DPP4/CD26 RESEARCH ASSAY FOR HUMAN SERUM AND PLASMA SAMPLES

Summary:

Sample: 2 mL clotted blood is preferred.

Alternatives: (i) edta blood. (ii) 0.1 mL minimum of serum or plasma. Temperature: Cooler than ambient.

Cost: No fee during 2013; conditions apply.

Contacts:

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Cost and Conditions: No fee during 2013 for fewer than 16 samples in a week. For greater numbers please contact us. If data is included in a publication, acknowledgement of MDG is required and sample provider is responsible for human ethics approval and informed consent. Please cite ref 1 for assay method.

Timing: Six business days, usually less.

Background

Dipeptidyl peptidase-4 (DPP4), also known as adenosine deaminase complexing protein 2 or CD26 (cluster of differentiation 26) is a protein that is encoded by the DPP4 gene. The substrates of this enzyme include growth factors, chemokines, neuropeptides, and vasoactive peptides [2]. DPP4 has a major role in glucose metabolism. It is responsible for the degradation of incretins such as GLP-1 and hence its inhibition by drugs such as sitagliptin, saxagliptin and linagliptin has been used for treatment of diabetes mellitus type 2 [2,3]. DPP4 levels in type 2 diabetic patients, however, are not currently monitored as a routine in the clinical practice. DPP4 activity has been reported to be higher in serum of patients with Hepatitis C [2-6], as well as in nonalcoholic steatohepatitis (NASH) [7], a severe form of Non-Alcoholic Fatty Liver Disease (NAFLD), and in heart failure patients [8].

Enzyme assay method:

Serum and plasma levels of DPP4 are measured using an in-house enzyme activity assay. The serum or plasma may be stored for several days refrigerated, or for any length of time frozen. Freeze/thaw ten times has been shown to have no effect on the assay data. The enzyme needs to remain below 40°C.

Clear 96-well plates (Falcon, Becton Dickinson, USA) containing duplicate chromogenic standards in a linear range of 0-60 nmol of p-nitroaniline (pNA) are used to interpolate DPP4 activity in all serum/plasma samples. Optical density is measured at 405 nm after incubating 10 µl of serum/plasma with the chromogenic substrate pNA at a final concentration of 1 mM. Negative control/blank wells contain Tris-EDTA buffer plus substrate only. Three in-house controls are included in each assay and are used to normalize the final data. Assay plates are read in a FluoStar plate reader (BMG Labtech, Germany) every 2 min for 30 min at 37°C. All samples are analysed within the linear range of the standard curve and are determined in triplicate.
References


22 October 2013.