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A novel strategy for comprehensive profiling and identification of acidic glycosphingolipids using ultra-high-performance liquid chromatography coupled with quadrupole time of flight mass spectrometry

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Acidic glycosphingolipids (AGSLs), which mainly consist of ganglioside and sulfatide moieties, are highly concentrated in the central nervous system. Comprehensive profiling of AGSLs has been historically challenging because of their high complexity and the lack of standards. In this paper, a novel strategy was developed to comprehensively profile AGSLs using ultra-high-performance liquid chromatography-quadrupole time of flight mass spectrometry (UPLC-Q-TOF-MS). Ganglioside isomers with different glycan chains such as GD1a/ GD1b were completely separated on a C18 column for the first time in our knowledge, facilitated by the addition of formic acid in the mobile phase. A mathematical model was established to predict the retention times (RTs) of all theoretically possible AGSLs based on the good logarithmic relationship between the ceramide carbon numbers of the AGSLs in the reference material and their RTs. A dataset was created with 571 theoretically possible AGSLs, including the ceramide carbon numbers, RTs and high-resolution quasi-molecular ions. A novel fast identification strategy was established for global AGSL profiling by comparing the high-resolution quasi-molecular ions and RTs of the tested peaks to the dataset of 571 AGSLs. Using this strategy, 199 AGSL candidates were identified in rat brain tissue. MS/MS fragments were further collected for these 199 candidates to confirm their identity as AGSLs. This novel strategy was employed to profile AGSLs in brain tissue samples from control rats and model rats with bilateral common carotid artery (2-VO) cerebral ischemia. Forty AGSLs were significantly different between the control and model groups, and these differences were further interpreted.

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