HYDRODYNAMICS ANALYSIS OF A SELF-ASSOCIATED BOWMAN-BIRK PROTEASE INHIBITOR BTCI

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The Bowman-Birk Inhibitors (BBIs) have been characterized as potential anticarcinogenic and chemopreventive agents. The Bowman-Birk protein purified from *Vigna unguiculata* seeds, named black eyed-pea trypsin/chymotrypsin inhibitor (BTCI), presents the same therapeutic properties due its ability to alter significantly the proliferation and viability of breast cancer cells. Hence, as an attempt to optimize the physicochemical BTCI conditions to specific further cancer studies, we investigated its self-association as a function of concentration and temperature. BTCI was purified from *V. unguiculata* seeds using TCA 2.5% and (NiH\(_4\))\(_2\)SO\(_4\) 50% precipitation and ion exchange chromatography through DEAE-cellulose resin. The superficial charge and dimension of the self assembled BTCI were determined as a hydrodynamic diameter and zeta potential on the dynamic light scattering zeta sizer detector (Malvern, laser 633 nm). The average hydrodynamic diameter ranged from 1.12 nm to 6.50 nm at concentration of 6.5-130 \(\mu\)M and temperature of 20-50 \(^{\circ}\)C. Monodisperse monomers of BTCI were found at concentrations of 13 \(\mu\)M and 26 \(\mu\)M at 50 \(^{\circ}\)C and 40 \(^{\circ}\)C conditions. The zeta potentials > - 30.0 mV were obtained showing the physicochemical stability of the oligomers, primarily at 26 mM condition. Based on these results, the best conditions of BTCI as a monomer or self-associated dimer, trimer and hexamer in solution were found at 13 \(\mu\)M and 26 \(\mu\)M. These conditions will be explored in further biotechnological investigation.

**Key-words:** Serine proteases inhibitors; self-association; dynamic light scattering; oligomerization.

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