PLOS ONE

RESEARCH ARTICLE

Dysregulation of *miR-122*, *miR-574* and *miR-375* in Egyptian patients with breast cancer

Eman A. Elghoroury[1], Esmat E. Abdelghafar[1], Solaf Kamel[1], Eman Awadallah[1], Aliaa Shalaby[1], Gamila S. M. EL-Saeed[2], Eman Mahmoud[1], Mahmoud M. Kamel[3]*, Asmaa Abobakr[3,4], Rasha Nazih Yousef[1]

1 Department of Clinical & Chemical Pathology, Medical Research and Clinical Studies Institute, National Research Center, Giza, Egypt, **2** Medical Biochemistry, Medical Research and Clinical Studies Institute, National Research Center, Giza, Egypt, **3** Clinical Pathology Department, National Cancer Institute, Cairo University, Cairo, Egypt, **4** Baheya Centre for Early Detection and Treatment of Breast Cancer, Giza, Egypt

* mahmoud.kamel@nci.cu.edu.eg, mm.kamel@yahoo.com

Check for updates

OPEN ACCESS

Citation: Elghoroury EA, Abdelghafar EE, Kamel S, Awadallah E, Shalaby A, EL-Saeed GSM, et al. (2024) Dysregulation of *miR-122*, *miR-574* and *miR-375* in Egyptian patients with breast cancer. PLoS ONE 19(5): e0298536. https://doi.org/10.1371/journal.pone.0298536

Editor: Muhammad Tarek Abdel Ghafar, Tanta University Faculty of Medicine, EGYPT

Received: June 22, 2023

Accepted: January 25, 2024

Published: May 31, 2024

Copyright: © 2024 Elghoroury et al. This is an open access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Data Availability Statement: All relevant data are within the paper and its Supporting Information files.

Funding: This study was supported by the National Research Centre-Egypt, grant number 12060146, Grant Recipient; Eman A. Elghoroury. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript. There was no additional external funding received for this study.

Abstract

Background

The early detection of breast cancer (BC) is receiving global attention, creating an urgent need for more sensitive and comprehensive strategies for preventive intervention, therapy assessment, and prognosis prediction. Aberrant expression of miRNAs has been observed in various malignancies and may be potential targets for therapy. Our study aims to examine the expression profiles of *miR-375*, *miR-574-3p*, and *miR-122* in the sera of Egyptian women with BC, benign breast lesions, and a control group. We hope to determine if these miRNAs can serve as minimally invasive biomarkers for BC.

Methods

This is a case-control study in which 77 patients with newly diagnosed BC, 20 patients with benign breast tumors, and 30 normal healthy subjects as controls were recruited from the outpatient clinic of the National Cancer Institute. The assessment of miRNAs was conducted using RT-PCR (Applied Biosystems).

Results

The expression level of *miRNA-122* was significantly upregulated in the BC group, while the expression levels of *miRNA-574* and *miRNA-375* showed significant downregulation in BC patients. Serum *miR-122* and *miRNA-375* were able to distinguish breast cancer from the benign and control groups in ROC curve analysis, with AUCs of 0.786 and 0.796, respectively. Our results also showed that serum *miR-122* and *miR-574* are significant predictor variables in the multivariate analysis, after adjusting for age.

Competing interests: The authors declare that they have no competing interests.

Abbreviations: BC, Breast cancer; miRNAs, MicroRNAs.

Conclusions

Our findings suggest that *miR-122* may act as an onco-microRNA, while *miR-574* and *miR-375* may have a main tumour suppressor role. The studied miRNAs may serve as minimally invasive biomarkers for cases of breast cancer and as promising potential therapeutic targets for breast cancer.

Background

In Egypt, breast cancer (BC) is the most prevalent form of cancer among women, making up 38.8% of all cases. It is estimated that there were about 22,700 cases of BC in 2020, and this number is expected to rise to about 46,000 by 2050. BC is the second leading cause of cancer-related deaths among women globally [1]. In Egypt, the average age for BC diagnosis is 50.4 years, with 57% of cases occurring in premenopausal or perimenopausal women. In comparison, the median age at diagnosis in Japan is 59.7 years and, in the US, it is 63 years, according to the Japanese Breast Cancer Society registry and US-based SEER data [2].

The age distribution in Egypt may be explained by the population pyramid, with only 8–9% of females over 60 years old. Additionally, 20% of BC patients in Egypt were under 40 years old, compared to 5% in SEER. In developed countries like Korea, the population of premenopausal females has decreased over time, from 59.4% in 2002 to 46.5% in 2015 [2]. Early detection of BC is gaining worldwide attention, so there is an urgent need to find more sensitive and comprehensive strategies for preventive intervention, assessment of therapy, and prediction of prognosis [3, 4].

MicroRNAs (miRNAs) are small non-coding RNAs that regulate gene expression. They can be secreted into extracellular fluids and transported to target cells via vesicles like exosomes or by protein binding. Extracellular miRNAs act as chemical messengers, mediating cell to cell communication [5]. They base pair with 3′untranslated region of the messenger RNA to be suppressed, causing halting, or slowing of the translation process. This step is essential for the targeted proteins production regulation [6]. Due to their multi-level regulatory action, miRNAs have attracted significant interest in the field of cancer research. Depending on the target gene that they regulate, miRNAs can act as ' tumour suppressor miR ' by suppressing oncogenes or 'onco-miR' by targeting tumor suppressor genes [7].

Abnormal miRNA expression has been found in different types of cancer, suggesting that they may be important in the development and progression of tumour. Additionally, miRNAs are easy to isolate, structurally stable, and not affected by sample isolation and processing, making them potential diagnostic and prognostic markers, as well as targets for therapy [8]. miRNAs expression levels are altered in BC due to several mechanisms including transcription factors, epigenetic factors, and protein mutations [9].

MiR-375 is beta cell-specific miRNA located on *Chr2* (*219,001,648–219,001,669*), it serves as a multifunctional regulator in various cellular pathways. Aberrant expressions of *miR-375* are commonly associated with pathological changes [10]. It has been suggested that it may act as a tumour suppressor or oncogene in different types of tumours, but its expression and regulatory role in breast cancer (BC) are still not fully understood [11]. *MiR-574-5p* play an important role in regulating the migration of tumour cells, including those in BC, thyroid cancer, non-small cell cancer, and colorectal cancer [12]. *miR-574-3p* was previously identified as a potential prognostic marker for BC [13]. It has been found to suppress proliferation,

migration, and epithelial mesenchymal transition in triple negative BC cells. Additional research has confirmed that *MiR-574-5p* reduces both tumour size and metastasis in vivo [13].

MiR-122 was initially identified as a tumour suppressor gene in hepatocellular carcinoma. It can suppress the expression of *SRF* and *ADAM10*, which play important roles in the development of various cancers. Over-expression of *miR-122* may promote cell death and halt the cell cycle in cancer cells by reducing the expression of *Bcl-W* and/or *CCNG1*. These findings indicate that *miR-122* may act as a tumour suppressor in cancer [14].

In BC, *miR-122* has been found to act as both a tumour suppressor, targeting the insulin-like growth factor 1 receptor, and as an oncogene, reprogramming glucose metabolism in the tumour microenvironment via exosome [15]. The role of *miR-122* has been considered both as tumor suppressor miRNA and onco miR in breast tumor phenotypes and *miR-122* may also increase the sensitivity of tumor cells to chemotherapy agents [15].

However, there is a lack of sufficient data on the expression of miRNAs and their role in Egyptian women with BC. Therefore, the objective of the present study was to examine the expression profiles of *miR-375*, *miR-574-3p*, and *miR-122* in the sera of Egyptian women with BC, benign breast lesions, and a control group. The aim is to determine if these miRNAs could potentially serve as minimally invasive biomarkers for BC.

Methods

Characteristics of patients

This case control study included 77 consecutive patients with newly diagnosed BC who were recruited from outpatient clinic of National Cancer Institute, Cairo University and Baheya Centre of Early Detection and Treatment of Breast Cancer, Giza, Egypt between February 2020 and March 2021, 20 patients with breast benign breast lesions and 30 normal healthy subjects as controls (age and sex matching). None of the healthy controls had been previously diagnosed with any malignancy. All the recruited BC patients have been pathologically diagnosed as BC.

General Inclusion criteria. Primary single malignancy and Subject age range 21–65

Exclusion criteria. Age <21 or >65, patients received chemotherapy.

This study was approved by the National Research Centre Ethics Committee on 16-9-2019 (Approval No. 19–206) and is in accordance with the ethical standards of the Declaration of Helsinki and written informed consent was obtained from all participants.

Extraction of serum miRNAs was done using miRNA assay Serum/Plasma Kit (Qiagen, Hilden, Germany). All serum RNA preparations were quantified by NanoDrop 1000 (Nanodrop, Wilmington, Delaware, USA). cDNA was done using TaqMan microRNA Reverse Transcription Kit (Applied Biosystems, Carlsbad, CA, USA) and microRNA-specific stem-loop primers (part of the TaqMan microRNA Assay Kit; Applied Biosystems). Real-time Quantitative PCR was performed using the Rotor gene Q Real Time PCR System (Qiagen, Valencia, CA, USA).

We selected miRNAs that had been shown to be dysregulated in BC. Researchers have found significant up regulation of *miR-574-3p* acting as a tumour promoter in breast cancer, osteosarcoma, and gastric cancer [16]. Cardinali B. and colleagues revealed the diagnostic potential of the seven miRNAs (*miR-375*, *miR-21-5p*, *miR-205-5p*, and *miR-194-5p*, upregulated, and *miR-376c-3p*, *miR-382-5p*, and *miR-411-5p*, downregulated), as well as (HR+) BC patients, suggested recurrence in TNBC [17]. *MiR-122a*, *miR- 191*, *miR-382-5p*, and *miR-213* had increased expression in a study on the aberrant expression of miRNAs in ER-positive breast cancer, while *miR-145*, *miR-125b-1*, and *miR-125b-2* had decreased expression levels [18].

The relative expression levels of *miR-375*, *miR-574-5p*, and *miR-122* were calculated and normalized to miR-16 (Applied Biosystems, Foster City, CA) using $2^{-\Delta\Delta ct}$ method [19]. Normalization is a key process in quantitative analysis of miRNA levels by qPCR. MiR-16 showed little overall and between-group variability and seemed to be dysregulated in myelodysplastic syndrome and many cancer types [20], so we used *MiR-16* as a standard miRNA to compare expression levels of other miRNAs in plasma of studied groups.

Statistical analysis

Data were analysed using SPSS win statistical package version 24. Numerical data will be expressed as mean and standard deviation (SD), median and range as appropriate. Qualitative data will be expressed as frequency and percentage. Chi-square (Fisher's exact) test will be used to examine the relation between qualitative variables as appropriate.

Multivariate analysis was done for variables statistically significant on univariate level to indicate independent predictive factors using logistic regression model. Calculation of sensitivity, specificity, positive predictive value, negative predictive value and total accuracy with their 95% confidence interval will be done. Correlation analysis was done by using Pearson correlation. P-value $\leq$0.05 will be considered statistically significant and all tests will be 2 tailed.

Results

Our study included 77 patients with primary BC, 20 patients with benign breast lesions, and 30 controls. Descriptive data for the BC group are presented in Table 1. We found no significant differences between the groups studied in terms of age or hormone receptor status, including *ERs*, *PRs*, *and HER2*.

The expression level of *miRNA- 122* was significantly up regulated in BC group than in the benign breast lesions and control groups: those of patients with benign breast lesions and controls however, on analysing the expression levels of *miRNA 574* and *miRNA 375*, patients with BC showed significant down regulation in comparison to the benign breast lesions and control groups as shown in Table 2. Relation Between miRNA 122,574, and 375 and Clinicopathologic Features of BC Patients were presented in S1 Table.

With an AUC of 0.786, serum *miR-122* was able to distinguish the benign breast lesions and control groups from BC in ROC curve analysis, with CI = 0.699 to0.873, p <0.001, with sensitivity = 76.6%, specificity = 70%, at a cut-off 2.5 The analysis of ROC curve also revealed that serum *miR-574* discriminated the benign breast lesions and control groups from BC with AUC = 0.800, 95% CI = 0.708 to 0.893, p < 0.0001, with sensitivity = 74%, specificity = 75.3%, at a cut-off 0.0139. Serum *miRNA-375* also distinguished BC from benign breast lesions and control groups with AUC = 0.796, 95% CI = 0.711to0.882, p < 0.001, with sensitivity = 70%, specificity = 68.8%, at a cut-off 0.1875 Fig 1. In addition to ROC curve analysis, multivariate logistic regression was done to select the independent predictor factors associated with BC risk among non-malignant groups. Our results showed that serum *miR-122* and *miR-574* are considered as significant predictor variables in the multivariate analysis, with adjustment for age as shown in Table 3.

In BC, we found no significant association between *miRNAs 122,574* and *375* and age subgroups (P = .236, .427, and .092 respectively). No significant difference was found in our study between hormone receptor status and studied miRNAs (P = .597, 334 and .497) for estragon receptors, (P = .586, .482, and.831) for progesterone receptors, and (P = .482,.694, and.301) for human epidermal growth factor receptor 2.

Table 1. Baseline characteristics of BC group (n = 77).

	N	%
Age		
Mean ± SD	52.92 ± 10.953	
Median (IQR)	53.00 (29.0–75.0)	
< = median value	39	50.6
>median value	38	49.4
Histopathology pattern		
ductal carcinoma insitu	3	3.9
Invasive duct carcinoma	62	80.5
Invasive lobular carcinoma,	6	7.8
Invasive micro papillary carcinoma	1	1.3
Invasive tubular/cribriform carcinoma	1	1.3
mixed invasive duct and invasive lobular carcinoma	1	1.3
Mucinous carcinoma	3	3.9
Grades		
grade I	5	6.5
grade II	52	67.5
grade III	17	22.1
Grade 0	3	3.9
ER status		
negative	2	2.6
positive	75	97.4
PR status		
negative	2	2.6
positive	75	97.4
HER2 status		
negative	65	84.4
positive	12	15.6

https://doi.org/10.1371/journal.pone.0298536.t001

Spearman correlation analysis showed a positive significant correlation between *mir574* and *mir375* ($p<0.001$) and weak negative significant correlation between *mir574* and *mir122* ($p = 0.013$) and *mir375* and *mir122* ($p = 0.017$) as shown in Table 4, Fig 2.

Discussion

The incidence of BC is rapidly increasing in developed countries. Early detection not only improves treatment outcomes, but also has a positive impact on the psychological, economic, and social complications of this disease [21]. Therefore, there is a significant focus on finding minimally invasive, cost-effective diagnostic biomarkers for BC. This research aims to identify and investigate their utility for diagnosing and prognosing different cancers, including BC [22]. Several miRNAs have appeared in the plasma or serum of patients with BC. MiRNAs

Table 2. Expression of miRNAs among studied groups.

Group		*mir122*	*mir574*	*mir375*
BC	Median (IQR) (Min -Max)	8.0 (1.0–64.0)	0.006 (0.0003–0.45)	0.1250 (0.0010–1.0)
Other groups	Median (IQR) (Min -Max)	2.0 (0.0078–32.0)	2.0 (0.0006–64.0)	4.0 (0.0020–128.0)
P value		**0.002**	**<0.001**	**<0.001**

https://doi.org/10.1371/journal.pone.0298536.t002

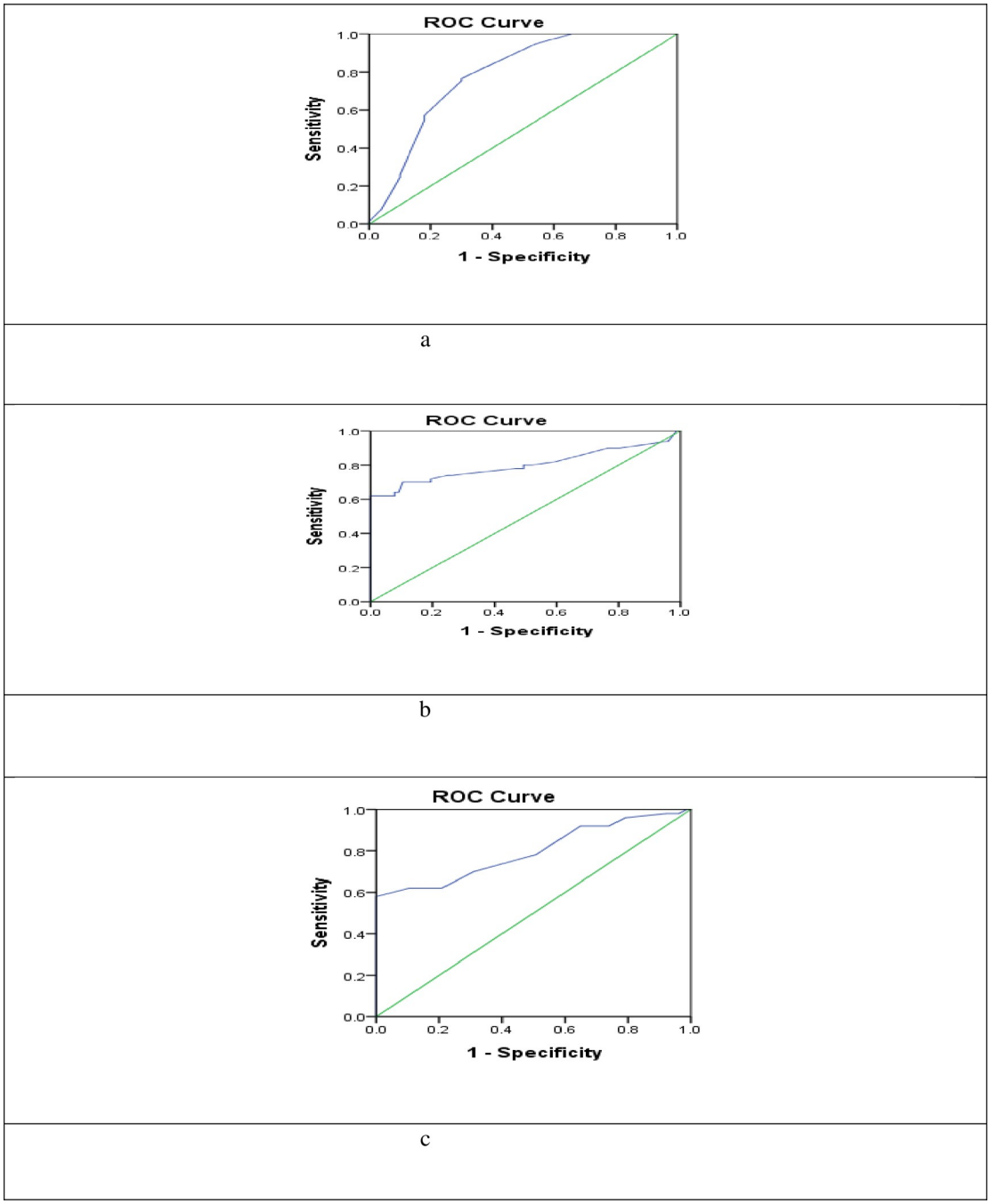

Fig 1. Diagnostic accuracy of serum mir122, mir574 and mir375. A: ROC curve of mir122, B: ROC curve of mir574, C: ROC curve of mir575.

https://doi.org/10.1371/journal.pone.0298536.g001

Table 3. Multivariate analysis (logistic regression) model.

Independent predictive factor	Beta coefficient	Standard error	p value	odds ratio	95% C.I. for odds ratio	
					Lower	Upper
mir122 (>2.5 v. < = 2.5)	1.432	0.463	0.002	4.185	1.689	10.371
mir574 (< = 0.0139 v.>0.0139)	1.560	0.474	0.001	4.758	1.880	12.041
mir375 (< = 0.1875 v >0.1875)	0.552	0.489	0.258	1.737	0.667	4.527

https://doi.org/10.1371/journal.pone.0298536.t003

Table 4. Correlation analysis between studied miRNAs.

Pearson correlation		*mir574*	*mir375*
mir375	Pearson Correlation coefficient	0.838	
	p value	**<0.001**	
mir122	Pearson Correlation coefficient	-0.219	-0.212
	p value	**.013**	**.017**

https://doi.org/10.1371/journal.pone.0298536.t004

profiling studies can categorize dysregulated miRNAs and group BC patients for treatments, this may demonstrate the potential of miRNA as a prognostic and therapeutic biomarker [23].

In BC patients, *miR-122* has been identified as a tumour suppressor that targets the insulin-like growth factor-1 receptor [24]. A previous study showed that *miR-122* acts as a tumour suppressor by regulating oncogenes such as *cyclin G1*, *AKT3*, *and CDK4* in hepatocellular carcinoma [25]. Downregulation of *miR-122* has been observed in hepatocellular carcinoma [26].

On the other hand, higher levels of circulatory *miR-122* correlate with hepatocellular carcinoma [27] and liver injury [28]. Previous study reported that *miR-122* Expression has been detected in primary fibroblasts, where it is involved in *p53mRNA* polyadenylation/translation by targeting cytoplasmic polyadenylation element binding protein [29].

A study by Fong et al. found that breast cancer (BC) secretes an excess amount of *miR-122*, suggesting that it may act as an onco-microRNA [30]. The physiological role of *miR-122* differs according to the type of cancer [19]. Our study demonstrated a significantly higher expression of serum *miRNA122* in BC patients compared to age-matched patients with benign breast lesions and healthy controls. To find a possible relationship between *miRNA 122* and the clinicopathological features of BC patients, we did not find any association between *miR-122* and any of these features. However, a study by Wu et al. found that higher levels of *miR-122* were associated with *HER2*-negative and non-inflammatory tumours [31]. This difference could be explained as most of our patients were *HER2*-negative.

A study by Zhang et al. found that overexpressing *miR-574-5p* significantly reduced tumor weight and size in vivo. Additionally, *miR-574-5p* overexpression inhibited pulmonary migration and significantly decreased the number of metastatic nodules. This suggests that *miR-574-5p* may prevent tumorigenesis and lung metastasis in vivo [13].

Our BC patients exhibited downregulation of *miR-574*, indicating its role as a tumour suppressor miRNA. *MiR-574* inhibits *SKIL* expression by targeting *SOX2 and BCL11A*, thereby inhibiting proliferation, migration, and epithelial-mesenchymal transition (EMT) in triple negative BC [32]. Similarly, *miR-375*, which acts as a tumour suppressor miRNA, is dysregulated in numerous cancers [33].

The results of this study revealed a downregulation of *miR-375* in the BC group, which is consistent with previous research that also reported a downregulation of *miR-375* in BC [34, 35]. However, some studies have indicated that *miR-375* is significantly upregulated in BC [36, 37]. Therefore, the expression level of miR-375 in BC tissues remains controversial, and further studies and research are needed to clarify its role in BC. Additionally, the clinical significance and potential targets of *miR-375* in BC have not been comprehensively investigated thus far. The discrepancy between results may be attributed to differences in sample sizes and types.

Study by Li,L et al. revealed that *miR-375* was significantly downregulated in HCC cell lines and tissues compared with normal hepatic cells and tissues. Further investigation by Li,L et al showed that the stimulation of *miR-375* can supress cell proliferation of HCC and stimulate apoptosis. Thus, *miR-375* regulation has key role in tumorigenesis of HCC *in vitro* [38]. Previous research have shown that upregulation of *ErbB2* gene contributes significantly to hepatocellular growth, and the upregulation of *ErbB2* was associated with the regulation of *miR-375* [39].

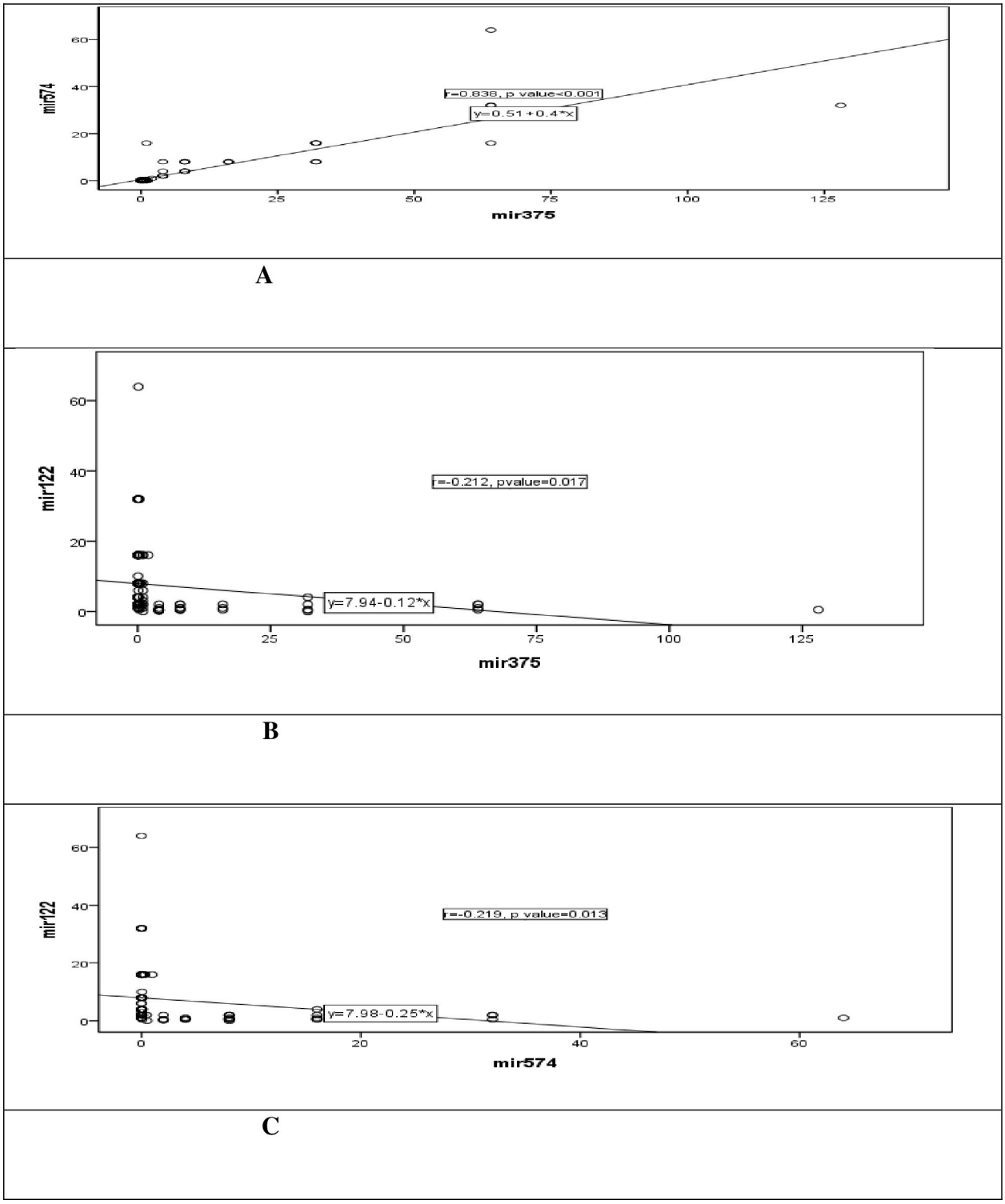

Fig 2. Scatter diagram showing linear relation between mir122, mir574 and mir375. A: linear relation between mir375 & mir3574, B: linear relation between mir122 & mir375, C: linear relation between mir122 & mir574.

https://doi.org/10.1371/journal.pone.0298536.g002

In our ROC analysis, serum *miR-122* had an AUC of 0.786 and was able to distinguish between benign breast lesions and control groups from BC with a sensitivity of 76.6% and specificity of 70% at a cut-off of 2.5. The analysis of the ROC curve also revealed that serum *miR-574* discriminated between benign breast lesions and control groups from BC with an AUC of 0.800 at a cut-off of 0.0139, with a sensitivity of 74% and specificity of 75.3%. Serum *miRNA-375* also distinguished BC from benign breast lesions and control groups with an AUC of 0.796, a sensitivity of 70%, and a specificity of 68.8% at a cut-off of 0.1875.

Limitations of the study

The main limitation of this study was the lack of a power analysis to calculate the sample size. Additionally, due to budget constraints, we were unable to have an equal number of participants in all groups.

Conclusion

In conclusion, we found that serum *miRNA-122* is upregulated in patients with BC, while *mir-574* and *miR-375* expression are downregulated. Our findings suggest that *miR-122* may act as an onco-microRNA, while *mir-574* and *miR-375* have a tumour suppressor role. These studied miRNAs may serve as minimally invasive biomarkers for BC and could represent promising potential therapeutic targets to increase patient survival and quality of life. However, validation and standardization of miRNA-based therapeutics are required to ensure their clinical efficacy.

Supporting information

S1 Table. Relation between miRNA 122,574, and 375 and clinicopathologic features of BC patients.
(DOCX)

Acknowledgments

We are thankful to all the research team for cooperation. All appreciation to participants.

Author Contributions

Conceptualization: Eman A. Elghoroury, Esmat E. Abdelghafar.

Data curation: Eman Awadallah.

Formal analysis: Gamila S. M. EL-Saeed, Rasha Nazih Yousef.

Funding acquisition: Eman A. Elghoroury.

Investigation: Solaf Kamel, Eman Awadallah, Aliaa Shalaby, Gamila S. M. EL-Saeed, Eman Mahmoud, Mahmoud M. Kamel, Rasha Nazih Yousef.

Methodology: Esmat E. Abdelghafar, Solaf Kamel, Aliaa Shalaby, Gamila S. M. EL-Saeed, Asmaa Abobakr, Rasha Nazih Yousef.

Project administration: Aliaa Shalaby, Eman Mahmoud.

Writing – original draft: Eman Awadallah, Rasha Nazih Yousef.

Writing – review & editing: Eman A. Elghoroury, Esmat E. Abdelghafar, Solaf Kamel, Aliaa Shalaby, Gamila S. M. EL-Saeed, Eman Mahmoud, Mahmoud M. Kamel, Asmaa Abobakr, Rasha Nazih Yousef.

References

1. Moaz I, Fouad FA, Elmasry H, Tarek G, Elzoheiry A, Elgamal M, et al. Associations Between Serum Soluble Toll-like Receptors 4 and 9 and Breast Cancer in Egyptian Patients. Cancer Control. 2023 Jan-Dec; 30: 10732748231204755. https://doi.org/10.1177/10732748231204755 PMID: 37771087; PMCID: PMC10541740.
2. Azim HA, Elghazawy H, Ghazy RM, Abdelaziz AH, Abdelsalam M, Elzorkany A, et al. Clinicopathologic Features of Breast Cancer in Egypt-Contemporary Profile and Future Needs: A Systematic Review and Meta-Analysis. JCO Glob Oncol. 2023 Mar; 9:e2200387. https://doi.org/10.1200/GO.22.00387 PMID: 36888929; PMCID: PMC10497263.
3. Khoury S, Tran N. Circulating microRNAs: potential biomarkers for common malignancies. Biomark Med. 2015; 9(2):131–51. https://doi.org/10.2217/bmm.14.102 PMID: 25689901.
4. Ge W, Yi M, Pak TR, Peng C, O'brien J, Hayder H, et al. Overview of MicroRNA Biogenesis, Mechanisms of Actions, and Circulation. Front Endocrinol | www.frontiersin.org. 2018; 1:402.
5. Yousef RN, Ramadan A, Awadallah E, Alnaggar AR, Khalil NM, E Behiry M, et al. Pro-apoptotic Bax mRNA expression: A novel predictor for systemic lupus erythematosus disease flare-up. Arch Rheumatol. 2022 Sep 20; 38(1):129–137. https://doi.org/10.46497/ArchRheumatol.2023.9448 PMID: 37235117; PMCID: PMC10208621.
6. Zografos E, Zagouri F, Kalapanida D, Zakopoulou R, Kyriazoglou A, Apostolidou K, et al. Prognostic role of microRNAs in breast cancer: A systematic review. Oncotarget. 2019 Dec 24; 10(67):7156–7178. https://doi.org/10.18632/oncotarget.27327 PMID: 31903173; PMCID: PMC6935258.
7. Acunzo M, Romano G, Wernicke D, Croce CM. MicroRNA and cancer—a brief overview. Adv Biol Regul. 2015 Jan; 57:1–9. https://doi.org/10.1016/j.jbior.2014.09.013. Epub 2014 Sep 28. Erratum in: Adv Biol Regul. 2015 May;58:53. PMID: 25294678.
8. Bertoli G, Cava C, Castiglioni I. MicroRNAs: New Biomarkers for Diagnosis, Prognosis, Therapy Prediction and Therapeutic Tools for Breast Cancer. Theranostics. 2015 Jul 13; 5(10):1122–43. https://doi.org/10.7150/thno.11543 PMID: 26199650; PMCID: PMC4508501.
9. Liu Y, Wang Q, Wen J, Wu Y, Man C. MiR-375: A novel multifunctional regulator. Life Sci. 2021 Jun 15; 275:119323. https://doi.org/10.1016/j.lfs.2021.119323. Epub 2021 Mar 17. PMID: 33744323.
10. Liu J, Wang P, Zhang P, Zhang X, Du H, Liu Q, et al. An integrative bioinformatics analysis identified miR-375 as a candidate key regulator of malignant breast cancer. J Appl Genet. 2019 Nov; 60(3–4):335–346. https://doi.org/10.1007/s13353-019-00507-w. Epub 2019 Aug 1. PMID: 31372832.
11. Lin Z, Chen M, Wan Y, Lei L, Ruan H. miR-574-5p Targets FOXN3 to Regulate the Invasion of Nasopharyngeal Carcinoma Cells via Wnt/β-Catenin Pathway. Technol Cancer Res Treat. 2020 Jan-Dec; 19:1533033820971659. https://doi.org/10.1177/1533033820971659 PMID: 33317407; PMCID: PMC7745553.
12. Krishnan P, Ghosh S, Wang B, Li D, Narasimhan A, Berendt R, et al. Next generation sequencing profiling identifies miR-574-3p and miR-660-5p as potential novel prognostic markers for breast cancer. BMC Genomics. 2015 Sep 29; 16:735. https://doi.org/10.1186/s12864-015-1899-0 PMID: 26416693; PMCID: PMC4587870.
13. Zhang KJ, Hu Y, Luo N, Li X, Chen FY, Yuan JQ, et al. miR-574-5p attenuates proliferation, migration and EMT in triple-negative breast cancer cells by targeting BCL11A and SOX2 to inhibit the SKIL/TAZ/CTGF axis. Int J Oncol. 2020 May; 56(5):1240–1251. https://doi.org/10.3892/ijo.2020.4995. Epub 2020 Feb 20. PMID: 32319565
14. Ma L, Liu J, Shen J, Liu L, Wu J, Li W, et al. Expression of miR-122 mediated by adenoviral vector induces apoptosis and cell cycle arrest of cancer cells. Cancer Biol Ther. 2010 Apr 1; 9(7):554–61. https://doi.org/10.4161/cbt.9.7.11267. Epub 2010 Apr 1. PMID: 20150764.
15. Faramin Lashkarian M, Hashemipour N, Niaraki N, Soghala S, Moradi A, Sarhangi S, et al. MicroRNA-122 in human cancers: from mechanistic to clinical perspectives. Cancer Cell Int. 2023 Feb 20; 23(1):29. https://doi.org/10.1186/s12935-023-02868-z PMID: 36803831; PMCID: PMC9940444.
16. Shen X, Xue Y, Cong H, Wang X, Ju S. Dysregulation of serum microRNA-574-3p and its clinical significance in hepatocellular carcinoma. Ann Clin Biochem. 2018 Jul; 55(4):478–484. https://doi.org/10.1177/0004563217741908. Epub 2017 Nov 15. PMID: 29065698
17. Cardinali B, Tasso R, Piccioli P, Ciferri MC, Quarto R, Del Mastro L. Circulating miRNAs in Breast Cancer Diagnosis and Prognosis. Cancers (Basel). 2022 May 7; 14(9):2317. https://doi.org/10.3390/cancers14092317 PMID: 35565446; PMCID: PMC9101355.
18. Shao X, Huang P, Shi L, Lei L, Cao W, Chen Z, et al. MicroRNA and LncRNA Expression Profiles in Human Estrogen Receptor Positive Breast Cancer. Clin Lab. 2019 Jan 1; 65(1). https://doi.org/10.7754/Clin.Lab.2018.180340 PMID: 30775882.

19. Schmittgen TD, Livak KJ. Analyzing real-time PCR data by the comparative C(T) method. Nat Protoc. 2008; 3(6):1101–8. https://doi.org/10.1038/nprot.2008.73 PMID: 18546601.

20. Masè M, Grasso M, Avogaro L, D'Amato E, Tessarolo F, Graffigna A, et al. Selection of reference genes is critical for miRNA expression analysis in human cardiac tissue. A focus on atrial fibrillation. Sci Rep. 2017 Jan 24; 7:41127. https://doi.org/10.1038/srep41127 PMID: 28117343; PMCID: PMC5259703

21. Koopaie M, Kolahdooz S, Fatahzadeh M, Manifar S. Salivary biomarkers in breast cancer diagnosis: A systematic review and diagnostic meta-analysis. Cancer Med. 2022 Jul; 11(13):2644–2661. https://doi.org/10.1002/cam4.4640. Epub 2022 Mar 22. PMID: 35315584; PMCID: PMC9249990.

22. Martins I, Ribeiro IP, Jorge J, Gonçalves AC, Sarmento-Ribeiro AB, Melo JB, et al. Liquid Biopsies: Applications for Cancer Diagnosis and Monitoring. Genes (Basel). 2021 Feb 27; 12(3):349. https://doi.org/10.3390/genes12030349 PMID: 33673461; PMCID: PMC7997281.

23. Afzal S, Hassan M, Ullah S, Abbas H, Tawakkal F, Khan MA. Breast Cancer; Discovery of Novel Diagnostic Biomarkers, Drug Resistance, and Therapeutic Implications. Front Mol Biosci. 2022 Feb 21; 9:783450. https://doi.org/10.3389/fmolb.2022.783450 PMID: 35265667; PMCID: PMC8899313.

24. Wang B, Wang H, Yang Z. MiR-122 inhibits cell proliferation and tumorigenesis of breast cancer by targeting IGF1R. PLoS One. 2012; 7(10):e47053. https://doi.org/10.1371/journal.pone.0047053. Epub 2012 Oct 8. PMID: 23056576; PMCID: PMC3466252.

25. Nassirpour R, Mehta PP, Yin MJ. miR-122 regulates tumorigenesis in hepatocellular carcinoma by targeting AKT3. PLoS One. 2013 Nov 7; 8(11):e79655. https://doi.org/10.1371/journal.pone.0079655. Retraction in: PLoS One. 2017 Sep 8;12 (9):e0184778. PMID: 24244539; PMCID: PMC3820664.

26. Kutay H, Bai S, Datta J, Motiwala T, Pogribny I, Frankel W, et al. Downregulation of miR-122 in the rodent and human hepatocellular carcinomas. J Cell Biochem. 2006 Oct 15; 99(3):671–8. https://doi.org/10.1002/jcb.20982 PMID: 16924677; PMCID: PMC3033198.

27. Xu J, Wu C, Che X, Wang L, Yu D, Zhang T, et al. Circulating microRNAs, miR-21, miR-122, and miR-223, in patients with hepatocellular carcinoma or chronic hepatitis. Mol Carcinog. 2011 Feb; 50(2):136–42. https://doi.org/10.1002/mc.20712. Epub 2010 Dec 10. PMID: 21229610.

28. Zhang Y, Jia Y, Zheng R, Guo Y, Wang Y, Guo H, et al. Plasma microRNA-122 as a biomarker for viral-, alcohol-, and chemical-related hepatic diseases. Clin Chem. 2010 Dec; 56(12):1830–8. https://doi.org/10.1373/clinchem.2010.147850. Epub 2010 Oct 7. PMID: 20930130.

29. Burns DM, D'Ambrogio A, Nottrott S, Richter JD. CPEB and two poly(A) polymerases control miR-122 stability and p53 mRNA translation. Nature. 2011 May 5; 473(7345):105–8. https://doi.org/10.1038/nature09908. Epub 2011 Apr 10. PMID: 21478871; PMCID: PMC3088779.

30. Fong MY, Zhou W, Liu L, Alontaga AY, Chandra M, Ashby J, et al. Breast-cancer-secreted miR-122 reprograms glucose metabolism in premetastatic niche to promote metastasis. Nat Cell Biol. 2015 Feb; 17(2):183–94. https://doi.org/10.1038/ncb3094. Epub 2015 Jan 26. PMID: 25621950; PMCID: PMC4380143.

31. Wu X, Somlo G, Yu Y, Palomares MR, Li AX, Zhou W, et al. De novo sequencing of circulating miRNAs identifies novel markers predicting clinical outcome of locally advanced breast cancer. J Transl Med. 2012 Mar 8; 10:42. https://doi.org/10.1186/1479-5876-10-42 PMID: 22400902; PMCID: PMC3342150.

32. Zhu Q, Le Scolan E, Jahchan N, Ji X, Xu A, Luo K. SnoN Antagonizes the Hippo Kinase Complex to Promote TAZ Signaling during Breast Carcinogenesis. Dev Cell. 2016 Jun 6; 37(5):399–412. https://doi.org/10.1016/j.devcel.2016.05.002. Epub 2016 May 26. PMID: 27237790; PMCID: PMC4902294.

33. Wei J, Lu Y, Wang R, Xu X, Liu Q, He S, et al. MicroRNA-375: potential cancer suppressor and therapeutic drug. Biosci Rep. 2021 Sep 30; 41(9):BSR20211494. https://doi.org/10.1042/BSR20211494 PMID: 34494089; PMCID: PMC8458691.

34. Hong S, Noh H, Teng Y, Shao J, Rehmani H, Ding HF, et al. SHOX2 is a direct miR-375 target and a novel epithelial-to-mesenchymal transition inducer in breast cancer cells. Neoplasia. 2014 Apr; 16 (4):279–90.e1-5. https://doi.org/10.1016/j.neo.2014.03.010. Epub 2014 Apr 18. PMID: 24746361; PMCID: PMC4094831.

35. Luo D, Wilson JM, Harvel N, Liu J, Pei L, Huang S, et al. A systematic evaluation of miRNA:mRNA interactions involved in the migration and invasion of breast cancer cells. J Transl Med. 2013 Mar 5; 11:57. https://doi.org/10.1186/1479-5876-11-57 PMID: 23497265; PMCID: PMC3599769.

36. Munagala R, Aqil F, Vadhanam MV, Gupta RC. MicroRNA 'signature' during estrogen-mediated mammary carcinogenesis and its reversal by ellagic acid intervention. Cancer Lett. 2013 Oct 10; 339 (2):175–84. https://doi.org/10.1016/j.canlet.2013.06.012. Epub 2013 Jun 18. PMID: 23791885; PMCID: PMC3775863.

37. de Souza Rocha Simonini P, Breiling A, Gupta N, Malekpour M, Youns M, Omranipour R, et al. Epigenetically deregulated microRNA-375 is involved in a positive feedback loop with estrogen receptor

alpha in breast cancer cells. Cancer Res. 2010 Nov 15; 70(22):9175–84. https://doi.org/10.1158/0008-5472.CAN-10-1318. Epub 2010 Oct 26. PMID: 20978187.

38. Li L, Jia L, Ding Y. Upregulation of miR-375 inhibits human liver cancer cell growth by modulating cell proliferation and apoptosis via targeting ErbB2. Oncol Lett. 2018 Sep; 16(3):3319–3326. https://doi.org/10.3892/ol.2018.9011. Epub 2018 Jun 22. PMID: 30127930; PMCID: PMC6096281.

39. Du Y, Zhu M, Zhou X, Huang Z, Zhu J, Xu J, et al. miR-20a enhances cisplatin resistance of human gastric cancer cell line by targeting NFKBIB. Tumour Biol. 2016 Jan; 37(1):1261–9. https://doi.org/10.1007/s13277-015-3921-1. Epub 2015 Aug 20. PMID: 26286834.