Effect of Hypoxia on Tumor Cell Proteins

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Culturing cancer cells in an incubator makes it easy for scientists to study them, but this technique does not exactly mimic the environment that the tumor cells grow in when the tumor is inside a person. Many tumors are often oxygen starved or hypoxic. The goal of my project is to study a human brain cancer cell line that is grown in an incubator containing 21% oxygen (normoxia) and compare this to the same cell line grown in a hypoxia chamber containing 1% oxygen (hypoxia). I hypothesized that hypoxia will cause changes in tumor proteins. For this study I used a glioblastoma cell line that I cultured in normoxic and hypoxic conditions for 96 hours. I analyzed the abundance of 3 membrane receptors (EGF-R, Erb-B2 and IL13R-2a) and three intracellular proteins (PCNA, HSP90 and caveolin-1) using Western Blot analysis. Since hypoxia may also cause proteins to move into or out of specialized membrane regions of the cell called "lipid rafts", I also isolated lipid rafts from normal and hypoxic cells and also compared the protein content in these special membrane regions using Western Blot analysis. I found that hypoxia caused a significant change in the protein composition of total cell lysates and that this change was also found at the subcellular level in lipid rafts. This data may demonstrate the importance of studying tumor cells in a hypoxic environment for the testing of new experimental drugs aimed at killing tumor cells in humans.