

Hsp60 from cancer cells can reach near and distant targets: A proposal for a multistage pathway

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Cancer cells have means to influence other cells in their vicinity and distant, and in this signal-delivering mechanisms the chaperonin Hsp60 plays a role, which is currently being recognized as potentially crucial for the growth and dissemination of at least certain types of tumors. In order to arrive at its destination, Hsp60, a typical resident of mitochondria in normal and tumor cells, leaves the organelle and reaches the blood. In the latter, Hsp60 can travel and arrive at targets situated far away from its origin. The details of the route followed by Hsp60 and their molecular mechanisms have not yet been fully elucidated. We investigated Hsp60 levels and secretion in normal and tumor cell lines. We traced Hsp60 in whole-cell lysates; culture medium; purified exosomes, membranes, and lipid rafts; and samples for electron microscopy (EM); using various complementary techniques, for example electrophoresis, ultracentrifugation, Western blotting, ELISA, enzymatic tests, EM and immuno-EM, and enzymatic digestions and determinations. Exosomes were purified and assessed by EM and by determining presence of ALIX, Hsp90 alpha and beta, and Hsp70, and by measuring ATPase and AChE activities. To obtain information on the secretion routes that might be involved, we used inhibitors of the lipid raft and exosome pathways. We found that the tumor but not the normal cell lines studied secreted Hsp60 actively (cell death and destruction did not play a role in the chaperonin exiting the tumor cells in our experiments). The data also showed that the lipid rafts-exosome pathways were definitely involved. We then established that Hsp60 associated with the cell membrane, lipid rafts, and exosomal membrane. We integrated all the data into a model that could serve as a blueprint for future experiments aiming at filling in the missing information. This working model includes the following stages: Hsp60 in the cytosol, coming either from mitochondria or from synthesis in it without entering the organelle, reaches the cell membrane and binds lipid rafts, and through lipid-raft mediated endocytosis is included in early endosomes and then in multivesicular bodies and, finally, in exosomes for secretion. Extracellular exosomes may then reach: a) inflammatory cells and/or other components of the immune system such as neutrophils and macrophages to elicit inflammation and adaptive-type of immune responses; b) endothelial cells to induce neoangiogenesis; and c) other tumor cells to act as anti-apoptotic factors and promote cancer survival.

Keywords: Chaperonins, Cellular secretion, Exosomes, Lipid rafts, Multivesicular bodies, Cell membrane