Involvement of Mutant p53, MAPK and STAT3 in Promotion of Metastatic Brain Tumors

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Disclosures

• None
Introduction

• Brain metastases are a leading cause of morbidity and mortality occurring in 40% of cancer patients with metastatic disease.
• Metastatic brain tumors arise from specific subsets of neoplastic cells originating from primary tumors which disseminate to the cerebral gray/white matter junction.
• Genetic and epigenetic mechanisms of complex metastatic transformations remained undefined.
• Inactivation of tumor suppressor gene p53 and activation of ERK and (STAT3) are observed in aggressive carcinomas.
• Here, we establish the role of p53, ERK1/2 and STAT3 in metastatic brain tumors to discover suitable therapeutic targets.
Materials and Methods

• Immunohistochemical (IHC) analysis for mutant-p53 (mt-p53), phosphorylated-ERK1/2 (pERK 1/2) and STAT3 expression was performed on IRB-approved metastatic brain tumors.

• A 22k-gene chip was used for gene expression profiling of the same tumors and genes associated with p53, MAPK and STAT3 were extracted.

• Metastatic breast cancer cells cultured in astrocytic media, with MEK (U0126) and STAT3 (STAT3 inhibitor IV, S31-201) inhibitors to further established their role in metastatic process.
Results

Expression of activated MAPK (pERK1/2), p53 and pSTAT3 in metastatic brain tumors.

- Representative photomicrographs of pERK1/2, mutant p53 and pSTAT3 Tyr705 expression in metastatic carcinoma. Nuclear pERK1/2 and pSTAT3 expression is observed surrounding the peripheral area of the tumor, while nuclear mt-p53 was expressed throughout the tumor section.
- Venn diagram representing the overlap in expression of p53mt, pERK Thr202/Tyr204, pSTAT3 Tyr705 in metastatic brain tumor samples. (A) 81% Samples displayed mutant p53 (B) 86% samples expressed pERK1/2 and 63% of tested metastatic tumor samples expressed pSTAT3 in the nucleus with significant overlap in expression.
Results

• Gene expression profiling showed that 17, 44 and 21 genes were altered, which were associated with p53, MAPK and STAT3 respectively.

• The identified genes are involved with cell cycle regulation, neurogenesis, differentiation, cell-cell interaction, migration, maintenance of cellular dynamics, and reprogramming.
Cell proliferation of MD-MBA-231 cells was increased in an astrocytic environment. MAPK inhibitor, U0126 (20uM) suppressed cell proliferation effectively. STAT3 inhibitor STATi VI S31-201 (300uM) was less effective. B. Cell motility of MD-MBA-230 cells was enhanced toward an astrocytic environment and STAT3 inhibitor STATi VI S31-201 (300uM) and MAPK inhibitor, U0126 (20uM) suppressed cell migration significantly.
Summary of Results:

• Results indicate that aberrant p53 with phospho-MAPK and phospho-STAT3 activation was seen in brain metastasis. Intense staining of activated ERK1/2 and STAT3 at the tumor margin indicates their role in an enhanced inflammatory response.

• Genes associated with p53, MAPK and STAT3, that regulate various cellular functions were altered.

• Astrocytic milieu promoted the metastatic potential of tumor cells, and co-culture analysis showed an amicable relation between tumor cells and astrocytes.

• Inhibition of STAT3 and MAPK signaling pathways reduced cell proliferation and halted cell migration of metastatic cancer cells.
Conclusions

These findings suggest that mt-p53 and aberrant MAPK and STAT3 play a crucial role in brain metastasis with cerebral milieu providing a suitable microenvironment for dissemination. Activation of STAT3 promotes metastasis to the brain, and which may serve as a target for therapeutic intervention.