Analysis of dimethyl arginines, nitric oxide synthase inhibitor, in biological samples by HPLC

Tudev Munkhtsetseg [a], Youn-Sun Lee [a], Jae-Myung Yoo [a], Yong-Moon Lee [a], Yeo-Pyo Yun [a], Young-Joo Kim [b], Seikwan Oh [b], Jeong-Euy Park [c], Hwan-Soo Yoo [a]

[a]College of Pharmacy and CBITRC, Chungbuk National University, Cheongju
[b]Department of Neuroscience, College of Medicine, Ewha Womans University, Seoul
[c]Cardiology Division, Samsung Medical Center, Seoul

ABSTRACT

Asymmetric dimethylarginine (ADMA) is an endogenous inhibitor of nitric oxide synthase. ADMA, symmetric dimethylarginine (SDMA), homoarginine and arginine was resolved in human plasma by high-performance liquid chromatography (HPLC) with an isocratic elution. Arginine metabolites in plasma following the extraction with a strong cation-exchange column, are derivatized with naphthalene-2, 3-dicarboxaldehyde (NDA) and separated simultaneously on a reversed-phase phenyl column. NDA fluorescence derivatives are stable for a week at 4℃ without a decrease in fluorescence intensity. Chromatograms showed a complete separation of ADMA and SDMA, and are linear within the range of 0.01-20.0 μM. *N*-methylarginine (NMMA) was used as an internal standard for simultaneous quantification of arginine metabolites. Coefficients of variation are less than 2.5% for within a day and less than 4.7% for day to day. Recoveries from exogenously added samples are between 90-105%. Thus, this method becomes reliable and easily reproducible for biological samples such as human plasma from normal volunteers and pregnancy-induced hypertention.