Plexin: Putting Together the Pieces

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Abstract

The cause of Proteus Syndrome, characterized by uncontrolled cell division leading to tumor formations, is currently not understood. Recent work suggests that cells from Proteus Syndrome patients express a higher level of a protein called Plexin D1. Plexin D1 has been found in angiogenic vessels during embryogenesis and may play an important role during embryonic development. Exploring the structure and function of Plexin D1 may contribute to a further understanding of Proteus Syndrome.

Mass spectrometry can be used to compare the types and amounts of proteins found in diseased and healthy cells. Using Trypsin, the proteins in a healthy control cell and a diseased cell can be cleaved at basic residue groups along the polypeptide chains, such as arginine and lysine. Performing trypsin digestion in presence of heavy water (H$_2$O) produces peptides with a +4 da mass shift. If $^{18}$O labeled peptides from diseased cells are mixed with unlabeled peptides from control cells, the ratio of labeled : unlabeled protein can indicate a change in protein expression between the two samples.

The molecular weight of peptides from Proteus cells can be found through the process of electrospray ionization mass spectrometry (ESI-MS). This process involves a fine spray of protein fragments that can be analyzed by a computer. The data is then recorded in order to find the molecular weight of the peptides. Comparing the observed masses to a database of known proteins allows researchers to identify the peptide fragments. Plexin D1 may be the key to unlocking the mystery of Proteus Syndrome; the higher frequency of Plexin D1 in Proteus cells versus healthy cells remains promising. By collecting and analyzing data through mass spectrometry the uncertainty surrounding this elusive protein and obscure disease could be dissolved.

Elephant Man

Joseph Carey Merrick (5 August 1862 – 11 April 1890) was an Englishman who became known as "The Elephant Man" because of his physical appearance, which was caused by a disorder, now believed to be Proteus Syndrome. Because of his disorder, he was recognized and sympathized by Britons in the Victorian Era. He died at the young age of 27.

Mass spectrometry

Electrospray Ionization (ESI) Source

During electrospray ionization, Plexin dissolved into a solvent and pumped through a narrow capillary. A high voltage is applied to the tip of the capillary, which disperses the solvent from the tip into a fine spray containing highly charged droplets.

The charged droplets diminish in size by solvent evaporation assisted by a flow of nitrogen gas. These charged ions enter into the source and get separated in the analyzer.

The ion trap uses Helium to cut the Plexin peptides into smaller recognizable pieces. The data regarding mass and amount of these polypeptide chains are sent to the detector, which are read as a mass spectrum on a computer.

Electron multiplier

Detector

Zoom Scan - Mass Spectrum

The first four peaks show increased expression of Plexin in contrast to the next four peaks, a healthy cell's expression.

The data system consists of a computer that organizes the data received from the detector. This data is graphed to show the mass and the amount of charged polypeptides. Often, the amount of a selected protein from a healthy cell is compared to the frequency of a diseased cell.

The disease cells are labeled with $^{18}$O, and when a mixture of protein from diseased and healthy cells is analyzed, the polypeptide fragments from the diseased sample appear with slightly more mass than the healthy. This allows for a comparison between the expression in the diseased cell and the healthy cells.

What does it mean?

It is possible to find the source of disease by finding an over expression of any given protein by comparing the expression of healthy cells to diseased cells.

In order to make a causal relation between Plexin D1 and Proteus Syndrome, future research should explain the molecular interaction occurring in diseased cells.

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