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Proteome and phosphoproteome analysis of stratified meningiomas

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Introduction:

Meningiomas are the most common primary tumours of the central nervous system in adults and despite the majority of them displaying benign features they can cause mild to severe morbidity. The current main therapeutic approach is complete resection commonly with adjunct radiotherapy. No pharmaceutical treatment is available and surgical resection is not always possible, so new therapeutic options are needed.

In the context of molecular targeted therapy, we aim to identify novel targets/biomarkers by deciphering both the proteome and phosphoproteome in stratified meningiomas.

Material and Methods:

Meningiomas were stratified based on their histological grade defined by the World Health Organization (WHO). A total of 5 grade I, 5 grade II and 4 grade III meningioma specimens were analysed and compared to 3 meninges used as normal control.

Equal amounts of total proteins were loaded on a gradient SDS-PAGE and evenly separated bands were fractionated and tryptic digested. Extracted peptides were purified and analysed by liquid chromatography–tandem mass spectrometry (LC-MS/MS). Phosphoprotein purification was performed using Qiagen® PhosphoProtein Purification Kit. Phosphoproteins were processed as before. Raw mass spectrometry files were analysed using MaxQuant™ and data were processed by using DAVID 6.7 and the Ingenuity® Pathway Analysis (IPA) software.

Results:

We have identified approximately 3000 proteins across the 14 samples with over 2000 quantified; similar numbers were obtained for phosphoproteins. Over 600 candidates were commonly identified across the grades for the total proteome, and over 800 for the phosphoproteome, which were not quantified in normal meningeal tissue. Additional proteins and phosphoproteins were found to be grade-specific. Gene ontology (GO) enrichment analysis of upregulated proteins (fold change 1.5; p-value <0.05) revealed several up-regulated biological processes. After uploading the datasets to IPA, comparative analysis of primary meningiomas vs. normal meningeal tissue showed several dysregulated pathways. Among the most affected pathways identified were the mTOR, integrin and ERK/MAPK signalling. Notably, using this tool we identified several novel targets associated with an already available drug, either in clinical practice or trial. These targets will be validated first as well as the efficacy of specific repositioned drugs in an effort of making this study time and cost effective.

Conclusions:

Proteome and phosphoproteome analysis of different grade meningiomas revealed several potential novel targets/biomarkers and dysregulated pathways, demonstrating the effectiveness of the proteomic approach. Our studies increased the overall knowledge of protein expression and regulation in meningiomas and will give the rationale for future drug testing on common or stratified targets.