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PLASMA MICRORNAS IN ASSESSING THE RISK OF RECURRENCE IN TRIPLE NEGATIVE BREAST CANCER

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INTRODUCTION & AIM

Triple-negative breast cancer (TNBC) accounts for 15-20% of all breast cancer cases and is more common in patients under 50, making its study socially significant. TNBC lacks known targets for targeted and hormonal therapy, and chemotherapy remains the leading treatment option. Given tumor heterogeneity and the invasiveness of core biopsy procedures, interest has grown in assessing the effect of neoadjuvant chemotherapy by liquid biopsy. Circulating in blood plasma microRNAs can serve as a more accurate and earlier marker for various conditions than biochemical markers, and their analysis can be implemented clinically. Our retrospective study analyzed the association between pre-treatment plasma circulating microRNA levels in patients with clinical stages II and III TNBC and disease outcome, including recurrence or metastasis.

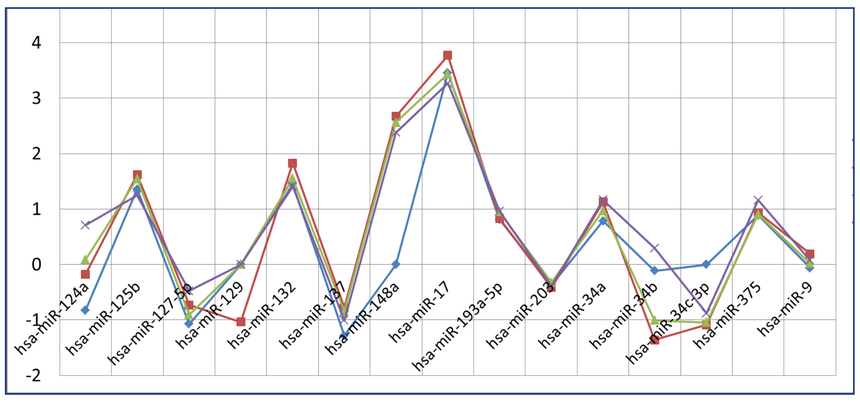
METHOD

The study included plasma samples from patients treated at the N.N. Blokhin National Medical Research Center of Oncology, collected before treatment and frozen at -80°C. Circulating microRNAs were isolated using the miRNeasy Serum/Plasma Advanced Kit (Qiagen, USA), reverse transcription was performed using the TaqMan® MicroRNA Reverse Transcription Kit (ThermoFisher Scientific, USA), and digital PCR was performed using a DropDX-2044HT thermocycler (RainSure, China) with RainSure_3.02.01.4066 TaqMan probe mix (China) and microRNA-specific TaqMan® MicroRNA Assay probes (ThermoFisher Scientific, USA). The results were processed using GeneCount Analysis (RainSure, China) and the Hiplot (ORG) cloud service (https://hiplot.cn/).

RESULTS & DISCUSSION

Fifteen circulating microRNAs were analyzed in a group of 25 patients. The largest changes in concentrations were found for miR-124a, miR-137, miR-148a, and miR-34c-3p (Figure 1). MiR-17 was excluded from the analysis because its concentrations reached several thousand copies per 1 µL of plasma, requiring additional sample dilution. Four patient groups were identified based on the analysis of circulating microRNAs concentrations (Figure 2). Group 1 was characterized by a decreased concentration of miR-124a, miR-137, and miR-148a to 0.0-0.5 copies per 1 µL of plasma, which was associated with cancer progression and lack of response to certain therapeutic agents. Group 1 included patients aged 40-42 years, T3-T4, N1-N3, regional progression was observed in 67% of cases, and brain metastases were observed in 5% of cases. Groups 2 and 3 were characterized by a gradual increase in miR-124a, miR-137, and miR-148a levels to 5-7 and 7-10 copies per 1 µL of plasma. Groups 2 and 3 included patients aged 48-67 years, metastasis was absent in these groups, and patients in group 3 did not show disease progression during 18 months of observation. Patients in Group 4 had miR-124a levels of 15-32 copies per 1 µL of plasma, miR-137 and miR-148a levels of 7-10 copies per 1 µL of plasma, and elevated miR-34c-3p levels (12-17 copies per 1 µL of plasma) compared to other groups. There was no disease progression in patients in Group 4 over an 18-month follow-up period, and all patients achieved a pathologic complete response following the administration of neoadjuvant chemotherapy.

Figure 1. Levels of circulating microRNAs (the Y axis is the Log10 of the concentration) by groups: blue - Group 1, red - Group 2, green - Group 3, and purple - Group 4.



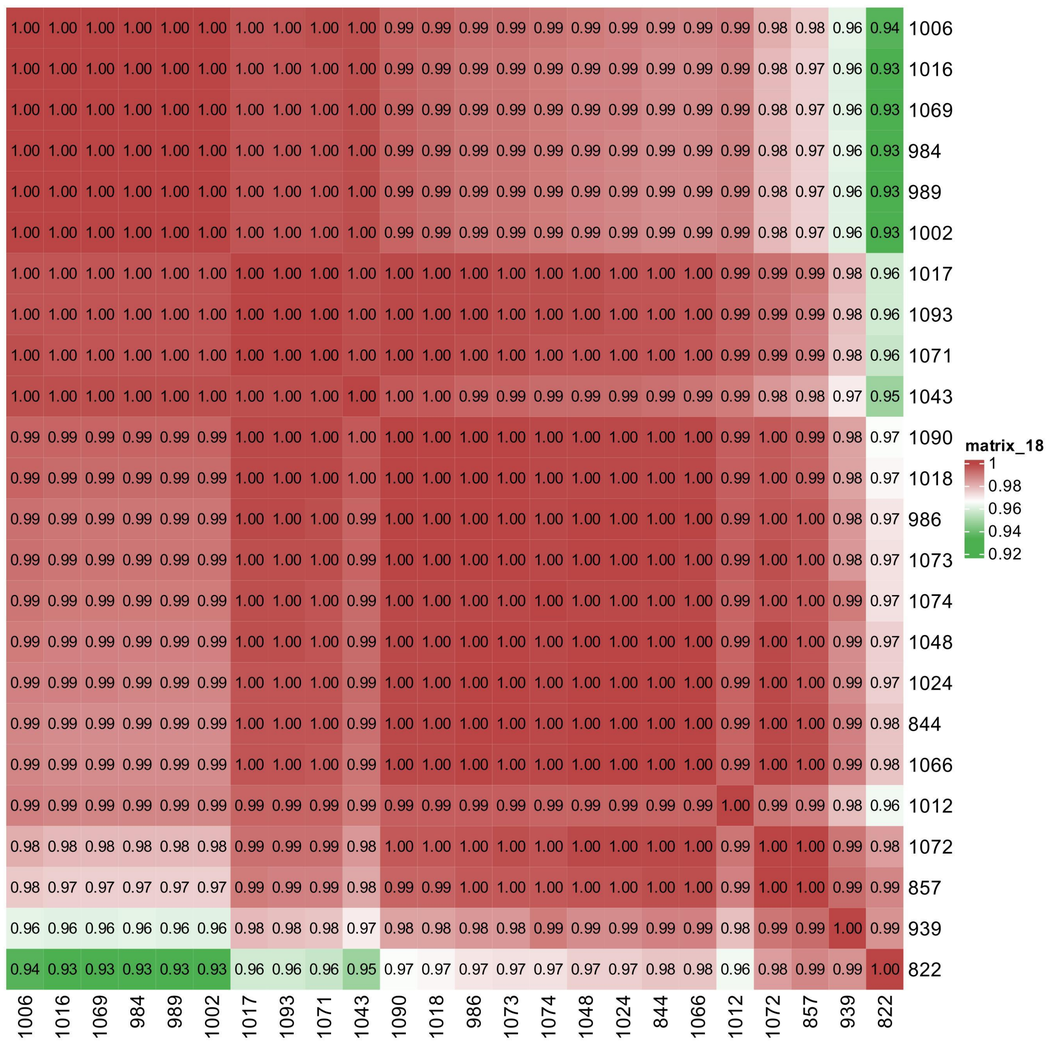


Figure 2. Clustering of patients by circulating plasma microRNA levels. The numbers inside the matrix are the levels of correlation of circulating microRNA concentrations. The numbers outside the matrix are patient identification numbers.

CONCLUSIONS

The high concentration of miR-17 in plasma may be an additional indirect marker of the absence of the estrogen receptor in the tumor, since miR-17 inhibits the expression of the ESR1 gene through its activator RUNX6 inhibition. Analysis of circulating microRNA levels, particularly miR-124a, miR-137, miR-148a, and miR-34c-3p, before treatment can be used to assess the risk of TNBC progression. However, further studies of these microRNA levels in the plasma of healthy volunteers and patients with other oncological diseases are needed.

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