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Altered Expression Levels of MicroRNA-132 and Nurr1 in Peripheral Blood of Parkinson's Disease: Potential Disease Biomarkers

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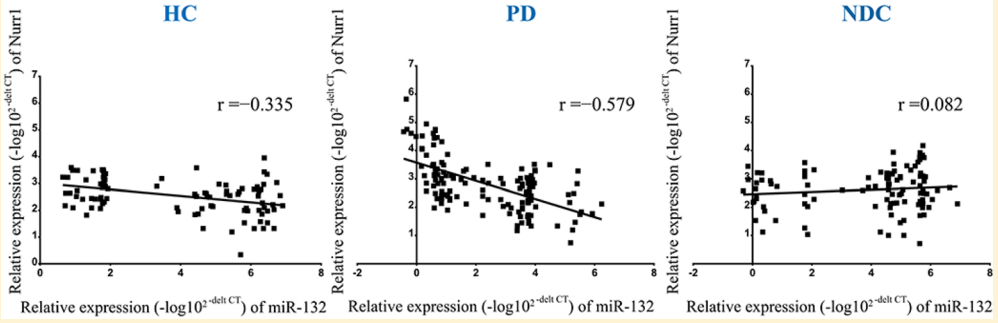
Supporting Information



ABSTRACT: MicroRNAs (miRNAs) are small and evolutionary conserved noncoding RNAs that are involved in post-transcriptional gene regulation. Differential expression levels of miRNAs can be used as potential biomarkers of disease. Previous animal studies have indicated that the expression level of miR-132 is negatively correlated with its downstream molecule nuclear receptor related 1 protein (Nurr1), which is one of the key factors for the maintenance of dopaminergic function and is particularly vulnerable in Parkinson's disease (PD). However, this correlation has not been confirmed in human patients with PD. Moreover, the possible involvement of miR-132 during the pathogenesis and progression of PD is not fully investigated. Therefore, in the present study, we determined the peripheral circulation levels of miR-132 and Nurr1 in patients with PD, neurological disease controls (NDC) and healthy controls (HC) by reverse transcription real-time quantitative PCR (RT-qPCR). Our data clearly demonstrated that the plasma miR-132 level in PD was significantly higher than those in HC (178%, $p < 0.05$) and NDC (188%, $p < 0.001$). When adjusted for gender and age, higher level of miR-132 expression was associated with the significantly increased risk for PD in males and was closely related with the disease stages and disease severity. Furthermore, peripheral Nurr1 was significantly decreased in PD compared with HC (56%, $p < 0.001$) and NDC (58%, $p < 0.001$). Much more interestingly, further analysis revealed a negative correlation between the decreased Nurr1 level and the elevated miR-132 level in PD. All these findings indicated that the combination of a high miR-132 level with the low level of its downstream Nurr1 might be a potential biomarker aiding in the diagnosis of PD and monitoring disease progression.

KEYWORDS: *Parkinson's disease, miR-132, Nurr1, biomarker, peripheral blood*

INTRODUCTION

Parkinson's disease (PD) is the second most common neurodegenerative disease in the world,[1,2] pathologically characterized by the selective loss of dopaminergic (DAergic) neurons in substantia nigra (SN) and the formation of Lewy bodies (LB).[3] Although great achievements have been made to clarify the disease pathogenesis and explore potent therapeutic strategies against PD for over 200 years, there is still no systematic understanding of this disease and the current treatments are mostly symptomatic.[4] Moreover, the current diagnosis of PD is based primarily on subjective clinical rating of motor functions. By the time of diagnosis, over 60% of

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Table 1. Demographic Characteristics of Subjects Enrolled in the miR-132 Study[a]

groups	number (%)	gender male/ female	P value	P value	age (years) (mean ± SEM)	P value	P value
HC	222 (33.3)	130:92	ref		66.16 ± 0.61	ref	
PD	269 (40.3)	157:112	NS	ref	66.10 ± 0.61	NS	ref
NDC	176 (26.4)	105:71	NS	NS	66.15 ± 0.74	NS	NS

[a] χ^2 test and Kruskal–Wallis test. ref, reference; NS, not significant; HC, healthy controls; PD, Parkinson's disease; NDC, neurological disease control.

Table 2. Demographic Characteristics of Subjects Enrolled in the Nurr1/miR-132 Correlation Study[a]

groups	number (%)	gender male/female	*P* value	*P* value	age (years) (mean ± SEM)	*P* value	*P* value
HC	98 (28.4)	54:44	ref		66.87 ± 0.91	ref	
PD	142 (41.2)	79:63	NS	ref	67.19 ± 0.75	NS	ref
NDC	105 (30.4)	56:49	NS	NS	67.44 ± 1.12	NS	NS

[a] χ^2-square test and Kruskal–Wallis test. ref, reference; NS, not significant; HC, healthy controls; PD, Parkinson's disease; NDC, neurological disease control.

DAergic neurons in the SN of PD patients are already lost.[5] Since there is no reliable quantitative diagnostic test for PD, the objective and measurable molecular biomarkers should be critical to serve as potential clinical tool to facilitate PD diagnosis and early intervention.

MicroRNAs (miRNAs) are a class of ∼22 nt endogenous RNA molecules.[6] Mature miRNAs recognize and bind to the 3′ untranslated region of the message RNA (mRNA) by specific complementary sequence, and then affect the expression of target genes.[7] Several miRNAs have been found to play important roles in DAergic neuronal differentiation, apoptosis and protection of PD.[8,9] For instance, miR-133b can regulate DAergic function negatively and affect PD pathogenesis by repressing the expression of Pitx3 gene, a homeobox transcription factor required for the development and mature of DAergic neurons;[8] miR-7 and miR-153 protect DAergic neurons from oxidative stress damage via altering α-synuclein expression;[10,11] miR-433 induces overexpression of α-synuclein indirectly by binding to the promoter region of fibroblast growth factor 20,[12] while miR-128 suppresses the accumulation of α-synuclein by targeting to transcription factor EB;[13] miR-132 dysregulation is associated with the changes of momoamine oxidase A (MAO-A), D1- and D2-dopamine receptor (DRD1 and DRD2) genes in dopamine receptor signaling in *Toxoplasma gondii* infected mice.[14]

Among these above-mentioned miRNAs, we were particularly interested in miR-132, which have been reported to be highly expressed in neurons, and significantly altered in various neurodegenerative disorders.[15−18] The level of mesencephalon miR-132 was found to be significantly increased in rat PD model, accompanied by a decreased Nurr1.[19] Nurr1 is a transcription factor[20−22] that plays crucial roles in the differentiation and functional maintenance of midbrain DAergic neurons and the microglia-mediated neuroinflammation.[23,24] Our previous study demonstrated that overexpression of miR-132 repressed embryonic stem (ES) cells differentiation into DAergic neurons by directly targeting Nurr1 gene,[25] and Nurr1 expression level was decreased in the patients with PD compared to health individuals.[26,27] However, miR-132 expression level in PD patients and controls, and the possible correlation between peripheral levels of miR-132 and Nurr1 in PD patients has not been determined.

In the present study, we recruited 269 patients with diagnosed PD, 222 healthy controls (HC), and 176 patients with various non-PD neurological disorder controls (NDC). The main aims of the present study were to determine (1) whether peripheral miR-132 expression level was significantly altered in patients with PD as compared with controls; (2) whether age, gender, disease severity and medications affect miR-132 expression; (3) whether peripheral miR-132 had a negative correlation with Nurr1 in PD patients.

RESULTS AND DISCUSSION

In order to prove our hypothesis, we measured the relative expression level of miR-132 in human plasma from patients with PD, NDC, and HC by RT-qPCR. All 667 subjects enrolled in this study were Han ethnic Chinese with the male/female ratio at 392/275. Their ages ranged from 40 to 89 years with a mean of 66.14 ± 0.66 years. The demographic characteristics including age and gender of each group were summarized in Table 1. There was no significant difference among PD, HC, and NDC groups for the distribution of age and gender in each group. In addition, total 345 available peripheral blood lymphocytes (PBL) samples (142 PD, 98 HC, and 105 NDC) were used for the further determination of Nurr1 expression and evaluation the correlation between Nurr1 and miR-132. There was no significant difference in both gender and age among all groups. The demographic characteristics of the cohort of subjects for the Nurr1 gene expression were summarized in Table 2.

miRNAs have been proposed as putative noninvasive biomarkers in diagnosis, prognosis, and response to treatment for several diseases, including neurodegenerative disorders such as PD.[28] In the present study, we aimed to measure the relative expression of miR-132 and Nurr1 in human peripheral blood from PD and controls. Our results showed that plasma miR-132 expression was significantly higher in PD than that in HC (Figure 1, 0.32 ± 0.03 vs 0.18 ± 0.03, $p < 0.05$) and in NDC (Figure 1, 0.32 ± 0.03 vs 0.17 ± 0.03, $p < 0.001$), suggesting that plasma miR-132 expression level may be specially altered.

In order to explore the possible impacts of age on the changes of plasma miR-132 expression level, we analyzed the correlation between miR-132 expression and age in both HC and PD groups using Spearman's correlation analysis. As shown in Figure 2A, the coefficient (R) results revealed no correlation between miR-132 and age in HC ($r = 0.117$, $p = 0.144$) and PD groups ($r = 0.048$, $p = 0.465$), suggesting that age did not influence the expression of plasma miR-132.

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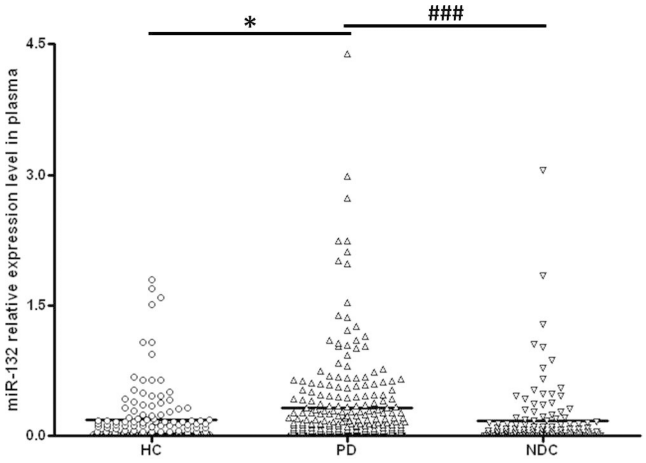

Figure 1. Scatter plots of miR-132 relative expression in plasma in different study groups. Fluorescent reading from real-time PCR reaction was quantitatively analyzed by determining the difference of Ct (ΔCt) between Ct of miR-132 and external control, and the miR-132 expression was determined by the formation of $2^{-\Delta Ct}$. The level of miR-132 expression was significantly increased in patients with PD (n = 269) as compared with healthy controls (HC, n = 222) and neurological disease controls (NDC, n = 176). Horizontal bar represent median value. The Kruskal–Wallis test was used to evaluate the differences in the relative miR-132 expression levels in individuals from each group. $^{*}p < 0.05$ vs HC, $^{\#\#\#}p < 0.001$ vs NDC.

Interestingly, we found a significantly higher plasma miR-132 level in male PD than that in HC, while there was no difference between PD and NDC (Figure 2B). It was reported that miRNAs expression profiles were differentially expressed between male and female in SN DAergic neurons of PD patients.[29] Moreover, the expression levels of miR-132 were varied among different regions of the brain and presented in a sex-dependent manner in mice with chronic *Toxoplasma gondii* infection.[30] It is conceivable that the significant higher miR-132 expression in male PD than female in our study may suggest that gender influence might be involved in miR-132 expression and might play a role in different ratio of male/female PD.[31]

We further analyzed disease duration and severity (H-Y score) in 269 patients with PD. We found that plasma miR-132 expression was associated with the disease duration. The Spearman correlation analysis results revealed a positive correlation between the relative expression of miR-132 and duration in PD (r = 0.187, p = 0.006) (Figure 3A). Consistently, similar association was also found between miR-132 expression and H–Y scores (Figure 3B). In our study, we analyzed plasma miR-132 expression level of 67 PD patients who were of recent-onset and not yet been treated with anti-PD medications ("naïve" PD), we found that the miR-132 expression in PD was significantly increased in comparison to that in HC (Figure 3C). We also analyzed the miR-132 expression in 183 PD patients who were regularly treated with first- or second-line anti-PD medications, including DA receptor agonists alone (DR, n = 16), levodopa alone (L-dopa) (n = 69), or the combination of L-dopa and DR (n = 98). Nineteen PD patients treated irregularly with several other medications were not included in the analysis. Interestingly, L-dopa treatment increased the miR-132 expression level of PD patients compared to HC, but there was no significant difference among the other four groups (PD, naïve, DR, L-dopa+DR, Figure 3C). Alieva et al.[32] reported that the level of miR-132 in PBL was increased in 18 L-dopa-treated patients with PD compared with nontreated PD (H-Y

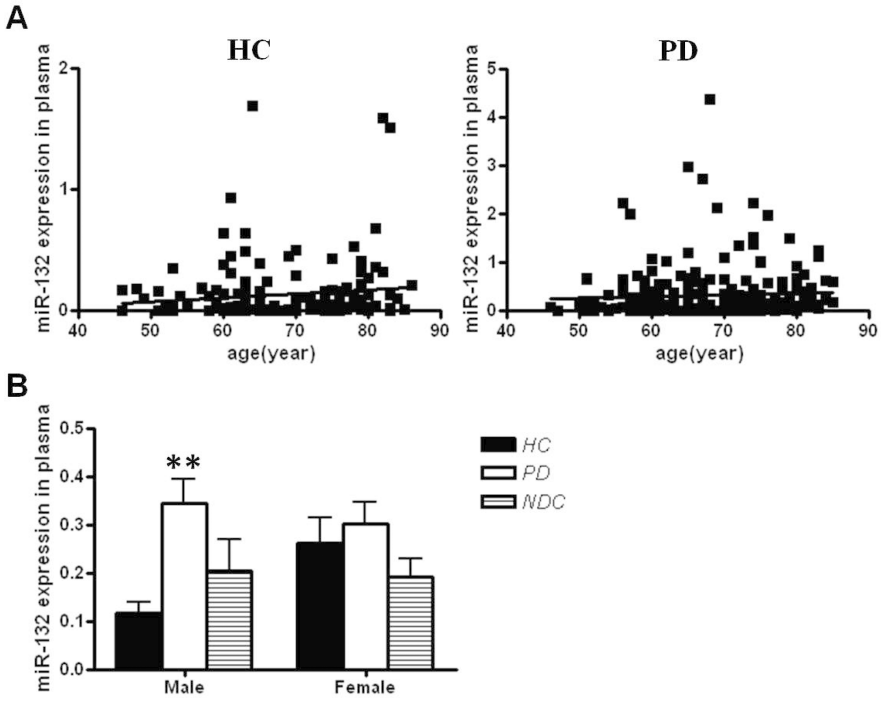

Figure 2. Age and gender effects on plasma miR-132 expression levels in PD and controls. (A) Correlation between plasma miR-132 and age in PD and HC groups. The Spearman correlation analysis results revealed no correlation between the relative expression of miR-132 and age in HC (r = 0.117, p = 0.144) and PD group (r = 0.048, p = 0.465). (B) Expression levels of miR-132 in male and female of HC, PD, and NDC groups. The Kruskal–Wallis test was used to evaluate the differences in the gender distribution in individuals from each group. $^{**}p < 0.01$ vs HC.

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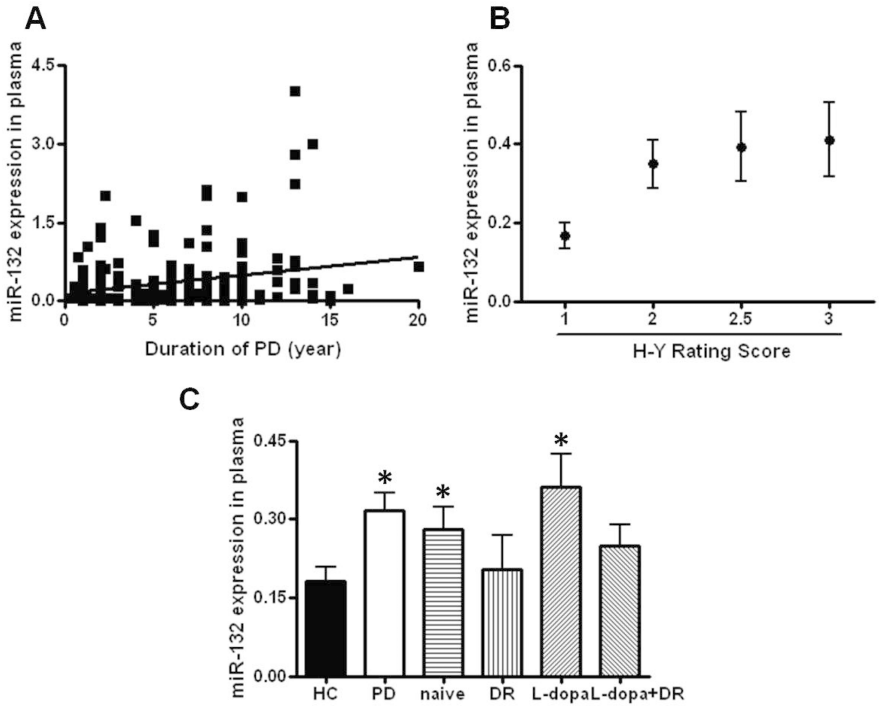

Figure 3. Expression levels of miR-132 in PD and HC. Effect of (A) disease course, (B) severity, and (C) anti-PD medications to the expression levels of miR-132. Spearman correlation analysis revealed a positive correlation between the relative expression of miR-132 and duration in PD (r = 0.187, p = 0.006). The Kruskal–Wallis test was used to evaluate the differences in the relative miR-132 expression levels in individuals from each group. *p < 0.05 vs HC.

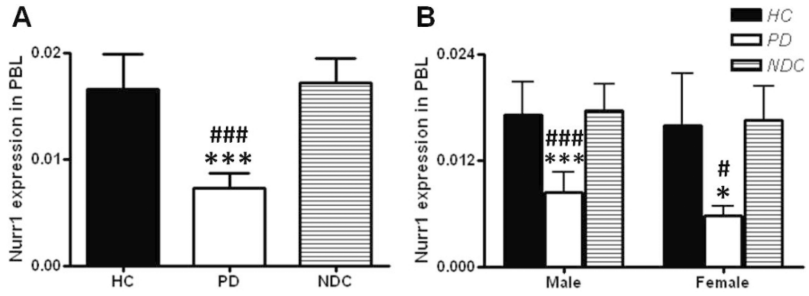

Figure 4. PBL Nurr1 expression levels in PD and controls (HC, NDC). Kruskal–Wallis test was used to evaluate the differences in the relative Nurr1 expression levels in individuals from each group. *p < 0.05 vs HC, $^{\#}p$ < 0.05 vs NDC, ***p < 0.001 vs HC, $^{\#\#\#}p$ < 0.001 vs NDC.

scores 1–2). We considered that the expression level of miR-132 were also affected by other factors, such as gender, duration and severity of disease in addition of medications. Receiver operating characteristic (ROC) analysis revealed that the areas under the ROC curve (AUC) were 0.72 (95% CI, 0.66–0.78; p < 0.0001) for miR-132 in plasma between PD and HC. The AUC of miR-132 were larger than 0.7, which means that miR-132 could be considered to have a discriminative value. Certainly, a larger population-based study is needed for the validation of this observation.

To further explore how miR-132 worked in the occurrence and development of PD, we measured the PBL expression level of Nurr1, which has been reported to be a target gene of miR-132.[25] We previously found that Nurr1 gene expression was significantly decreased in PBL of patients with PD as compared with both HC and NDC.[26,27] Furthermore, Nurr1 mRNA level in PBL of PD was significantly lower than that in HC (0.007 ± 0.001 vs 0.017 ± 0.003, p < 0.001), and NDC (0.007 ± 0.001 vs 0.017 ± 0.002, p < 0.001). There was no difference of Nurr1 expression between HC and NDC (Figure 4A), which was in correlation with the changes of plasma miR-132. Besides, in contrast to the higher plasma miR-132 expression level in male PD, we also found a significantly lower level of Nurr1 in male PD, as compared to HC and NDC (Figure 4B). We further documented that Nurr1 was significantly decreased in naïve PD comparing to HC (Figure S1).

Research on human peripheral blood is common in a clinic study to identify disease biomarkers and evaluate disease progression. miRNAs have been reported to be encapsulated in microvesicles, which are secreted by circulating blood cells and other cells from different tissues.[33,34] Kim and colleagues[8] determined pre-miR-132 by qPCR for a panel of 224 precursor miRNAs in RNAs samples from PD and normal brains. Their results showed that pre-miR-132 was enriched and high expressed in the midbrain of PD brains compared to HC. Moreover, it is known that Nurr1 is expressed predominantly in the central nervous system (CNS), especially in the midbrain and limbic areas.[20] The decline of Nurr1-immunoreactive neuronal number and optical density have been observed in the SN neurons of PD midbrain.[35] In addition

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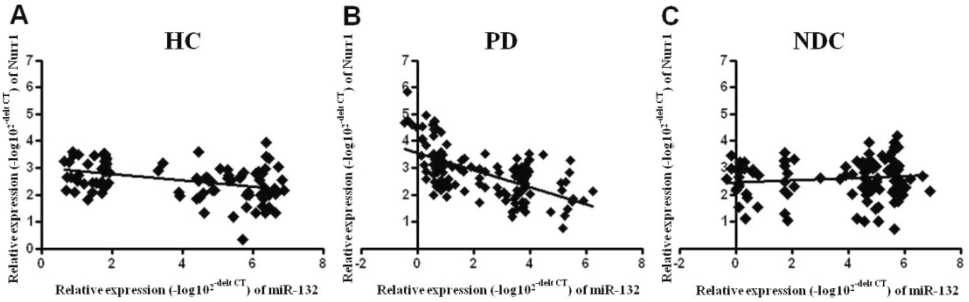

Figure 5. Correlation between plasma miR-132 and PBL Nurr1 levels in the peripheral blood of HC (n = 98), PD (n = 142), and NDC (n = 105). Spearman correlation analysis revealed a negative correlation between the relative expression of miR-132 and Nurr1. (A) HC group: $r = -0.335$, $p < 0.001$. (B) PD group: $r = -0.579$, $p < 0.001$. (C) NDC group: $r = 0.082$, $p = 0.411$. Values are displayed on a $\log10^{2-\Delta CT}$ scale on the X- and Y-axes.

to the biological effects on CNS, Nurr1 has been reported to be associated with pro-inflammatory cytokines in peripheral blood mononuclear cell including PBL in patients with PD and type 2 diabetes.[36,37] Here, we measured Nurr1 expression in PBL and analyzed the correlation between PBL Nurr1 and plasma miR-132, showing a negative correlation in PD patients ($r = -0.579$, $p < 0.001$) (Figure 5). Interestingly, no correlation was found in NDC patients ($r = 0.082$, $p = 0.411$) (Figure 5), suggesting that miR-132/Nurr1 signaling was specifically involved in PD. Moreover, to explore the correlation of miR-132 and Nurr1 in blood, we determined if miR-132 can affect the Nurr1 expression in PBL. We transfected with miR-132 mimic or normalized control (NC) mimic in cultured PBL for 24 h, and found that the Nurr1 level was significantly decreased in PBL transfected with miR-132 mimic (Figure S2), suggesting a regulative effect of miR-132 on Nurr1 expression in PBL. Our previous studies and others have documented that Nurr1 was critical not only in the midbrain DAergic neuron differentiation and functional maintenance, but also in regulating glia- and lymphocyte-mediated inflammation.[24,36,37] Collectively, Nurr1 defect might play an important role in PD pathogenesis.

Recently, growing evidence has shown that miR-132 may be one of the promising miRNAs in the diagnosis and prognosis of neurological disease. In AD, miR-132 was found to be downregulated in patients' brains and cerebrospinal fluid,[38] whereas it was upregulated in their serum[39] and plasma.[15] However, these reports did not compare miR-132 expression to NDC. In this study, we found that miR-132 measurement in plasma may help in differentiating PD from various non-PD NDC, and the combination of Nurr1 with miR-132 may enhance the sensitivity and specificity for differential diagnosis of PD.

Nevertheless, the results from our present study may merit the attention that miR-132 may be associated with PD, as evidenced by the progressive increasing of plasma miR-132 level in PD patients in a disease stage- and disease severity-dependent manner. In conclusion, our study may provide the following information: (1) miR-132 expression level was significantly increased in patients with PD compared to both HC and NDC; (2) the changes in plasma miR-132 expression level were correlated with disease progression and severity; (3) the difference of miR-132 level in male PD vs controls was greater than that in female PD vs controls; (4) Nurr1 gene expression was significantly decreased in PBL of PD as compared with HC and NDC, showing negative correlation with miR-132 expression level in PD patients. Collectively, the combined measurements of miR-132 and Nurr1 expression levels in blood may help the diagnosis and differential diagnosis of PD, and may also assist the monitoring of the disease progression.

METHODS

Participants and Blood Sampling. In this study, we recruited a total of 667 participants including 269 patients with sporadic PD, 222 HC, and 176 various non-PD NDC which consist of 38 cerebrovascular disease, 38 epilepsy, 25 peripheral neuropathy, 20 essential tremor, 16 myasthenia gravis, 15 migraine, 14 motor neuron disease, and 10 multiple sclerosis. PD patients were examined and diagnosed by at least two experienced neurologists from the First Affiliated Hospital of Dalian Medical University, according to the Movement Disorder Society Clinical Diagnostic Criteria for Parkinson's disease.[40] Subjects with secondary parkinsonism, neoplastic or metabolic disorders, alcoholism were excluded. PD disease severity was assessed by Modified Hoehn and Yahr (H-Y) staging. HC group were recruited from the Health Examination Center of the First Affiliated Hospital of Dalian Medical University, and were either diagnosed without a neurological disorder or with a systemic disease without neurological manifestations. All subjects (or their caregivers) recruited to our studies provided a written informed consent agreeing to participate the project. This study has been granted ethical approval by the Ethics Committee of the First Affiliated Hospital of Dalian Medical University (approval number: LCKY2014-29).

Blood samples used in this study were collected by direct venipuncture at the First Affiliated Hospital of Dalian Medical University. Briefly, two milliliters of peripheral blood was drawn from cubital vein into a vacuum blood tube with ethylenediaminetetraacetic acid (EDTA) and then the plasma was aliquoted (200 μL) into sterile tube and stored at −80 °C. PBL were separated from gradient Ficoll/Paque by centrifugation at 450 g for 20 min at room temperature (20 ± 2 °C), resuspended with Sample Protector of RNA/DNA (TaKaRa, Dalian, China) and stored at −80 °C until RNA extraction.

Plasma miRNA and PBL RNA Extraction and Quantification. Total miRNAs were extracted from plasma using miRNA isolation system (Tiangen Biotech (Beijing) Co., Ltd., Beijing, China). Five microliters of miRNA from plasma was reverse transcribed into first strand cDNA by Tiangen miRCute miRNA cDNA synthesis kit (Tiangen Biotech (Beijing) Co., Ltd.). Total RNAs from PBL were extracted using the mirVana miRNA Isolation Kit (Ambion, Carlsbad, CA) following the manufacturer's instructions, and 1 μg RNA from PBL was reverse transcribed into first strand cDNA by PrimeScript RT reagent Kit with gDNA Eraser (TaKaRa, Dalian, China).

The plasma miR-132 and PBL Nurr1 mRNA levels were determined by RT-qPCR. For plasma, considering the lack of suitable internal control for normalization, artificial synthetic external miRNA (Tiangen Biotech (Beijing) Co., Ltd., Beijing, China) was used for normalization. Besides, the reverse primer of miR-132 was provided by the miRcute miRNA qPCR Detection Kit (Tiangen Biotech

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(Beijing) Co., Ltd., Beijing, China). The up-primer targeting plasma miR-132-3p was as follows: 5′-GCT AAC AGT CTA CAG CCA TGG TCG-3′. For PBL, human GAPDH gene was used as an internal control. The specific primers targeting PBL Nurr1 and GAPDH as follows: Nurr1-forward: 5′-TCC AAC GAG GGG CTG TGC G-3′, Nurr1-reverse: 5′-CAC TGT GCG CTT AAA GAA GC-3′; GAPDH-forward: 5′-GAA GGT GAA GGT CGG AGT C-3′, GAPDH-reverse: 5′-GAA GAT GGT GAT GGG ATT TC-3′. Relative quantification was performed using the $2^{-\Delta Ct}$ method. PCR was carried out using ABI 7500 fast real-time PCR system (Applied Biosystems, Foster City, CA) in a total volume of 20 μL for each reaction.

Statistical Analysis. Quantitative data were expressed as mean ± SEM, or median depending on the distribution of the data. The χ^2 test was used to evaluate the statistical differences of gender distribution between PD patients and control subjects. The Kruskal−Wallis test followed by Dunn's multiple comparison test as a post hoc test using the GraphPad Prism software version 4 (GraphPad Inc., San Diego, CA) was used to evaluate the differences in the age distribution and relative miRNA expression levels in individuals from each group. Exact 95% confidence intervals (CIs) were reported for estimation of sensitivity and specificity. ROC analysis was performed using MedCalc software version 17.2 (MedCalc Software Inc., Mariakerke, Belgium), and predictive performance of the putative biomarker (miRNA-132 or Nurr1 or both) for the presence of PD was quantified by using AUC. Correlations were assessed using Spearman's correlation coefficient. The correlations were reported at á level of 0.05. The other statistical analysis in this research was performed with the SPSS software version 13.0 (SPSS Inc., Chicago, IL). All statistical checks were carried out two-sided and a p-value < 0.05 was considered as statistical significance.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acschemneuro.8b00460.

> Effect of miR-132 on Nurr1 expression in cultured PBL and effect of medications on the Nurr1 expression in PBL of PD patients (PDF)

Special Issue Paper

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Author Contributions

W.L. and F.P. designed the project. Z.Y. and T.L. carried out all the experiments. Z.Y., T.L., and B.S. contributed to statistical analyses and results interpretation. Z.Y., T.L., S.L., M.W., and B.S. contributed to drafting of the manuscript. Z.Y., T.L., S.L., M.W., H.Q., R.C.-C.C, W.L., and F.P. revised the paper. All authors edited and approved the final version of the manuscript.

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Notes

The authors declare no competing financial interest.

■ REFERENCES

(1) Xie, Y., and Chen, Y. (2016) microRNAs: Emerging Targets Regulating Oxidative Stress in the Models of Parkinson's Disease. *Front. Neurosci. 10*, 298.
(2) Sarkar, S., Raymick, J., and Imam, S. (2016) Neuroprotective and Therapeutic Strategies against Parkinson's Disease: Recent Perspectives. *Int. J. Mol. Sci. 17* (pii), E904.
(3) Kalia, L. V., and Lang, A. E. (2015) Parkinson's disease. *Lancet 386*, 896−912.
(4) Li, S., and Le, W. (2017) Milestones of Parkinson's Disease Research: 200 Years of History and Beyond. *Neurosci. Bull. 33*, 598−602.
(5) Dauer, W., and Przedborski, S. (2003) Parkinson's disease: mechanisms and models. *Neuron 39*, 889−909.
(6) Bartel, D. P. (2004) MicroRNAs: genomics, biogenesis, mechanism, and function. *Cell 116*, 281−297.
(7) Wang, Y., Yang, Z., and Le, W. (2017) Tiny But Mighty: Promising Roles of MicroRNAs in the Diagnosis and Treatment of Parkinson's Disease. *Neurosci. Bull. 33*, 543−551.
(8) Kim, J., Inoue, K., Ishii, J., Vanti, W. B., Voronov, S. V., Murchison, E., Hannon, G., and Abeliovich, A. (2007) A MicroRNA feedback circuit in midbrain dopamine neurons. *Science 317*, 1220−1224.
(9) Chmielarz, P., Konovalova, J., Najam, S. S., Alter, H., Piepponen, T. P., Erfle, H., Sonntag, K. C., Schütz, G., Vinnikov, I. A., and Domanskyi, A. (2017) Dicer and microRNAs protect adult dopamine neurons. *Cell Death Dis. 8*, No. e2813.
(10) Junn, E., Lee, K. W., Jeong, B. S., Chan, T. W., Im, J. Y., and Mouradian, M. M. (2009) Repression of alpha-synuclein expression and toxicity by microRNA-7. *Proc. Natl. Acad. Sci. U. S. A. 106*, 13052−13057.
(11) Doxakis, E. (2010) Post-transcriptional regulation of alpha-synuclein expression by mir-7 and mir-153. *J. Biol. Chem. 285*, 12726−12734.
(12) Wang, G., van der Walt, J. M., Mayhew, G., Li, Y. J., Züchner, S., Scott, W. K., Martin, E. R., and Vance, J. M. (2008) Variation in the miRNA-433 binding site of FGF20 confers risk for Parkinson disease by overexpression of alphasynuclein. *Am. J. Hum. Genet. 82*, 283−289.
(13) Decressac, M., Mattsson, B., Weikop, P., Lundblad, M., Jakobsson, J., and Björklund, A. (2013) TFEB-mediated autophagy rescues midbrain dopamine neurons from α-synuclein toxicity. *Proc. Natl. Acad. Sci. U. S. A. 110*, E1817−26.
(14) Xiao, J., Li, Y., Prandovszky, E., Karuppagounder, S. S., Talbot, C. C., Jr., Dawson, V. L., Dawson, T. M., and Yolken, R. H. (2014) MicroRNA-132 dysregulation in Toxoplasma gondii infection has implications for dopamine signaling pathway. *Neuroscience 268*, 128−138.
(15) Sheinerman, K. S., Tsivinsky, V. G., Abdullah, L., Crawford, F., and Umansky, S. R. (2013) Plasma microRNA biomarkers for detection of mild cognitive impairment: biomarker validation study. *Aging 5*, 925−938.
(16) Salta, E., and De Strooper, B. (2017) microRNA-132: a key noncoding RNA operating in the cellular phase of Alzheimer's disease. *FASEB J. 31*, 424−433.
(17) Sheinerman, K. S., Tsivinsky, V. G., Crawford, F., Mullan, M. J., Abdullah, L., and Umansky, S. R. (2012) Plasma microRNA biomarkers for detection of mild cognitive impairment. *Aging 4*, 590−605.
(18) Kumar, S., and Reddy, P. H. (2016) Are circulating microRNAs peripheral biomarkers for Alzheimer's disease? *Biochim. Biophys. Acta, Mol. Basis Dis. 1862*, 1617−27.

DOI: 10.1021/acschemneuro.8b00460
ACS Chem. Neurosci. 2019, 10, 2243−2249

(19) Lungu, G., Stoica, G., and Ambrus, A. (2013) MicroRNA profiling and the role of microRNA-132 in neurodegeneration using a rat model. *Neurosci. Lett. 553*, 153−158.
(20) Jankovic, J., Chen, S., and Le, W. D. (2005) The role of Nurr1 in the development of dopaminergic neurons and Parkinson's disease. *Prog. Neurobiol. 77*, 128−138.
(21) Dong, J., Li, S., Mo, J. L., Cai, H. B., and Le, W. D. (2016) Nurr1-Based Therapies for Parkinson's Disease. *CNS Neurosci. Ther. 22*, 351−359.
(22) Wang, Z., Benoit, G., Liu, J., Prasad, S., Aarnisalo, P., Liu, X., Xu, H., Walker, N. P., and Perlmann, T. (2003) Structure and function of Nurr1 identifies a class of ligand-independent nuclear receptors. *Nature 423*, 555−560.
(23) Decressac, M., Volakakis, N., Björklund, A., and Perlmann, T. (2013) NURR1 in Parkinson disease−from pathogenesis to therapeutic potential. *Nat. Rev. Neurol. 9*, 629−636.
(24) Saijo, K., Winner, B., Carson, C. T., Collier, J. G., Boyer, L., Rosenfeld, M. G., Gage, F. H., and Glass, C. K. (2009) A Nurr1/CoREST pathway in microglia and astrocytes protects dopaminergic neurons from inflammation-induced death. *Cell 137*, 47−59.
(25) Yang, D., Li, T., Wang, Y., Tang, Y., Cui, H., Tang, Y., Zhang, X., Chen, D., Shen, N., and Le, W. (2012) miR-132 regulates the differentiation of dopamine neurons by directly targeting Nurr1 expression. *J. Cell Sci. 125*, 1673−1682.
(26) Le, W., Pan, T., Huang, M., Xu, P., Xie, W., Zhu, W., Zhang, X., Deng, H., and Jankovic, J. (2008) Decreased NURR1 gene expression in patients with Parkinson's disease. *J. Neurol. Sci. 273*, 29−33.
(27) Liu, H., Wei, L., Tao, Q., Deng, H., Ming, M., Xu, P., and Le, W. (2012) Decreased NURR1 and PITX3 gene expression in Chinese patients with Parkinson's disease. *Eur. J. Neurol. 19*, 870−875.
(28) Batistela, M. S., Josviak, N. D., Sulzbach, C. D., and de Souza, R. L. (2017) An overview of circulating cell-free microRNAs as putative biomarkers in Alzheimer's and Parkinson's Diseases. *Int. J. Neurosci. 127*, 547−558.
(29) Briggs, C. E., Wang, Y., Kong, B., Woo, T. U., Iyer, L. K., and Sonntag, K. C. (2015) Midbrain dopamine neurons in Parkinson's disease exhibit a dysregulated miRNA and target-gene network. *Brain Res. 1618*, 111−21.
(30) Li, Y. E., Kannan, G., Pletnikov, M. V., Yolken, R. H., and Xiao, J. (2015) Chronic infection of Toxoplasma gondii downregulates miR-132 expression in multiple brain regions in a sex-dependent manner. *Parasitology 142*, 623−32.
(31) Taylor, K. S., Cook, J. A., and Counsell, C. E. (2007) Heterogeneity in male to female risk for Parkinson's disease. *J. Neurol., Neurosurg. Psychiatry 78*, 905−906.
(32) Alieva, A., Filatova, E. V., Karabanov, A. V., Illarioshkin, S. N., Limborska, S. A., Shadrina, M. I., and Slominsky, P. A. (2015) miRNA expression is highly sensitive to a drug therapy in Parkinson's disease. *Parkinsonism. Rrelat. Disord. 21*, 72−74.
(33) Ding, H., Huang, Z., Chen, M., Wang, C., Chen, X., Chen, J., and Zhang, J. (2016) Identification of a panel of five serum miRNAs as a biomarker for Parkinson's disease. *Parkinsonism. Relat. Disord. 22*, 68−73.
(34) Zhang, Y., Liu, D., Chen, X., Li, J., Li, L., Bian, Z., Sun, F., Lu, J., Yin, Y., Cai, X., Sun, Q., Wang, K., Ba, Y., Wang, Q., Wang, D., Yang, J., Liu, P., Xu, T., Yan, Q., Zhang, J., Zen, K., and Zhang, C. Y. (2010) Secreted monocytic miR-150 enhances targeted endothelial cell migration. *Mol. Cell 39*, 133−144.
(35) Chu, Y., Le, W., Kompoliti, K., Jankovic, J., Mufson, E. J., and Kordower, J. H. (2006) Nurr1 in Parkinson's disease and related disorders. *J. Comp. Neurol. 494*, 495−514.
(36) Li, T., Yang, Z., Li, S., Cheng, C., Shen, B., and Le, W. (2018) Alterations of NURR1 and cytokines in the peripheral blood mononuclear cells: combined biomarkers for Parkinson's disease. *Front. Aging Neurosci. 10*, 392.
(37) Xu, Y., Huang, Q., Zhang, W., Wang, Y., Zeng, Q., He, C., Xue, J., Chen, J., Hu, X., and Xu, Y. (2015) Decreased expression levels of Nurr1 are associated with chronic inflammation in patients with type 2 diabetes. *Mol. Med. Rep. 12*, 5487−5493.
(38) Cogswell, J. P., Ward, J., Taylor, I. A., Waters, M., Shi, Y., Cannon, B., Kelnar, K., Kemppainen, J., Brown, D., Chen, C., Prinjha, R. K., Richardson, J. C., Saunders, A. M., Roses, A. D., and Richards, C. A. (2008) Identification of miRNA changes in Alzheimer's disease brain and CSF yields putative biomarkers and insights into disease pathways. *J. Alzheimer's Dis. 14*, 27−41.
(39) Xie, B., Zhou, H., Zhang, R., Song, M., Yu, L., Wang, L., Liu, Z., Zhang, Q., Cui, D., Wang, X., and Xu, S. (2015) Serum miR-206 and miR-132 as potential circulating biomarkers for mild cognitive impairment. *J. Alzheimer's Dis. 45*, 721−731.
(40) Postuma, R. B., Berg, D., Stern, M., Lewis, S., Lang, A. E., Halliday, G., Goetz, C. G., Chan, P., Slow, E., Seppi, K., Schaffer, E., Rios-Romenets, S., Mi, T., Maetzler, C., Li, Y., Heim, B., Bledsoe, I. O., and Berg, D. (2018) Validation of the MDS clinical diagnostic criteria for Parkinson's disease. *Mov. Disord. 33*, 1601.

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ACS Chem. Neurosci. 2019, 10, 2243−2249