

Original Paper

Inhibitory effect of asymmetric dimethylarginine and NG-Monomethyl-L-arginine methyl ester on nitric oxide synthase activity

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Abstract

Background and Objective: Nitric oxide synthase (NOS) play a role in nitric oxide (NO) generation. Despite the beneficial effects of NO on different body systems its overproduction of produce reactive nitrogen species (RNS) and nitrosilation of proteins. This study was done to evaluate the effect of asymmetric dimethylarginine (ADMA) and NG-Monomethyl-L-arginine methyl ester (L-NMMA) on inhibition of nitric oxide synthase activity.

Materials and Methods: In this laboratory study, Nitric oxide synthase was extracted from 500 grams of sheep kidney by homogenization, ammonium sulphate precipitation and column chromatography on DEAE-32 Cellulose and 2', 5'-ADP-agarose. During purification, protein content was measured according to the Bradford and enzyme activity was assayed using the Griess reactions the inhibitory effects of 25 μ M concentrations of ADMA and L-NMMA on purified enzyme were determined.

Results: Specific activity and yield of NOS were 0.6 units/mg protein and 0.9%, respectively. Molecular weight of purified enzyme was 54 KD with SDS-PAGE. ADMA and L-NMMA in 25 μ M concentrations reduced enzyme activity by 76 and 61.2%, respectively. Km values for NOS in absence and in presence of ADMA and L-NMMA were 5.32 μ M, 31.25 μ M ($P < 0.05$) and 14.29 μ M ($P < 0.05$), respectively. Vmax for NOS in absence and presence of inhibitors was not changed.

Conclusion: ADMA and L-NMMA have competitive inhibitory effect on NOS activity and ADMA have higher inhibitory effect than L-NMMA.

Keywords: Nitric oxide synthase, Asymmetric dimethyl Arginine, NG-monomethyl-L-arginine methyl ester, Kidney

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