

HUMAN SPECTRIN SRC HOMOLOGY 3 DOMAIN BINDING PROTEIN 1 (HSSH3BP1) IS A POTENTIAL REGULATOR OF MACROPINOCYTOSIS AND A PUTATIVE TUMOR SUPPRESSOR

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Macropinocytosis is an endocytic process that occurs through non-clathrin coated vesicles larger than 0.2 μm in diameter. Macropinocytosis is often upregulated in macrophages and tumor cell lines following stimulation with growth factors and mitogenic agents. Although macropinosomes are readily visualized in cultured cells by the introduction of fluorescent, water-soluble dyes into the culture medium, protein markers associated with this type of vesicles have not yet been well defined. Here, we report that human spectrin SH3 domain binding protein 1, or Hssh3bp1 [1], associates with macropinosomes in NIH 3T3 fibroblasts. Hssh3bp1 macropinosomes are heterogeneous in morphology and size, ranging between 0.5 and 2 μm . Hssh3bp1 macropinosomes do not endocytose transferrin and are resistant to brefeldin A treatment. Cytochalasin D, and wortmannin block endocytosis of fluorescent dyes into the Hssh3bp1 macropinosomes and dramatically affect their morphology. Overexpression of Hssh3bp1-green fluorescent protein (GFP) decreased endocytosis of a fluorescent dye suggesting a potential regulatory role of Hssh3bp1 in macropinocytosis. In fact Hssh3bp1-GFP macropinosomes do not take up fluorescence dyes in contrast to endogenous Hssh3bp1 macropinosomes. Time lapse confocal microscopy experiments indicate that Hssh3bp1-GFP macropinosomes do not move or fuse with each other or with vesicles containing a fluorescent dye. Experiments using nocodazole suggest that these effects are likely to be due to an altered association of Hssh3bp1-GFP macropinosomes with microtubules. In the macropinosomes of NIH 3T3 cells, Hssh3bp1 associates with a 200-kDa protein that crossreacts with a mAb to the erythroid α -spectrin SH3 domain. Thus macropinosomes in cells may contain a spectrin-like protein. We will present data indicating that the spectrin-like protein is ubiquitously expressed in nonerythroid cells [2]. In addition to these data we observed loss of expression of Hssh3bp1 in some primary prostate tumors and found mutations of the gene in cancer cell lines. These suggest the possibility that Hssh3bp1 is tumor suppressor in prostate cancer [3].

REFERENCES

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