

Deteksi DNA adduct 8-OHDG secara in vitro dan in vivo dalam urin pasien kanker paru = Detection of DNA adduct 8-OHDG in vitro and in vivo in urine of lung cancer patients.

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Abstrak

Deteksi DNA adduct dapat dijadikan sebagai pendekatan untuk mendeteksi dini risiko kanker. Salah satu produk kerusakan oksidatif DNA adalah 8-hidroksi-2'-deoksiganosin (8-OHdG). Penelitian ini dilakukan untuk mendeteksi 8-OHdG secara in vitro dan in vivo. Studi in vitro dilakukan dengan inkubasi 2'-deoksiganosin dengan H₂O₂ dan akrilamida pada variasi pH 7,4 dan 8,4 selama 24 jam dalam suhu 37 oC. Kemudian hasil inkubasi dianalisis menggunakan HPLC. Sedangkan secara in vivo dilakukan deteksi 8-OHdG dalam urin pasien kanker paru stadium III-IV, urin perokok, dan urin non perokok dengan menggunakan LCMS/MS. Pada validasi instrumen HPLC diperoleh nilai regresi linier 0,9985, nilai LOD dan LOQ sebesar 6,108 ppb dan 20,361 ppb. Sedangkan untuk LCMS/MS diperoleh nilai regresi linier sebesar 1, nilai LOD dan LOQ sebesar 1,819 ppb and 6,066 ppb. Hasil penelitian menunjukkan bahwa paparan H₂O₂ dan akrilamida dapat membentuk 8-OHdG. Konsentrasi 8-OHdG yang terbentuk dari inkubasi 2-deoksiganosin dan H₂O₂ serta 2-deoksiganosin, H₂O₂, dan akrilamida maksimum pada pH 8,4 yakni sebesar 2,151 ppm dan 2,617 ppm. 8-Hidroksi-2'-Deoksiganosin terdeteksi dalam urin pasien kanker paru, perokok, dan non perokok masing-masing sebesar 4,668 – 19,919 ppb, 6,873 – 12,111 ppb, - 0,502 – 6,578 ppb. Nilai rata-rata konsentrasi 8-OHdG dalam sampel urin pasien kanker paru, perokok dan non perokok masing-masing sebesar 9,710 ppb, 10,226 ppb, dan 3,080 ppb.

.....DNA adduct detection could be an approach to early detection of risk cancer. One of oxidative DNA damage products is 8-hydroxy-2'-deoxyguanosine (8-OHdG). This study was conducted to detect DNA adduct 8-OHdG in vitro and in vivo. In vitro study was started to incubate 2'-deoxyguanosine added by H₂O₂ and acrylamide in various pH for 24 hours at 37 oC. Then the result was analyzed with HPLC. In vivo study was conducted detection 8-OHdG in urine of lung cancer patients with stage III-IV disease, smokers and non smokers using LCMS/MS. Instrument validation (HPLC) was yielded linear regression value 0,9985, LOD and LOQ as much as 6,108 ppb and 20,361 ppb as well as validation instrument of LCMS/MS was yielded linier regression value 1, LOD and LOQ as much as 1,819 ppb and 6,066 ppb. The results of study found exposure of H₂O₂ to 2-deoxyguanosine induced 8-OHdG formation and the addition of acrylamide increased 8-OHdG formation. The highest 8-OHdG level obtained by incubation of 2'-deoxyguanosine and H₂O₂ then 2'deoxyguanosine, H₂O₂ and acrylamide at pH 8,4 as much as 2,151 ppm and 2,617 ppm. 8-Hydroxy-2'-Deoxyguanosine was detected in urine of lung cancer patients, smokers and non smokers respectively 4,668 – 19,919 ppb, 6,873 – 12,111 ppb, -0,502 – 6,578 ppb. The mean value of 8-hydroxy-2'-deoxyguanosine in urine of lung cancer patients, smokers and non smokers as much as 9,710 ppb, 10,226 ppb, and 3,080 ppb.