Abstract

The treatment of epilepsy in pregnant women represents a major clinical challenge, primarily due to the teratogenic effects of most antiepileptic drugs, which may adversely affect fetal nervous system development. In the absence of therapies that are fully safe during pregnancy, intensive research is being conducted on alternative treatment strategies, including the ketogenic diet (KD), which has demonstrated high efficacy in the management of drug-resistant epilepsy in both children and adults.

This doctoral dissertation employed advanced spectroscopic techniques—Fourier-transform infrared microspectroscopy (FTIRM), Raman microscopy (RM), and synchrotron radiation based X-ray fluorescence microscopy (micro-SR-XRF)—to evaluate biomolecular alterations in the central nervous system of offspring born to females maintained on a KD during pregnancy. Analyses were performed on postnatal days 2, 6, 14, 30, and 60. For each analytical method, optimized sample preparation protocols and measurement conditions were developed to minimize physical artifacts and ensure high data quality.

FTIRM analysis revealed an increased relative abundance of carbonyl-containing compounds in the cerebral cortex and hippocampal cell layers of 14-day-old male offspring, accompanied by a reduction in lipid content and alterations in lipid structure within the white matter. In addition, the area of the internal capsule was significantly reduced compared to controls. In 60-day-old males prenatally exposed to KD, further decreases in lipid-related biochemical parameters were observed, suggesting impairments in lipid metabolism, oligodendrocyte function, or myelination processes.

Additionally, in the hippocampal and cortical regions of this group of animals, inclusions rich in creatine and cholesterol were detected and confirmed by Raman spectroscopy. Quantitative analysis demonstrated a significant increase in the area of cholesterol inclusions in the ketogenic group, while an increasing trend was observed for creatine. This may reflect adverse effects of altered energy metabolism and/or neuroadaptive mechanisms. In contrast to males, female offspring did not exhibit long-term biochemical changes in the brain, which may be related to the protective effects of estrogens regulating creatine metabolism, mitochondrial function, and lipid homeostasis.

Elemental mapping using micro-SR-XRF in 30- and 60-day-old male offspring demonstrated a reduction in the area of brain regions with elevated phosphorus and sulfur levels corresponding to white matter structures, indicating compromised myelin integrity. These findings are consistent with FTIRM results.

In conclusion, the applied spectroscopic approaches enabled a comprehensive biochemical and elemental characterization of the brains of offspring from mothers fed standard and ketogenic diets, revealing a potential age- and sex-dependent impact of maternal KD during pregnancy on central nervous system development.

12.05.26 r., Marzena Rygiel