PLEKHA5 regulates tumor growth in metastatic melanoma

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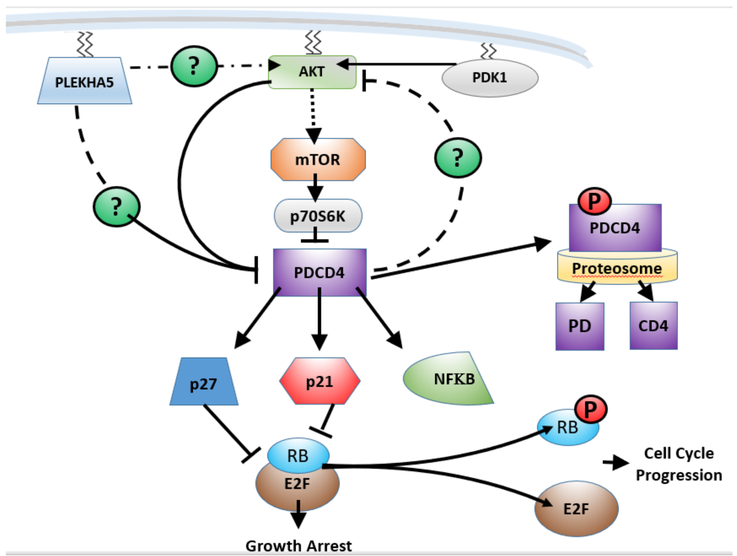
Background

- Brain metastasis (BM) is a common feature of late stage melanoma with significant morbidity and mortality among melanoma patients. While several new systemic therapies are available, very few have been evaluated in untreated brain metastases. Therefore, the identification of new markers as drug targets remains a priority in the treatment of this disease.
- Our earlier studies identified PLEKHA5, a protein involved in normal brain development, as a likely mediator of melanoma brain metastasis. Using both transcript profiling and cell-based BM models, we identified PLEKHA5 as among the most differentially expressed gene in extra-cranial tumors of patients who developed early brain metastases compared to patients who did not. These findings implicate PLEKHA5 as a mediator of melanoma brain homing and a likely drug target.
- However, the precise mechanism of action of PLEKHA5 in this metastatic process is largely opaque and is the primary focus of this study.

Conclusion

- Our findings demonstrate the significance of PLEKHA5 as a likely upstream mediator of cell cycle and the Akt/mTOR signaling pathway driving the proliferation and growth of brain-tropic melanoma.



Results

- Stable PLEKHA5 knockdown negatively regulated cell proliferation in brain tropic melanoma cell lines. Conversely, overexpression of long *PLEKHA5* isoform in non-brain tropic melanoma cells lines which do not express PLEKHA5, A375P and YUSIK, resulted in increased cell proliferation (Fig. 1a).
- This decreased proliferation coincided with decreased transition into S-phase (Fig. 1b)

Fig. 1a

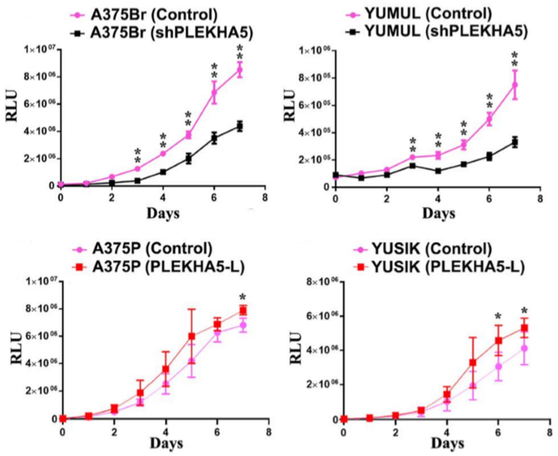
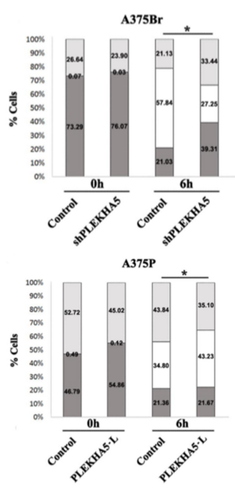
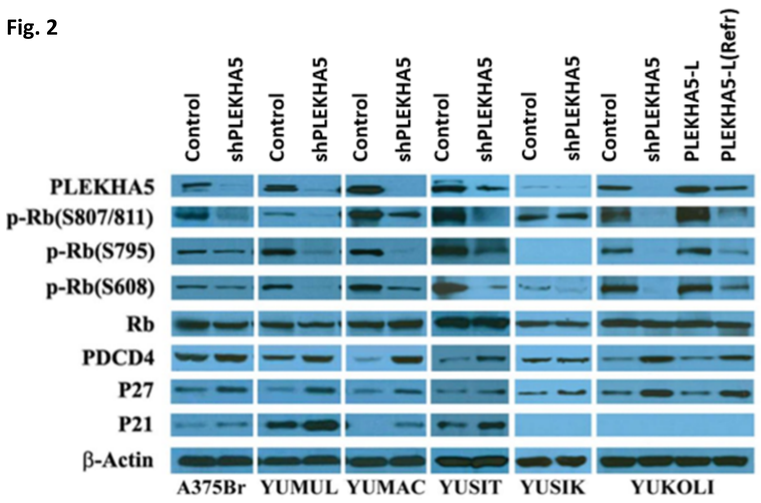


Fig. 1b



- This decreased proliferation and G1-S transition stagnation coincided with up-regulation of PDCD4, p21, and p27, as well as the downregulation of pRb protein. Conversely, the ectopic re-expression of PLEKHA5-L in YUKOLI cells had an inverse effect as shown in Fig. 2.

Fig. 2



- *In vivo*, both subcutaneous (Fig. 3a) and direct intracranial (Fig. 3b) injections with PLEKHA5 knockdown cells resulted in slow tumor growth
- The overall incidence of tumor growth in the brain was similar among cell lines and ranged between 90 - 100% of injected mice.

Fig. 3a

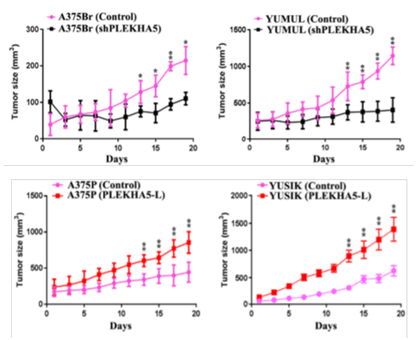
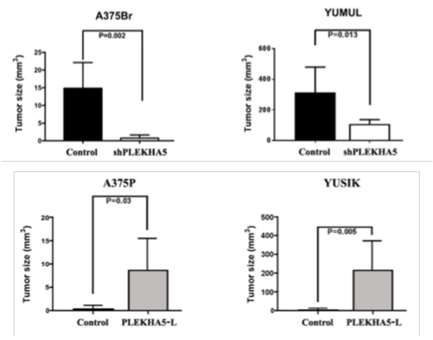
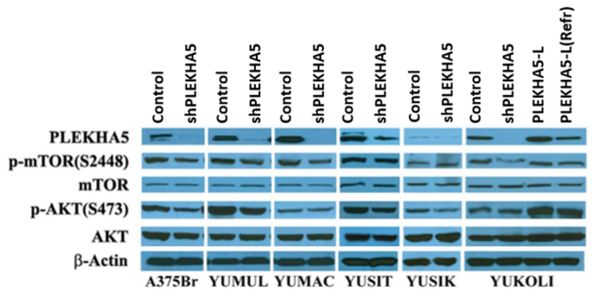


Fig. 3b



- This reduction in tumor growth *in vivo* could be attributed to decreased phosphorylation of Akt (S473) and mTOR (S2448) kinases, key regulators of PDCD4 stability.

Fig. 4



- Upregulation of PLEKHA5 in patient tumors inversely correlated with PDCD4 expression.
- Depending on its localization (nuclear or cytoplasmic), it is a likely indicator of disease progression.
- Low PDCD4 expression in cerebral specimens was associated with poor overall survival

Fig. 5