

## Prostate-Specific antigen value and micro RNAs as potential diagnostic biomarkers for prostate cancer

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### ABSTRACT

**Objective:** It is necessary to provide PSA alternatives or methods that can be used in conjunction with PSA to regress complications rising from negative biopsies and to increase diagnostic value.

**Patients and Methods:** The study is consisting of 59 men as the sample group. Blood samples from the individuals are grouped as prostate cancer and BPH (benign prostatic hyperplasia) groups. 27 prostate cancer patients whom some of them also operated are assembled in the patients group and the other 32 individuals are grouped as BPH group. Micro RNA expression levels evaluated by RT-PCR.

**Results:** Prostate cancer group when compared with the control group, it is observed that expression levels of miRNA-221 and miRNA-432 increased while expression levels of miRNA-17-5p, miRNA-30c, miRNA-107, miRNA-141, miRNA-145, miRNA-181a-2, miRNA-331-3p, miRNA-574-3p decreased and expression levels of miRNA-21 and miRNA-375 are quite similar between the groups.

**Conclusion:** The prospect of strong and sensitive serum miRNA expression levels in prostate cancer cases which are easily detectable by non-invasive methods as biomarkers is a promising field of study. Nevertheless, it is currently necessary to work in conjunction with both tissue and serum to enhance both sensitivity and specificity of miRNAs as biomarkers. As such, expression levels of the same miRNAs in tissue and serum provide different expression values which in turn make it difficult to indicate a common biomarker.

**Keywords:** Biomarker; miRNA; cancer; BPH; prostate.

### INTRODUCTION

Prostate cancer is the second most commonly diagnosed malignancy in men and is the second leading cause of cancer death (1). The clinical behavior of prostate cancer ranges from a microscopic, well-differentiated tumor that may never be clinically significant to an aggressive, high-grade cancer that ultimately causes metastases, morbidity, and death. Clinically, prostate cancer is diagnosed with a transrectal ultrasound-guided prostate biopsy (TRUS) because of a suspicious serum prostate-specific antigen (PSA) and /or abnormal digital rectal examination findings usually used biomarker for the detection of prostate cancer, is limited by its lack of sensitivity and specificity for prostate cancer because benign prostatic hyperplasia (BPH), prostatic inflammation and infection may cause elevated PSA, therefore not considered an ideal biomarker. Additionally prostate cancer screening and prevention trials, such as The Prostate, Lung, Colorectal, and Ovarian cancer screening trial (PLCO), the European Randomised Study of Screening for Prostate cancer (ERSPC) trial, Prostate Cancer Prevention Trial (PCPT) and Reduction by Dutasteride of Prostate Cancer Events (REDUCE) trial have highlighted that despite an increase in the diagnosis of prostate cancer using PSA and DRE (digital rectal examination), there is still no clear improvement in mortality (2).

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To reduce unnecessary biopsies and to improve the effect of screening modalities on mortality of prostate cancer are major goals of researchers. As a result, a search for a novel, minimally invasive, clinically relevant biomarkers for the detection of prostate cancer is required. MiRNAs are ~20–22 nucleotide long noncoding RNAs, which regulate gene expression at the post-transcriptional level by mRNA repression and/or degradation. It is becoming clear that miRNAs represent a vast, previously unrecognized layer of molecular signaling in eukaryotes, and that miRNAs play an important role in the regulation of protein expression (3). Several studies have investigated miRNAs as possible diagnostic or prognostic biomarkers for malignancies and other diseases. The experimental over-expression or inhibition of specific miRNAs in cellular and animal models can result in tumorigenesis, or in more aggressive cancer phenotypes, indicating the potential of miRNAs to function as oncogenes and tumor suppressors (4). Many miRNAs are known to be dysregulated in prostate cancer (5). For instance, miRNAs are differentially expressed between benign prostate and prostate cancer tissues: in a genome-wide microarray-based microRNA expression study, 25 miRNAs were found to be deregulated (6). Some of the miRNAs are dysregulated in prostate cancer tissue and some of the miRNAs are dysregulated detectable in the circulation of patients with prostate cancer (7). Consequently miRNAs may be novel, useful, stable, non-invasive biomarkers. This study aimed to assess and compare the levels of specific miRNA between men with prostate cancer and BPH.

## MATERIAL AND METHODS

In our study, 2 groups were formed, including the BPH group (control group) of 32 individuals without prostate cancer and the prostate cancer group of 27 individuals with prostate cancer patients whom some of them also operated. The descriptive data as PSA, prostate volume (PV), age, body mass index (BMI), and Gleason score are collected from the individuals in this study. All the individuals are diagnosed by Cumhuriyet University Medical School Application and Expertise Hospital Urology Department between dates June 2014 and November 2014. Ethical approval has been granted before study by Medical School Ethical Commission (Date of Approval: 24.12.2013, Approval Number: 2013-12/16).

### Isolation of Serum Samples

Blood samples taken into 3cc biochemical tubes are centrifuged by 10 min. at 4000 rpm. Serum portions of samples are taken into sterile 2 ml Eppendorf tubes and centrifuged under 100C cooling by 5 min. centrifuge at 10000 rpm. Supernatant taken into sterile 2 ml Eppendorf tubes and second high-speed centrifugation is done to remove the remaining cell remnants. The resulting serum samples are taken into sterile micro centrifuge tubes and stored at -800C until RNA extraction (8).

### RNA Isolation and Reverse Transcription (RT) Reaction

Total RNA extracts from serum samples were obtained by following the manufacturer's directives using miRNeasy Mini Kit (Qiagen, cat. no: 217004). Total RNA samples are converted to cDNA by using Qiagen MiScript reverse transcription kit (Qiagen, Cat. No: 218161). cDNA samples are stored under -800C until PCR. 125 to 250 ng total RNA is

used as starter for miRNA panel. For inactivation of MiScript Reverse Transcriptase, hold at 950C for 5 min. and 200 µl water is added then protocol is followed (8).

### Pre-amplification of cDNA and qPCR

Twelve different miRNAs (hsa-miR-17-5p, hsa-miR-21-3p, hsa-miR-30c-1-3p, hsa-miR-107, hsa-miR-141-5p, hsa-miR-145-3p, hsa-miR-181a-2-3p, hsa-miR-221-5p, hsa-miR-331-3p, hsa-miR-375, hsa-miR-432-3p, and hsa-miR-574-3p) are selected for the study. These sequences are formed into a panel by a commercial service and bought as qPCR kit (Qiagen). In addition to pre-emptive 12 miRNAs, 4 control miRNA wells are used in custom miScript miRNA PCR Array (Qiagen, cat. no: 218300). For pre-amplification purposes miScript SYBR Green PCR kit is used. 2 µl cDNA sample is transferred into a clean well on 96-well plate and 9 µl DNA suspension buffer is added onto the well which then reacts thoroughly mixed. The 1/5 diluted 2 µl RT sample is pre-amplified in a PCR tube using 2x quantiTec SYBR Green master mix and 10x miScript Universal Primers. Real-Time PCR plate formation includes selected 12 miRNAs with specific forward-reverse primers for each and 4 universal control primers (Applied Biosystems). All the conditions are optimized in accordance with manufacturer directives to cover all samples (8).

### Statistical Analysis

Obtained data uploaded into SPSS v.22.0 and analyzed. When parametrical test defaults are met Kolmogorov–Simirnov significance test of the difference between the two average is concluded, whereas when parametrical test defaults are not met, Mann Whitney U Test and Correlation Analyses are concluded. The error rate for analyses is chosen as 0.05 (P value).

## RESULTS

In this study, a total of 59 patients of 27 prostate cancer and 32 BPH patients were contrasted by both clinical and genetic data using 12 miRNA regions compiled from the literature. The mean age was  $65.35 \pm 9.01$  for prostate cancer patients and  $64.29 \pm 4.61$  for BPH patients, of which age contrast between groups is insignificant ( $P > 0.05$ ). Gleason scores of patients with prostate cancer range between 6 to 9 and the respective average is  $7.85 \pm 0.948$ . Also, some metastasis conditions were observed in the different individuals within the prostate cancer group. Indeed 19 patients with prostate cancer (70.4%) showed no metastasis whereas 8 patients (29.6%) have shown metastasis to the bone of which 6 patients (22.2%) with single point metastasis and 2 patients (7.4%) with multiple point metastasis. The expression level of miRNA-30c is found to be the lowest value in both groups. The highest expression levels are obtained from miRNA-221 ( $7.459 \pm 3.887$ ) value in patients with prostate cancer group and miRNA-181a-2 value ( $9.711 \pm 14.604$ ) in BPH patients group. There are statistically significant differences between the groups according to the expression levels of the different miRNAs except for miRNA-21 and miRNA-375 levels (Table 1). It is observed that the expression levels of miRNA-17-5p, miRNA-30c, miRNA-107, miRNA-141, miRNA-145, miRNA-181a-2, miRNA-331-3p, and miRNA-574-3p are found to be lower in the patients with prostate cancer than the individuals with BPH.

Table 1. Comparing the groups according to miRNA expression value (\*: P&lt;0.05, miR: miRNA: micro RNA)

miRNA name	Prostate Cancer			Benign Prostatic Hyperplasia			Statistical Value
	Min	Max	Mean±SD	Min	Max	Mean±SD	P
miR17-5p	0.404	15.616	2.281±2.866	0.125	17.267	3.418±3.601	<b>0.034*</b>
miR21	0.062	4.723	1.608±1.167	0.059	7.412	1.632±1.397	0.922
miR30c	0.003	0.027	0.007±0.005	0.004	0.56	0.023±0.014	<b>0.001*</b>
miR107	0.188	24.16	3.599±5.553	0.079	34.775	6.495±7.178	<b>0.008*</b>
miR141	0.061	4.084	0.139±0.941	0.014	3.797	1.315±1.007	<b>0.027*</b>
miR145	0.034	4.69	0.835±0.956	0.015	3.810	1.279±0.889	<b>0.007*</b>
miR181a-2	0.142	11.004	2.896±2.762	0.126	68.119	9.711±14.604	<b>0.009*</b>
miR221	0.208	16.564	7.459±3.887	0.003	22.471	3.109±4.405	<b>0.050*</b>
miR331-3p	0.639	15.030	2.298±4.193	0.096	12.728	5.423±3.491	<b>0.047*</b>
miR375	0.360	7.972	2.441±1.914	0.062	10.966	2.864±2.848	0.814
miR432	0.088	8.027	4.765±1.885	0.008	2.445	0.799±0.648	<b>0.046*</b>
miR574-3p	0.213	4.469	1.035±1.098	0.049	11.004	3.508±2.575	<b>0.034*</b>

Table 2. Comparing PSA value and prostate volume between the groups (\*: P&lt;0.05, PSA: prostate-specific antigen, PV: prostate volume)

	Prostate Cancer			Benign Prostatic Hyperplasia			Statistical value
	Min	Max	Mean±SD	Min	Max	Mean±SD	P
PSA (ng/ml)	5.43	1778.2	111.52±335.05	3.89	19.48	8.06±3.70	<b>0.001*</b>
PV (ml)	17.00	136.0	52.77±34.80	20.00	138.00	65.61±30.42	<b>0.032*</b>

However, expression levels of miRNA-221 and miRNA-432 are found to be higher in the individuals with prostate cancer. Nevertheless, these three miRNA (miRNA-221, miR-331-3p and miRNA-432) expression levels have a low significance (P=0.05; P=0.047 and P=0.046 respectively) compared to others (Table 1). The average of PSA values are found as 111.52 ± 335.05 ng/mL in prostate cancer patients and 8.06 ± 3.70 ng/mL in BPH group. PSA values in the prostate cancer group were statistically higher than the BPH (control) group values (Table 2).

The difference about PSA values between the two groups was found to be statistically significant (P<0.05) (Table 2). Prostate volume (PV) values are 52.77 ± 34.80 mL in the cancer patients group and 65.62 ± 30.42 mL in the BPH group. PV values of the BPH group were higher than the prostate cancer patients group (Table 2). In terms of PV comparison, the difference between both groups was found to be statistically significant (P<0.05) (Table 2). Prostate volume values and PSA values are usually used for the cancer diagnostic and these are the important data for determining the prostate cancer. Therefore, finding a relationship between PSA and PV values and any miRNA values will be important in terms of finding a marker that can be used practically in the diagnosis of prostate cancer (Table 3). There was a positive correlation between PSA and PV values in prostate cancer group (correlation coefficient: 0.393) and this correlation is found to be statistically significant (P<0.05, Table 3).

However, no correlation was found between PSA and PV values in the BPH group (Table 4). Also, a significant positive correlation was found between PSA values and miR-432 in the prostate cancer group (correlation coefficient: 0.435).

In addition a positive correlation was found between PV values and miR-30c expression level in this group (correlation score: 0.502). On the other hand, a negative correlation is observed between Gleason score and miRNA-30c with the correlation coefficient -0.387 and a positive correlation is observed between Gleason score and miRNA-574-3p with the 0.464 correlation coefficients (Table 3). Even though these correlations are statistically significant, their relationship values are found to be weak.

Additionally there are no general correlation between Gleason score, PSA and PV values and other miRNAs' expression levels (Table 3). Among the 12 miRNAs we investigated in our study, serious correlations were detected between some micro RNAs. These results have shown that no direct relation is present between PSA and respective miRNA expressions. There is a weak positive correlation observed between PV and miRNA-30c expression (correlation score: 0.502) but no correlation has been seen for other miRNAs. Detailed correlation analyses between miRNAs are shown in Table 3.

Two-way correlation analyses were conducted on the individuals consisting BPH using PSA, PV and miRNA expression and no correlation has been observed between PSA and PV in this group. While PSA and miRNA-145 provided a positive and weak correlation (correlation coefficient: 0.391). There is another positive and weak correlation found between PV and miRNA-30c (correlation coefficient: 0.481), but no correlation has been observed for other miRNAs. Detailed correlation analyses between miRNAs are shown in Table 4. Also when we made 2 groups according to PSA value under and over 10; just miRNA-30c is statistically significant but has a weak correlation (P: 0,092; P<0,10)

**Table 3.** The correlation analysis between Gleason score, miRNA expression level, PSA, and prostate volume inside the prostate cancer group. (miR: miRNA; micro RNA, PSA: prostate-specific antigen, PV: prostate volume)

	Correlation Value	Gleason	miR 17-5p	miR 21	MiR 181a-2	miR 30c	miR 145	miR 107	miR 141	miR 331-3p	miR 574-3p	miR 432	miR 221	miR 375	PSA	PV
Gleason	1	0.122	1	0.122	-0.25	-0.387*	0.309	0.101	-0.062	0.078	0.464*	-0.08	0.023	0.052	0.25	0.351
miR17-5p	0.122	1	<b>0.401*</b>	<b>0.648**</b>	-0.031	-0.034	<b>0.840**</b>	0.258	<b>0.618**</b>	0.163	0.098	-0.09	-0.11	-0.05	-0.26	-0.26
miR21	-0.254	<b>0.401*</b>	1	0.289	0.359	0.025	<b>0.624**</b>	<b>0.488*</b>	<b>0.722**</b>	0.082	<b>0.602**</b>	-0.09	0.136	0.091	0.119	0.119
miR181a-2	-0.001	<b>0.648**</b>	0.29	1	-0.043	0.008	<b>0.668**</b>	0.243	<b>0.675**</b>	0.069	-0.04	0.16	0.073	-0.04	-0.2	-0.2
miR30c	<b>-0.387**</b>	-0.03	0.36	-0.04	1	0.075	0.297	0.204	0.095	-0.14	0.029	-0.13	0.217	-0.06	<b>0.502**</b>	<b>0.502**</b>
miR145	0.309	-0.03	0.03	0.008	0.075	1	-0.1	0.328	0.014	0.349	-0.17	0.018	<b>0.703**</b>	0.049	0.143	0.143
miR 107	0.101	<b>0.840**</b>	<b>0.624**</b>	<b>0.668**</b>	<b>0.297</b>	-0.103	1	0.292	<b>0.760**</b>	0.024	0.093	-0.12	-0.21	-0.1	-0.1	-0.1
miR141	-0.062	0.258	<b>0.488*</b>	0.243	0.204	0.328	0.292	1	0.203	0.224	<b>0.408*</b>	-0.04	<b>0.482*</b>	-0.08	0.126	0.126
miR331-3p	0.078	<b>0.618**</b>	<b>0.722**</b>	<b>0.675**</b>	0.095	0.014	<b>0.760**</b>	0.203	1	0.219	0.031	-0.19	-0.24	-0.08	-0.3	-0.3
miR574-3p	<b>0.464**</b>	0.163	0.08	0.069	-0.136	0.349	0.024	0.224	0.219	1	0.087	0.008	<b>0.393*</b>	0.046	-0.15	-0.15
miR432	-0.08	0.98	<b>0.602**</b>	-0.04	0.029	-0.165	0.093	<b>0.408*</b>	0.031	0.087	1	-0.08	0.125	<b>0.435**</b>	-0.06	-0.06
miR221	0.023	-0.09	-0.09	0.16	-0.127	0.018	-0.12	-0.04	-0.194	0.008	-0.08	1	0.276	-0.11	0.252	0.252
miR375	0.052	-0.11	0.14	0.073	0.217	<b>0.703**</b>	-0.214	<b>0.482*</b>	-0.244	<b>0.393*</b>	0.125	0.276	1	-0.01	0.323	0.323
PSA	0.25	-0.05	0.09	-0.04	-0.061	0.049	-0.1	-0.081	-0.08	0.046	<b>0.435*</b>	-0.11	-0.01	1	<b>0.393*</b>	<b>0.393*</b>
PV	0.351	-0.26	0.12	-0.2	<b>0.502**</b>	0.143	-0.1	0.126	-0.302	-0.15	-0.06	0.252	0.323	<b>0.393*</b>	1	1

## DISCUSSION

It is known that various oncogenes and tumor-suppressing genes are taking place in prostate cancer and their respective expressions increase or decrease by epigenetic regulations (8, 9). Since the last decade, many studies have been conducted on miRNAs from the point that they may take place in regulations of cancer-related genes and many studies are still ongoing (10). Many studies on prostate cancer have shown that some of miRNAs exhibit increased expression whereas other miRNAs decreased in expression. It has been stated in some studies that certain miRNA expressions are correlated with Gleason scores and might be related to clinical recurrences (11). According to CAPRA scoring, patients with severe risk have provided increased expression levels for miRNA-20a and miRNA-21 while patients with mild to severe risks have provided increased expression levels for miRNA-21, miRNA-17-5p and miRNA-145 (12, 13). It is also pointed out that they can be used to differentiate patients with mild, medium and severe risks and to estimate the aggressiveness of prostate cancer (12). We have observed 35% depressed expression of miRNA-17-5p among prostate cancer patients compared to BPH patients. There is a negative correlation observed between PSA and miRNA-17-5p expression and no significant correlation with Gleason score. Also, it is showed increased expression of miRNA-181a-2 in both tumoral tissue and serum (14). It has been stated that many different miRNAs have shown increased expression in tumors with Gleason scores ranging 8 (4+4) to 9 (4+5) (17). Some of studies have pointed out that miRNA-181 is functioning in the differentiation of hemopoietic cells and might take place in the formation of leukemia and other solid tumors (15, 16). BPH patients in the present study have shown 60% increased expression for miRNA-181 which is in accordance with previous studies. For this miRNA, there is a negative and weak relationship observed. Nevertheless, some researchers have stated that miRNA-181a-2 expression has increased among patients with higher Gleason score (14) we have not observed such a significant correlation in the present study. A significant relationship has been reported between the expression of miRNA-375 and miRNA-141 in various studies conveying metastatic prostate cancer patients that exhibit lymph node metastasis and higher Gleason score (17). Expression of miRNA-375 is also increased in breast cancer and since these two cancer kinds are similar in endocrinological and physiopathological aspects it has been stated that this miRNA is an oncogene for both cancers (18). Expression levels of miRNA-375 in the present study are found similar to each other. Additionally, no significant relationship has been observed for this miRNA expression with PSA and Gleason score in our study. It has been shown that miRNA-331-3p in prostate cancer cells reduces PSA expression by inhibiting androgen-sensitive promoter region of PSA gene which in turn results in lower expression in cancer cells compared to normal cells (19). Expression of miRNA-331-3p has been observed 60% reduced in prostate cancer patients compared to BPH patients in our present study which is in accordance with previous studies. Also there is a negative weak correlation observed between miRNA-331-3p expression and PSA levels. Some studies have reported diffuse reduced regulation of miRNAs in prostate cancer tissues (20). Expression levels of miRNAs are quite variable

in tumoral tissue and serum and therefore differential diagnosis of these values improves findings. Tissue expression levels of miRNA-21 have shown an increase in many kinds of cancer including prostate cancer (21). Expression levels of miRNA-21 in the present study for both prostate cancer patients and BPH patients groups are similar. Also, there is a positive but weak correlation observed between miRNA-21 and PSA (Table 3). The contrast observed to other studies might be due to the expression of miRNA-21 is evaluated only in serum in the present study. Increased expression of miRNA-221 in prostate cancer has been reported (20). In contrast, some other studies indicated that miRNAs might be oncomirs and progressive reduction in expression of miRNA-221 in aggressive prostate cancer is possibly due to Gleason score, progression level, metastasis, and clinical recurrence (22). In present study, expression of miRNA-221 in prostate cancer patients was observed 2 times increased compared to BPH patients, and the expression difference is found to be significant ( $P < 0.05$ ) (Table 1). Also, there is a negative and weak correlation observed between miRNA-221 and PSA. MiRNAs which secreted from tumoral tissue into the blood stream can be used as biomarkers both in serum and plasma samples. Increased regulation of miRNA-141 in metastatic prostate cancer patients is also reported (23). It has been reported that miRNA-141 and miRNA-375 can be used as circulatory markers for metastatic prostate cancer patients both in serum and plasma (24). Conversely, in the present study such increase is not present and 85% actual depression in the expression of miRNA-141 in prostate cancer patients observed compared to BPH patients ( $P < 0.05$ ). Expression of miRNA-145 in the present study for prostate cancer patients observed as 30% reduced compared to BPH patients ( $P < 0.05$ ; Table 1) and showed a positive yet weak correlation with PSA. In another study where the evaluation of serum/plasma levels of different miRNAs in prostate cancer patients has shown reduced regulation of miRNA-30c (25). Another study has indicated that one of the 34 miRNAs with increased expression in prostate cancer tissue is miRNA-30c (26). For prostate cancer patients in the present study miRNA-30c exhibit, 70% reduced expression and provide a negative weak correlation with PSA. Also, a negative correlation between miRNA-30c and Gleason score and a positive correlation between miRNA-30c and PV are observed in our present study. Various studies have pointed out that miRNA-107 and miRNA-574-3p are being prominent among miRNAs with increased expression in prostate cancer (27). Also, it is reported that the expression of miRNA-574-3p is greatly depressed in prostate tumoral tissue and this depression is found to be correlated with both Gleason score and progressed tumoral state (28). In our study, the expression level of miRNA-107 is found to be 45% lower in prostate cancer patients compared to BPH patients, and also a negative and very weak correlation with PSA observed (correlation coefficient: -0.1). In additionally the expression level of miRNA-574-3p in prostate cancer patients is obtained with a depressed value with 70% ratio compared to BPH patients and provided a positive weak correlation with PSA (correlation coefficient: 0.046) and a strongly positive correlation with Gleason score (correlation coefficient: 0.464). On the other hand, the expression level of miRNA-432 in prostate cancer patients observed as 85% increase compared to BPH patients and provided a positive strong

correlation with PSA (correlation coefficient: 0.435). Notably there is not much report on the relationship of this miRNA with prostate cancer. Porkka et al. have conducted a study using 319 different miRNAs and have reported that 51 of these miRNAs exhibited either increased or decreased expression in cancerous tissue compared to normal tissue. Also indicated that reduced expression is observed for 22 of these 51 miRNAs in all prostate cancer cases while 15 of them only in hormone-resistant cancers. In another study, it has been reported that 8 miRNAs are found to be increased in expression for all prostate cancer cases while 6 miRNAs are found to be increased in expression only in hormone-resistant prostate cancer cases (9). Upregulation of miRNA expression levels in prostate cancer cases has been generally supported by different comparative studies. In the present study, it is observed that expression levels of miRNA-221 and miRNA-432 are increased whereas expression levels of miRNA-17-5p, miRNA-30c, miRNA-107, miRNA-141, miRNA-145, miRNA181-a2, miRNA-331-3p, miRNA-574-3p are decreased for prostate cancer patients contrasted to BPH patients. Expression levels of miRNA-21 and miRNA-375 are found to be similar for both groups. As a summary, it is observed that of 12 miRNAs selected for our present study expression levels of 2 miRNAs (17%) observed as similar, another 2 miRNAs (17%) observed as increased and the remaining 8 miRNAs (66%) observed as decreased for prostate cancer patients compared to BPH patients (Table 1).

## CONCLUSION

The prospect of strong and sensitive serum miRNA expression levels in prostate cancer cases which are easily detectable by non-invasive methods as biomarkers is a promising field of study. Nevertheless, it is currently necessary to work in conjunction with both tissue and serum in order to enhance both sensitivity and specificity of miRNAs as biomarkers. As such, expression levels of the same miRNAs in tissue and serum provide different expression values which in turn makes it difficult to indicate a common biomarker. Within the scope of this research, it was possible to work with a small sample group and the expression level of only 12 miRNAs was investigated. Although these are restrictive, our results are still informative and guiding for scientists working in this field. Therefore studies conveying comparative analysis of miRNAs in prostate cancer cases, BPH patients, and healthy individuals would hopefully provide determinative results into the subject for the future.

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