Differential cerebrospinal fluid signatures revealed by metabolomic profiling of primary progressive and secondary progressive multiple sclerosis patients.

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INTRODUCTION

- Identification of biomarkers for diagnosis and therapeutic monitoring of patients with multiple sclerosis (MS) has proven difficult due to the varied clinical course and complex pathophysiology associated with this disease. Furthermore, disease mechanisms associated with progression in MS, are not defined.
- Here we used metabolomics to identify the chemical fingerprint of specific cellular processes associated with progressive MS.
- Cerebrospinal fluid (CSF) provides a unique interface between the periphery and the central nervous system (CNS) to investigate MS pathogenesis.

OBJECTIVE

The purpose of this study was to determine and compare the global metabolic profiles in human CSF associated with disease progression in MS and allow stratification of the two progressive disease subtypes.

DESIGN & METHODS

- Cerebrospinal fluid (CSF) obtained from controls with no disease (n=15), patients with clinically diagnosed primary progressive multiple sclerosis (PPMS; n=15), and patients with secondary progressive multiple sclerosis (SPMS; n=15) were analyzed on the GC/MS and LC/MS/MS platforms conducted by Metabolon (Durham, NC).
- All samples and clinical information were obtained with IRB approval and informed consent.
- An equivalent volume of CSF for each sample was extracted and loaded onto the platform with no additional data normalization performed prior to statistical analysis..

RESULTS

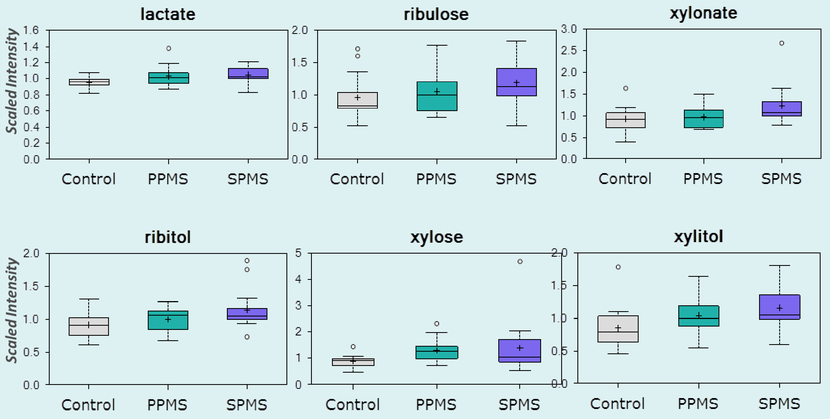
- A total of 198 compounds of known identity were included in the analysis.
- 26 biochemicals were found to be significantly affected in the MS cohort as compared to the controls ($p \leq 0.05$).
- Notably, the analysis also revealed differences between the primary progressive and secondary progressive populations (18 biochemicals) ($p \leq 0.05$).
- Of particular interest are the changes in carbohydrate metabolism, creatine and creatinine metabolism, extracellular matrix (ECM) remodeling and neuroactive amino acids.

RESULTS

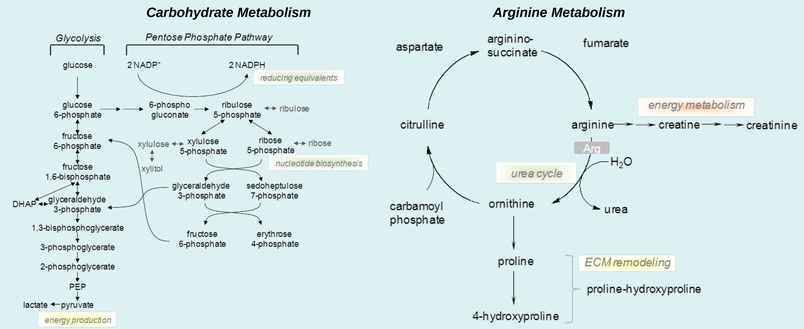
Statistical Comparisons
ANOVA Contrasts

Significantly Altered Biochemicals	PPMS Control	SPMS Control	All MS Control	SPMS PPMS
Total biochemicals $p \leq 0.05$	26	25	26	18
Biochemicals (↑|↓)	21|5	20|5	19|7	7|11
Total biochemicals $0.05 < p < 0.10$	17	16	13	15
Biochemicals (↑|↓)	9|8	11|5	10|3	12|3

Carbohydrate metabolism and Energy Generation: A consistent *accumulation of metabolites related to carbohydrate metabolism, including glucose utilization through glycolysis and the pentose phosphate pathway (PPP), in CSF from patients with PPMS or SPMS,* was observed. In particular, increased levels of the biochemical indicator of glucose utilization, lactate, as well as elevations in several pentose sugars that may be obtained from the diet or synthesized through the PPP (ribulose, ribitol, xylonate, xylose, xylitol, arabinose, and threitol) were noted in both MS patient groups, with slightly *more pronounced changes in patients with SPMS as compared to PPMS*. Moreover, higher levels of metabolites involved in alternative pathways of glucose utilization, such as sorbitol and fructose, along with increased levels of the sugar alcohol mannitol and its derivative mannose were also observed in the MS groups.

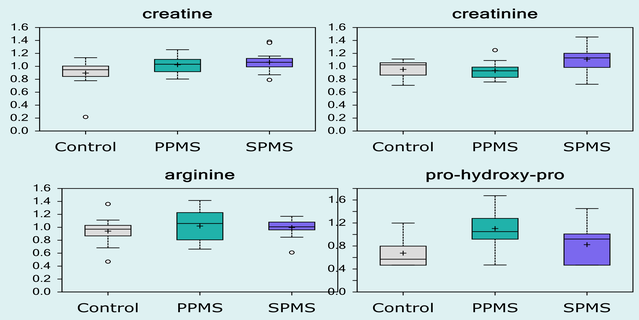


RESULTS



Arginine Metabolism: *Elevations in the energy-related metabolites creatine and creatinine*, which are derived from the amino acid arginine, were observed in CSF from patients with PPMS or SPMS.

Additional changes in biochemical markers of extracellular matrix (ECM) remodeling were also identified in the PPMS group (and to a lesser extent in the SPMS group). This may reflect a differential contribution of ECM remodeling to the PPMS and SPMS disease processes.



CONCLUSIONS

- ✓ Results from this global profiling study revealed perturbations in the CSF metabolome that were both consistent and different when comparing patients with PPMS or SPMS.
- ✓ Changes in carbohydrate metabolism, arginine metabolism, ECM remodeling, neuroactive amino acids, and additional "isolated" metabolites provide insight into the pathogenesis of these two subtypes of MS.
- ✓ Although validation of results from the current study in an independent cohort is necessary, consideration of differential CSF signatures may assist with diagnosis of patients with PPMS or SPMS as well as elucidate novel disease mechanisms.

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