عنوان مقاله:

Determination of ۸-OHGua by LC-MS/MS after Acid Hydrolysis of Oxidative Damaged DNA

محل انتشار:

مجله تحقیقات شیمی تجزیه و تجزیه زیستی, دوره 10, شماره 4 (سال: 1402)

تعداد صفحات اصل مقاله: 7

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خلاصه مقاله:

A variety of influences can cause DNA damage to the genome. The hydroxyl radical attacks the C-۸ atom of guanine, forming ۸-hydroxyguanine (۸-OHGua) or ۸-hydroxy-۲ ' -deoxyguanosine (۸-OH-۲'dG) which are an important indicator of oxidative damage on DNA. Determining these damaged base products can be accomplished through measurement by LC-MS/MS after enzymatic hydrolysis or measurement by GC-MS/MS after chemical hydrolysis. In this study, it was aimed to hydrolyze DNA using various strong acids and to measure ۸-hydroxyguanine by LC-MS/MS. In the first stage of the study, the nucleoside ۸-hydroxy-۲'-deoxyguanosine was treated with HCl (۲ M and ۶ M), TCA (۱۰%), TFA (۱۰%), o-phosphoric acid (۲ M), methanesulfonic acid (۲ M), and formic acid (۸۰%). The amounts of ۸-hydroxyguanine were determined by LC-MS/MS. It has been identified that formic acid with the highest yield (۷۰%) hydrolyzes the β-glycosidic linkage between the base and the sugar. Subsequently, oxidative damage was induced on calf thymus DNA by producing hydroxyl radicals via Fenton reaction. The resulting oxidative damaged DNA, was hydrolyzed using formic acid. The amount of ۸-hydroxyguanine was then determined using by LC-MS/MS. Based on the results obtained, it was observed that the acidic hydrolysis applied was effective in breaking the N-glycosidic bond, but not effective in breaking the phosphodiester bond of oxidatively damaged DNA.

کلمات کلیدی:

OHG, ۸-OH-۲'dG, LC-MS/MS, Acidic hydrolysis, Thymus DNA-۸