[PD4-12] [10/19/2000 (Thr) 15:00 - 16:00 / [Hall B]]

Amperometric Enzymatic Determination of Ethanol Using Potassium ferrocyanide as the Mediator

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Alcohol oxidase specifically reacts with alcohols producing aldehydes and hydrogen peroxide. Hydrogen peroxide itself can be measured amperometrically at a relatively high positive potential but ascorbic acid and other drugs in the biological fluids may interfere. In this study potassium ferrocyanide(Fo) is coupled to hydrogen peroxide using peroxidase(PO) enzyme so that the rate of overall enzymatic reactions is determined by the alcohol oxidase(AO) reaction. Then potassium ferricyanide(Fi) produced by the peroxidase reaction is measured at - 50 mV which is low enough not to have interference. A thin layer glassy carbon flow cell is connected to the flow injection system for the measurement. According to the kinetic studies, the enzymatic reactions reached to equilibrium in two minutes at 25oC with a solution containing 9.0 U/ml AO, 50 U/ml PO, 2.5 x 10-4 M Fo and 2.5 x 10-5 M ethanol. Ethanol was also completely converted to Fi. The present analytical method offers the advantage of quantification using Fi standard addition and requiring small volume(200 μl) of solution per measurement.

[PD4-13] [10/19/2000 (Thr) 15:00 - 16:00 / [Hall B]]

Spectrophotometric Studies of Hydrogen peroxide -Producing Oxidase Reactions Using Tetramethylbenzidine as the Mediator

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Glucose, cholesterol and ethanol are the clinically important substances which are also the substrates of glucose oxidase(GO), cholesterol oxidase and alcohol oxidase, respectively. These oxidases commonly produce hydrogen peroxide(HP) which can be coupled to 3,3',5,5'-tetramethylbenzidine(TMB) speedily by peroxidase(PO). As the preliminary studies to develop a visual sensor for ethanol, we have studied absorption spectra of the reaction mixtures(reacting period: 2 min) consisting of 0.4~1600μM HP, 0.4 mM TMB and 36 U/ml PO. At the low concentrations of HP(0.4~40μM), the reaxtion mixtures developed yellow color and showed λmax at 405nm. At the higher concentrations the solutions turned into green ~ blue green showing two λmax at 370nm and 650 nm. Similar spectrophotometric studies were performed with the mixtures of 0.16~112μM glucose, 0.4U/ml GO, 0.4 mM TMB and 36 U/ml PO reacted for one minute. At the glucose concentrations less than 8μM, yellow colors were developed showing λmax at 400nm At the higher concentrations, two λmax at 370nm and 650 nm were also appeared. Absorbances at λmax were linearly increased as the concentrations of HP or glucose were increased. Spectrophotometric studies of ethanol-alcohol oxidase reaction will be presented.

[PD4-14] [10/19/2000 (Thr) 15:00 - 16:00 / [Hall B]]

Differential Pulse Polarographic Studies of Cephalosporins

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Many organic functional groups including hydroxyl, carbonyl, carboxyl, nitro and azomethine are contained in the structures of the antibiotic substances of which potencies are measured by microbial assays. Although differential pulse polarography(DPP) offers the advantages of speed and accuracy for the analyses of antibiotics which posess the electrochemically reducible functional groups, DPP has not been employed in the official assay methods in Korea. Cefotaxime sodium, ceftriaxone sodium and ceftazidime have been investigated to develop their DPP analytical procedures using phosphate and tartarate buffers of various pH values. DP polarograms of these cephalosporins showed one, two or three peaks which were shifted toward the negative potentials as the pH values of the buffers were increased. This indicates protons are involved in their electrochemical reduction. Cefotaxime sodium exhibited a stable and sensitive peak at -1.016 V in a phosphate buffer of pH 7.0. Ceftriaxone sodium and ceftazidime were stable and sensitive in a phosphate buffer of pH 6.0: Ep1;-0.852 V, Ep2;-1.208 V with ceftriaxone sodium and Ep;-0.628 V with ceftazidime. The peak currents(Ip) were linearly increased as the concentrations of each cephalosporin antibiotic were increased.

[PD4-15] [10/19/2000 (Thr) 15:00 - 16:00 / [Hall B]]

Quality control of Polygonati Rhizoma

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Polygonati Rhizoma, the root of Polygonatum sibiricum, has been used as an antibiotic, antifungal and hypotensive in traditional medicine. Azetidine-2-carboxylic acid, homoserine, diaminobutylic acid are reported from Polygonati Rhizoma. However quality control method of Polygonati Rhizoma is not reported so far. Therefore, we studied quality control method of Polygonati Rhizoma. Azetidine-2-carboxylic acid (1) was selected as the analytical marker since it is the major and specific compound of Polygonati Rhizoma. Powdered Polygonati Rhizoma was extracted and (1) was derivatized with trimethylsilylating reagent to increase volatility. Trimethylsilyl derivatized (1) was analyzed with capillary gas chromatography using methylsilicon fluid stationary phase. The content of (1) in 49 samples collected over the country was 0.67±0.34 %.

[PD4-16] [10/19/2000 (Thr) 15:00 - 16:00 / [Hall B]]

Metabolites of Aloesin

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Aloe is a perennial evergreen herbaceous plant or tropical woody plant and more than 360 species are known to the whole world. Its leaves have long been used in folk medicine for the treatment of burns and dermatitis. Reported activities of aloe include antibacterial, antigastric ulcer, antidiabetic, antitumor, tyrosinase inhibition, and anti-inflammatory activities. Several anthrones, anthraquinones, anthronoles, chromones, pyrones, polysaccharries, and their C-glycosyl compounds have been isolated from various Aloe species. Aloesin, the most abundant C-glycosylated chromone in A. barbadensis, is reported to have whitening and kidney-protection activities. In this study, we isolated and identified metabolites of aloesin. Aloesin was administered to rats through oral administration or i.v. injection, and its metabolites in blood, urine and bile were studied. Two metabolites were isolated. Their structure were elucidated to be 8-C-glucosyl-7-hydroxy-2-(2-hydroxypropyl)-5-methylchromone (1) and 7-hydroxy-2-(2-hydroxypropyl)-5-