ABSTRACT

Some Aspects of The Flow Kinetics and Applications
of Free and Immobilized Urease

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This thesis deals with the immobilization of urease on to 2-fluoro-1-methylpyridinium salt activated support. The study seeks to characterise this immobilized urease in terms of its kinetic parameters and properties. It also looks at the potential use of immobilized urease in bioreactors and biosensors for the determination of urea in serum samples using flow injection analysis.

An evaluation of three different detection methods (potentiometric, colorimetric and fluorometric) for urea serum analysis using immobilized urease bioreactor was done. Of these fluorosence method appears to be the most efficient with a sample throughput rate of about 25 per hour. Its within-day CV was only 1.12% and the day-to-day CV was 1.25% for serum containing 10.50 mg urea nitrogen dL⁻¹. Also the urease bioreactor shows

excellent storage (at 4° C) and operational stabilities (at 37° C).

An FMP-activated PVA urease electrode is also described for the analysis of urea in serum samples. Recovery yields with this biosensor varied between 97%-102%. The within-day CV was 1.2 - 2.7%, whereas the day-to-day CV was 2.0 - 4.5%. Up to 30 serum samples can be analysed per hour. There is only a 14% decrease in biosensor activity over a period of 28 days.

The possibility of L-arginine determination using co-immobilized arginase/urease bioreactor was also investigated. Up to 160 samples can be analysed per 8 hour work day with a detection limit of 3 x $10^{-6} \mathrm{M}$ L-arginine using potentiometric detection incorporating a flow injection analysis system.

Finally a method for the potential use of free urease activity measurements as a basis of a screening method for the detection of trace amounts of mercury and silver is also described. Detection limits of 0.10 ng/g and 2 ng/g respectively were obtained for silver and mercury analyses. A sample throughput state of 30 samples per hour were obtained under flow injection analysis conditions utilizing fluorescence detection.