ABSTRACT

Glutathione *S*-transferases (GSTs) are among the enzymes involved in the phase II detoxification metabolism of wide range exogenous and endogenous compounds in living cells. Bivalves GSTs often proposed as biomarker for marine pollution detection for several reasons; filter feeder, sessile, wide range distribution, and not affected by some biotic factors. In this study, GSTs from remis *Donax* sp. was purified by using two affinity column; GSTrap^{TM}HP and GSH-agarose (C_3). The total recovery of CDNB-active GSTs was 12% and 3% for GSTrap^{TM}HP and GSH-agarose (C_3), respectively. SDS-PAGE of GSTrap^{TM}HP purified extract revealed two subunits with apparent molecular masses (MW) of 29 and 26 kDa while GSH-agarose (C_3) showed three subunits corresponding to 29, 28, and 26 kDa. Two-dimensional electrophoresis (2-DE) of GSTs purified from GSH-agarose (C_3) discovered nine similar spots to GSTs purified using GSTrap^{TM}HP but with additional six distinct spots. Analysis by isoelectric focusing (IEF) illustrated most GSTs purified from both column resolved at *p*I in between 4.5 to 6.9. Apart from this cluster, there were also GSTs appearing each at *p*I 4.2 and 8.3. Purified GSTs from both columns exhibited activity towards 1-chloro-2,4-dinitrobenzene (CDNB), 1,2-dichloro-4-nitrobenzene (DCNB), sulfobromophthalein (BSP) and ethacrynic acid (EA). GSH-agarose (C_3) showed less specific activity in all substrates compared to GSTrap^{TM}HP, except for EA which count about 10- fold. However, GSTs eluted from both columns did not show any activity with *p*-nitrobenzylchloride (NBC), trans-4-phenyl-3-butene-2-one (PBO), and nitrocinnamaldehyde (NCA). However, mass spectrometry analysis did not show any match with the available database. Therefore based on the current data, GSTs obtained in this study were summarized belong to pi- and mu-class.