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HOW COPPER GETS INTO BACTERIA

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Copper is a toxic element, but is also required in small quantities by all living cells. Therefore, cells have evolved systems to control intracellular copper. *Escherichia coli*, the model organism we used in this work, expels excess copper by a copper pump, CopA, which pumps copper out of the cytoplasm. However, it is still not known how copper enters bacteria. The research of the interaction of bacterial cells and copper using modern methods is the focus of this work.

A strain of *E. coli* without the *copA* gene, and thus unable to export excess copper was used. The strain also contained a biosensor plasmid, pUA615. When copper enters the cells, the biosensor leads to the generation of luminescence. This allows to test under which conditions copper enters cells. According to our sources such researches have never been conducted.

It was determined which chemicals or nutrients affect copper entry into cells. The following compounds were tested: casamino acid (hydrolyzed casein; contains all 20 amino acids), 17 different DL-amino acids, and chloride. Bacteria were grown in liquid media (LB), which is composed of NaCl, yeast extract, peptone. Cells were grown to mid-log phase, centrifuged, and washed with a solution of sucrose. Then, the cells were suspended in 0.8% NaCl, 1% glucose, 0.333 mM CuSO₄, and 0.5% casamino acids. Cells were distributed to wells of 96-well microtiter plates. Different amino acids were added to the wells to test their effect on bioluminescence. After incubation for 1 h at 37 °C, the plates were placed in a BioRad ChemiDoc MP instrument, to record bioluminescence, induced by copper entry into cells.

As a result, it was found that copper enters cells more rapidly in the presence of some amino acids, such as alanine, cystine, methionine, asparagine, serine, and threonine. This suggests that copper is co-transported into cells with these amino acids.

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