Expression, Purification, and Characterization of Functional Recombinant Human Daintain/AIF-1 in *Escherichia coli*

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Z. Naturforsch. **65 c**, 726–732 (2010); received March 8/June 29, 2010

Daintain/AIF-1 was identified from injured rat carotid arteries and porcine intestine in the mid 1990s. It is involved in autoimmune disorders, chronic rejection of allografts, gliomas, and breast cancer. Since it is convenient and economical to obtain such a peptide biologically, in this study, we describe the expression, purification, and characterization of recombinant human daintain/AIF-1 (rhdaintain/AIF-1). The backbone of vector pET32a, a high-level expression plasmid, was used to construct the pET32a-daintain/AIF-1 plasmid for daintain/AIF-1 expression in *Escherichia coli*. The recombinant daintain/AIF-1 protein was solubly expressed in the BL21 (DE3) strain and was purified by Ni²⁺ affinity chromatography. After purification, the recombinant protein showed the expected size of 18 kDa on Tricine-SDS-PAGE gels which was further confirmed by Western blotting. A total of 34.0 mg of high purity (over 98%) rhdaintain/AIF-1 was obtained from 1 L culture. The recombinant peptide was able to increase blood glucose elimination rates and enhance the proliferation of human MCF-7 cells. These results suggest that biological activity of the recombinant peptide was preserved after purification.

Key words: Daintain/AIF-1, Blood Glucose Elimination, MCF-7 Proliferation