ADVANCED MASS SPECTROMETRY TECHNIQUES FOR DETERMINATION OF BRAIN GANGLIOSIDE EXPRESSION AND THEIR FUNCTIONAL INTERACTIONS IN HEALTHY AND DISEASED CENTRAL NERVOUS SYSTEM

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Abstract
A pivotal role in the brain development is played by the cellular membrane. Since glycolipids (GLs) are the predominant components of plasma membrane, a direct correlation of GLs with crucial processes and neurological disorders exists [1]. Therefore, GLs, formed by a ceramide moiety attached to an oligosaccharide chain, possibly mono- to polysialylated, are important biomarkers in early diagnosis of central nervous system (CNS) pathologies, being in the focus of our research as potential therapeutic targets [2]. Here, we have developed here an approach based on nanoelectrospray (nanoESI) Orbitrap mass spectrometry (MS) in combination with ion fragmentation by collision induced dissociations (CID) for profile comparison and structural characterization of native GL mixtures extracted and purified from histopathologically-defined anencephalic brain remnants originating from fetuses in different intrauterine developmental stages [3]. The native GL extracts dissolved in pure methanol up to a concentration of 5 pmol/µl were infused at 2µL/min flow rate on a LTQ Orbitrap MS. Based on high resolution mass spectrometry capability for a reliable determination of glycopatterns, changes in diversity and number of GLs with age were observed. Over 150 distinct GL structures were identified in the three samples of anencephalic fetal brain remnants. The high resolution of the instrument and the newly developed methodology allowed not just the ionization and detection of low-abundance species, such as polysialylated GLs, but also revealed the presence of different components modified by fucosylation, acetylation and *N*-acetyl galactosamine attachment. Tandem MS (MS/MS) experiments carried out in the LTQ by CID in the manual mode of ion selection and fragmentation using variable collision energy within 25-30 eV confirmed the incidence of potential biomarker species in the investigated anencephalic fetal brain remnants.

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