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Uplc-Ms/Ms Method for The Simultaneous Determination of Plasma Catecholamines And Their Metabolites

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Introduction

Phaeochromocytomas and paragangliomas (PPGLs) are rare tumors of adrenal chromaffin cells or extra-adrenal paraganglia. they're difficult to diagnose due to the non-specific clinical symptoms like hypertension, palpitations, flushing and sweating. Inappropriate catecholamines (CAs) production was normally used for the diagnosis of PPGLs within the clinical laboratory. Traditionally, 24-hour urine was used for analysis to permit for intermittent secretion of CAs by PPGLs. The guideline recommends screening of fractionated urinary metanephrines (MNs, the metabolites of CAs) or plasma free MNs using high performance liquid chromatography with electrochemical detection (HPLC-ECD) or liquid chromatography including tandem mass spectrometry (LC-MS/MS) as MNs are continuously secreted by the tumor. CAs and 3-methoxytyramine (3-MT) weren't determined in most cases, due to their high polarity and instability in plasma for LC-MS/MS analysis. The analysis time must be elongated if eluting CAs [including epinephrine (E), norepinephrine (NE), dopamine (DA)] and MNs, [including metanephrine (MN), normetanephrine (NMN), 3-MT] in one run, which to an outsized extent, will restrict the sample analysis throughput during a clinical laboratory. The opposite hand, CAs and 3-MT useful for the diagnosis of some special PPGLs that only secrete DA but not E and NE.

Here, we report a rapid UPLC-MS/MS method with only 3-min for one run and with high sensitivity to simultaneously quantify MNs and CAs in human plasma. We further validated the diagnostic efficiency of the developed UPLC-MS/MS method using 415 patient plasma samples (PPGLs: non-PPGLs = 7:408). This UPLC-MS/MS method will function a crucial tool to avoid the danger for missing patients with PPGLs.Certified reference materials of E, NE, DA, MN, NMN and 3-MT were purchased from Sigma-Aldrich. Isotope-labeled internal standards (IS): E-d3, NE-d6, DA-d4, MN-d3, NMN-d3, and 3-MT-d4 were purchased from Cambridge Isotope Laboratories. L-Ascorbic acid was purchased from Sigma-Aldrich. The rapid and sensitive ultra-performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS) method for catecholamines and their metabolites, and to know the efficiency for the diagnosis of phaeochromocytomas and paragangliomas (PPGLs). Samples are pretreated with solid-phase extraction, to 3-min analysis to quantify epinephrine (E), norepinephrine (NE), dopamine (DA), metanephrine (MN), simultaneously. The method was comprehensively verified and its diagnostic efficiency on PPGLs was tested plasma samples.

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