

Abstract

N-acetylated alpha linked acidic dipeptidase like 2 protein (Naaladase L2) is a transmembrane protein possessing large extracellular as well as intracellular part. The function of this protein has not been determined yet. It was suggested to be associated with a wide range of diseases such as Kawasaki syndrome, an autoimmune disease causing inflammation of vessels, and others. Its homologue, glutamate carboxypeptidase II (GCPII), also called N-acetylated alpha linked acidic dipeptidase (Naaladase), is a transmembrane protein with large extracellular part possessing protease activity. GCPII was suggested as a diagnostic and prognostic marker of prostate cancer.

According to a recent study, Naaladase L2 mRNA levels correlate with Gleason score used for evaluating the prognosis of prostate cancer. Cells overexpressing Naaladase L2 showed higher invasivity, migration and formed more colonies than the negative controls. Additionally, a difference in mRNA expression between benign hyperplasia and tumour was found. Naaladase L2 is thus believed to have a potential in serving as a diagnostic or prognostic marker of prostate cancer.

Only a few reports have been published concerning investigation of Naaladase L2 and none of them focus on exploration of the protein itself. Therefore, we decided to recombinantly prepare and characterise this protein. We expressed the extracellular part of Naaladase L2 in insect cells and optimised its purification using affinity chromatography and gel filtration. We found that similarly to its homologue GCPII, Naaladase L2 undergoes posttranslational glycosylation. On the other hand, in contrast to GCPII, no proteolytic activity of extracellular part of Naaladase L2 has been observed. We also expressed the intracellular part of Naaladase L2 in HEK cells and investigated its potential interaction partners using pull-down experiments and mass spectrometry. The preliminary data suggest that Naaladase L2 might be associated with cytoskeleton organisation and cytoskeletal changes in cell division by interacting with proteins involved in these processes. However, more experiments need to be performed to confirm this hypothesis.

Key words: Naaladase L2, GCPII, homologue, protein-protein interaction