INTERACTION OF CRATABL, A MULTIFUNCTIONAL PROTEIN, WITH BIOMOLECULES SECRETED BY THE PROSTATE CANCER CELL LINES (DU145, PC3)

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INTRODUCTION. CrataBL, a multifunctional protein, induces prostate cancer cell line death by apoptosis mechanism resulting in the release of mitochondrial cytochrome *c* and activation of caspase-3. **OBJECTIVES**: Characterization of biomolecules detected by secretome of prostate cancer cell lines treated and no treated with CrataBL. **MATERIALS AND METHODS**: Secretomes of the prostate cancer cell line DU145 and PC3 were incubated with p-nitrophenyl phosphate, H-D-Pro-Phe-Arg-AMC, Ac-Asp-Glu-Val-Asp-AMC in the presence or absence of CrataBL and/or heparin for screening the hydrolytic activities. The inhibitory activity of CrataBL on tissue kallikrein 3 (KLK3) was also assayed using as substrate Abz-KLYSSKQ-EDDnp. Cytokines APF, CA125, CEA, FGF2, HGF, IL-6, IL-8, sFas, sFas-L, TGF-alpha, Tpsa, VEGF were quantified. CrataBL conformacional modifications in the presence of heparin was monitored by circular dichroism. **DISCUSSION AND RESULTS**: Cell secretomes did not hydrolyze p-nitrophenyl phosphate and Asp-Glu-Val-Asp-AMC, but released AMC from H-D-Pro-Phe-Arg-AMC. Heparin significantly increased this peptidase activity and CrataBL inhibited this hydrolytic activity in a dose-dependent manner, either in presence or absence of heparin. KLK3 was similarly inhibited. CrataBl neutralized the potentiation of the hydrolytic activity promote by heparin. The association of CrataBL- heparin was confirmed by the conformational modification of CrataBL in presence of this glycosaminoglycan. Vascular endothelial growth factor (VEGF) and Transforming growth factor alpha (TGF –alpha) concentration increased in DU145 but not in PC3 secretome while in tPSA (total prostate specific antigen) was significantly reduced compared to that of control and tPC3. In contrast, decrease of APF (alpha-fetoprotein), CA125 (cancer antigen 125) and FGF2 (Fibroblast growth factor 2) in PC3 but not in DU145 treated cells were observed. **CONCLUSIONS**: CrataBL treatment of DU145 and PC3 cells altered the release of their cytokines, and these effects suggest that CrataBL could be useful for biomarker identification.

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