Circulating miRNAs can affect the melanoma microenvironment and outcome

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Abstract

Metastatic melanoma is an aggressive and deadly disease, with high capacity for metastasis and resistance to treatment, resulting in high number of patients dying within 5 years of diagnosis. Various tumors, including melanoma, can interact with their microenvironment and modulate it to enable disease progression, presenting immune evasion characteristics and facilitating metastatic behaviour. To delve the impact of the crosstalk between melanoma cells and tumor microenvironment (TME) on patient's outcome, we accessed RNA-Seq data from 164 metastatic melanoma samples from The Cancer Genome Atlas to characterize their TME using the CIBERSORT. Next, samples were separated into 3 groups by unsupervised hierarchical clustering analysis based on their TME profiles (Jaccard bootstrap mean: G1 = 0.56, G2 = 0.75, G3 = 0.84). The TME profile in each group was distinct, with G1 enriched in naïve, memory and plasma B cells and depleted in resting natural killer (NK) cells, G2 enriched in T CD8 cells, monocytes and M1 macrophages and G3 enriched in M0 macrophages and depleted in plasma cells, T CD8 cells, memory activated T CD4 cells, follicular T helper cells, activated NK cells, monocytes, and resting dendritic cells (p = 0.05, Mann-Whitney test - MW). Overall survival of the groups was compared and G2 patients presented a significantly better prognosis than G3 (p = 0.01, log-rank test, Hazard Ratio (HR) = 0.49, CI.95 = 0.28 0.85). To better understand the interplay between tumor and its TME, we investigated putative interactions between differentially expressed miRNA (miR) and target genes (mRNAs) in G3 compared to G2. We selected miR-targets pairs (MTP) that were predicted in at least one of the databases available in the multimiR package and that were highly negatively correlated to each other(r = -0.4 and p = 0.5), ending up with a list of 139 MTP. We use igraph to represent the network of MTPs, including additional information of gene expression levels, impact on survival, and possible origins of the miRNA. We found interactions that suggested inter- and intracellular regulations, with tumor modulating gene expression on microenvironment cells and vice-versa. One interesting example is the MTP mir-149/NLRC5. The mir-149 is upregulated in G3 and does not impact patients' survival. However, downregulation of its target, NLRC5, has a negative impact on patients overall survival (p = 0.0047, log-rank test, HR = 0.46, CI.95 = 0.27 0.8). NLRC5 is a transcription coactivator that regulates the expression of genes belonging to the antigen presentation pathway. We found many of its targets such as HLA-C, TAP1 and B2M also downregulated in G3, with significant impact on overall survival. Moreover, mutations in the antigen processing and presenting pathway in G3 were associated with increased number of neoepitopes - new and potentially immunogenic peptides generated by mutations (p = 0.05, MW). This can help explain why the immune infiltrate in G3 is so poor on effector cells. Our results point to a crosstalk between melanoma and the TME that can impact the cell types present within the microenvironment and the capacity of the tumor to evade immune surveillance, favouring metastasis and a worse patient's outcome. This knowledge can be used in the future

for melanoma and treatment assessment by looking into circulating molecules that can inform on TME constitution.

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