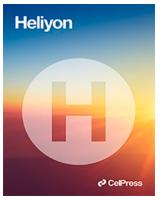
Heliyon 10 (2024) e26588

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.cell.com/heliyon



Research article

Pyruvate is modified by tea/coffee metabolites and reversely correlated with multiple system atrophy and Parkinson's disease

Xu-Ying Li [a,b,1], Teng Xue [c,d,1], Hong Lai [a], Jing Dai [e], Fangda Peng [e], Fanxi Xu [a], Junge Zhu [a], Xian Li [a], Junya Hu [f], Wei Li [f], Raoli He [g], Lina Chen [g], Ying Chen [g], Chunguang Ding [e], Guoguang Zhao [h], Xianyang Chen [d,i,****], Qinyong Ye [g,***], Zhiheng Xu [b,**], Chaodong Wang [a,*]

[a] *Department of Neurology and Neurobiology, Xuanwu Hospital of Capital Medical University, National Clinical Research Center for Geriatric Diseases, Beijing, 100053, China*
[b] *Institute of Genetics and Developmental Biology, Chinese Academy of Sciences, Beijing, 100101, China*
[c] *Zhongyuanborui Key Laboratory of Genetics and Metabolism, Guangdong-Macao In-depth Cooperation Zone in Hengqin, China*
[d] *Zhongguancun Biological and Medical Big Data Center, Beijing, China*
[e] *National Center for Occupational Safety and Health, NHC (National Center for Occupational Medicine of Coal Industry), Beijing, China*
[f] *Department of Neurobiology, Xuanwu Hospital of Capital Medical University, Beijing, 100053, China*
[g] *Department of Neurology, Fujian Medical University Union Hospital, Institute of Neuroscience, Fujian Key Laboratory of Molecular Neurology, Fujian Medical University, Fuzhou, China*
[h] *Department of Neurosurgery, Xuanwu Hospital of Capital Medical University, Clinical Research Center for Epilepsy Capital Medical University, Beijing, China*
[i] *Bao Feng Key Laboratory of Genetics and Metabolism, Beijing, China*

ARTICLE INFO

Keywords:
Multiple system atrophy
Parkinson's disease
Pyruvate
Tea/coffee metabolites
Diagnostic and differential diagnostic model

ABSTRACT

Introduction: Multiple system atrophy (MSA) is a rapidly progressing neurodegenerative disorder. Although diverse biomarkers have been established for Parkinson's disease (PD), no widely accepted markers have been identified in MSA. Pyruvate and lactate are the end-product of glycolysis and crucial for brain metabolism. However, their correlation with MSA remains unclear. Moreover, it is elusive how lifestyles modify these metabolites.

Methods: To investigate the correlation and diagnostic value of plasma pyruvate and lactate levels in MSA and PD. Moreover, we explored how lifestyle-related metabolites interact with these metabolites in determining the disease risk. We assayed the 3 metabolites in pyruvate/lactate and 6 in the tea/coffee metabolic pathways by targeted mass spectrometry and evaluate their interactions and performance in diagnosis and differentiation between MSA and PD.

Results: We found that 7 metabolites were significantly different between MSA, PD and healthy controls (HCs). Particularly, pyruvate was increased in PD while significantly decreased in MSA patients. Moreover, the tea/coffee metabolites were negatively associated with the pyruvate level in HCs, but not in MSA and PD patients. Using machine-learning models, we showed that the combination of pyruvate and tea/coffee metabolites diagnosed MSA (AUC = 0.878) and PD (AUC

* Corresponding author.
** Corresponding author.
*** Corresponding author.
**** Corresponding author. Bao Feng Key Laboratory of Genetics and Metabolism, Beijing, China
E-mail addresses: cxyibcas@163.com (X. Chen), unionqyye@163.com (Q. Ye), zhxu@genetics.ac.cn (Z. Xu), cdongwang01@126.com (C. Wang).
[1] These authors contributed equally to this work.

https://doi.org/10.1016/j.heliyon.2024.e26588
Received 11 January 2024; Received in revised form 7 February 2024; Accepted 15 February 2024
Available online 22 February 2024

= 0.833) with good performance. Additionally, pyruvate had good performance in distinguishing MSA from PD (AUC = 0.860), and the differentiation increased (AUC = 0.922) when combined with theanine and 1,3-dimethyluric acid.
Conclusions: This study demonstrates that pyruvate correlates reversely with MSA and PD, and may play distinct roles in their pathogenesis, which can be modified by lifestyle-related tea/coffee metabolites.

1. Introduction

Parkinson's disease (PD) and multiple system atrophy (MSA) are progressive neurodegenerative disorders characterized by motor and non-motor symptoms. Contrasting to PD, MSA patients follow a rapid progression for 6–10 years from symptomatic onset to death, without effective therapy [1]. However, due to the overlapping symptomatology and pathology between PD and MSA, it is difficult to make a differential diagnosis, especially in the early stage.

Table 1
Study participant demographic features and clinical characteristics.

Items	Variables	HCs	MSA	*P*-value (MSA *vs.* HCs)	*PD*	*P*-value (PD *vs.* HCs)
Demographic characteristics	Gender, Female (%)	65 (53.3%)	58 (47.5%)	0.442	67 (54.9%)	0.891
	Age, Mean ± SD	62.6 ± 6.90	60.4 ± 9.29	0.075	61.1 ± 8.67	0.403
	BMI, Mean ± SD	24.7 ± 3.62	24.3 ± 2.39	0.818	23.3 ± 2.90	0.051
	Disease duration, Mean ± SD	–	4.30 ± 2.89	–	2.81 ± 3.78	–
	UPDRS III/UMSARS II, Mean ± SD	–	19.45 ± 11.18 (UMSARS II)	–	25.15 ± 14.88 (UPDRS III)	–
	H–Y stage, Median (min~max)	–	3 (2,5)	–	2 (1,5)	–
Epidemiological information	Smoking, Y (%)	45 (36.9%)	34 (27.9%)	0.221	27 (22.1%)	**0.017**
	Alcohol-drinking, Y (%)	33 (27.0%)	36 (29.5%)	0.177	18 (14.8%)	**0.028**
	Frequency-Alcohol-drinking Median (min~max)	0 (0, 1)	0 (0, 0.75)	**0.003**	0 (0, 0)	**0.031**
	Tea intake, Y (%)	87 (71.3%)	48 (39.3%)	**9.92E-07**	70 (57.4%)	**0.023**
	Frequency-Tea Median (min~max)	1 (0, 1)	0 (0, 1)	**5.21E-06**	1 (0, 2)	**4.99E-04**
	Coffee intake, Y (%)	29 (23.8%)	5 (4.10%)	**2.12E-05**	13 (10.7%)	**0.007**
	Frequency-Coffee Median (min~max)	0 (0, 0)	0 (0, 0)	**0.000572**	0 (0, 0)	**0.003**
No-motor symptoms	Uroclepsia, Y (%)	38 (31.1%)	61 (50.0%)	**0.004128**	61 (50.0%)	**0.003**
	Constipation, Y (%)	11 (9.02%)	23 (18.9%)	**0.042005**	23 (18.9%)	**0.038**
	MoCA, Mean ± SD	25.2 ± 3.41	22.2 ± 5.41	**2.12E-07**	22.2 ± 5.41	**2.12E-07**
	ESS, Mean ± SD	2.92 ± 2.66	4.79 ± 4.25	**0.00074**	4.79 ± 4.25	**7.40E-04**
	RBDQ-HK, Mean ± SD	7.98 ± 8.44	21.4 ± 16.2	**2.56E-15**	21.4 ± 16.2	**2.56E-15**
	SCOPA-AUT, Mean ± SD	4.82 ± 3.66	12.6 ± 9.46	**2.04E-15**	12.6 ± 9.46	**2.04E-15**
	HAMD, Mean ± SD	3.13 ± 3.11	5.97 ± 4.39	**9.35E-09**	5.97 ± 4.39	**9.35E-09**
	HAMA, Mean ± SD	5.14 ± 3.78	6.76 ± 4.94	**0.011362**	6.76 ± 4.94	**0.011**
	NMSS-Total, Mean ± SD	19.4 ± 15.3	40.3 ± 25.0	**3.41E-13**	40.3 ± 25.0	**3.41E-13**

Abbreviations: HCs = healthy controls; MSA = multiple system atrophy; PD = Parkinson's disease; BMI = body mass index; MoCA = Montreal Cognitive Assessment; ESS = Epworth sleepiness scale; RBDQ-HK = rapid eye movement sleep behavior disorder questionnaire-Hong Kong; SCOPA-AUT = scales for outcome in Parkinson's disease-autonomic dysfunction; HAMD = Hamilton Depression Scale; HAMA = Hamilton Anxiety Scale; NMSS = non-motor symptoms scale. Statistical significance was assessed by Mann-Whitney *U* test and Chi-square test.

Various molecules, such as α-syn, DJ-1, and neuroinflammatory cytokines, have been widely tested for utility in diagnosing PD and MSA [2]. There were also approaches, including clinical decision trees [3] and imaging technologies [4] reported to differentiate MSA from PD. However, the sample sizes in these reports were small and the results were controversial. The difficulty of finding an ideal differential biomarker lies in the complex disease pathogenesis, which involves genetic, environmental factors and aging. Most recently, metabolome, the collection of entities of small chemical molecules involved in metabolism, has been explored for potentials biomarkers for diseases. Among the metabolites, pyruvate and lactate, the end-product of glycolysis, have been implicated as the crucial molecules for brain metabolism, especially in coordination of neurons, astrocytes and oligodendrocytes.

In addition, environmental factors or lifestyles have been implicated as the important etiology of neurodegenerative disorders in recent years. Tea, coffee or alcohol drinking and smoking have been commonly recognized as the protective factors for synucleinopathies. The neuroprotective effects of tea/caffeine had been established in patients with Alzheimer's disease (AD), PD, amyotrophic lateral sclerosis (ALS) and Huntington's disease (HD) [5]. However, the metabolic profiles of MSA and their association with environmental exposures have not been well investigated.

To investigate the pyruvate/lactate and tea/coffee metabolic profiles in PD and MSA and their performance in diagnosis and differential diagnosis of the two diseases, we performed targeted MS spectrometry for these metabolites in age- and sex-matched MSA, PD patients and healthy controls (HCs). We revealed significant differences in metabolite levels between different groups and identified pyruvate as a strong differential marker for MSA and PD, which can be modified by tea/coffee metabolites and lifestyles such as smoking and alcohol-drinking.

2. Participates and methods

2.1. Study design and participants

The study cohort of MSA and PD patients were recruited from the outpatient or inpatient department who were admitted to Xuanwu hospital of Capital Medical University from Aug 2021 to Sep 2022. The HC subjects were enrolled from Xinjiekou Community Medical Health Service Center. The diagnosis was based on the international clinical guidelines. All patients completed detailed clinical interviews and physical examinations. The symptomatic scores for clinical assessments included: Unified Multiple System Atrophy Rating Scale [UMSARS] for MSA and Unified Parkinson's disease Rating Scale [UPDRS] for PD, and non-motor symptoms scale (NMSS), Montreal Cognitive Assessment (MoCA), Epworth sleepiness scale (ESS), rapid eye movement sleep behavior disorder questionnaire-Hong Kong (RBDQ-HK), scales for outcome in Parkinson's disease-autonomic dysfunction (SCOPA-AUT), Hamilton Depression Scale (HAMD), and Hamilton Anxiety Scale (HAMA) scores for patients of both diseases. In addition, the demographic information including age, age at onset, smoking, alcohol-drinking and tea/coffee-taken was also inquired (Table 1).

The study was approved by the Institutional Review Board and Ethics Committees of the Xuanwu Hospital of Capital Medical University ([2020]060). Written informed consent was obtained from each participant or their legal guardians before they were included in the study.

2.2. Chemicals

Analytical standards of citrate, lactate, 1,3-dimethyluric acid, 1-methylxanthine, citric acid d4, theobromine-d6 and 1-methylxanthine-d3 were purchased from Toronto Research Chemicals (TRC). Caffeine and theophylline were purchased from National Institutes for Food and Drug Control, China. Lactic acid-13C3, theophylline-d6 were purchased from CMASS Science/Technology. Caffeine 13C3 was supplied by Sigma-Aldrich (St. Louis, MO). Theobromine was supplied by Aladdin, China. Acetonitrile, methanol, and formic acid were supplied by Thermo Fisher Scientific (Houston, TX, USA). All the chemicals were of LC-MS grade. Ultra-pure water was obtained from the Milli-Q system (Millipore, Bedford, MA, USA).

2.3. Sample preparation for metabolomic analysis

Sampling times are assumed to be random. 10 ml of whole blood from each subject was drawn into an ethylenediaminetetraacetic acid (EDTA) anticoagulant tube (1.8 mg EDTA/mL blood). The blood was allowed to stand at 4–8 °C for 30 min before being centrifuged at 4 °C, 1500×*g* for 15 min. The upper and middle layers, containing plasma was collected. After the serum was thawed at 4 °C, 200 μL sample was aliquoted into a 1.5 mL microfuge tube, and 10 μL of the 10 μg/ml internal standard solution was precisely introduced. To improve protein precipitation, 600 μL pre-cooled methanol was added to 200 μL serum sample, vortexed for 2 min, incubated at room temp for 10min. The sample was then centrifuged at 14,000g for 20min at 4 °C, where after 2 μL supernatant was injected into the liquid chromatography-mass spectrometry (LC-MS) system. All samples were kept at −80 °C for uses.

2.4. Apparatus and operation conditions

Samples were analyzed on a TQ-S micro (Waters, Millford, MA, USA) mass spectrometer (MS) coupled with an Acquity UPLC system (LC) (Waters, Millford, MA, USA). The analysis of 2 μL of the sample was injected into an analytical column (Waters Acquity UPLC HSS T3, 100 × 2.1 mm column; Waters, Millford, MA, USA). 0.05% formic acid in water constituted the mobile phase A and 0.05% formic acid in acetonitrile constituted the mobile phase B. The rate of flow was 0.3 mL/min. The gradient elution was applied and programmed as follows: the gradient started with 99% eluent A in 1.0 min and decreased linearly down to 90% in 0.5 min. It

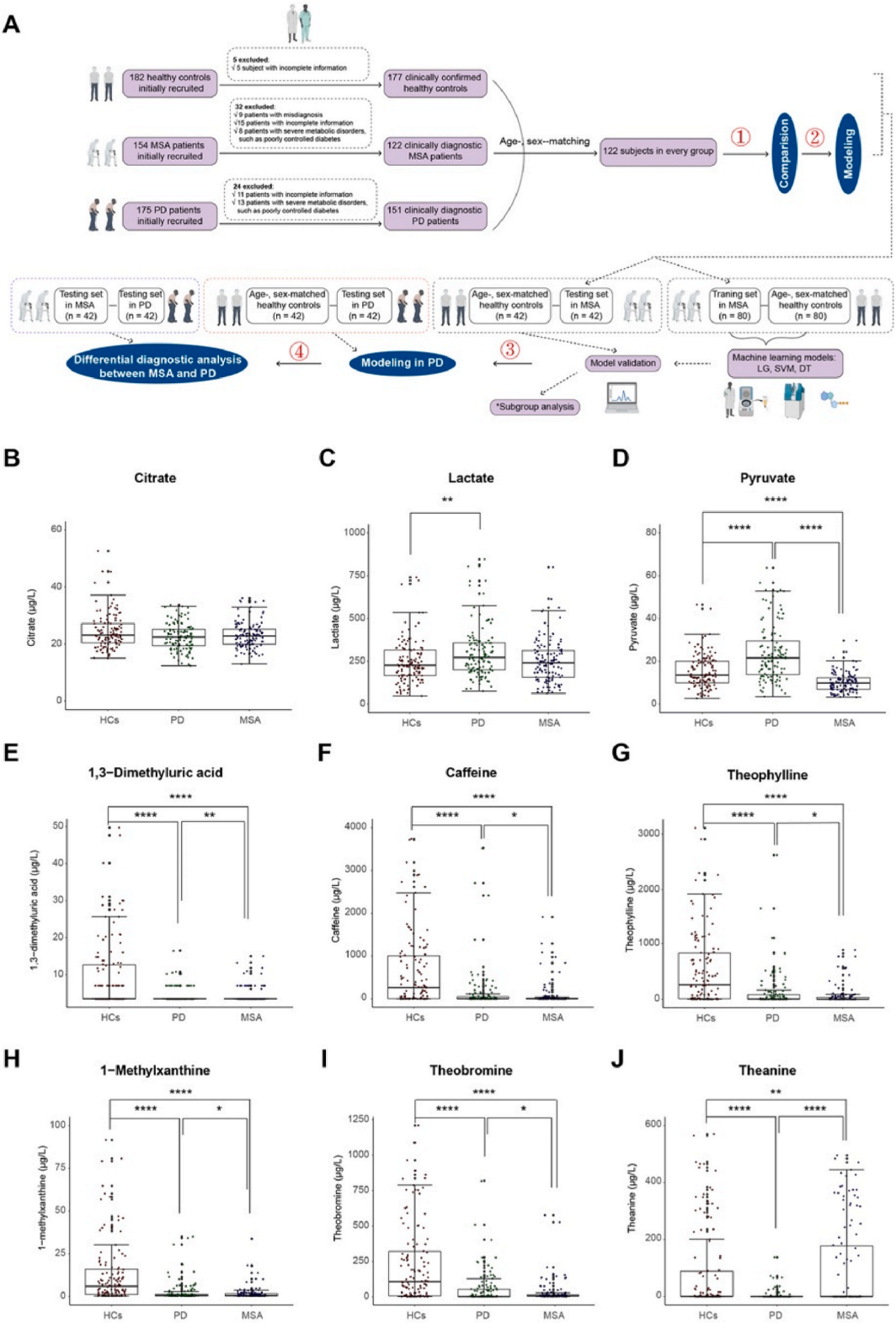


Fig. 1. Comparison of the concentrations of targeted metabolites detected in MSA, PD patients and HC subjects. A The design and flowchart of the study. **B** citrate, **C** lactate, **D** pyruvate, **E** 1,3-edimethyluric acid, **F** caffeine, **G** theophylline, **H** 1-methylxanthine, **I** theobromine, **J** theanine. Statistical significance was assessed by Mann-Whitney *U* test. $^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.001$, $^{****}p < 0.0001$.

decreased linearly again down to 70% in 1 min and held for 0.5 min. Then, it returned to 99% of eluent A in 0.5 min immediately, followed by a re-equilibration for 1.5 min. The total run time for each sample analysis was 5 min.

The LC-MS system were conducted in positive and negative ionization modes on a tandem quadrupole mass spectrometer (Acquity TQD; Waters) equipped with an ESI source. The optimal mass spectrometry settings were: source temperature and capillary voltage were 150 °C and 3.5 kV (positive ionization modes)/2.5 kV (negative ionization modes), respectively, the rate of flow of cone gas was 50 L/h, and the rate of flow of desolvation gas was 650 L/h. Data analyses were performed with QuanLynx 4.0 software (Waters Corporation). The mass transitions of all the analytes are listed in Supplementary Table 1.

2.5. Quality control procedure

A procedural blank and a spiked sample were run periodically for each batch of 20 samples to evaluate the possible contamination of samples and the accuracy of analytical procedures. The procedural blanks for target analytes were below the limit of detection (LOD). A series of calibration samples were assessed before and after the quality control samples and the unknown samples. Besides, solvent control was assessed once or twice in each batch. A typical sample batch per day consisted of 48 samples including the calibration samples.

2.6. Statistical analysis

R software version 3.6.3 was utilized for univariate and multivariate analyses, and permutation of ANOVA. One-way ANOVA permutation and unpaired Student's t-test were conducted to evaluate the significance of the differences among groups, and the Kruskal–Wallis test was used for one-way analysis of variance. Receiver operator characteristic (ROC) curve analysis was also used to evaluate the discriminative capability of valid machine learning classifiers, including support vector machine (SVM), decision tree (DT) and logistic regression (LG). The relative importance of the variables in the optimized model was ranked by the GINI index (for DT) and measure input importance coefficient (for SVM). Ten-fold cross analysis was used to further validate the representativeness and robustness of models.

3. Results

3.1. Demographic, epidemiological data and non-motor symptoms

The cohorts included age- and sex-matched HCs, and MSA and PD patients. As shown in Figs. 1A and 154 clinically diagnosed MSA patients were initially recruited, and 32 were excluded because of misdiagnosis ($n = 9$), incomplete information ($n = 15$) or complicating with severe metabolic disorders ($n = 8$) via re-evaluation by neurological experts. We also recruited 175 PD patients, and 24 were excluded because of incomplete information ($n = 11$) and complicating with severe metabolic disorders ($n = 13$). A total of 182 HCs were initially recruited and 5 were excluded because of incomplete information ($n = 5$). For the next analyses, the 122 MSA cases were age- and sex-matched for PD patients and HCs with the same sample sizes. For diagnosis and differential diagnosis modeling, 80 individuals were randomly selected for training and the remaining 42 for testing analysis in the MSA and PD patients.

The demographic and clinical characteristics were summarized in Table 1. There was no significant difference in age and sex between the three groups. The frequency of tea-intake in the MSA (39.3%) and PD (57.4%) cases was significantly lower than the HCs (71.3%) (MSA *vs* HCs, $P = 9.92 \times 10^{-7}$; PD *vs* HCs, $P = 0.023$). Similarly, the frequency of coffee-intake in the MSA (4.10%) and PD (10.7%) cases was significantly lower than the HCs (23.8%) (MSA *vs* HCs, $P = 2.12 \times 10^{-5}$; PD *vs* HCs, $P = 0.007$). For non-motor symptoms, uroclepsia occurred significantly more frequently in the MSA (50.0%) and PD (50.0%) cases than HCs (31.1%) (MSA *vs* HCs, $P = 0.004$; PD *vs* HCs, $P = 0.003$). Similarly, constipation occurred significantly more frequently in the MSA and PD cases than HCs. Moreover, the scales for clinical assessment were all significantly different in the MSA and PD cases from the HCs ($P < 0.001$).

3.2. Targeted metabolic profiling for MSA and PD patients

Comparing metabolic profiles among these three cohorts, no difference was observed in citrate between the MSA and PD patients, or controls (Fig. 1B). However, the lactate level was significantly higher in PD patients (310.54 ± 168.81 μg/L) than in the controls (247.91 ± 126.93 μg/L) ($P = 0.002$), but absent in MSA patients (250.67 ± 125.23 μg/L) ($P = 0.40$) (Fig. 1C). Moreover, the pyruvate in MSA patients (10.21 ± 4.40 μg/L) was significantly lower than that in controls (15.25 ± 7.80 μg/L) ($P = 8.43E-08$), while it was significantly higher in PD patients (25.41 ± 21.52 μg/L) than that in the controls ($P = 4.47E-09$) and MSA patients ($P = 2.20E-16$) (Fig. 1D). In addition, the levels of the 6 tea/coffee metabolites were all significantly lower in MSA patients than in controls (1,3-dimethyluric acid, $P = 2.65E-07$; caffeine, $P = 1.30E-16$; theophylline, $P = 2.76E-15$; 1-methylxanthine, $P = 7.52E-13$; theobromine, $P = 1.52E-11$; theanine, $P = 0.004$), which was consist with PD patients ($P < 0.05$) (Fig. 1E–J).

3.3. Correlation between pyruvate/lactate, tea/coffee metabolites and lifestyles

Correlation between the concentrations of the tea/coffee metabolites and lifestyles (smoking and alcohol-drinking) was analyzed and compared between three groups with and without tea/coffee intake. To avoid bias that might be caused by the number of individuals in different groups, we selected 31 samples, the least number of the "MSA with tea/coffee" group, and matched the same

number of individuals from the other groups. Interestingly, the tea/coffee metabolites in cluster A (red triangle) were positively correlated with each other in controls (upper and lower rows in the left panel), whereas the correlations were significantly reduced in MSA and PD patients (upper and lower rows in the middle and right panel). Moreover, the concentrations of tea/coffee metabolites (cluster B, blue rectangle) were positively correlated with smoking and alcohol-drinking in controls (especially those with tea/coffee intake), but not in MSA or PD patients. Furthermore, the pyruvate concentration was negatively correlated with those of the tea/coffee metabolites and smoking and alcohol-drinking in controls with tea/coffee intake (lower left), but not in the MSA or PD patients (cluster C, green rectangle) (Fig. 2A). These results suggest that the tea/coffee metabolites can interact with each other in determining the risk for MSA and PD. In addition, lifestyles may modify the risk-reducing effect of the metabolites for the diseases, especially the pyruvate level.

3.4. Diagnostic and differential diagnostic models for MSA and PD

To assess the utility of metabolites as potential biomarkers for the disease diagnosis, we selected the variables showing significant difference for the internal model validation. Comparing the area under curve (AUC) of the ROCs, the models with metabolites alone did not show a good effect for diagnosing MSA (Supplementary Table 2), while the model combing all the significant variables showed a good performance (AUC = 0.908 for the SVM model, 0.907 for DT and 0.878 for LG) (Fig. 2B and Supplementary Fig. 1A). Furthermore, the 10-fold cross-validation was carried out to validate the representativeness and robustness of models, we found the AUC was 0.862 for LG, 0.813 for DT, and 0.627 for SVM, indicating that good performance was LG model with chosen variables (Table 2). We also tested the model for diagnosing PD using these combined variables. All the models showed good prediction

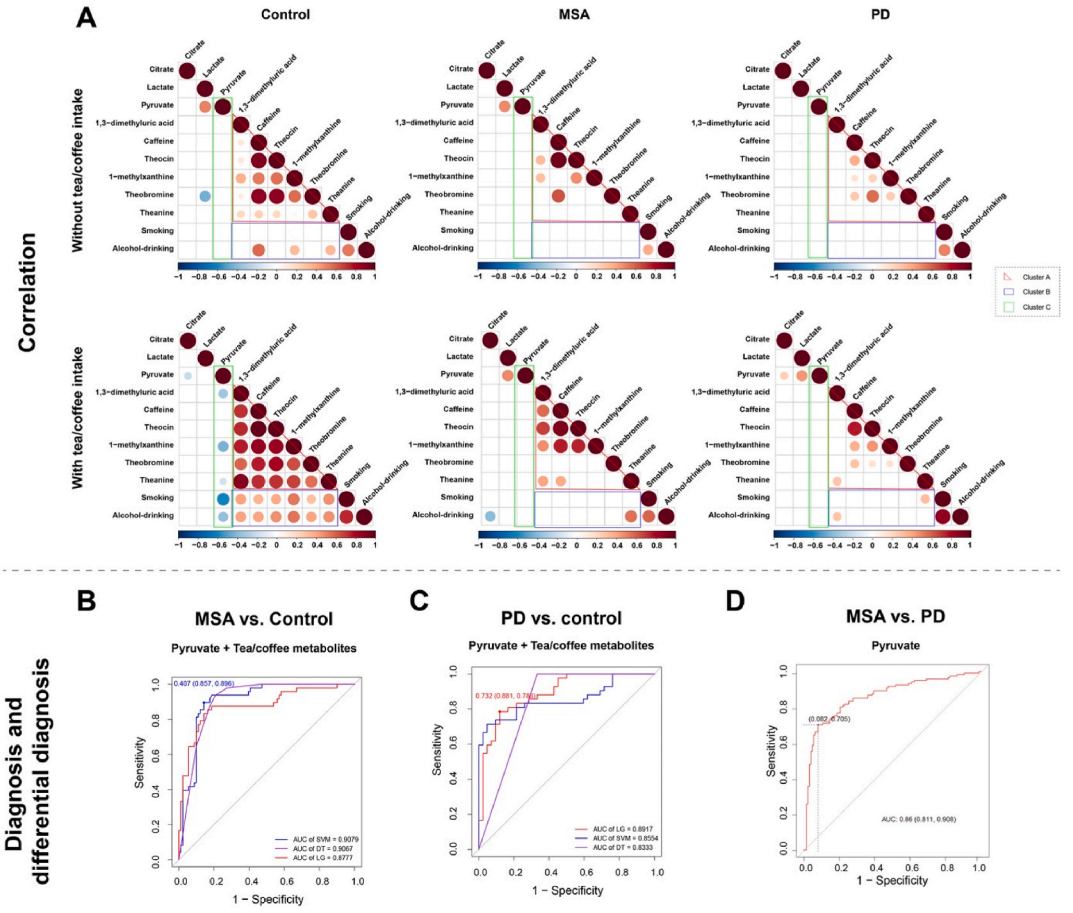


Fig. 2. Inter-correlations between the pyruvate/lactate and tea/coffee metabolites and the lifestyles. A The columns indicated healthy controls, MSA and PD patients. The upper rows included individuals without tea/coffee intake and the lower rows indicate those with tea/coffee intake. The correlation was assessed by Spearman rank correlation coefficients. Blue dots indicate the negative, while the red dots indicate positive correlations. The inter-correlations between the variables were categorized into three clusters: tea/coffee metabolites (cluster A), red triangle; lifestyle + tea/coffee metabolites (cluster B), blue rectangle; pyruvate + tea/coffee metabolites + lifestyle (cluster C), green rectangle. **B–C** The diagnostic models (DT, LG, SVM) for MSA and PD using pyruvate and the tea/coffee metabolites. **D** The differential diagnostic model for MSA and PD by using pyruvate.

performance (AUC = 0.892 for the LG model, 0.908 for SVM and 0.833 for DT) (Fig. 2C and Supplementary Fig. 1B). Also, by 10-fold cross-validation analysis, we found the AUC was 0.911 for LG, 0.987 for DT, and 0.726 for SVM, indicating that good performance was DT model with chosen variables (Table 2).

Additionally, a ternary diagram of the eight factors in HCs, PD and MSA was used to investigate the overall variation proximity and skewing trends. As shown in Supplementary Fig. 2A, caffeine, theophylline, 1-methylxanthine and theobromine were close and skewed to HCs, suggesting that individuals with high levels of these metabolites tend to be healthy. In contrast, the other three markers, theanine, 1,3-dimethyluric acid and pyruvate, were skewed to the PD or MSA dimension, indicating the potential for discriminating the two diseases. Using the three markers as independent variables for differentiating MSA and PD, we found that pyruvate had the best performance (ROC = 0.860), with validation with 10-fold cross-validation data analysis (AUC was 0.872) (Fig. 2D–Table 2 and Supplementary Figs. 2B–C). Furthermore, combining all the three markers, the AUCs for the ROC was significantly increased, which reached 0.922 by DT algorithm (Supplementary Fig. 2D).

4. Discussion

To date, the diagnosis and differential diagnosis model for MSA and PD using metabolites has not been well established in the clinical practice. Our results suggest that pyruvate and tea/coffee metabolites have good performance in disease diagnosing, and pyruvate can differentiate MSA from PD. Moreover, tea/coffee metabolites and lifestyles, including smoking and alcohol-drinking, may negatively modify the effect of pyruvate to reduce the risk for disease. One of the crucial findings of the study was the identification of pyruvate for differentiating PD from MSA with a good performance. Pyruvate is the end-product of glycolysis and ultimately destined for transport into mitochondria as a master fuel for TCA cycle. As is well known, glucose from the blood is taken up by neurons, astrocytes and oligodendrocytes, and can be metabolized via glycolysis, giving rise to pyruvate. In this model, the plasma pyruvate was significantly elevated in PD patients, while dramatically decreased in MSA patients. Our findings of increased plasma pyruvate level in PD patients are consistent with many previous studies [6]. Since pyruvate is an endogenous scavenger of reactive oxidants hydrogen peroxide, superoxide, and peroxynitrite, many genes regulating pyruvate metabolism are expressed in dopaminergic neurons [7]. Moreover, pyruvate prevents dopaminergic neurodegeneration and motor deficits in the 1-Methyl-4-Phenyl-1,2,3,6-Tetrahydropyridine model of PD [8]. All these observations suggested that pyruvate may play a protective role in PD pathology.

However, what triggers the reduction of pyruvate level in MSA remain unclear. MSA is a disorder characterized by the emergence of glial cytoplasmic inclusions (GCIs) within oligodendrocytes [1]. Oligodendroglia are glial cells to provide support for neurons [9]. As reported, oligodendrocytes transfer energy metabolites, like pyruvate and lactate, to neurons through the "myelinic" channels and monocarboxylate transporters. Transport occurs through monocarboxylate transporter 1 (MCT1) expressed at the axonal oligodendrocyte process and moves substrates into the periaxonal space, where metabolites can be taken up by neurons through monocarboxylate transporter 2 (MCT2) and processed for ATP synthesis [9]. Therefore, we hypothesize that the pyruvate shortage may compromise the metabolic support of oligodendrocyte for neurons under the pathological conditions and promote the disease process, which deserve in depth investigation.

Similar to previous studies, we suggested that coffee/tea metabolites are protective factors for neurodegenerative disorders [5,10]. In this study, we also observed that the level of pyruvate can be negatively modified by the tea/coffee intake and metabolites in HCs, but not in MSA or PD patients. Previous studies demonstrated that caffeine could diminished the expression of pyruvate kinase [11], an enzyme that catalyzed the conversion of phosphoenolpyruvate to pyruvate. In addition, pyruvate dehydrogenase kinase complex (PDK1-4) that promotes pyruvate conversion into acetyl-coenzyme A were significantly lower in the liver of coffee polyphenols-fed than in high-fat mice [12]. Moreover, we showed that lifestyles like smoking and alcohol-drinking may modify the disease-reducing effect of these metabolites. These observations suggested that the modification effect has been counteracted or be deleted by the unknown mechanisms in PD and MSA patients. The results also implied that the compromized pyruvate modification might be one of the mechanisms of these diseases. All these observations suggest that lifestyles, including coffee/tea, smoking or alcohol drinking may reduce the risk for PD and MSA by modifying the pyruvate level.

There were also several limitations in this study. First, the data of other lifestyles (including exercise, milk-uptake et al.) that may be

Table 2
The results of 10-fold cross-validation.

	Accuracy	Sensitivity	Specificity	AUC
HCs *vs.* MSA				
Result_multinom_avg (LG)	79.116	0.714	0.871	**0.862**
Result_rpart_avg (DT)	77.818	0.762	0.796	0.813
Result_svm_avg (SVM)	79.116	0.698	0.887	0.627
HCs *vs.* PD				
Result_multinom_avg (LG)	86.866	0.861	0.878	0.911
Result_rpart_avg (DT)	98.349	0.984	0.984	**0.987**
Result_svm_avg (SVM)	88.083	0.871	0.893	0.726
MSA *vs.* PD				
Result_multinom_avg (LG)	79.866	0.738	0.859	**0.872**

Abbreviations: HCs = healthy controls; MSA = multiple system atrophy; PD = Parkinson's disease; LG = Logistic regression; DT = Decision tree; SVM = Supporting vector machine; AUC = Area under curve.

potentially related with synucleinopathies were incomplete. Second, we lacked validation of the findings in another cohort. Further, well-designed experiments are needed to address the molecular pathways concerning the metabolites and their associations with environment exposures and lifestyles.

In summary, the results provide evidence supporting the central roles of pyruvate in differentiating PD from MSA, and the opposite changes of pyruvate within the two diseases suggested a distinct role in the pathogenesis of disease. Moreover, pyruvate may be modified by lifestyles and the related tea/coffee metabolites. The data provide important insights into the mechanisms, diagnosis, and interventions of the two neurodegenerative diseases.

Ethics statement

This study was approved by the Institutional Review Board of Xuanwu Hospital of Capital Medical University, with the approval number: [2020]060. All participants or legal guardians provided informed consent to participate in the study.

Funding source declaration

This work was supported by grants from National Key Research and Development Program (2022YFC2009600, 2022YFC2009601) to Dr.Xu-Ying Li; National Natural Science Foundation of China, China (NSFC) (82171412) to Prof. Chaodong Wang, NSFC (No. 31921002, 31730108, 32061143026) and the Strategic Priority Research Program and Innovation Program of the Chinese Academy of Sciences (No. XDB32020100, YJKYYQ20200052) to Prof. Zhiheng Xu, and Beijing Municipal Medical Science Institute-Public Welfare Development Reform Pilot Project (Capital Medical Research No. 2019-2) to Prof. Guoguang Zhao.

Data availability statement

All data had been listed in this study. Any additional information required to reanalyze the data reported in this paper is available from the lead contact upon request, and we will do this without unnecessary delay and at a reasonable cost.

CRediT authorship contribution statement

Xu-Ying Li: Writing – review & editing, Writing – original draft, Visualization, Funding acquisition, Formal analysis, Conceptualization. **Teng Xue:** Writing – original draft, Visualization, Methodology, Investigation, Formal analysis. **Hong Lai:** Data curation. **Jing Dai:** Resources, Formal analysis. **Fangda Peng:** Methodology, Formal analysis. **Fanxi Xu:** Resources, Data curation. **Junge Zhu:** Resources, Data curation. **Xian Li:** Resources, Data curation. **Junya Hu:** Resources, Data curation. **Wei Li:** Resources, Data curation. **Raoli He:** Resources, Data curation. **Lina Chen:** Resources, Data curation. **Ying Chen:** Resources, Data curation. **Chunguang Ding:** Validation, Supervision, Methodology. **Guoguang Zhao:** Supervision, Funding acquisition. **Xianyang Chen:** Writing – review & editing, Supervision, Project administration, Methodology, Investigation, Conceptualization. **Qinyong Ye:** Validation, Supervision, Resources, Project administration, Conceptualization. **Zhiheng Xu:** Writing – review & editing, Supervision, Project administration, Investigation, Funding acquisition, Conceptualization. **Chaodong Wang:** Writing – review & editing, Validation, Supervision, Resources, Project administration, Investigation, Funding acquisition, Conceptualization.

Declaration of AI and AI-assisted technologies in the writing process

There were no AI and AI-assisted technologies in the writing process of our manuscript

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.

Acknowledgements

We appreciated the patients and their families for their cooperation and the colleagues for their work in this study.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2024.e26588.

References

[1] A. Fanciulli, G.K. Wenning, Multiple-system atrophy, N. Engl. J. Med. 372 (3) (2015) 249–263.

[2] N.K. Magdalinou, R.W. Paterson, J.M. Schott, N.C. Fox, C. Mummery, K. Blennow, K. Bhatia, H.R. Morris, P. Giunti, T.T. Warner, R. de Silva, A.J. Lees, H. Zetterberg, A panel of nine cerebrospinal fluid biomarkers may identify patients with atypical parkinsonian syndromes, J. Neurol. Neurosurg. Psychiatry 86 (11) (2015) 1240–1247.

[3] Y. Miki, E. Tsushima, S.C. Foti, K.M. Strand, Y.T. Asi, A.K. Yamamoto, C. Bettencourt, M.C.B. Oliveira, E. De Pablo-Fernandez, Z. Jaunmuktane, A.J. Lees, K. Wakabayashi, T.T. Warner, N. Quinn, J.L. Holton, H. Ling, Identification of multiple system atrophy mimicking Parkinson's disease or progressive supranuclear palsy, Brain 144 (4) (2021) 1138–1151.

[4] V. Chelban, M. Bocchetta, S. Hassanein, N.A. Haridy, H. Houlden, J.D. Rohrer, An update on advances in magnetic resonance imaging of multiple system atrophy, J. Neurol. 266 (4) (2019) 1036–1045.

[5] S. Camandola, N. Plick, M.P. Mattson, Impact of coffee and cacao purine metabolites on neuroplasticity and neurodegenerative disease, Neurochem. Res. 44 (1) (2019) 214–227.

[6] S. Koga, D.W. Dickson, Recent advances in neuropathology, biomarkers and therapeutic approach of multiple system atrophy, J. Neurol. Neurosurg. Psychiatry 89 (2) (2018) 175–184.

[7] B. Zheng, Z. Liao, J.J. Locascio, K.A. Lesniak, S.S. Roderick, M.L. Watt, A.C. Eklund, Y. Zhang-James, P.D. Kim, M.A. Hauser, E. Grunblatt, L.B. Moran, S. A. Mandel, P. Riederer, R.M. Miller, H.J. Federoff, U. Wullner, S. Papapetropoulos, M.B. Youdim, I. Cantuti-Castelvetri, A.B. Young, J.M. Vance, R.L. Davis, J. C. Hedreen, C.H. Adler, T.G. Beach, M.B. Graeber, F.A. Middleton, J.C. Rochet, C.R. Scherzer, P.D.G.E.C. Global, PGC-1alpha, a potential therapeutic target for early intervention in Parkinson's disease, Sci. Transl. Med. 2 (52) (2010) 52ra73.

[8] Y.M. Kim, S.Y. Choi, O. Hwang, J.Y. Lee, Pyruvate prevents dopaminergic neurodegeneration and motor deficits in the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine model of Parkinson's disease, Mol. Neurobiol. 59 (11) (2022) 6956–6970.

[9] T. Philips, J.D. Rothstein, Oligodendroglia: metabolic supporters of neurons, J. Clin. Invest. 127 (9) (2017) 3271–3280.

[10] M. Fujimaki, S. Saiki, Y. Li, N. Kaga, H. Taka, T. Hatano, K.I. Ishikawa, Y. Oji, A. Mori, A. Okuzumi, T. Koinuma, S.I. Ueno, Y. Imamichi, T. Ueno, Y. Miura, M. Funayama, N. Hattori, Serum caffeine and metabolites are reliable biomarkers of early Parkinson disease, Neurology 90 (5) (2018) e404–e411.

[11] F.J. Zapata, M. Rebollo-Hernanz, J.E. Novakofski, M.T. Nakamura, E. Gonzalez de Mejia, Caffeine, but not other phytochemicals, in mate tea (Ilex paraguariensis St. Hilaire) attenuates high-fat-high-sucrose-diet-driven lipogenesis and body fat accumulation, J. Funct.Foods 64 (2020).

[12] T. Murase, K. Misawa, Y. Minegishi, M. Aoki, H. Ominami, Y. Suzuki, Y. Shibuya, T. Hase, Coffee polyphenols suppress diet-induced body fat accumulation by downregulating SREBP-1c and related molecules in C57BL/6J mice, Am. J. Physiol. Endocrinol. Metab. 300 (1) (2011) E122–E133.