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Development of solid phase assays for quantitative, semi-quantitative and qualitative determination of urinary glucose

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Determination of urinary glucose is important in management of diabetes mellitus and fanconi syndrome. Commercial test strips available for this have used O- toludine as the colour development reagent. With restrictions on the use of O- toludine due to its carcinogenic properties, new regents are being developed for glucoses assays. Use of 4 - methoxy - 1 - napthol and 5', 5 - dimethyl cyclohexane dione has been demonstrated in our laboratory. In this paper, we describe the use of this assay for quantitative, semi-quantitative and qualitative determination of urinary glucose. The dry

reagent element proposed for quantitative and semi-quantitative determination of urinary glucose was prepared by immobilisation of glucose oxidase, peroxidase, 4-methoxy-1-naphthol, 5,5-dimethyl cycolohexanedione in gelatin with phosphate buffer pH 6, followed by uniform coating using the Meyer rod technique on Whatmann filter paper. The analytical elements undergo a double sequential enzymatic reaction which results in a blue colored oxidised dye conjugate. Dye can be quantified by recording transmittance intensity in solid phase by optical densitometry. The proposed dry reagent strip can be used in quantitative determination of urinary glucose by comparing transmittance intensity developed after 2 minutes from the time of dipping the enzyme coated area in urine at room temperature. No interference was observed with ascorbic acid, ketones, urobilinogen, creatinine, fructose and galactose. Semi-quantitative measurements can also be done with enzyme coated paper strips by comparing the blue colour developed after 1 minute with the rating scale we have developed. Dry reagent element proposed for qualitative determination of urinary glucose was also prepared by immobilisation colour developing reagents on processed coconut kernel. The test detects the presence of glucose with a sensitivity of 99.12 % and a specificity of 100% at a detection limit of 2 mg/dl. Both types of test strips can be stored in closed bottles with a desiccant with or without refrigeration for more than 4 months with no detectable loss of sensitivity.

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