalgal strains from different taxa over a period of several weeks. By means of the new immobilisation technique a microalgal biosensor system was constructed. The signal of the biosensor was measured by a PAM chlorophyll fluorometer in terms of photosynthetic fluorescence induction of immobilised algae. The simultaneous application of different algal strains was enabled by the development of array plate biochips (algal sensor chip: ASC) in association with a fluorescence imaging technique.

To characterise the biosensor system, gaseous and water soluble toxicants were used in toxicologically relevant concentrations: Methanol (50-500 ppm) and formaldehyde (0.01-10 ppm) vapours were significantly detectable within minutes by a concentration-dependent biosensor signal derived from the green alga *Klebsormidium*. Additionally, repeated exposure experiments revealed a reversible and reproducible signal of the biosensor during 30 days of operation in gas phase. Detection of the herbicides atrazine, simazine, diuron, isoproturon and paraquat was also performed rapidly in aqueous solutions using several algal strains at detection limits from 0.1 to 5 $\mu g\, l^{-1}$, depending on the herbicide and strain used. Diverse sensitivities of microalgae to toxicants were employed to obtain selectivity of the biosensor. Methanol and formaldehyde were significantly identified by the compound-specific response rate as a ratio of the *Klebsormidium*-biosensor's responses of two different strains. By the same method, a complex herbicide-specific response pattern (RP1) was generated for each of five different herbicides using the biosensor signals of nine ASC-immobilised algal strains. For evaluation of selectivity, an identical set of RPs (RP2) was recorded. Comparing RP1 and RP2, a relatively high congruence was observed between RPs of identical herbicides, while RPs of different herbicides showed higher deviations.

This study suggested that the diversity-based, selective real-time detection of toxicants can be realised employing the algal sensor chip developed here. However, further improvements will be needed.