Comparison of miR Expression Patterns in Plasma, Serum, and Cell Line Microvesicles

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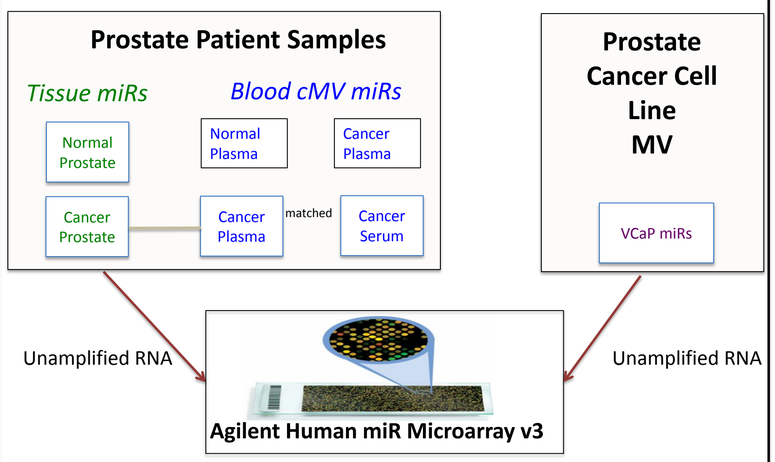
Abstract

Circulating microvesicles (cMVs) isolated from either blood plasma or serum can yield genetic material sufficient for informative microRNA (miR) expression analysis. The ability to purify RNA from MV isolated from several sources was compared, and the miR expression patterns were determined. cMVs were isolated from the blood plasma and serum of men with and without prostate cancer, as well as from healthy and prostate tumor tissue and prostate cancer cell lines. Total RNA was extracted from the plasma and cell line microvesicles (MVs) using the Qiagen miReasy kit and from the serum cMVs using the ExoMir extraction method. The miR expression was determined for each source using Agilent V3 miRNA microarrays. Each sample (300ng) was hybridized to the microarrays, and the acquired data were analyzed with the GeneSpring software package. Expression patterns for miRs from cMVs isolated from the plasma and serum of men with and without prostate cancer were evaluated for significant differences. Hierarchical clustering on both samples and genes demonstrated a distinct expression pattern for miRs from plasma cMVs compared with miRs from cell line MVs and tumor tissue. These findings demonstrate that cMVs isolated from the peripheral blood offer a unique and potentially clinically relevant source of miR for blood-based analysis.

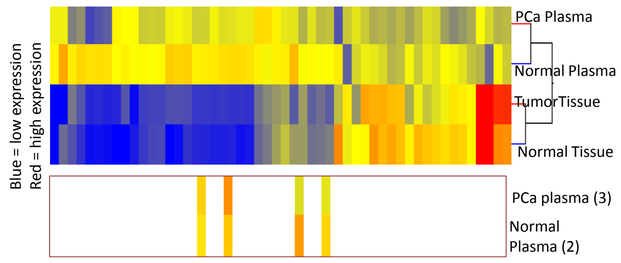
Methods



Objective: compare the miR profile of human blood cMV, and prostate cancer cell line MV and prostate tissue

Results

Comparison of plasma cMVs to tissue



43 miRs in common to plasma and tissue

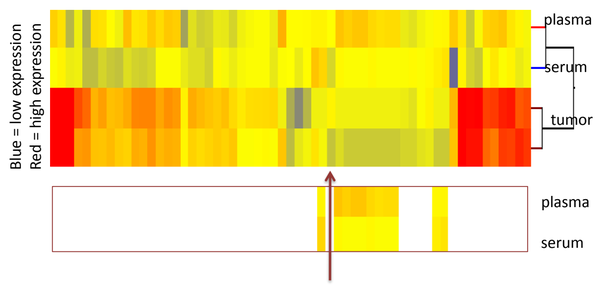
4 miRs unique to plasma

VCaP Cell line and plasma MV expression comparison

51 miRs in common

One miR unique to plasma

MVs from 6 matched prostate cancer serum and plasma samples comparison



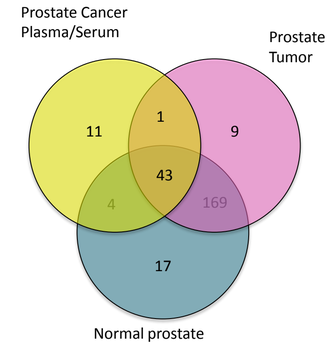
59 miRs expressed

43 in common to tumor tissue

11 miRs unique to prostate cancer serum/plasma cMVs

One miR common to tumor and plasma/serum not found in normal tissue

Many miRs found to be in common between all sample types – one miR unique to cancer plasma/serum and prostate tumor - 4 unique to MVs



Comparing plasma/serum to VCaP

miRs common to microvesicles from plasma, serum and VCaP	Description
ebv-miR-A	Viral miR
hsa-miR-a	122 potential targets
hsa-miR-b	minor miR in let 7 family
hsa-miR-c	7 potential targets including RAP2B

Conclusions

- miRs are present in smaller numbers in cMVs from plasma/serum than in tissue
- We found small, distinct populations of miRs in different sources
- Four miRs were specific to cMVs and cell line microvesicles
- Microarrays offer the advantage of direct hybridization but sacrifice detection of low abundance miRs
- Study of miRs from blood-based cMVs is possible with microarrays but may be better suited to more sensitive methods
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