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The interplay between miR-579-3p and microphthalmia-associated transcription factor (mitf) controls melanoma progression and resistance to targeted therapies

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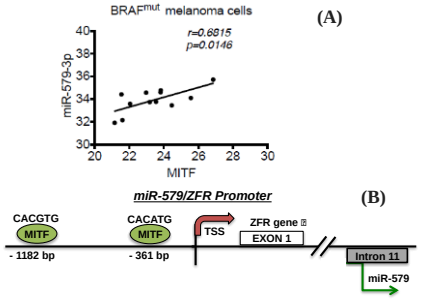
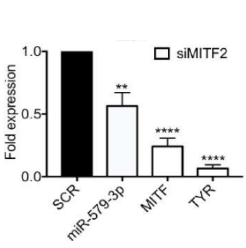
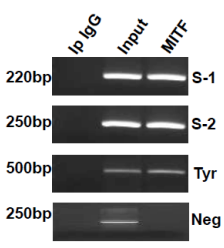
BACKGROUND

Therapy of metastatic melanoma has improved dramatically over the last years thanks to the development of targeted therapies (MAPK inhibitors) and immunotherapies. Drug resistance continues to be a major limitation to the efficacy of these therapies. Our research group has provided robust evidence as to the involvement of a set of microRNAs in the development of non-genetic resistance to target therapy in BRAF-mutated melanoma cell lines. Among them, a pivotal role is played by the newly oncosuppressor miR-579-3p. This miRNA controls the expression of oncoproteins BRAF and MDM2 and is down-regulated in MAPKi-resistant melanoma cells. However, the molecular mechanisms driving miR-579-3p down-regulation during the onset of drug resistance is still open.

I° RESULTS

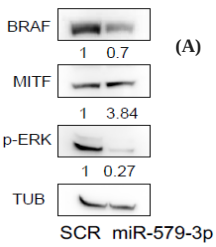


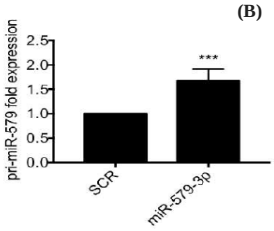




miR-579-3p and Microphthalmia-associated Transcription Factor (MITF) are positively correlated in BRAF-mutant melanoma cell lines **(A)**. MITF has two binding sites within the promoter of miR-579-3p **(B)**. qRT-PCR showed the reduction of miR-579-3p after MITF silencing **(C)**. ChIP results confirmed that MITF was able to bind the two consensus sites within miR-579-3p promoter **(D)**

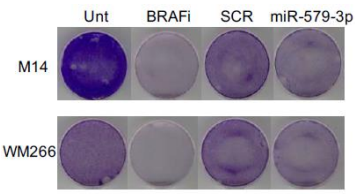
II° RESULTS





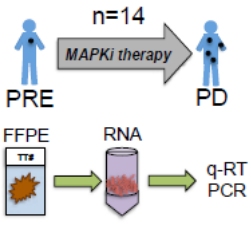
BRAF-MAPK signaling control MITF protein expression levels promoting its degradation. miR-579-3p overexpression stabilized MITF protein **(A)** and induced its own transcription in a positive feedback regulatory loop **(B)**

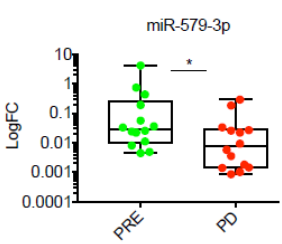
III° RESULTS

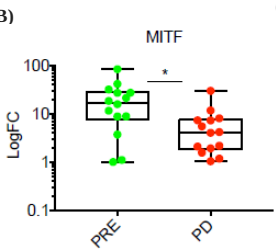


miR-579-3p overexpression inhibited M14 and WM266 BRAF-mutant melanoma cell proliferation.

IV° RESULTS







(C)

Total RNA was extracted from 14 matched formalin-fixed paraffin-embedded (FFPE) melanoma samples before starting targeted therapies (Pre) and after disease progression has occurred (PD) **(A)**. qRT-PCR confirmed the down-regulation of both miR-579-3p **(B)** and MITF **(C)** levels in PD as compared to pre-therapy samples.

CONCLUSIONS

1. miR-579-3p is under the transcriptional control of MITF.
2. miR-579-3p increases MITF protein and induces its own transcription in a positive feedback regulatory loop by targeting BRAF-V600-MAPK signaling in melanoma cells.
3. The interplay between MITF/miR-579-3p induces block of proliferation in BRAF-mutant melanoma cells.
4. The levels of miR-579-3p and MITF are lost in patients' biopsies taken upon disease progression.