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microRNA profile of endometrial cancer from Indian patients-identification of potential biomarkers for prognosis

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A B S T R A C T

Endometrial cancer is one of the major cancers in women throughout the world. If diagnosed early, these cancers are treatable and the prognosis is usually good. However, one major problem in treating endometrial cancer is accurate diagnosis and staging. Till date, the choice method for diagnosis and staging is histopathology. Although there are few molecular markers identified, they are not always sufficient in making accurate diagnosis and deciding on therapeutic strategy. As a result, very often patients are under treated or over treated. In this study, our group has profiled microRNAs from Indian patients using NGS-based approach. We have identified 212 differentially expressed microRNAs in endometrial cancer. Among these, there are 17 novel miRNAs. Since this data represents only Indian cohort and also lacks survival data, validation across other populations is necessary before being considered as biomarkers. As one approach towards this, these microRNAs have also been compared to data from TCGA, which represent other populations and also correlated to relevance in overall survival. Using *in-silico* approaches, mRNA targets of the miRNAs have been predicted. After comparing with TCGA, we have identified 16 miRNA-mRNA pairs which could be potential prognostic biomarkers for endometrial cancer. This is the first miRNA profiling report from Indian cohort and one of the very few studies which have identified potential biomarkers of prognosis in endometrial cancer.

1. Introduction

Endometrial cancer, arising from the lining of the uterus is the most common cancer of the female reproductive organs in the United States. It is estimated by The American Cancer Society that in 2023, there would be about 66,200 new cases and 13,030 women would die of this cancer. About 90 % of the cancers of endometrium are carcinomas. 5 year survival rate for these cancers is 81 % [1]. Endometrial cancers are broadly classified as Type 1 lesions, which are the most common and are usually hormone sensitive and have good prognosis. These tumors are usually low-grade and have a background of hyperplasia. Type 2 lesions are higher grade and have higher recurrence frequencies [2]. Most endometrial cancers present with symptoms such as vaginal bleeding and pelvic pain. These cancers are usually treatable if diagnosed early. For a long time, histopathology was the choice method for staging endometrial cancer and also for deciding on the course of therapy. However, studies have shown that cancers of the same stage and histology can have very distinct molecular and genomic profiles (reviewed in Ref. [3]). Identification of such molecular markers would aid in developing more effective and personalized therapies. Studies have shown that pembrolizumab (anti-PD-1), an immune checkpoint inhibitor, would be beneficial to endometrial cancer patients who have defective DNA mismatch repair [4,5] (reviewed in Ref. [3]). A better understanding of other molecular changes in endometrial cancer, such as hormone receptor status, could lead to more tailored therapies in the future.

Several biomarkers of prognostic/predictive relevance have been studied in terms of survival and therapeutic response in women with endometrial cancer. This includes hormone receptors, microRNAs (miRNA), and other molecules (Ex: HER2 [6], p21 [7], HE4 [8], PTEN [9], p27 [10], ANCCA [11], and ANXA2 [12]), which have emerged as potentially useful biomarkers (reviewed in Ref. [13]). However, for a

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better understanding and classification of the disease and for making therapeutic decisions, it is essential to identify and critically evaluate more biomarkers and their relevance in prognosis.

miRNAs (micro RNAs) have been reported to have a substantial association with many diseases including cancer. miRNAs, particularly circulating and exosomal miRNAs have a huge potential to be used as diagnostic markers (reviewed in Ref. [14]). miRNAs have been shown to be stable in formalin-fixed, paraffin-embedded (FFPE) tissues in multiple cancer types. Also, miRNAs can be isolated and analyzed from body fluids such as serum, pancreatic juice, urine, semen etc., which makes it possible to be analyzed from liquid biopsies, hence useful in diagnosis as well as forensic analyses [15,16]. Differential miRNA profiles have been seen in pancreatic juice of chronic pancreatitis vs pancreatic adenocarcinoma patients [17]. Differential profiles of miRNA have been seen in sera of patients with malignant and non-malignant lung disease [18]. Analysis of urine samples have identified miRNAs which are potential diagnostic and prognostic biomarkers [19,20]. Besides cancer, miRNAs have been found to be differentially expressed in several other disease conditions. miRNAs are expressed in cell-type specific manner and due to the feasibility of analyzing these with non-invasive procedures, have a huge potential to be developed as biomarkers.

In cases of endometrial cancer, the classification is based mostly on histopathology. The major drawback of this approach is interobserver variability and this substantially influences the therapeutic approaches leading to under or over treatment. Molecular markers would overcome this problem to some extent. Based on genomic abnormalities, TCGA has identified 4 major molecular subtypes namely POLE (DNA polymerase epsilon catalytic subunit) mutated, mismatch repair (MMR)–deficient, p53 abnormal, and no specific molecular profile [21]. This requires a combination of immunohistochemical tests and mutation analysis. The efficacy of these molecular markers in a diagnostic/prognostic test needs evaluation [22]. Also, better molecular markers are needed to stage and sub-type endometrial cancers and decide on the best therapeutic options.

There have been a few studies which have profiled miRNAs in endometrial cancer [23], compared different stages and sub-types [24] and also compared data available from TCGA to identify markers relevant in prognosis [25]. However, all of the above studies are done on populations restricted to one region or data from TCGA. The study shown here presents miRNA profiling data from Indian population which is mostly not represented even in public databases. The data has also been compared to TCGA data, which shows relevance in other populations. Besides, we have also identified mRNA targets of the differentially expressed miRNAs and identified miRNA-mRNA combinations that would be more relevant to be used as biomarkers. Although this study has mainly focused on one sub-type of endometrial cancer, this approach would also be useful in identifying sub-type specific profiles as well.

2. Materials and methods

2.1. Collection of clinical samples

Samples used for this study have been collected from Kidwai Memorial Institute of Oncology (KMIO), Bengaluru and Vani Vilas Hospital, Bengaluru, between the period of 2018–2022. This study was approved by the Institutional Ethics Committee at Kidwai Memorial Institute of Oncology, Vani Vilas Hospital and Centre for Human Genetics where most of the molecular work was carried out (KMIO/MEC/023/05. January 2018 dated 17th Jan 2018; BMCRI/PS/254/2020-21 dated 4th January 2021 and CHG/077(b)/IEC/2018-19/001 dated 21st May 2018). Informed consent has been obtained from the participants and the identity has been kept confidential. The women diagnosed with pathological stage I-IV endometroid type of endometrial carcinoma at KMIO between 2018 and 2022 were included in this study. The control samples were collected from patients undergoing hysterectomy for non-neoplastic conditions. Upon receiving the samples at the histopathology laboratory at KMIO, Hematoxylin and Eosin-stained sections was examined by the pathologist. Immunohistochemistry was performed wherever necessary to arrive at the diagnosis. Based on the histopathology report, endometroid carcinoma samples along with controls were taken for miRNA profiling. Any other histology like polyps, hyperplasia and stromal tumors were rejected. However, a couple of serous carcinomas have been sequenced too. The details of these samples are summarized in Supplementary Table 1.

2.2. RNA isolation

All tissue samples were collected in RNA later (Sigma Life Sciences) and stored at −80 °C till further processing. Before isolation of RNA, the tissues were rinsed with PBS and homogenized with RLT Buffer (QIAGEN GmbH) using a hand-held homogenizer. RNA was isolated using the Qiagen RNeasy Mini Kit (QIAGEN GmbH) according to manufacturer's instructions. On column DNase digestion was also performed for all samples. The isolated RNA was quantified using NanoDrop (Thermo Scientific, USA). The integrity of RNA was assessed and samples with RIN$>/=$ 4 were used for further analysis.

2.3. RNA – Sequencing and analysis

The RNA-sequencing was outsourced to Molsys Pvt Ltd, Bengaluru and Nucleome Informatics Pvt Ltd, Hyderabad.

44 small RNA libraries generated using NEB NextR Multiplex Small RNA Library Prep Set for IlluminaR were sequenced and the resultant fastq files showed the phred score greater than 30 for all samples. The sequencing was performed on Illumina Novaseq 6000 platform. Single end reads were obtained for the samples. The reads were trimmed using the adapter sequence (AGATCGGAAGAGCACACGTCT). Reads that mapped to rRNA sequences were removed using Bowtie [26]. Reads were size-selected for 18-25bp long reads using BBTools [27,28]. The size-selected reads had less than 1.5 % rRNA reads and these were not removed as there was no rfam alert. Samples with a minimum of one million mapped reads were used for DE analysis. Obtained sequences were then collapsed using the mapper tool from miRDeep2 [29–31] and used for identification and quantification of miRNA. DESeq2 function in R [32,33] was used to identify the differentially regulated miRNAs in endometrial cancer vs control samples. Differentially regulated novel miRNAs were identified at FDR$<$0.001 and four-fold change in expression between endometrial cancer samples versus control samples.

In DESeq2, the p-values attained by the Wald test (*pvalue*) are corrected for multiple testing using the Benjamini and Hochberg (BH) method by default and reported as *padj*. The BH method for controlling the FDR is implemented in DESeq2 in the following way: rank the genes/transcripts by p-value, then multiply each ranked p-value by m/rank, where m = total number of tests. The p-adjusted values were used to determine significant transcripts.

2.4. Identification of novel miRNAs

miRDeep2 analysis has been used for identification of novel miRNAs. Two parameters are used to obtain high confidence novel miRNAs namely randfold value and miRDeep score.

The randfold value represents the p-value of the stability of the precursor structure. Whether a novel miRNA passes this threshold or not is indicated by "yes" or "no". We have considered "high_confidence_novel_miRNAs" only when they pass randfold p-value threshold set by miRDeep2.

The second parameter is miRDeep score. Higher the score, greater the confidence that the identified miRNA is real. We have set a miRDEep2 cut-off of 5, which is quite stringent since we don't have experimental validation of these results.

Also, none of the novel miRNAs were part of the Rfam database.

2.5. Retrieval of data from The Cancer Genome Atlas (TCGA)

RNA-Seq data on endometrial cancer was retrieved from The Cancer Genome Atlas [34] database using the Xena Browser [35]. The data from TCGA Endometrioid Cancer (UCEC) containing 26 datasets was selected for our study. This dataset had miRNA sequenced from 429 patient samples using IlluminaHiSeq miRNASeq. Information about 2238 sequenced miRNAs was obtained from TCGA for our analysis.

2.6. Comparison of data obtained from miRNA seq and TCGA

Differential expression data obtained from our cohort (Indian population) identifies the miRNA using gene ID, whereas the data from TCGA identifies using accession numbers. In order to make the comparison, the first step was to convert gene IDs to their respective accession numbers which was done as described in Ref. [36]. The differentially expressed miRNAs were compared to TCGA data to obtain a common list of miRNAs which could be then used for further analysis such as survival analysis and target prediction.

2.7. Survival analysis

The TCGA Endometrioid Cancer (UCEC) cohort had a survival dataset for 583 patients in total. The survival data has endpoints such as Overall survival (OS), Progression-free interval (PFI), Disease-specific survival (DSS), Disease free interval (DFI). The survival data was retrieved from TCGA and compared with the patients for which miRNA expression data was available. 398 patients had both the relevant data and hence only these were used for further analysis.

For prognostic relevance of each miRNA, the patients were grouped into high expressers and low expressers based on the median expression value (as per TCGA data) and the survival probability was plotted against time using K-M plotter [37–39].

If the Hazard ratio was >1, higher expression of the miRNA corresponds to greater risk/lower survival; and if the Hazard ratio <1, higher expression corresponds to less risk/better survival.

2.8. Statistical analysis

All the statistical analyses were performed using the R statistical package. In all the statistical tests, $p < 0.05$ was considered significant [40]. Hazard ratio was also taken into consideration for the selection of significant K-M plots of respective miRNAs [41].

2.9. miRNA target prediction

Gene targets for miRNA were predicted using multiple platforms such as DIANA-microT [42], TargetScan [43], miRDB [44], RNA22 [45]. Highest stringency was applied in these tools while predicting the targets and only those targets were considered which were predicted by at least two tools. Further, the expression of these target mRNAs was checked using NCBI [46] GTEx portal [47] and BioGPS [48] to consider only those relevant in endometrial tissue (Supplementary methods).

3. Results

3.1. Differential expression of miRNAs in endometrial cancer (Indian population)

12 control and 32 endometrial cancer samples representing Indian population were subjected to small RNA sequencing and upon differential expression analysis using miRDeep2 algorithm 927 putative novel miRNAs were identified. Differential expression analysis with a false discovery rate (FDR) < 0.001 and 4-fold change (>/ = 2 log2 fold) in expression, 114 known miRNAs were overexpressed and 81 under expressed in endometrial cancer samples as compared to controls. With

Table 1A
List of overexpressed miRNAs in endometrial cancer in Indian population.

Sl No	miRNA ID	log2FoldChange	p value
1	hsa-miR-10394-3p	4.05408812821108	2.70892647029174E-05
2	hsa-miR-10396b-3p	6.64203044385496	2.78185279346778E-08
3	hsa-miR-10401-3p	2.52064623111676	2.16007211844158E-08
4	hsa-miR-10527-5p	2.15897773298719	1.09066383121924E-10
5	hsa-miR-10a-3p	2.02612950196752	1.48399683441376E-05
6	hsa-miR-1180-3p	2.05966798142879	4.97771335203261E-08
7	hsa-miR-1224-5p	3.83718639800823	2.53905567471859E-08
8	hsa-miR-1226-3p	2.73693385915086	3.49485565794623E-06
9	hsa-miR-1226-5p	3.97332553860335	6.50653130901821E-07
10	hsa-miR-1229-3p	2.62156524768231	6.08569322122924E-05
11	hsa-miR-1246	6.30848477098956	2.99676596646739E-22
12	hsa-miR-1268a	3.91563876962638	2.77537073343144E-14
13	hsa-miR-1268b	3.9458373012541	5.08166859251836E-15
14	hsa-miR-1269a	7.55585207637933	2.89466346340276E-08
15	hsa-miR-1276	3.02346699204678	6.54486648253563E-06
16	hsa-miR-1285-3p	2.69161633029994	1.68192755298267E-14
17	hsa-miR-1285-3p	2.60136385630366	1.54307577458282E-13
18	hsa-miR-1290	4.43946503043541	4.66439067168231E-11
19	hsa-miR-1293	4.90771558121099	2.26699622900911E-08
20	hsa-miR-1301-3p	2.09328299692938	4.18488917893996E-10
21	hsa-miR-1303	3.58458014553992	1.85626587833221E-22
22	hsa-miR-1307-3p	2.1343783418261	1.18672346030074E-16
23	hsa-miR-1307-5p	2.81435136653829	1.01178321586704E-12
24	hsa-miR-135b-3p	3.0667323715462	4.11781765141214E-07
25	hsa-miR-135b-5p	3.40739346449744	2.19357134365905E-11
26	hsa-miR-147b-3p	2.13682031411326	9.90080854995144E-06
27	hsa-miR-16-2-3p	2.36429596959004	2.95510880632931E-08
28	hsa-miR-182-3p	2.57556442671659	4.42955297969504E-06
29	hsa-miR-183-3p	3.44569450157141	9.35744656001801E-12
30	hsa-miR-183-5p	3.83824591967294	7.14261792343733E-24
31	hsa-miR-1908-3p	3.43618155083958	4.54295141571509E-05
32	hsa-miR-190b-5p	2.37304721275059	0.000122298176606933
33	hsa-miR-200a-5p	3.05084224174411	3.86675073391566E-16
34	hsa-miR-200b-3p	3.33816652016192	2.61440466268318E-19
35	hsa-miR-200b-5p	2.51855670298452	1.05220984388054E-08
36	hsa-miR-200c-5p	2.36460933809586	2.54907673906881E-08
37	hsa-miR-205-5p	4.52359013096449	7.45450371343785E-14
38	hsa-miR-2276-3p	2.0472476653875	4.47400463611318E-08
39	hsa-miR-3074-5p	2.76810071804243	3.20341235302688E-07
40	hsa-miR-3127-3p	2.48406790633332	1.47090590117161E-08
41	hsa-miR-3176	4.64483618840627	8.4180067496182E-18
42	hsa-miR-3177-3p	2.06881650070979	4.7917750412006E-10
43	hsa-miR-3180	5.36357497111174	2.34052761458961E-10
44	hsa-miR-3180-5p	4.03712362669603	5.39219249102783E-07
45	hsa-miR-33b-3p	2.98390136361142	3.80020687571396E-05
46	hsa-miR-3614-5p	3.007329912184	1.28405101317073E-08
47	hsa-miR-3615	3.29914085420372	4.36164254855927E-12
48	hsa-miR-3620-5p	3.51797043364398	1.02889973050558E-08
49	hsa-miR-363-5p	2.36797995291836	0.000143931913013642
50	hsa-miR-3651	2.12300343968433	2.51466103688661E-06
51	hsa-miR-3661	3.55114764402196	2.44045903894401E-05
52	hsa-miR-3679-5p	2.22073616995678	2.19064467755307E-07
53	hsa-miR-3691-5p	2.6552827807177	9.53627584259016E-07
54	hsa-miR-3934-5p	2.07831143776761	8.88627096162426E-11
55	hsa-miR-3944-3p	3.29033518875596	6.14265193374122E-11
56	hsa-miR-3960	5.35686102368536	8.83664649538607E-17
57	hsa-miR-429	3.00457065047811	4.41854801894761E-10
58	hsa-miR-4301	3.82574932006719	9.82632701947428E-06
59	hsa-miR-4326	2.01109091190411	5.79637071799888E-05
60	hsa-miR-4443	2.86321615166268	3.11670382869458E-05
61	hsa-miR-4446-3p	3.56757855027163	1.99829398723837E-08
62	hsa-miR-4448	4.56925928311121	2.93965666121221E-12
63	hsa-miR-4449	2.786502500286	2.80519148081319E-07
64	hsa-miR-4474-3p	2.41931094886354	2.93369482080835E-05
65	hsa-miR-4485-3p	3.20217325091375	4.69597215687623E-06
66	hsa-miR-4488	6.24110248345137	6.99901049338773E-22
67	hsa-miR-4498	3.95124591459793	2.20759634087092E-07
68	hsa-miR-449a	3.37974733541941	7.05086961070405E-05
69	hsa-miR-449c-3p	5.4808569069542	3.30575014138808E-05
70	hsa-miR-449c-5p	3.83605784190935	3.53533919676045E-06
71	hsa-miR-4508	2.81036351764211	1.92587479345072E-05
72	hsa-miR-4516	3.80027297920497	1.09716365580978E-06
73	hsa-miR-4652-5p	4.70758973973522	3.60857085971404E-09
74	hsa-miR-4664-3p	3.32566877047998	1.05076948177747E-14

(continued on next page)

Table 1A (*continued*)

Sl No	miRNA ID	log2FoldChange	p value
75	hsa-miR-4690-3p	2.47213750107946	2.92686210266164E-05
76	hsa-miR-4724-5p	3.34780665680445	3.57147098571115E-08
77	hsa-miR-4726-5p	3.26643593492107	7.25133156565705E-05
78	hsa-miR-4741	2.218386693758	2.62469590868086E-05
79	hsa-miR-4746-5p	3.48776977224806	1.62565785544972E-14
80	hsa-miR-4758-3p	2.95865268201134	8.50370395355878E-07
81	hsa-miR-4767	3.58756695255244	7.69676783563651E-05
82	hsa-miR-4800-3p	2.65230899958965	3.73474084919887E-05
83	hsa-miR-508-5p	3.02325369575263	4.516707687124E-06
84	hsa-miR-509-5p	2.60474693756901	3.50177042314105E-05
85	hsa-miR-514a-3p	4.87104861060073	2.87932004887106E-12
86	hsa-miR-522-3p	3.84046059299443	3.41981948973063E-07
87	hsa-miR-548ah-3p	3.35004864653017	7.41127150502119E-09
88	hsa-miR-548p	3.31947770858415	9.31189430847719E-09
89	hsa-miR-5571-3p	3.90151251169962	4.3760158535582E-08
90	hsa-miR-5585-3p	2.77343143921205	2.07116263282558E-08
91	hsa-miR-592	2.48514351622425	4.22680404118158E-06
92	hsa-miR-615-3p	5.29400879855919	3.1980785682538E-10
93	hsa-miR-619-5p	2.81471727223885	7.07333873621349E-09
94	hsa-miR-636	2.39072315162245	1.41994802446829E-05
95	hsa-miR-6499-5p	4.08541269596827	2.99326177305601E-05
96	hsa-miR-6510-3p	3.58752810256029	4.10286449060687E-08
97	hsa-miR-6511b-5p	2.46652826855943	0.000111313565815777
98	hsa-miR-6515-5p	2.02077698498877	9.46104058954359E-05
99	hsa-miR-6741-3p	2.63491077671236	4.73493703035576E-06
100	hsa-miR-6783-3p	3.21893542144498	3.24745802524751E-05
101	hsa-miR-6811-5p	4.05670034750473	0.000124766870251714
102	hsa-miR-6877-5p	2.42555991657886	5.26458435730932E-07
103	hsa-miR-7110-3p	3.91138181436889	1.72799525520476E-05
104	hsa-miR-7114-5p	4.46698702438156	2.7660285236524E-05
105	hsa-miR-744-5p	2.05024254269854	1.65856261490537E-07
106	hsa-miR-760	2.45727729402395	1.07803218494679E-10
107	hsa-miR-7974	2.73579323338091	2.41278215573592E-06
108	hsa-miR-8485	3.07401259012259	7.98471645567668E-07
109	hsa-miR-877-3p	2.23231922935153	6.23469814218819E-07
110	hsa-miR-891a-5p	4.75282674818038	9.74938050592614E-07
111	hsa-miR-892a	6.80979775410319	7.59762221715838E-09
112	hsa-miR-92b-5p	2.23075445681714	1.15377295578123E-07
113	hsa-miR-937-3p	3.54715406186854	3.76545847130846E-11
114	hsa-miR-940	2.67288135935662	1.46776676481919E-08

Table 1B
List of under-expressed miRNAs in endometrial cancer in Indian population.

Sl No	miRNA ID	log2FoldChange	p value
1	hsa-miR-133b	−4.77559548705149	3.62837358209596E-09
2	hsa-miR-6507-5p	−4.2590677491803	4.95481290330244E-10
3	hsa-miR-133a-3p	−3.9625838297427	4.42941333076803E-08
4	hsa-miR-873-3p	−3.7934859801017	8.74029005962238E-08
5	hsa-miR-424-5p	−3.78187014780614	1.18121122230841E-11
6	hsa-miR-99a-5p	−3.77412365846921	1.43846183508716E-14
7	hsa-miR-542-3p	−3.74847956941136	1.82613142158411E-11
8	hsa-miR-450b-5p	−3.74400034127361	1.83604917638782E-11
9	hsa-miR-127-3p	−3.67111499853014	5.53550008739789E-19
10	hsa-miR-548ba	−3.63590340609386	7.72922877848349E-06
11	hsa-miR-503-3p	−3.55910617827279	1.08734401752894E-07
12	hsa-miR-7978	−3.52810452558055	5.35128700898092E-05
13	hsa-miR-145-5p	−3.50507754673605	3.70143771244119E-12
14	hsa-miR-424-3p	−3.37989395803264	4.65862647017077E-12
15	hsa-miR-376c-3p	−3.34708893121866	1.79005484475227E-07
16	hsa-miR-1245b-5p	−3.30709849218289	8.46776457553963E-08
17	hsa-miR-379-5p	−3.29548569179275	1.09712761937035E-15
18	hsa-miR-542-5p	−3.25057294232792	1.80133342015073E-10
19	hsa-miR-199b-5p	−3.21539184456613	2.64728273502414E-17
20	hsa-miR-10b-5p	−3.20994337624249	2.06015955409292E-15
21	hsa-miR-450a-5p	−3.18801821094432	1.63674846397548E-09
22	hsa-miR-450a-5p	−3.1878950736227	1.34790442874315E-09
23	hsa-miR-299-5p	−3.17423052643186	9.94668902342721E-13
24	hsa-miR-143-5p	−3.17254080791684	4.7425932844118E-11
25	hsa-miR-2682-5p	−3.1036877752164	0.000131537472499683
26	hsa-miR-1-3p	−3.00550128364442	1.09683785726244E-07
27	hsa-miR-1-3p	−3.02640310466597	1.43677833216923E-07
28	hsa-miR-758-3p	−2.97367654647158	1.6918354592864E-13

Table 1B (*continued*)

Sl No	miRNA ID	log2FoldChange	p value
29	hsa-miR-494-3p	−2.9687768829988	1.94814197818927E-09
30	hsa-miR-450a-2-3p	−2.95299510194245	1.04153623405706E-09
31	hsa-miR-543	−2.92132025456557	2.46356542754701E-09
32	hsa-miR-337-5p	−2.91954394903171	2.69263248824243E-08
33	hsa-miR-6130	−2.91754931731692	0.000171306219456584
34	hsa-miR-379-3p	−2.91325877831179	4.08613667422692E-11
35	hsa-miR-195-5p	−2.90813586343039	4.09591613433413E-14
36	hsa-miR-152-3p	−2.85342660083679	3.024681164583 99E-14
37	hsa-miR-1247-3p	−2.83885689509533	2.3619771678957E-07
38	hsa-miR-668-3p	−2.82104019441439	3.86774666190839E-07
39	hsa-miR-127-5p	−2.809341694179	5.35463935399 19E-09
40	hsa-miR-145-3p	−2.76312242318583	1.32581369993171E-11
41	hsa-miR-431-3p	−2.74112425527549	3.55277006990286E-09
42	hsa-miR-4510	−2.74049310573019	2.63768891073768E-09
43	hsa-miR-889-3p	−2.73655084116807	4.54928200424253E-06
44	hsa-miR-410-3p	−2.73492304707428	5.06709326132631E-07
45	hsa-miR-548ag	−2.72442862917611	0.000149063309425882
46	hsa-miR-337-3p	−2.71926117708146	1.23732742732394E-07
47	hsa-miR-99a-3p	−2.70206723968973	5.23550260194107E-08
48	hsa-miR-4636	−2.69454665766088	1.04186512780747E-07
49	hsa-miR-381-3p	−2.66399848357141	1.36059536622461E-12
50	hsa-miR-377-5p	−2.6571615294674	5.11751319583271E-09
51	hsa-miR-204-5p	−2.65006381908225	6.49493042836585E-06
52	hsa-miR-487b-3p	−2.64806893985963	3.31610286893566E-14
53	hsa-miR-493-5p	−2.58824536815565	1.335997197437E-08
54	hsa-miR-455-5p	−2.58029855866442	2.34457510916723E-11
55	hsa-miR-1247-5p	−2.55145963305044	5.49587013655587E-07
56	hsa-miR-323a-3p	−2.55014504017967	2.16826148428022E-09
57	hsa-miR-370-5p	−2.54728217307635	1.76598471596164E-05
58	hsa-let-7c-5p	−2.51624217748811	7.50236956900454E-09
59	hsa-miR-409-5p	−2.48671809141588	6.35227806327678E-09
60	hsa-miR-125b-5p	−2.47550637922178	1.43962940989305E-09
61	hsa-miR-125b-5p	−2.4695827191568	1.70625056626765E-09
62	hsa-miR-329-3p	−2.45540288369757	1.48784872004363E-07
63	hsa-miR-136-5p	−2.41138769393291	2.44802907053943E-05
64	hsa-miR-139-5p	−2.39764052971719	7.86005011468326E-12
65	hsa-miR-134-5p	−2.34726761978274	2.83955566481395E-10
66	hsa-miR-140-3p	−2.34474529194242	5.75563986491247E-10
67	hsa-miR-432-5p	−2.31391520923814	3.47811874418365E-07
68	hsa-miR-503-5p	−2.30019741048466	2.08506932866013E-09
69	hsa-miR-382-3p	−2.28995153960109	6.50007904453887E-05
70	hsa-miR-504-5p	−2.28525720173758	2.78608315645544E-06
71	hsa-miR-497-5p	−2.28113627514731	3.33424075074446E-08
72	hsa-miR-143-3p	−2.26408566716571	5.680585043022E-09
73	hsa-miR-139-3p	−2.2539671776198	2.81256915252215E-05
74	hsa-miR-100-5p	−2.23482546990088	1.61076093811086E-06
75	hsa-miR-199a-3p	−2.18792966100933	4.81227506882991E-10
76	hsa-miR-199a-3p	−2.18710226134233	4.90997399296973E-10
77	hsa-miR-199b-3p	−2.18710226134233	4.90997399296973E-10
78	hsa-miR-675-3p	−2.14415753627	4.29053299203045E-05
79	hsa-let-7c-3p	−2.11654209052122	3.54981450768396E-05
80	hsa-miR-342-3p	−2.11465922616237	7.03430105111959E-06
81	hsa-miR-214-3p	−2.01104279731792	2.37243683813047E-06

similar parameters, 17 novel miRNAs were found to be overexpressed in endometrial cancer.

Table 1 shows a list of differentially expressed miRNAs between control and endometrial cancer samples along with the fold changes in expression. Table 2 shows a list of novel miRNAs that are differentially expressed in endometrial cancer.

3.2. Comparison with TCGA data

In order to understand the relevance of the differentially expressed miRNAs in prognosis, the list of differentially expressed miRNAs in Indian population was compared to the differentially expressed miRNA data from TCGA. Among the two endometrial cancer data sets available from TCGA, the one which had information on miRNA expression as well as clinical follow up was chosen (dataset ID: TCGA.UCEC.sampleMap/miRNA_HiSeq_gene). The TCGA endometrioid cancer cohort comprises of data collected from 429 patient samples using the

Table 2
List of differentially expressed novel miRNAs in endometrial cancer.

Sl No	miRNA	log2Fold Change	p-adj (FDR)	Consensus mature sequence	Consensus precursor sequence	Provisional ID
1	Novel_25	2.04	0.00014792	agccuggaagcuggagccugcagu	agccuggaagcuggagccugcagugagcuaugaucaugucccuguacucuagccugggc	10_dna:chromosome_chromosome: GRCh38:10:1:133797422:1_REF_4622
2	Novel_34	2.67	0.000296074	uuuuguugucagggugcaggu	aggccuccagacacaccgcagccaguuccugagggcuuuuguugucagggugcaggu	14_dna:chromosome_chromosome: GRCh38:14:1:107043718:1_REF_12901
3	Novel_42	2.68	0.000248885	aucugggucugcgugaguaacu	aucugggucugcgugaguaacuuucuugcauaaucaagguuacucaaguagaagccugaaaa	6_dna:chromosome_chromosome: GRCh38:6:1:170805979:1_REF_38497
4	Novel_51	4.64	2.97E-08	ucggggagaugagagacgugu	ucggggagaugagagacguguuggcuaaccagcugcagauauucaccucacaucucuugcucucccccagg	6_dna:chromosome_chromosome: GRCh38:6:1:170805979:1_REF_38141
5	Novel_218	4.01	1.57E-08	ucgggcgggcgggaggug	ucgggcgggcgggaggugcaccagcggcggcggcggcgguaaaauccucccggccgcucacagc	11_dna:chromosome_chromosome: GRCh38:11:1:135086622:1_REF_8681
6	Novel_63	3.67	4.13E-05	aacaggcuggcgcucuaggacgcu	aacaggcuggcgcucuaggacgcucuguguucacugaugaugaagaaaccuagaacuccaagccuguuu	16_dna:chromosome_chromosome: GRCh38:16:1:90338345:1_REF_16527
7	Novel_227	4.00	1.21E-06	uuggcugggccucgggcucc	agcuucccacuaaaaacauguuuugaugcugugugcuuuuggcugggccucgggcucc	9_dna:chromosome_chromosome: GRCh38:9:1:138394717:1_REF_45434
8	Novel_65	4.57	0.000956314	acaaagagcugugguagaaagu	acaaagagcugugguagaaaguaagguugcaaaucacuuucuaccacagcucuuuguu	10_dna:chromosome_chromosome: GRCh38:10:1:133797422:1_REF_6336
9	Novel_85	3.74	0.000508503	uucaggaguggggacaucaggg	uucaggaguggggacaucagggauguauggcagcccuggugcauccacuucugaacuca	5_dna:chromosome_chromosome: GRCh38:5:1:181538259:1_REF_34690
10	Novel_88	3.11	0.000403907	acuccuggugcugagauggacgg	acuccuggugcugagauggacggaucaugauuuccuucugucugcaccaggcuuugc	11_dna:chromosome_chromosome: GRCh38:11:1:135086622:1_REF_7420
11	Novel_221	3.27	0.000807564	augugugcaugugcugugugagu	augugugcaugugcugugugagugagcacacccgugugcacacucauacacaugugcacacaca	1_dna:chromosome_chromosome: GRCh38:1:1:248956422:1_REF_2453
12	Novel_90	3.92	0.000427363	gcugggaaguuugggcugcaggaac	Ugcugcuggcugaggagaggcggcugcuggggucucaagcacuuccucagcugcugcugggaaguuugggcugcaggaac	19_dna:chromosome_chromosome: GRCh38:19:1:58617616:1_REF_20519
13	Novel_102	4.02	0.00012697	ccggaguuggaccugggacuug	agucccagguccaauccugaguggccaaaagcaggagacccggaguuggaccugggacuug	6_dna:chromosome_chromosome: GRCh38:6:1:170805979:1_REF_37650
14	Novel_262	3.54	0.000343493	cucucgguucggaccugggcucg	Cucucgguucggaccugggcucggagcggcagccuccaccacugcucguuucuuuaccgcucaaggccgccccuucaugcu	7_dna:chromosome_chromosome: GRCh38:7:1:159345973:1_REF_41083
15	Novel_264	4.53	0.000921258	ccugcuggcuggaaauucuccug	ccugcuggcuggaaauucuccugagaucgguuggagucuauuugcaggcaugcaggca	6_dna:chromosome_chromosome: GRCh38:6:1:170805979:1_REF_36714
16	Novel_257	3.94	0.000112397	aaguguggguucuagaguugggc	aaguguggguucuagaguugggccagccuucuuuucagccucaccuccacuguccacacuugcuu	5_dna:chromosome_chromosome: GRCh38:5:1:181538259:1_REF_34506
17	Novel_233	3.77	0.000796819	cacccucucccagaacccauu	aggguuuuguggggguggaggucgucaacccuaaccguggccaccccucucccagaacccauu	19_dna:chromosome_chromosome: GRCh38:19:1:58617616:1_REF_20375

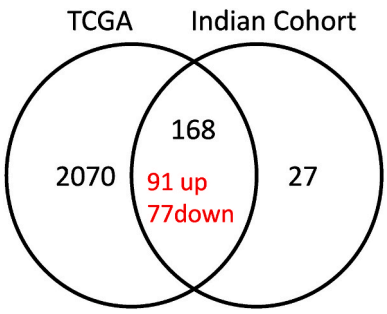

Fig. 1. Differentially expressed miRNAs in control and cancerous endometrial tissue in TCGA data set and Indian cohort. 168 miRNAs are found in common to the data sets.

Table 3A
List of common miRNAs overexpressed in endometrial cancer.

Sl No	miRNA Accession ID	miRNA ID
1	MIMAT0004555	hsa-miR-10a-3p
2	MIMAT0005825	hsa-miR-1180-3p
3	MIMAT0005458	hsa-miR-1224-5p
4	MIMAT0005577	hsa-miR-1226-3p
5	MIMAT0005584	hsa-miR-1229-3p
6	MIMAT0005898	hsa-miR-1246
7	MIMAT0018925	hsa-miR-1268b
8	MIMAT0005923	hsa-miR-1269a
9	MIMAT0005930	hsa-miR-1276
10	MIMAT0005876	hsa-miR-1285-3p
11	MIMAT0005797	hsa-miR-1301-3p
12	MIMAT0005951	hsa-miR-1307-3p
13	MIMAT0022727	hsa-miR-1307-5p
14	MIMAT0004698	hsa-miR-135b-3p
15	MIMAT0000758	hsa-miR-135b-5p
16	MIMAT0004928	hsa-miR-147b-3p
17	MIMAT0004518	hsa-miR-16-2-3p
18	MIMAT0000260	hsa-miR-182-3p
19	MIMAT0004560	hsa-miR-183-3p
20	MIMAT0000261	hsa-miR-183-5p
21	MIMAT0026916	hsa-miR-1908-3p
22	MIMAT0004929	hsa-miR-190b-5p
23	MIMAT0001620	hsa-miR-200a-5p
24	MIMAT0000318	hsa-miR-200b-3p
25	MIMAT0004571	hsa-miR-200b-5p
26	MIMAT0004657	hsa-miR-200c-5p
27	MIMAT0000266	hsa-miR-205-5p
28	MIMAT0011775	hsa-miR-2276-3p
29	MIMAT0019208	hsa-miR-3074-5p
30	MIMAT0019201	hsa-miR-3127-3p
31	MIMAT0015053	hsa-miR-3176
32	MIMAT0015054	hsa-miR-3177-3p
33	MIMAT0018178	hsa-miR-3180
34	MIMAT0004811	hsa-miR-33b-3p
35	MIMAT0017992	hsa-miR-3614-5p
36	MIMAT0017994	hsa-miR-3615
37	MIMAT0022967	hsa-miR-3620-5p
38	MIMAT0003385	hsa-miR-363-5p
39	MIMAT0018071	hsa-miR-3651
40	MIMAT0018082	hsa-miR-3661
41	MIMAT0018104	hsa-miR-3679-5p
42	MIMAT0018120	hsa-miR-3691-5p
43	MIMAT0018349	hsa-miR-3934-5p
44	MIMAT0018360	hsa-miR-3944-3p
45	MIMAT0001536	hsa-miR-429
46	MIMAT0016888	hsa-miR-4326
47	MIMAT0018961	hsa-miR-4443
48	MIMAT0018965	hsa-miR-4446-3p
49	MIMAT0018968	hsa-miR-4449
50	MIMAT0019022	hsa-miR-4488
51	MIMAT0019033	hsa-miR-4498
52	MIMAT0001541	hsa-miR-449a
53	MIMAT0013771	hsa-miR-449c-3p
54	MIMAT0010251	hsa-miR-449c-5p
55	MIMAT0019045	hsa-miR-4508
56	MIMAT0019053	hsa-miR-4516
57	MIMAT0019716	hsa-miR-4652-5p
58	MIMAT0019738	hsa-miR-4664-3p
59	MIMAT0019780	hsa-miR-4690-3p
60	MIMAT0019841	hsa-miR-4724-5p
61	MIMAT0019845	hsa-miR-4726-5p
62	MIMAT0019871	hsa-miR-4741
63	MIMAT0019880	hsa-miR-4746-5p
64	MIMAT0019904	hsa-miR-4758-3p
65	MIMAT0019919	hsa-miR-4767
66	MIMAT0019979	hsa-miR-4800-3p
67	MIMAT0004778	hsa-miR-508-5p
68	MIMAT0004779	hsa-miR-509-5p
69	MIMAT0002883	hsa-miR-514a-3p
70	MIMAT0002868	hsa-miR-522-3p
71	MIMAT0022258	hsa-miR-5571-3p
72	MIMAT0003260	hsa-miR-592
73	MIMAT0003283	hsa-miR-615-3p
74	MIMAT0003306	hsa-miR-636
75	MIMAT0025450	hsa-miR-6499-5p
76	MIMAT0025477	hsa-miR-6510-3p
77	MIMAT0025486	hsa-miR-6515-5p
78	MIMAT0027384	hsa-miR-6741-3p
79	MIMAT0027467	hsa-miR-6783-3p
80	MIMAT0027654	hsa-miR-6877-5p
81	MIMAT0028118	hsa-miR-7110-3p
82	MIMAT0028125	hsa-miR-7114-5p
83	MIMAT0004945	hsa-miR-744-5p
84	MIMAT0004957	hsa-miR-760
85	MIMAT0031177	hsa-miR-7974
86	MIMAT0004950	hsa-miR-877-3p
87	MIMAT0004902	hsa-miR-891a-5p
88	MIMAT0004907	hsa-miR-892a
89	MIMAT0004792	hsa-miR-92b-5p
90	MIMAT0004980	hsa-miR-937-3p
91	MIMAT0004983	hsa-miR-940

IlluminaHiseq platform. Out of these, 398 were tumor samples, and 31 were control samples. Clinical information included sample type, histological type, histological grade, age and year of initial pathological diagnosis, height, weight, clinical stage, pregnancy status, menopause status, birth control pill usage history, diabetes, additional treatment completion success outcome, vital status, etc. 2238 miRNAs were found to be differentially expressed between control and endometrial cancer according to the TCGA data. When compared to data from our cohort, 91 overexpressed and 77 under-expressed miRNAs were also found in the TCGA data (Fig. 1 and Table 3). However, in the common list, 35 miRNAs were overexpressed in both data, 63 miRNAs were under-expressed in both data. For the remaining miRNAs, the fold changes did not match between our data and TCGA data.

3.3. Survival analysis

The survival analysis of common miRNAs was performed as described in the methods. This revealed 40 overexpressed miRNAs (out of 91) and 25 under-expressed miRNAs (out of 77) had a significant effect on survival. 20 miRNAs show better survival when overexpressed in tumors (Fig. 2A, Table 4A), and 45 show poorer survival when overexpressed in tumors (Fig. 2B,Table 4B).

3.4. Prediction of targets of the miRNAs

mRNA targets were predicted for the miRNAs which had significant impact on survival. These targets have been predicted using multiple tools such DIANA-microT, TargetScan and miRDB, RNA22 as described in methods (Fig. 3). Targets which were predicted by at least two tools were considered for further analysis. The mRNA targets were further

Table 3B
List of common miRNAs under-expressed in endometrial cancer.

Sl No	miRNA Accession ID	miRNA ID
1	MIMAT0002890	hsa-miR-299-5p
2	MIMAT0019047	hsa-miR-4510
3	MIMAT0019693	hsa-miR-4636
4	MIMAT0024614	hsa-miR-6130
5	MIMAT0000448	hsa-miR-136-5p
6	MIMAT0000446	hsa-miR-127-3p
7	MIMAT0000447	hsa-miR-134-5p
8	MIMAT0026472	hsa-let-7c-3p
9	MIMAT0001545	hsa-miR-450a-5p
10	MIMAT0022721	hsa-miR-1247-3p
11	MIMAT0003150	hsa-miR-455-5p
12	MIMAT0002171	hsa-miR-410-3p
13	MIMAT0000736	hsa-miR-381-3p
14	MIMAT0000733	hsa-miR-379-5p
15	MIMAT0002820	hsa-miR-497-5p
16	MIMAT0000461	hsa-miR-195-5p
17	MIMAT0031181	hsa-miR-7978
18	MIMAT0003389	hsa-miR-542-3p
19	MIMAT0031074	hsa-miR-450a-2-3p
20	MIMAT0000720	hsa-miR-376c-3p
21	MIMAT0003881	hsa-miR-668-3p
22	MIMAT0004604	hsa-miR-127-5p
23	MIMAT0004601	hsa-miR-145-3p
24	MIMAT0004511	hsa-miR-99a-3p
25	MIMAT0000265	hsa-miR-204-5p
26	MIMAT0000263	hsa-miR-199b-5p
27	MIMAT0000064	hsa-let-7c-5p
28	MIMAT0003180	hsa-miR-487b-3p
29	MIMAT0003340	hsa-miR-542-5p
30	MIMAT0019950	hsa-miR-1245b-5p
31	MIMAT0000271	hsa-miR-214-3p
32	MIMAT0002814	hsa-miR-432-5p
33	MIMAT0002816	hsa-miR-494-3p
34	MIMAT0002813	hsa-miR-493-5p
35	MIMAT0000416	hsa-miR-1-3p
36	MIMAT0000098	hsa-miR-100-5p
37	MIMAT0000097	hsa-miR-99a-5p
38	MIMAT0004757	hsa-miR-431-3p
39	MIMAT0018969	hsa-miR-548ag
40	MIMAT0025470	hsa-miR-6507-5p
41	MIMAT0004749	hsa-miR-424-3p
42	MIMAT0000427	hsa-miR-133a-3p
43	MIMAT0000423	hsa-miR-125b-5p
44	MIMAT0004563	hsa-miR-199b-3p
45	MIMAT0022697	hsa-miR-382-3p
46	MIMAT0002875	hsa-miR-504-5p
47	MIMAT0002874	hsa-miR-503-5p
48	MIMAT0000254	hsa-miR-10b-5p
49	MIMAT0000250	hsa-miR-139-5p
50	MIMAT0000438	hsa-miR-152-3p
51	MIMAT0000435	hsa-miR-143-3p
52	MIMAT0000437	hsa-miR-145-5p
53	MIMAT0004552	hsa-miR-139-3p
54	MIMAT0004954	hsa-miR-543
55	MIMAT0022925	hsa-miR-503-3p
56	MIMAT0013517	hsa-miR-2682-5p
57	MIMAT0006790	hsa-miR-675-3p
58	MIMAT0001638	hsa-miR-409-5p
59	MIMAT0000232	hsa-miR-199a-3p
60	MIMAT0003879	hsa-miR-758-3p
61	MIMAT0026483	hsa-miR-370-5p
62	MIMAT0004690	hsa-miR-379-3p
63	MIMAT0031175	hsa-miR-548ba
64	MIMAT0001629	hsa-miR-329-3p
65	MIMAT0005899	hsa-miR-1247-5p
66	MIMAT0000754	hsa-miR-337-3p
67	MIMAT0000755	hsa-miR-323a-3p
68	MIMAT0000753	hsa-miR-342-3p
69	MIMAT0004921	hsa-miR-889-3p
70	MIMAT0001341	hsa-miR-424-5p
71	MIMAT0004597	hsa-miR-140-3p
72	MIMAT0004599	hsa-miR-143-5p
73	MIMAT0004689	hsa-miR-377-5p
74	MIMAT0000770	hsa-miR-133b
75	MIMAT0004909	hsa-miR-450b-5p
76	MIMAT0004695	hsa-miR-337-5p
77	MIMAT0022717	hsa-miR-873-3p

filtered using information from NCBI, GTEx and BioGPS for its expression in endometrium. After these two filters we found 40 mRNA targets for overexpressed miRNAs (Table 5A) and 25 mRNA targets for underexpressed miRNAs (Table 5B). The mRNA targets were then compared to TCGA data set for their status in endometrial cancer. From this it was found that 59 mRNA (targets of 34 overexpressed and 25 underexpressed miRNAs) targets were present in TCGA data. We then analyzed the miRNA-mRNA pairs and found that a total 16 pairs of miRNA-mRNA showed inverse correlation, which could be potential biomarkers for endometrial cancer prognosis (Table 6).

4. Discussion

Endometrial cancer is the 6th most common cancer in women worldwide and the 15th most common cancer worldwide [49]. If diagnosed early (stage 1 or 2), patients with endometrial cancer have a good prognosis, while patients diagnosed at later stages don't respond well to chemotherapy and have poor outcomes. The categorization of patients as early stage or late stage is mainly based on histopathological evaluation mostly post-surgery. However, this is not a very reliable method in endometrial cancer since it has been seen that cancers of the same stage and histology can have very different molecular profiles (reviewed in Ref. [3]). Moreover, classification based on histopathology would be subject to variability between observers, which in many cases can lead to lapses in patient care, either under or over treatment. Therefore, classification/staging requires better molecular markers so that therapeutic approaches can be better strategized.

Currently, markers such as mutations in POLE, MMR and p53 are being used for endometrial cancer classification [21]. However, these tests require a combination of histopathology and molecular techniques and hence may not be very feasible in routine clinical settings. Studies on multiple cancers including endometrial cancer have identified miRNAs as good diagnostic and prognostic markers [50]. miRNAs have good potential to be diagnostic and prognostic markers since it is seen that the miRNAs expression is cell-type specific, de-regulated in many disease conditions, and also because they can be analyzed with samples procured with non-invasive procedures [14]. Based on studies in other cancers such as breast cancer, miRNAs are good markers for early and sensitive diagnosis of cancers [51].

In the context of endometrial cancer, miRNA expression data is available from other populations but not from Indian population. Our data shows a miRNA profile from Indian patients. We have identified 195 differentially expressed miRNAs in endometrial cancers as compared to control samples. Further, we have compared our miRNA profile with that available from TCGA and found that 169 miRNAs were common to Indian population and the other populations on TCGA (Fig. 1, Table 3). Since ours is a limited time study, we did not have survival data for the Indian population. Hence, the differential expression data from this population has been correlated to survival data from TCGA to get a miRNA signature relevant in prognosis. From this analysis, we have identified 65 miRNAs in endometrial cancer whose expression levels may have a relevance in overall survival of patients. We have also identified potential mRNA targets for these miRNAs. This information could further be used to understand the mechanism of action of the miRNAs and how they may affect the behavior of the tumor cells. Also, this study has identified 59 miRNA-mRNA pairs which are relevant in endometrial cancer. Out of these 16 miRNA-mRNA pairs could be used as potential biomarkers with prognostic relevance (Table 6). Using a combination of miRNA and mRNA pairs may be more

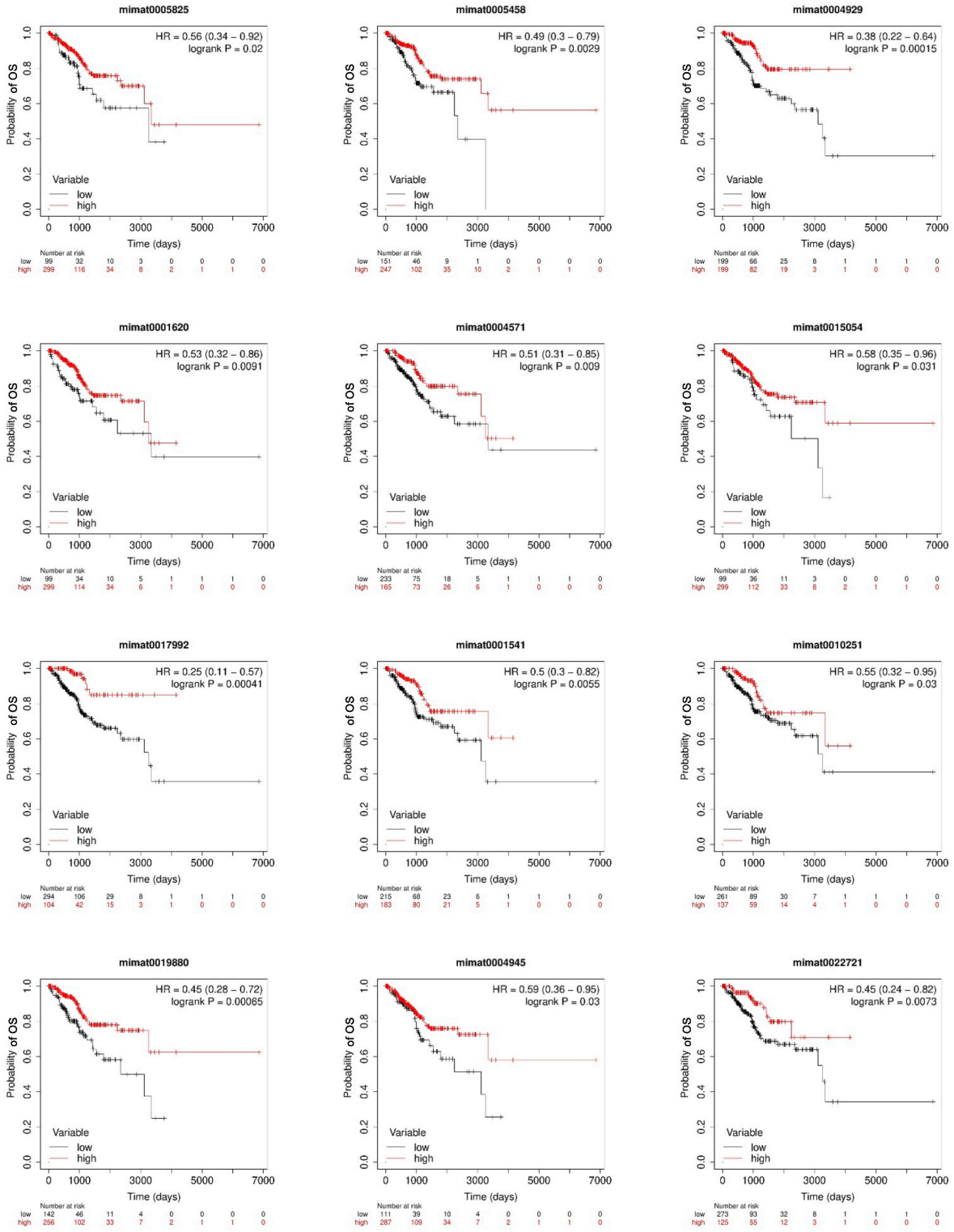

Fig. 2. Kaplan Meier Plots of miRNAs which have an impact on overall survival
2A. K-M plots of miRNAs which are positively correlated with survival (higher expression correlates with better survival)
2B. K-M plots of miRNAs which are negatively correlated with survival (higher expression correlates with poor survival).

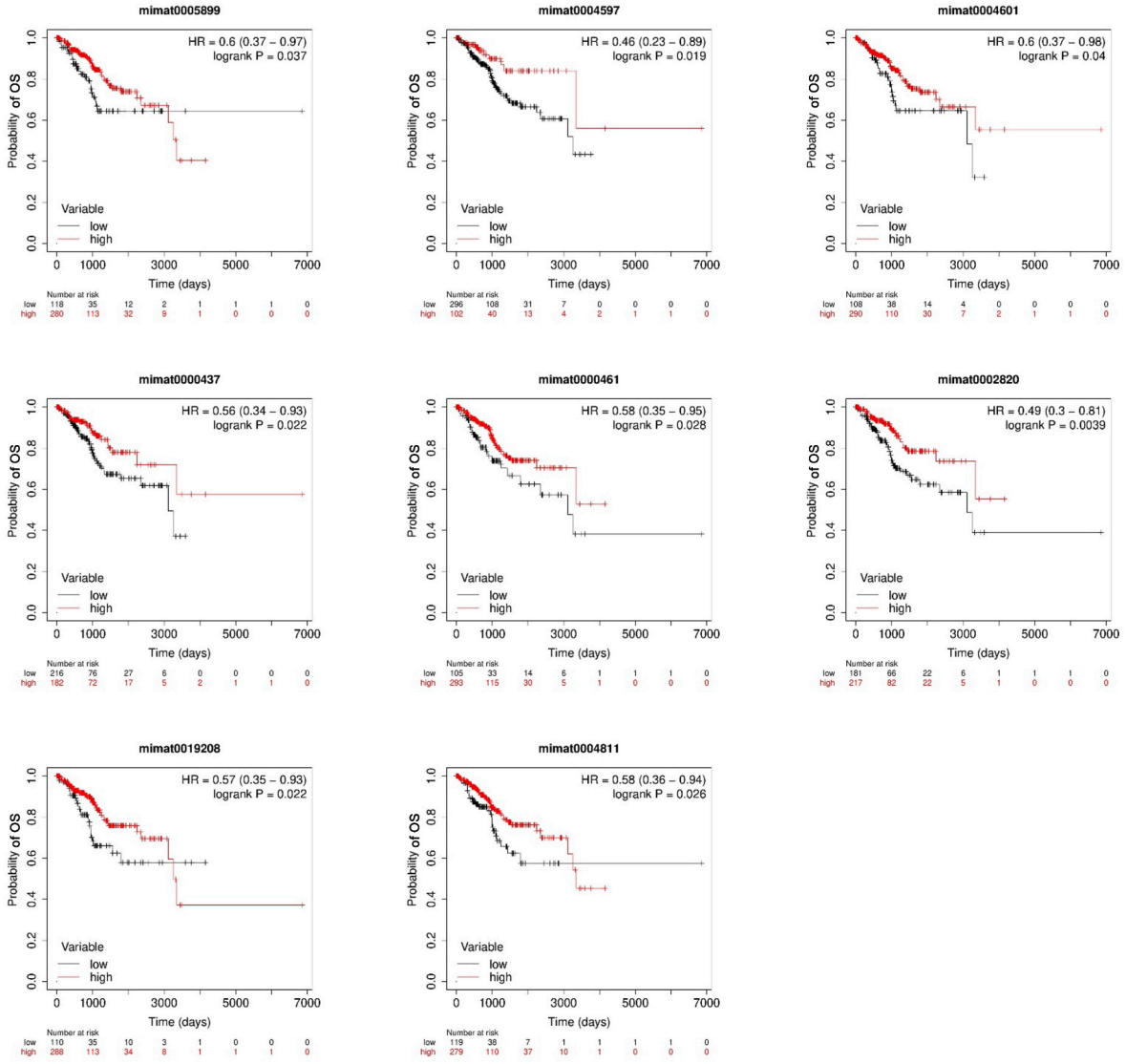

Fig. 2. (*continued*).

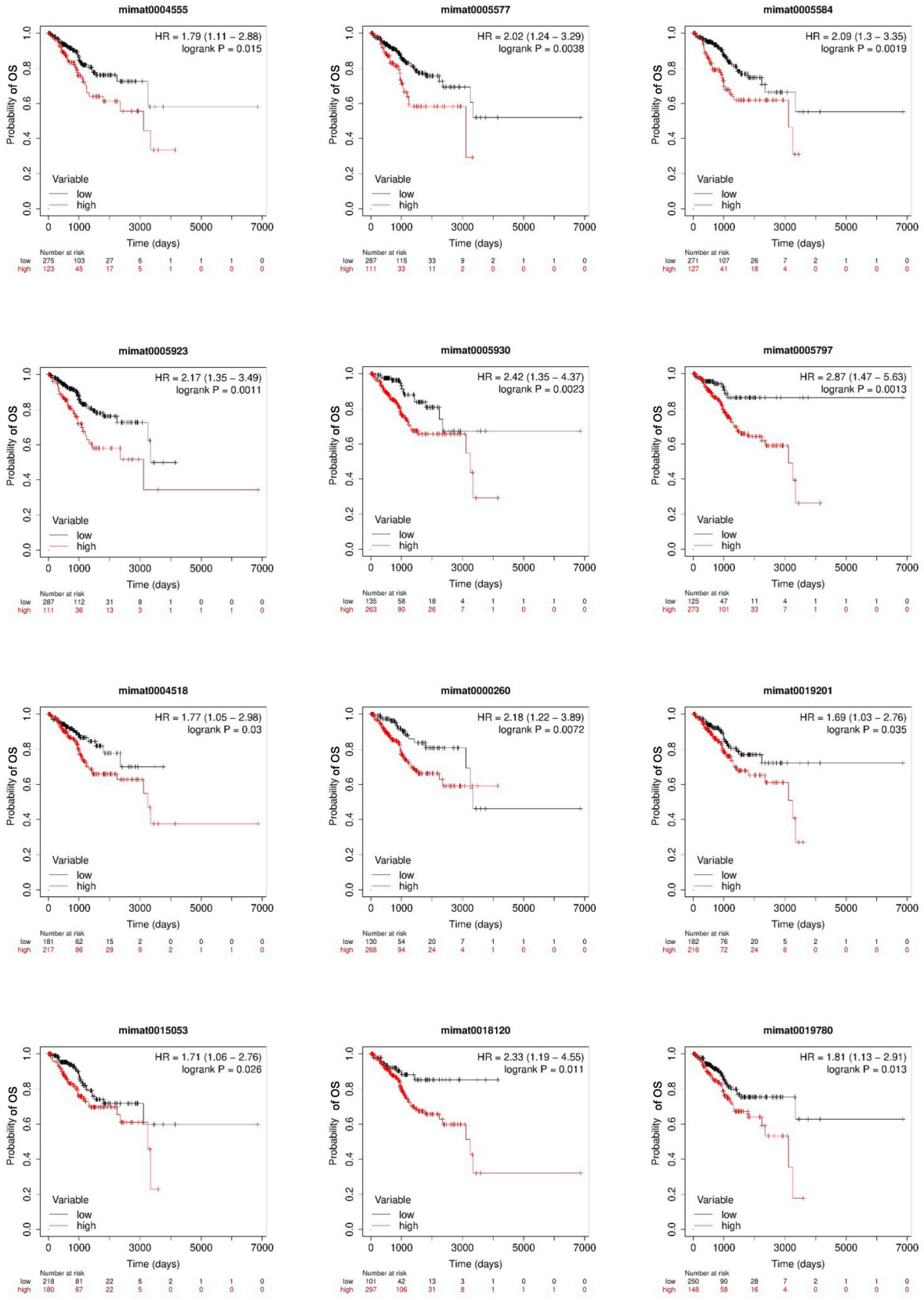

Fig. 2. (*continued*).

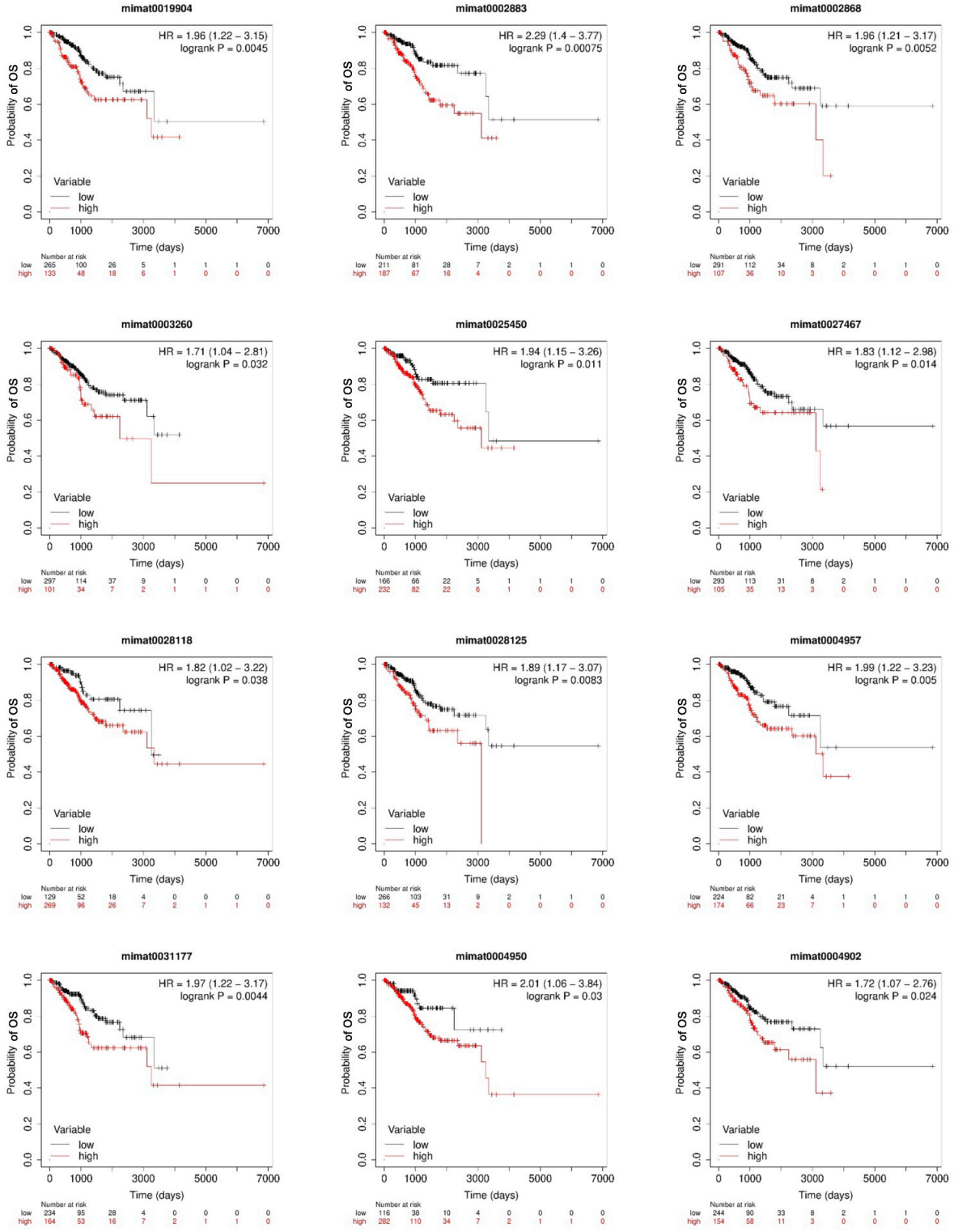

Fig. 2. (*continued*).

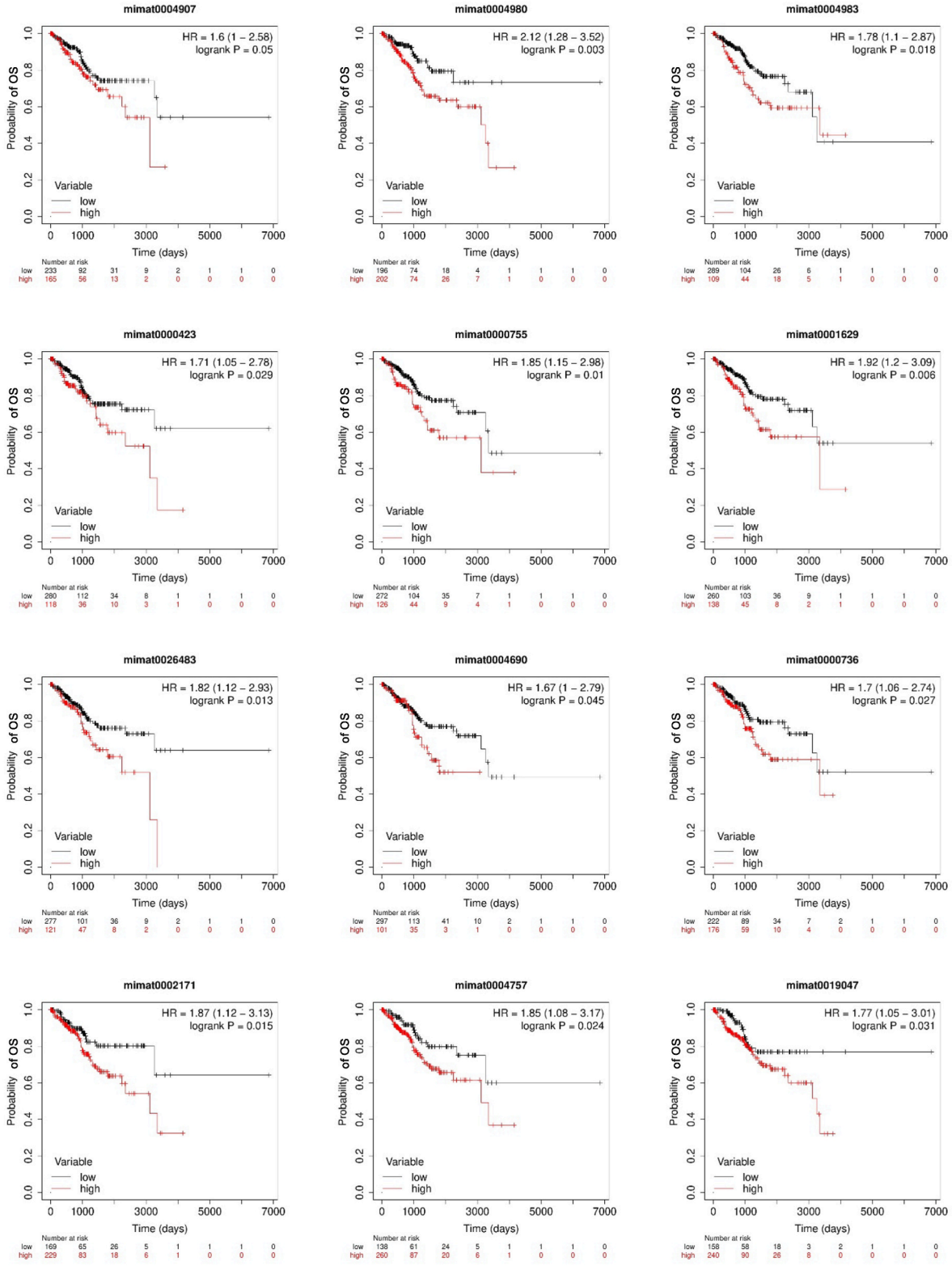

Fig. 2. (*continued*).

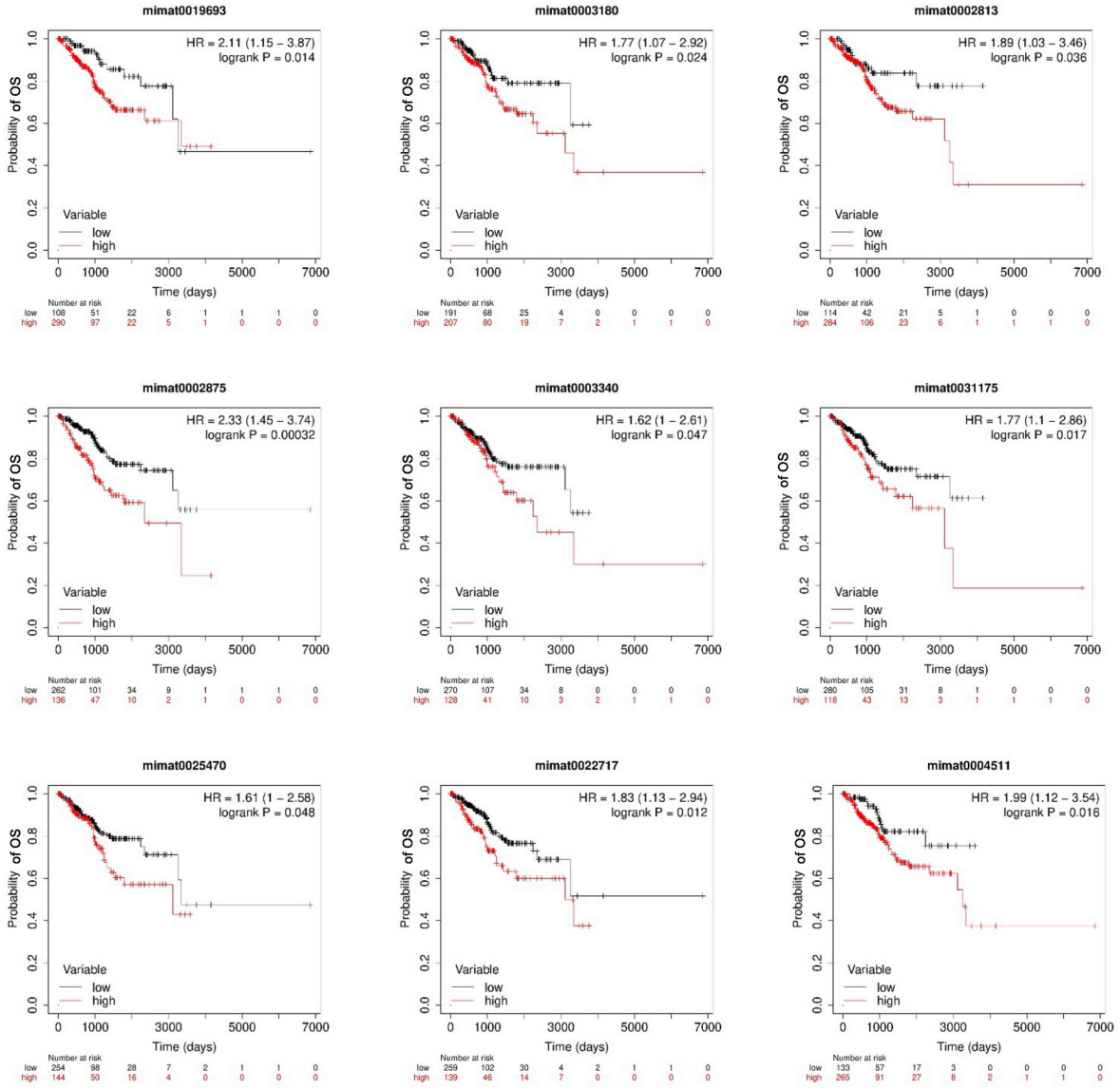

Fig. 2. (*continued*).

Table 4A
List of miRNAs which show positive correlation with overall survival (Higher expression, better survival).

Sl No	miRNA Accession ID	miRNA ID	log2FoldChange (TCGA)	Hazard Ratio	Class Interval
1	MIMAT0005825	hsa-miR-1180-3p	1.67607174384131	0.56	0.34-0.92
2	MIMAT0005458	hsa-miR-1224-5p	1.60613552720857	0.49	0.3-0.79
3	MIMAT0004929	hsa-miR-190b-5p	0.733345776343162	0.38	0.22-0.64
4	MIMAT0001620	hsa-miR-200a-5p	4.73026986908058	0.53	0.32-0.86
5	MIMAT0004571	hsa-miR-200b-5p	3.79951365383049	0.51	0.31-0.85
6	MIMAT0019208	hsa-miR-3074-5p	2.51502113618569	0.57	0.35-0.93
7	MIMAT0015054	hsa-miR-3177-3p	0.328884247468312	0.58	0.35-0.96
8	MIMAT0004811	hsa-miR-33b-3p	0.517516098543945	0.58	0.36-0.94
9	MIMAT0017992	hsa-miR-3614-5p	1.81451886659982	0.25	0.11-0.57
10	MIMAT0001541	hsa-miR-449a	2.60289572431898	0.5	0.32-0.86
11	MIMAT0010251	hsa-miR-449c-5p	2.06785136417921	0.55	0.32-0.95
12	MIMAT0019880	hsa-miR-4746-5p	2.02883196568186	0.45	0.28-0.72
13	MIMAT0004945	hsa-miR-744-5p	1.70536823137802	0.59	0.36-0.95
14	MIMAT0022721	hsa-miR-1247-3p	−2.95731444324289	0.45	0.24-0.82
15	MIMAT0005899	hsa-miR-1247-5p	−1.8262855174504	0.6	0.37-0.97
16	MIMAT0004597	hsa-miR-140-3p	−1.41576717792797	0.46	0.232-0.89
17	MIMAT0004601	hsa-miR-145-3p	−2.51528521913376	0.6	0.37-0.98
18	MIMAT0000437	hsa-miR-145-5p	−2.56918993483357	0.56	0.34-0.93
19	MIMAT0000461	hsa-miR-195-5p	−1.70879662262955	0.58	0.35-0.95
20	MIMAT0002820	hsa-miR-497-5p	−0.846226611959127	0.49	0.3-0.81

Table 4B
List of miRNAs which show negative correlation with overall survival (Higher expression, poor survival).

Sl No	miRNA Accession ID	miRNA ID	log2FoldChange (TCGA)	Hazard Ratio	Class Interval
1	MIMAT0004555	hsa-miR-10a-3p	1.94255175133079	1.79	1.11-2.88
2	MIMAT0005577	hsa-miR-1226-3p	1.32546170751203	2.02	1.24-3.29
3	MIMAT0005584	hsa-miR-1229-3p	0.770005091245478	2.09	1.3–3.35
4	MIMAT0005923	hsa-miR-1269a	3.15636302157153	2.17	1.35-3.49
5	MIMAT0005930	hsa-miR-1276	0.512369898112941	2.42	1.35-4.37
6	MIMAT0005797	hsa-miR-1301-3p	2.65631244224354	2.87	1.47-5.63
7	MIMAT0004518	hsa-miR-16-2-3p	1.90524387864307	1.77	1.05-2.98
8	MIMAT0000260	hsa-miR-182-3p	0.503988850722352	2.18	1.22-3.89
9	MIMAT0019201	hsa-miR-3127-3p	0.431996383409594	1.69	1.03-2.76
10	MIMAT0015053	hsa-miR-3176	0.432343250231378	1.71	1.06-2.75
11	MIMAT0018120	hsa-miR-3691-5p	0.6044296569339	2.33	1.19-4.55
12	MIMAT0019780	hsa-miR-4690-3p	0.20151348533183	1.81	1.13-2.91
13	MIMAT0019904	hsa-miR-4758-3p	0.550324869231555	1.96	1.22-3.15
14	MIMAT0002883	hsa-miR-514a-3p	0.420447080209811	2.29	1.4-3.77
15	MIMAT0002868	hsa-miR-522-3p	0.938142270147642	1.96	1.21-3.17
16	MIMAT0003260	hsa-miR-592	2.20998879245585	1.71	1.04-2.81
17	MIMAT0025450	hsa-miR-6499-5p	0.685171007241738	1.94	1.15-3.26
18	MIMAT0027467	hsa-miR-6783-3p	0.313200902411379	1.83	1.12-2.98
19	MIMAT0028118	hsa-miR-7110-3p	0.271280283126148	1.82	1.02–3.22
20	MIMAT0028125	hsa-miR-7114-5p	0.295414767956948	1.89	1.17-3.07
21	MIMAT0004957	hsa-miR-760	1.22604087653704	1.99	1.22-3.23
22	MIMAT0031177	hsa-miR-7974	0.620317931343121	1.97	1.22-3.17
23	MIMAT0004950	hsa-miR-877-3p	0.547782613870977	2.01	1.06-3.84
24	MIMAT0004902	hsa-miR-891a-5p	2.41710168785045	1.72	1.07-2.76
25	MIMAT0004907	hsa-miR-892a	0.942158295298464	1.6	1–2.58
26	MIMAT0004980	hsa-miR-937-3p	1.62337175148142	2.12	1.26-3.52
27	MIMAT0004983	hsa-miR-940	2.15497963429945	1.78	1.1–2.87
28	MIMAT0000423	hsa-miR-125b-5p	−1.4447269977861	1.71	1.05-2.78
29	MIMAT0000755	hsa-miR-323a-3p	0.306883981541982	1.85	1.15-2.98
30	MIMAT0001629	hsa-miR-329-3p	−0.0574029470693129	1.92	1.22-3.09
31	MIMAT0026483	hsa-miR-370-5p	−0.590254896140642	1.82	1.12-2.93
32	MIMAT0004690	hsa-miR-379-3p	−0.0592534362776794	1.67	1–2.79
33	MIMAT0000736	hsa-miR-381-3p	−2.06447284637611	1.7	1.06-2.74
34	MIMAT0002171	hsa-miR-410-3p	−0.699074668983452	1.87	1.12-3.13
35	MIMAT0004757	hsa-miR-431-3p	−0.21872733860158	1.85	1.08-3.17
36	MIMAT0019047	hsa-miR-4510	−0.0516976524353977	1.77	1.05-3.01
37	MIMAT0019693	hsa-miR-4636	0.148741073256622	2.11	1.15-3.87
38	MIMAT0003180	hsa-miR-487b-3p	−0.469981718229338	1.77	1.07-2.92
39	MIMAT0002813	hsa-miR-493-5p	0.0137593497076636	1.89	1.03–3.46
40	MIMAT0002875	hsa-miR-504-5p	0.288675863741899	2.33	1.45-3.74
41	MIMAT0003340	hsa-miR-542-5p	−1.481009839948	1.62	1–2.61
42	MIMAT0031175	hsa-miR-548ba	0.0153119093961629	1.77	1.1–2.86
43	MIMAT0025470	hsa-miR-6507-5p	−0.121791707634778	1.61	1–2.58
44	MIMAT0022717	hsa-miR-873-3p	0.162175439828722	1.83	1.13-2.94
45	MIMAT0004511	hsa-miR-99a-3p	−1.00590532200546	1.99	1.12-3.54

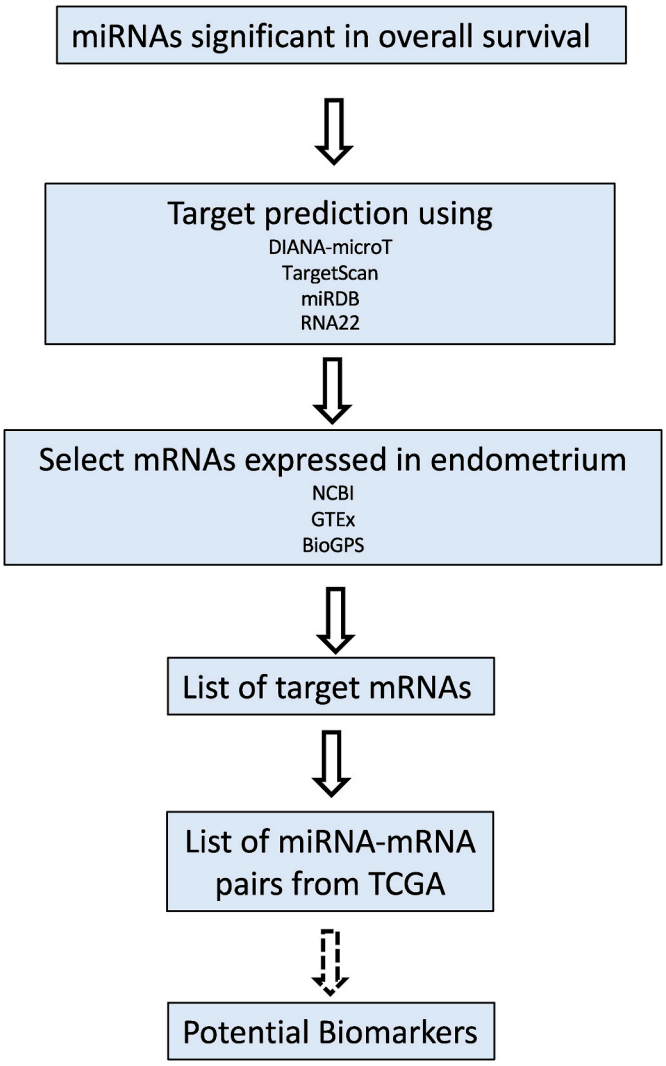

Fig. 3. Schematic work flow for identification of mRNA targets of the miRNAs.

useful in deciding the course of therapy or in sub-typing of cancers rather than using miRNA or mRNA alone, as these combinations may also shed light on the mechanism of action. However, this approach needs further validation on larger cohorts for prognostic significance as well as on cell-lines for mechanism of action.

There have been some studies which have identified differentially expressed miRNAs in endometrial cancer and their potential role in prognosis. Using archival FFPE samples, 84 cancer specific miRNAs have identified as differentially expressed in endometrial cancers. Some of these miRNAs also have an implication in prognosis [23]. When this data is compared with our cohort, only 2 miRNAs (hsa-miR-195-5p and hsa-miR-125b-5p) were in common. This could be due to the fact that the above study was PCR based and analyzed a limited number of miRNAs and our study being NGS based could identify even novel miRNAs.

Another study has performed a meta-analysis of miRNA data from TCGA has identified some potential miRNAs as well as miRNA-mRNA pairs that could have prognostic significance in endometrial cancer [25]. A study conducted at Ohio State University Medical Center, Columbus, OH, from January 1997–July 2003 has done a microRNA profiling of surgically staged endometrial cancers. They have identified differentially expressed miRNA between normal and tumor and also some miRNAs like miR199 which have an implication in patient outcome [24].

Besides these, there have been other studies which have profiled the miRNAs in normal and malignant endometrial tissues (reviewed in Ref. [52]. We find that hsa-miR-10a-3p [53], hsa-miR-135b-3p, hsa-miR-135b-5p [54], hsa-miR-182-3p [55], hsa-miR-183-3p, hsa-miR-183-5p ([55,56], hsa-miR-200a-5p, hsa-miR-200b-5p, hsa-miR-200b-3p [57,58], hsa-miR-200c-5p, hsa-miR-205-5p [9,58,59], hsa-miR-429 [57](56) [58] hsa-miR-522-3p [60], which are overexpressed in endometrial cancer in our data is also reported to be overexpressed in other cohorts. Also, miRNAs such as hsa-miR-133b [61], hsa-miR-424-5p, hsa-miR-127-3p [62], hsa-miR-99a-5p [63](24), hsa-miR-542-3p [64]), hsa-miR-503-3p [65], hsa-miR-424-3p [56](62), hsa-miR-376c-3p, hsa-miR-542-5p [56]), hsa-miR-199b-5p, hsa-miR-10b-5p [56](63) [62], hsa-miR-758-3p [56], hsa-miR-195-5p [54], hsa-miR-152-3p [62](66)), hsa-miR-431-3p [67]), hsa-miR-410-3p [62](58), hsa-miR-99a-3p [63,68], hsa-miR-381-3p [62], hsa-miR-377-5p

Table 5A
List of predicted mRNA targets for the overexpressed miRNAs which have significant impact on overall survival.

Sl No	miRNA Accession ID	miRNA ID	Target Gene	
1	MIMAT0004555	hsa-miR-10a-3p	PTEN	Phosphatase and tensin homolog
2	MIMAT0005825	hsa-miR-1180-3p	BCL6	B-cell lymphoma 6 protein
3	MIMAT0005458	hsa-miR-1224-5p	SIRT6	Sirtuin 6
4	MIMAT0005577	hsa-miR-1226-3p	NRAS	Neuroblastoma RAS viral oncogene homolog
5	MIMAT0005584	hsa-miR-1229-3p	CCNT1	Cyclin T1
6	MIMAT0005923	hsa-miR-1269a	KMT2A	Lysine methyltransferase 2A (MLL)
7	MIMAT0005930	hsa-miR-1276	DDX5	DEAD-box helicase 5 (p68)
8	MIMAT0005797	hsa-miR-1301-3p	ANK2	Ankyrin 2
9	MIMAT0004518	hsa-miR-16-2-3p	CDK6	Cyclin-dependent kinase 6
10	MIMAT0000260	hsa-miR-182-3p	RUNX1T1	RUNX1 partner transcriptional co-repressor 1
11	MIMAT0004929	hsa-miR-190b-5p	TNRC6B	Trinucleotide repeat-containing gene 6B protein
12	MIMAT0001620	hsa-miR-200a-5p	ZEB2	Zinc finger E-box-binding homeobox 2 (SIP1)
13	MIMAT0004571	hsa-miR-200b-5p	CKS2	Cyclin-dependent kinases regulatory subunit 2
14	MIMAT0019208	hsa-miR-3074-5p	THBS1	Thrombospondin 1
15	MIMAT0019201	hsa-miR-3127-3p	WNT5A	Wingless-type MMTV integration site family 5A
16	MIMAT0015053	hsa-miR-3176	BCL6	B-cell lymphoma 6 protein
17	MIMAT0015054	hsa-miR-3177-3p	PRKCG	Protein kinase C gamma
18	MIMAT0004811	hsa-miR-33b-3p	PTGER3	Prostaglandin E receptor 3 (subtype EP3)
19	MIMAT0017992	hsa-miR-3614-5p	MLH1	MutL homolog 1
20	MIMAT0018120	hsa-miR-3691-5p	RASGRP2	RAS guanyl-releasing protein 2
21	MIMAT0001541	hsa-miR-449a	LEF1	Lymphoid enhancer-binding factor 1
22	MIMAT0010251	hsa-miR-449c-5p	CCNG1	Cyclin G1
23	MIMAT0019780	hsa-miR-4690-3p	PKM	Pyruvate kinase M1/M2
24	MIMAT0019880	hsa-miR-4746-5p	LMO2	LIM domain only protein 2

(*continued on next page*)

Table 5A (*continued*)

Sl No	miRNA Accession ID	miRNA ID	Target Gene	
25	MIMAT0019904	hsa-miR-4758-3p	MMP16	Matrix metalloproteinase-16
26	MIMAT0002883	hsa-miR-514a-3p	RBX1	Ring-box 1 (ROC1)
27	MIMAT0002868	hsa-miR-522-3p	PDK1	Pyruvate dehydrogenase kinase isoform 1
28	MIMAT0003260	hsa-miR-592	PIK3CA	Phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha
29	MIMAT0025450	hsa-miR-6499-5p	ARHGAP23	Rho GTPase-activating protein 23
30	MIMAT0027467	hsa-miR-6783-3p	ITGA5	Integrin alpha-5
31	MIMAT0028118	hsa-miR-7110-3p	PDGFA	Platelet-derived growth factor subunit A
32	MIMAT0028125	hsa-miR-7114-5p	RUNX1T1	RUNX1 partner transcriptional co-repressor 1
33	MIMAT0004945	hsa-miR-744-5p	ATXN2	Ataxin-2
34	MIMAT0004957	hsa-miR-760	HIST2H3C	Histone H3.2
35	MIMAT0031177	hsa-miR-7974	CREB3L3	Cyclic AMP-responsive element-binding protein 3-like protein 3
36	MIMAT0004950	hsa-miR-877-3p	MMP16	Matrix metalloproteinase-16
37	MIMAT0004902	hsa-miR-891a-5p	EN1	Engrailed homeobox 1
38	MIMAT0004907	hsa-miR-892a	ZEB1	Zinc finger E-box-binding homeobox 1 (TCF8)
39	MIMAT0004980	hsa-miR-937-3p	PTGFRN	Prostaglandin F2 receptor inhibitor (TIR8 or SIGIRR)
40	MIMAT0004983	hsa-miR-940	IGF1	Insulin-like growth factor 1

Table 5B
List of predicted mRNA targets for the underexpressed miRNAs which have significant impact on Overall survival.

Sl No	miRNA Accession ID	miRNA ID	Target Gene	
1	MIMAT0022721	hsa-miR-1247-3p	ALKBH4	Alkb Homolog 4, Lysine Demethylase
2	MIMAT0005899	hsa-miR-1247-5p	SNX14	Sorting Nexin 14
3	MIMAT0000423	hsa-miR-125b-5p	MFHAS1	Multifunctional ROCO Family Signaling Regulator 1
4	MIMAT0004597	hsa-miR-140-3p	CBL	Cbl Proto-Oncogene
5	MIMAT0004601	hsa-miR-145-3p	DGKH	Diacylglycerol Kinase Eta
6	MIMAT0000437	hsa-miR-145-5p	TRIM2	Tripartite Motif Containing 2
7	MIMAT0000461	hsa-miR-195-5p	CCNE1	Cyclin E1
8	MIMAT0000755	hsa-miR-323a-3p	APPL1	Adaptor Protein, Phosphotyrosine Interacting With PH Domain And Leucine Zipper 1
9	MIMAT0001629	hsa-miR-329-3p	SLC44A1	Solute Carrier Family 44 Member 1
10	MIMAT0026483	hsa-miR-370-5p	TNRC6B	Trinucleotide Repeat Containing Adaptor 6B
11	MIMAT0004690	hsa-miR-379-3p	IGF1R	Insulin Like Growth Factor 1 Receptor
12	MIMAT0000736	hsa-miR-381-3p	ZFPM2	Zinc Finger Protein, FOG Family Member 2
13	MIMAT0002171	hsa-miR-410-3p	ETS1	ETS Proto-Oncogene 1, Transcription Factor
14	MIMAT0004757	hsa-miR-431-3p	INTS1	Integrator Complex Subunit 1
15	MIMAT0019047	hsa-miR-4510	MAX	MYC Associated Factor X
16	MIMAT0019693	hsa-miR-4636	KLC1	Kinesin Light Chain 1
17	MIMAT0003180	hsa-miR-487b-3p	GLCCI1	Glucocorticoid Induced 1
18	MIMAT0002813	hsa-miR-493-5p	ANK2	Ankyrin 2
19	MIMAT0002820	hsa-miR-497-5p	FGF2	Fibroblast Growth Factor 2
20	MIMAT0002875	hsa-miR-504-5p	UBE2I	Ubiquitin Conjugating Enzyme E2 I
21	MIMAT0003340	hsa-miR-542-5p	SGCZ	Sarcoglycan Zeta
22	MIMAT0031175	hsa-miR-548ba	ACBD3	Acyl-Coa Binding Domain Containing 3
23	MIMAT0025470	hsa-miR-6507-5p	XIAP	X-Linked Inhibitor Of Apoptosis
24	MIMAT0022717	hsa-miR-873-3p	PBX1	PBX Homeobox 1
25	MIMAT0004511	hsa-miR-99a-3p	GSK3B	Glycogen Synthase Kinase 3 Beta

[56], hsa-miR-204-5p [61,69], hsa-miR-487b-3p [64], hsa-miR-1247-5p, hsa-miR-370-5p [56]), hsa-let-7c-5p [68]), hsa-miR-409-5p [24], hsa-miR-432-5p [62], hsa-miR-503-5p [65]), hsa-miR-100-5p (21897839 [62,63]), hsa-miR-199b-3p [62]), hsa-let-7c-3p [66], hsa-miR-214-3p [56] are underexpressed both in our data as well as other data sets. There are also few miRNA which show opposite expression patterns in our study as compared to others. This could be because of differences in the detection methods as well as the kind of controls used (reviewed in Ref. [52].

Although some of these studies are similar to our study, they have analyzed patients from one particular region. Our data represents population of a different ethnicity, which is not very well represented in other studies including TCGA.

The major limitation of our study is that the profiling has been done on mainly one sub-type of endometrial cancer as identified by histopathology. Also, the samples collected here are prospective samples collected between 2018 and 2022 and hence we do not have survival data for these patients. However, the approach used here (summarized in Fig. 4), namely identifying the differentially expressed miRNAs and correlating to prognosis, also needs to be extended across other subtypes of endometrial cancer would be very valuable in deciding the best course of treatment for the patients. The 16 miRNA-mRNA pairs as identified by this study, if validated across a larger sample size has the potential to be developed into a diagnostic tool.

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Table 6
List of miRNA-mRNA pairs which could be potential biomarkers for prognosis.

Sl No	miRNA Accession ID	miRNA ID	Target mRNA	miRNA foldchange	mRNA foldchange
1	MIMAT0001620	hsa-miR-200a-5p	ZEB2	4.730269869	-2.779743371
2	MIMAT0004571	hsa-miR-200b-5p	CKS2	3.799513654	2.809840152
3	MIMAT0005797	hsa-miR-1301-3p	ANK2	2.656312442	-3.018816098
4	MIMAT0001541	hsa-miR-449a	LEF1	2.602895724	-0.235760038
5	MIMAT0019208	hsa-miR-3074-5p	THBS1	2.515021136	-2.783713258
6	MIMAT0004902	hsa-miR-891a-5p	EN1	2.417101688	1.34772381
7	MIMAT0003260	hsa-miR-592	PIK3CA	2.209988792	-0.279831429
8	MIMAT0004983	hsa-miR-940	IGF1	2.154979634	-2.854605114
9	MIMAT0010251	hsa-miR-449c-5p	CCNG1	2.067851364	-0.676169129
10	MIMAT0019880	hsa-miR-4746-5p	LMO2	2.028831966	-0.824667045
11	MIMAT0004555	hsa-miR-10a-3p	PTEN	1.942551751	-0.911062689
12	MIMAT0004518	hsa-miR-16-2-3p	CDK6	1.905243879	-0.941919508
13	MIMAT0017992	hsa-miR-3614-5p	MLH1	1.814518867	-0.826159659
14	MIMAT0004945	hsa-miR-744-5p	ATXN2	1.705368231	-0.139092803
15	MIMAT0004980	hsa-miR-937-3p	PTGFRN	1.623371751	-0.364245265
16	MIMAT0005458	hsa-miR-1224-5p	SIRT6	1.606135527	0.516670076
17	MIMAT0005577	hsa-miR-1226-3p	NRAS	1.325461708	0.844986364
18	MIMAT0004957	hsa-miR-760	HIST2H3C	1.226040877	1.664180411
19	MIMAT0002868	hsa-miR-522-3p	PDK1	0.93814227	1.084155871
20	MIMAT0004907	hsa-miR-892a	ZEB1	0.942158295	-3.605897727
21	MIMAT0005584	hsa-miR-1229-3p	CCNT1	0.770005091	-0.691964985
22	MIMAT0004929	hsa-miR-190b-5p	TNRC6B	0.733345776	-0.409451515
23	MIMAT0025450	hsa-miR-6499-5p	ARHGAP23	0.685171007	-1.784235985
24	MIMAT0031177	hsa-miR-7974	CREB3L3	0.620317931	-0.075698693
25	MIMAT0018120	hsa-miR-3691-5p	RASGRP2	0.604429657	-2.89991875
26	MIMAT0004950	hsa-miR-877-3p	MMP16	0.547782614	-1.81678469
27	MIMAT0004811	hsa-miR-33b-3p	PTGER3	0.517516099	-5.797349415
28	MIMAT0005930	hsa-miR-1276	DDX5	0.512369898	-0.245766856
29	MIMAT0019201	hsa-miR-3127-3p	WNT5A	0.431996383	-1.198701894
30	MIMAT0015053	hsa-miR-3176	BCL6	0.43234325	-0.833138447
31	MIMAT0002883	hsa-miR-514a-3p	RBX1	0.42044708	0.529799621
32	MIMAT0015054	hsa-miR-3177-3p	PRKCG	0.328884247	0.600708891
33	MIMAT0027467	hsa-miR-6783-3p	ITGA5	0.313200902	-0.825731818
34	MIMAT0028125	hsa-miR-7114-5p	RUNX1T1	0.295414768	-4.213303962

Sl No	miRNA Accession iD	miRNA ID	Target mRNA	miRNA foldchange	mRNA foldchange
1	MIMAT0022721	hsa-miR-1247-3p	ALKBH4	-2.957314443	0.223219508
2	MIMAT0000437	hsa-miR-145-5p	TRIM2	-2.569189935	0.321045455
3	MIMAT0004601	hsa-miR-145-3p	DGKH	-2.515285219	0.046150568
4	MIMAT0000736	hsa-miR-381-3p	ZFPM2	-2.064472846	-5.050758712
5	MIMAT0005899	hsa-miR-1247-5p	SNX14	-1.826285517	0.266484091
6	MIMAT0000461	hsa-miR-195-5p	CCNE1	-1.708796623	4.0485625
7	MIMAT0003340	hsa-miR-542-5p	SGCZ	-1.48100984	-1.359653846
8	MIMAT0000423	hsa-miR-125b-5p	MFHAS1	-1.444726998	0.275103598
9	MIMAT0004597	hsa-miR-140-3p	CBL	-1.415767178	-0.616743371
10	MIMAT0004511	hsa-miR-99a-3p	GSK3B	-1.005905322	0.261000947
11	MIMAT0002820	hsa-miR-497-5p	FGF2	-0.846226612	-4.195162689
12	MIMAT0002171	hsa-miR-410-3p	ETS1	-0.699074669	-1.377860038
13	MIMAT0026483	hsa-miR-370-5p	TNRC6B	-0.590254896	-0.409451515
14	MIMAT0003180	hsa-miR-487b-3p	GLCCI1	-0.469981718	-0.11034053
15	MIMAT0004757	hsa-miR-431-3p	INTS1	-0.218727339	0.402824053
16	MIMAT0025470	hsa-miR-6507-5p	XIAP	-0.121791708	-0.17249053
17	MIMAT0004690	hsa-miR-379-3p	IGF1R	-0.059253436	-1.15964697
18	MIMAT0001629	hsa-miR-329-3p	SLC44A1	-0.057402947	0.058539583
19	MIMAT0019047	hsa-miR-4510	MAX	-0.051697652	-0.585193561
20	MIMAT0002813	hsa-miR-493-5p	ANK2	0.01375935	-3.018816098
21	MIMAT0031175	hsa-miR-548ba	ACBD3	0.015311909	-0.043739394
22	MIMAT0019693	hsa-miR-4636	KLC1	0.148741073	-0.403395265
23	MIMAT0022717	hsa-miR-873-3p	PBX1	0.16217544	-2.082156629
24	MIMAT0002875	hsa-miR-504-5p	UBE2I	0.288675864	0.094574432
25	MIMAT0000755	hsa-miR-323a-3p	APPL1	0.306883982	-0.559366667

Note: The table shows the log2 fold changes of the miRNA and the target mRNA according to TCGA data. The highlighted cells are the pairs which show inverse correlation between miRNA and the corresponding mRNA target.
Note: Only those miRNAs which show similar pattern of expression in Indian data and TCGA data have been considered.

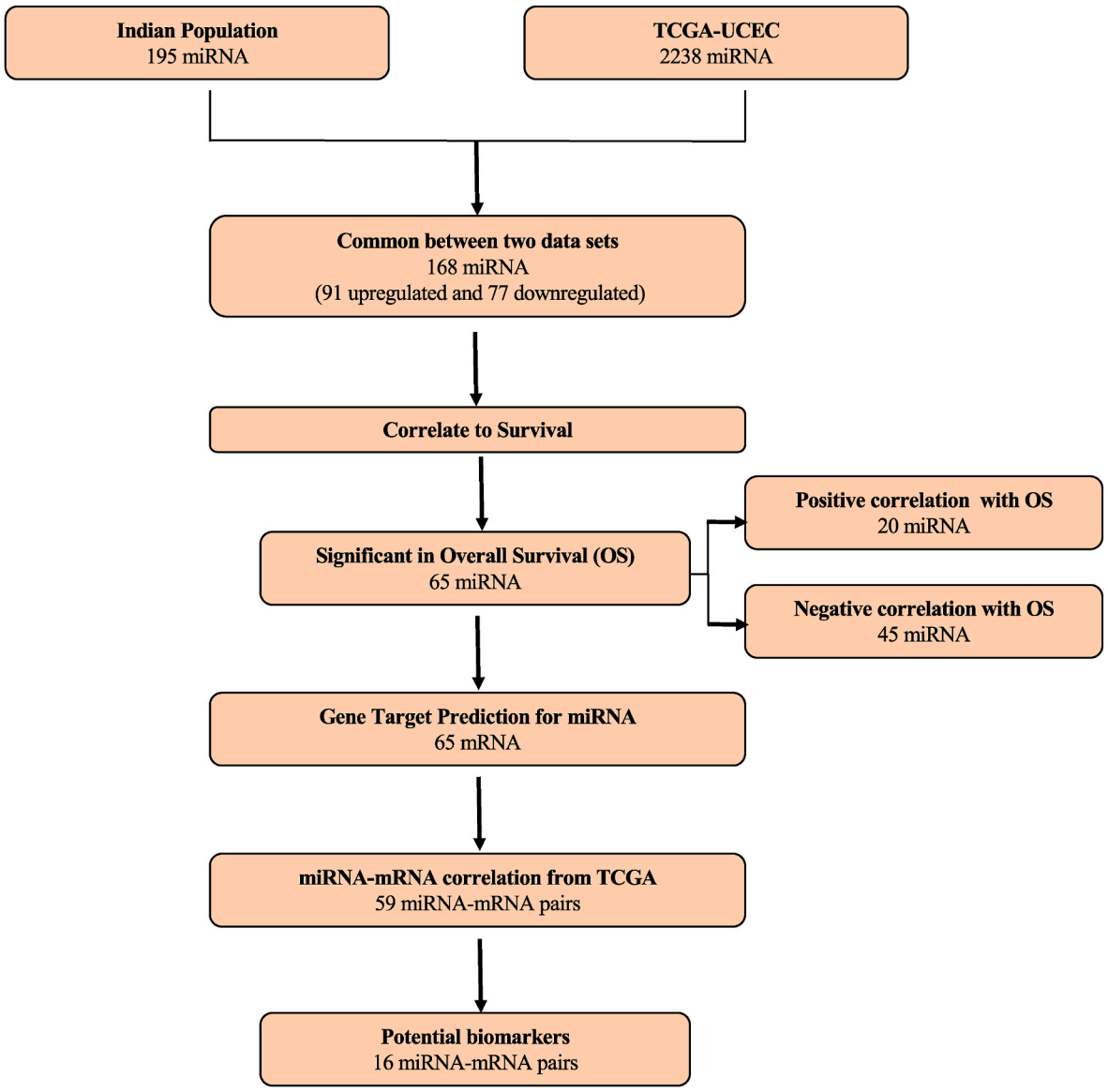

Fig. 4. Schematic work flow for identification of potential biomarkers for endometrial cancer.

CRediT authorship contribution statement

Shraddha Hegde: Writing – original draft, Methodology, Formal analysis. **Kalpesh Wagh:** Writing – original draft, Methodology, Formal analysis. **Suma Mysore Narayana:** Writing – review & editing, Resources, Funding acquisition, Conceptualization. **Apoorva Abikar:** Writing – original draft, Formal analysis. **Sughosha Nambiar:** Resources, Methodology. **Shriraksha Ananthamurthy:** Resources, Methodology. **Navyashree Hosahalli Narayana:** Resources, Methodology. **Pallavi Venkateshaiah Reddihalli:** Resources. **Savitha Chandraiah:** Resources. **Prathibha Ranganathan:** Writing – review & editing, Writing – original draft, Methodology, Investigation, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The raw data is submitted to GEO and the details shared in the manuscript

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.bbrep.2024.101812.

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