Development of personalized health monitoring using ultra-weak photon emission based on systems medicine concepts

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Chapter 3

Measuring ultra-weak photon emission as a non-invasive diagnostic tool for detecting early-stage type 2 diabetes: a step toward personalized medicine

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Abstract

The global prevalence of type 2 diabetes is estimated to reach 4.4% by 2030, placing a significant burden on our healthcare system. Therefore, the ability to identify patients in early stages of the disease is essential for both prevention and effective management, and diagnostic methods based on traditional Chinese medicine (TCM) may be suitable for identifying patients with early-stage type 2 diabetes. Here, a panel of three physicians trained in TCM classified 44 pre-diabetic subjects into three syndrome subtypes using TCM-based diagnostics. In addition, ultra-weak photon emission (UPE) was measured at four anatomical sites in each subject. Ten properties encompassing 40 parameters were then extracted from the UPE time series. Statistical analyses, including multinomial logistic regression, were performed using the results of each parameter measured at the four sites. Sixteen UPE parameters were then selected and used to discriminate between the three subtypes of pre-diabetic subjects. Next, Spearman's correlation coefficient was used to quantify the correlation between the 16 UPE parameters and the TCM-based diagnoses. The resulting correlation networks accurately reflected the differences between the three syndrome subtypes. These results suggest that UPE is a suitable tool for detecting subtypes in early-stage type 2 diabetes. In addition, our results provide evidence that TCM may represent an important step toward personalized medicine.

Key words: traditional Chinese medicine–based diagnostics, personalized medicine, type 2 diabetes, ultra-weak photon emission (UPE), correlation network

1. Introduction

Type 2 diabetes (T2D) is multifactorial in origin and is associated with genetic factors, metabolic disorders, and lifestyle-related risk factors such as obesity, inactivity, and poor diet.[1–4] Currently, the oral glucose tolerance test and established glucose criteria are the golden standard for diagnosing T2D.[5] However, T2D can be present in an early, undetected form for more than ten years, during which dysglycemia increases the risk of severe complications,[6] including hypertension, blindness, renal failure, and cardiovascular disease.[3] Glycemic control can prevent some—but usually not all—of the aforementioned complications.[7,8] It has therefore been argued that a more personalized diagnostic approach may provide the opportunity to effectively manage T2D in its early stages, before the onset of complications.[9,10]

The diagnostic approach used in traditional Chinese medicine (TCM) is highly personalized and takes into account the interactions between the patient and his/her environment and pathogenic factors.[10] The patient's response with respect to these interactions provides a functional profile of the disease-related signs and/or symptoms and can be used to identify specific "syndrome subtypes" within a specific disease.[11,12] Thus, using the principles of TCM, several syndrome subtypes have been identified within T2D.[13] Recent studies combined TCM-based syndrome subtyping with modern metabolomics technologies, which are commonly used in Western medicine,[14] thereby combining TCM-based subtyping of pre-diabetes with metabolomics-based medicine.[15,16] The ability to identify and characterize these syndrome subtypes is an important step toward encouraging changes in unhealthy lifestyle on a personalized basis.

Measuring ultra-weak photon emission (UPE) is a non-invasive method for recording changes in metabolic processes within the human body and can reflect the dynamics of metabolic organization.[17] Photobiology (the metabolic effects of absorbed photons) and low-level biological luminescence (the production and

emission of photons) are complementary manifestations of the photons' role in metabolism.[17,18] Thus, recording photon emissions provides a measure of the net activity of these types of metabolic reactions, which reflects the body's current physiological state. We hypothesized that this technology may provide an alternate method for subtyping early-stage T2D in combination with TCM-based concepts. The equipment needed to continuously measure UPE in human subjects is relatively simple and includes a sensitive photomultiplier tube (PMT) in a sealed dark environment.[19] Importantly, the current technology for measuring UPE is rapid, highly sensitive, relatively inexpensive, and non-invasive.[20] In addition, several studies have calculated parameters (e.g., mean signals and signal variance) in UPE signals measured in both healthy subjects and in the pathological state,[21–30] providing a baseline for comparison.

Here, we asked whether UPE parameters can be used as a tool for identifying syndrome subtypes in subjects with early-stage T2D diagnosed using TCM-based diagnostics. An explorative, non-intervention urine metabolomics study at TNO (https://clinicaltrials.gov/ct2/show/NCT00469287) was designed in which 44 pre-diabetic male subjects were diagnosed with three distinct syndrome subtypes by a panel of three TCM-trained physicians.[15] We then measured UPE parameters in this same cohort of 44 pre-diabetic subjects in order to investigate the relationship between UPE signal parameters and the syndrome subtypes identified using TCM-based diagnostics. Our results suggest that measuring UPE represents a non-invasive method for distinguishing between the three syndrome subtypes of pre-diabetes. Thus, UPE may provide key insight into personalized diagnostics and personalized medicine, ultimately improving the management of diabetes and other diseases.

2. Materials and Methods

2.1 Subjects and pre-diabetes subtypes

A total of 44 male Dutch subjects were recruited and screened as described previously.[15] Each subject provided written informed consent, and the study was approved by the Medical Ethics Committee of Tilburg, the Netherlands. Pre-diabetes was defined as a fasting glucose level of 6.1–6.9 $mmol \cdot L^{-1}$ in the absence of other clinical evidence of diabetic complications measured during several examinations.[15] Three physicians who were trained in TCM independently diagnosed the subjects in a blinded fashion.[15] The physicians asked the subjects questions about their various symptoms, and then grouped these symptoms into 26 TCM-based diagnostic items (Table 1). Next, the three physicians assigned specific scores to these diagnostic items based on their severity and frequency.[15] The consistency of TCM based diagnosis between the three physicians was 85% based on the generalized procrustes analysis (GPA),[15] resulting in the following three syndrome subtypes of pre-T2D: subtype A ("Qi-Yin deficiency", n=15 subjects), subtype B ("Qi-Yin deficiency with dampness", n=20 subjects), and subtype C ("Qi-Yin deficiency with stagnation", n=9 subjects).15 In addition, UPE measurements and urine metabolomics analysis were carried out by the other two independent teams in a blinded fashion.

2.2 Photomultiplier system and UPE measurements

For this study, a tabletop photomultiplier system specifically designed for measuring UPE signals in the hands (Type PMS06.1) was provided by Meluna Research (Geldermalsen, the Netherlands). The detection head is located at the top of a dark chamber and includes a custom-designed shutter system. To measure UPE, we used a model 9235QA photomultiplier tube (ET Enterprises, Uxbridge, UK) fitted with a 48-mm diameter window, with spectral sensitivity in the range of 200–650 nm. The background noise measured by the PMT was 4-5 counts/s. A ring was constructed at the end of the PMT. Subjects were asked to position their hand against the ring in order to fix their hands at a specific placement to secure that the same areas on subjects' hands were measured. This avoids errors related to the positioning of the hands below the PMT. The distance between the ring and skin was 27 mm. The PMT

Table 1. The 26 diagnostic items (symptoms) associated with T2D based on traditional Chinese medicine.

ID	Diagnostic item (symptom)
C1	Blood stagnation
C2	Damp heat in the liver
C3	Damp Heat in the Middle Jiao
C4	Damp heat in the Spleen
C5	Damp heat in the stomach
C6	Damp Heat
C7	Dry Heat consumes Yin
C8	Heart Qi deficiency
C9	Heart Yin deficiency
C10	Heat in the Heart
C11	Kidney Yin deficiency
C12	Liver fire
C13	Liver Qi stagnation
C14	Liver Yang ascending
C15	Liver Yin deficiency
C16	Lung Yin deficiency
C17	Lung Qi deficiency
C18	Qi and Yin deficiency in the Middle Jiao
C19	Spleen Qi deficiency
C20	Stomach Qi deficiency
C21	Stomach Yin deficiency
C22	Yin deficiency
C23	Yin deficiency in the Middle Jiao
C24	False heat consumes Yin
C25	Qi deficiency
C26	Lung fire run

was operated in the single-photon counting mode, and the signals were recorded using a model 6602 PCI card (National Instruments, Austin, TX). The temperature within the measuring chamber was maintained at 20±1.0°C.

All UPE signals were recorded between 11 a.m. and 3 p.m. in order to minimize any possible effects of diurnal rhythms.[31,32] Each subject wore light-tight gloves on both hands for 30 min prior to recording the signal in order to minimize the effect of

ambient light exposure. During the measurements, the subject placed the hand below the PMT, and the following four sites were recorded, resulting in a total of four measurements per subject: right dorsal (RD), right palm (RP), left dorsal (LD), and left palm (LP). Each signal was recorded for 5 min by counting the number of photons emitted in 6000 consecutive 50-ms bins. Background noise was recorded in the same manner for each subject.

2.3 Gas chromatography-mass spectrometry analysis of urine metabolomics

Gas chromatography-mass spectrometry (GC-MS) analysis was used to study the urine metabolomics in the 44 pre-diabetic subjects. GC-MS was performed using an Agilent 6890 GC system with an Agilent 5973 MS detector (Agilent Technologies, Palo Alto, CA). The urine sample preparation and GC-MS analysis methods have been reported previously.[15] The concentration of urine metabolites was processed using an Agilent ChemStation software.[15]

2.4 Data processing and statistical analysis

2.4.1 Statistical analysis of UPE parameters

Ten properties of the UPE signal—strength, FF0, FF1, FF2, alpha, rho, theta, phi, SSI, and SSR—were calculated for each signal recorded. The specific calculations used to obtain these UPE properties have been reported previously.[25–27] The number of photons detected in a time series is to indicate the strength of the UPE signal. It is the photon emission intensity subtracted by the background noise. The photon distribution characteristics can reflect using the Fano Factor.[33] The three parameters FF0, FF1 and FF2 can be extracted from a Fano factor curve fitted by a second order polynomial function.[27] FF0 is the intercept of the Fano Factor curve, and determines theoretically the feature of photon distribution at zero bin size (time series).[27] FF1 and FF2 are parameters to express the slope of the Fano factor curve which is an indication for the structural patterns in the signal dynamics. FF1 is the slope of the Fano Factor curve, and determines the photon distribution changes in the different

time series compared to the photon distribution in the zero-time series.[27] FF2 is a coefficient in the second order polynomial function, and influences the shape of Fano Factor curve in specific samples.[27] The photon number distribution was also analyzed for non-classicality by procedures that have been developed to link quantum features to photon statistics, hence constructing portraits of quantum optical states. The approach followed in [25,27] and resulting in the parameters alpha, rho, theta, phi, SSI and SSR have been used in the present study. The use of these parameters is not meant to argue about optical characteristics of UPE because reliable evidence for coherence or non-classicality is missing.[34] Instead, they are only used as ways to analyze the photo count distribution

All ten properties were calculated in the signals obtained from each of the four anatomical sites for each subject, yielding a total of 40 UPE parameters for each subject. Thus, signal strength (for example) was defined as follows: Str_RD, Str_RP, Str_LD and Str_LP, corresponding to the signal strength measured in each of the four anatomical sites. The other nine parameters were defined using this same nomenclature.

The 40 UPE parameters were used to identify possible predictors in the multinomial logistic regression.[35] To build the model, we used forward neural networks to identify the maximum of a joint posterior distribution over the candidate predictors' coefficients, using the R package nnet (version 3.2.2).[36–38] To reduce the number of possible predictors, backward elimination was performed using the Akaike information criterion for variable selection.[39] The multinomial logistic regression was validated using cross-validation in order to avoid overfitting of the model.[40]

2.4.2 Statistical analysis of urine metabolites

In the previous urine metabolomics study,[15] both principal component analysis (PCA) and partial least square discriminant analysis (PLS-DA) were used to identify a total

of 24 urine metabolites which contributed to the discrimination of the three TCM-based syndrome subtypes in the 44 pre-diabetic subjects.[15]

2.4.3 Statistics of the correlation network

To determine the correlation between the UPE parameters and TCM-based diagnoses, the median score was calculated for each of the 26 diagnostic items based on the scores assigned by the three physicians. Spearman's rank correlation coefficient (ρ) was used to quantify the correlation between the value of the predicted UPE parameters and the median score of each TCM-based diagnostic item using SPSS version 23.0 (IBM Corp., Armonk, NY). A linear relationship was defined as Spearman's $|\rho| > 0.30$.[41] Thereafter, Cytoscape version 3.2.1 (www.cytoscape.org) was used to draw a network view, which was used to visualize these correlations.[42] The Spearman's rank correlation coefficient was also used to study the correlation between the UPE parameters and the urine metabolites, the identified correlations were visualized using Cytoscape.

3. Results and Discussion

The three physicians trained in traditional Chinese medicine classified the 44 pre-diabetic subjects into three distinct syndrome subtypes. To predict these subtypes using UPE parameters, we used these subtypes as the response in multinomial logistic regression. From the original 40 UPE parameters (corresponding to ten parameters measured at four anatomical sites each; see Materials and Methods), 16 parameters were identified in the logistic regression model as effective predictors (Table 2). “Qi-Yin deficiency with dampness” was used as the reference category for the dummy coding in multinomial logistic regression.[43] Compared to this reference category, the estimated coefficient values indicate the features of each UPE parameter in the other two categories (“Qi-Yin deficiency” and “Qi-Yin deficiency with stagnation”). The coefficient values of these 16 UPE parameters accurately

identified the three syndrome subtypes identified by the TCM-trained physicians, and cross-validation of the model revealed predictive accuracy of 97.81%.

Table 2. Estimated coefficients for the multinomial logistic regression model for the pre-diabetes subtypes based on traditional Chinese medicine–based diagnostics.

UPE Parameter[1]	Estimated coefficients[2]		
	Qi-Yin deficiency (subtype A)	Qi-Yin deficiency with stagnation (subtype C)	Total effect[3]
Alpha_LD	-11704.69	7397.46	19102.15
Str_RD	4027.06	-12698.77	16725.83
Str_LD	8629.85	2795.39	11425.24
Alpha_LP	-3331.47	4651.03	7982.5
Alpha_RP	2718.85	-4134.38	6853.23
SSI_LP	2205.62	-2990.89	5196.51
SSI_RP	-3523.47	1590.54	5114.01
FF0_RP	2633.09	1759.27	4392.36
Phi_RP	-2284.12	1631.2	3915.32
Theta_RD	1692.18	971.66	2663.84
SSI_LD	1583.92	817.23	2401.15
FF1_RP	-1514.28	323.49	1837.77
Rho_RD	-343.77	-1066.63	1410.4
FF0_RD	1121	-89.01	1210.01
FF0_LP	-444.23	668.98	1113.21
FF2_LD	-373.06	53.1	426.16

1 LD, left dorsal; LP, left palm; RD, right dorsal; RP, right palm.

2 Qi-Yin deficiency with dampness (subtype B) was used as the reference category.

3 Total effect refers to the sum of the absolute values of the estimated coefficients in subtype A and subtype C.

In traditional Chinese medicine, syndrome subtype is the outcome reached by the physician after analyzing the patients' symptoms and signs comprehensively by inspection, listening, smelling, inquiry, and palpation.[12,13] Syndrome type is used to interpret the body's holistic state at both the large-scale and systematic organization levels in order to guide the choice of treatment in each patient using acupuncture and/or Chinese herbal medicines.[12,21] Similar to syndrome subtype, UPE is a highly sensitive holistic measurement that accurately reflects the dynamics of the human

body and the disease status at the systems level.[17,21] Importantly, some TCM concepts (for example, the body's response to acupuncture or herbal medicines) have been characterized using UPE intensity, particularly with respect to basic syndrome subtypes in animal models.[21] Therefore, UPE can provide a close approximation of the system level structures identified using TCM-based diagnostics. This may explain—at least in part—why the three syndrome subtypes of pre-diabetic subjects could be classified using UPE parameters measured in this study. These UPE parameters, which were derived from ten properties of the UPE signal, have been found that most of these UPE properties can predict pre-T2D subtype. This finding may indicate that obtaining a comprehensive UPE profile may be useful for predicting syndrome subtypes identified using TCM concepts.

In traditional Chinese medicine, many diseases can have two or more syndrome subtypes, and dynamic changes are likely to occur among these subtypes under specific conditions.[12,13] This suggests that TCM-based syndrome subtypes are not independent but rather are interrelated.[12] This notion supports our finding that 16 of the 40 UPE parameters (40%) were needed in order to identify the three syndrome subtypes with high predictive accuracy. This also underscores the importance of studying the relationships between syndrome subtypes in further detail. Therefore, we used correlation network-based analyses in order to identify the relationship between UPE parameters and TCM-based diagnostic items (symptoms) in the three pre-diabetic syndrome subtypes.

Network-based analysis is an emerging approach in biomedical research, and correlation networks have been used to discriminate between disease phenotypes and to understand the interactions that reflect parts of a complex biological system.[44,45] Fig. 1 illustrates the differences in the overall profile of correlations in the three pre-diabetes syndrome subtypes. Different distributions of correlations between UPE parameters and diagnostic items are observed in the three subtypes, with relatively fewer correlations in subtype B ("Qi-Yin deficiency with dampness"). This indicates

Fig. 1 Correlation network of the UPE parameters and traditional Chinese medicine–based diagnostic variables for three pre-diabetes syndrome types. Visualization of the data is concentrated on the correlations between UPE parameters and TCM-based diagnostic items (symptoms) using Cytoscape. Linear relationships are drawn for correlations in which Spearman's $|\rho|$ was >0.30. Negative correlations are indicated with dotted lines, and positive correlations are indicated with solid lines; thicker lines indicate a higher correlation (i.e., a bigger Spearman's $|\rho|$). The length of each line has no meaning. Specific diagnostic items are indicated with the letter "C" followed by the ID corresponding to the diagnostic item based on TCM (for example, C1 corresponds to blood stagnation); see also Table 1. Fifteen UPE parameters are correlated with 12 diagnostic items resulting in 61 correlations in subtype A. Fourteen UPE parameters are correlated with 11 diagnostic items, resulting in 39 correlations in subtype B. Sixteen UPE parameters are correlated with 10 diagnostic items resulting in 66 correlations in subtype C.

that TCM-based syndrome subtypes can also be characterized using UPE properties in a systematic network. According to Chinese medicine, “Qi deficiency” and “Yin deficiency” are key factors that can underlie the onset of T2D.[46] The combined deficiency of both Qi and Yin—which generally exists in patients with T2D—can be used to reflect a reduced functional level resulting in specific symptoms due to pathological factors such as heat, dampness, and stagnation.[13,15,46] Therefore, the three pre-diabetic subtypes identified by TCM-based diagnostics share a common basic, cross-biological background as well as individual features with respect to overall symptoms identified.

To interpret this pattern further, we focused on the identical and unique correlations between the three syndrome subtypes with respect to their correlation network structures. Table 3 summarizes the correlations that were common among the three syndrome subtypes. In these identical pairwise correlations, specific UPE parameters were correlated with nine diagnostic symptoms, most of which (e.g., C7, C11, C18, C19, and C22) reflect the functional effect of Qi deficiency and/or Yin deficiency measured at specific body locations. From the perspective of traditional Chinese medicine, pre-diabetic patients often show aggravated emotional tension, which is closely associated with liver-related T2D symptoms (C12, C13, and C14).[13,47] In particular, liver Qi stagnation (symptom C13) has been proposed as the first stage in the onset of diabetes.[13] These liver-related T2D symptoms were also observed in the identical pairwise correlations with UPE parameters among the three syndrome subtypes.

To visualize the unique pairwise correlations in each syndrome subtype, we removed the identical correlations and integrated the remaining pairwise correlations using Cytoscape. Fig. 2 shows that different syndrome subtypes can also share diagnostic symptoms; however, the pairwise correlations have non-overlapping, unique correlated UPE parameters with different correlation strength and/or direction (e.g., positive or negative correlations). Each subtype appears to correspond to a unique

set of diagnostic symptoms as reflected by the pairwise correlations. For example, the correlation between UPE parameters and the diagnostic item C21 was present in subtype A, but not in subtype B or subtype C. Thus, these unique pairwise correlations could potentially be used to identify the individual features of each pre-diabetes syndrome subtype, and UPE could be used to standardize the TCM-based symptoms, reflecting distinct syndrome subtypes.

Table 3. Overview of identical correlations between UPE parameters and diagnostic items among the three syndrome types.

Pairwise correlation		Subtype A	Subtype B	Subtype C	Pairwise correlation		Subtype A	Subtype B	Subtype C
UPE parameter	Diagnostic item				UPE parameter	Diagnostic item			
Alpha_RP	C11	+	+	+	SSI_LD	C7	+		+
Alpha_LP	C11	+	+	+	Str_RD	C11	+		+
FF2_LD	C11	+	+	+	Alpha_LD	C13	+		+
Str_RD	C14	+	+	+	Alpha_RP	C13	+		+
FF0_RP	C19	+	+	+	Str_LD	C13	+		+
FF0_LP	C19	+	+	+	Str_RD	C13	+		+
FF2_LD	C12	+	+		FF0_LP	C14	+		+
Alpha_LD	C14	+	+		FF0_RD	C18	+		+
Alpha_LD	C18	+	+		FF2_LD	C19	+		+
Alpha_RP	C18	+	+		Alpha_LP	C22	+		+
Str_LD	C18	+	+		FF1_RP	C3		+	+
Str_RD	C18	+	+		FF1_RP	C11		+	+
FF2_LD	C18	+	+		Alpha_LP	C14		+	+
SSI_RP	C19	+	+		FF0_RP	C22		+	+
					FF0_LP	C22		+	+

"+" indicates specific syndrome types that have the same correlations between UPE properties and TCM-based diagnostic items.

Although the physical meaning of UPE parameters has been well interpreted previously,[25–27] understanding the biochemical meaning of UPE parameters is of great importance to identify potential applications for UPE in disease characterization. A strategy of combination between UPE and TCM-based diagnostics may improve our understanding of UPE patterns in the biochemical relevance. In traditional Chinese medicine, the syndrome of "Qi-Yin deficiency" has similarities with "chronic fatigue syndrome" and/or "mild inflammatory status", both of which are induced by hyper-metabolism.[15] Qi-Yin deficiencies with a

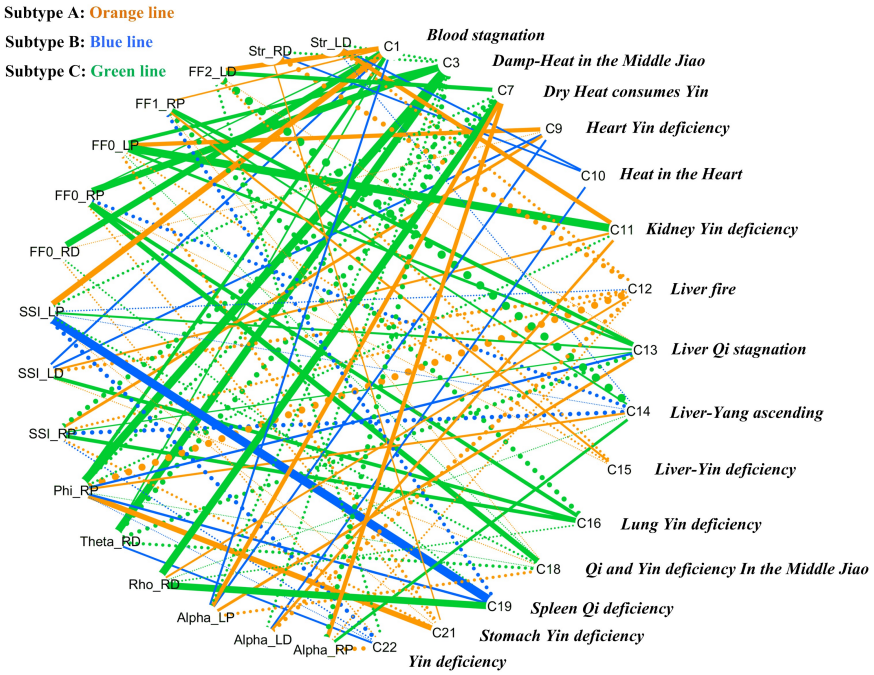

Fig. 2 Unique correlations between UPE parameters and traditional Chinese medicine–based diagnostic variables for the three pre-diabetes syndrome types. Negative correlations are indicated with dotted lines, and positive correlations are indicated with solid lines; thicker lines indicate a higher correlation (i.e., a bigger Spearman's $|\rho|$). The length of each line has no meaning. Specific diagnostic items are indicated with the letter "C" followed by the ID corresponding to the diagnostic item based on TCM (for example, C1 corresponds to blood stagnation); see also Table 1.

pathological factor is closely associated with an unhealthy lifestyle, and these subjects usually prefer raw, cold, greasy, and/or high-energy food.[13,15] Dampness is a sub-health status and can lead to development of metabolic syndrome and hypertension.[15] Another pathological factor is stagnation which may lead to emotional problems, impaired blood circulation, and diabetic peripheral neuropathy.[15,48] TCM syndrome-based biological relevance can be linked with TCM-based diagnostic symptoms (Table 1), the UPE parameters correlated with these diagnostic symptoms (Fig. 1) may therefore link to the biological interpretation of T2D from the disease cause, progression, to potential complications. Thereby, UPE parameters may be used to predict disease in a personalized basis.

Moreover, the biological and molecular interpretation of UPE parameters requires further study, and one promising strategy is to link UPE to metabolomics.[17,21,49] Metabolomics provides a comprehensive analysis of various low molecular weight compounds in biological systems based on specific metabolomics platform.[50] In addition, UPE results from the metabolic processes occurring in living organisms.[51,52] Therefore, UPE parameters may theoretically correlate to specific metabolites identified in metabolomics analysis. To test this hypothesis, we carried out a Spearman's correlation analysis between the identified 16 UPE parameters and the concentration of 24 urine metabolites in the 44 pre-diabetic subjects. The selected 24 urine metabolites contributed to the discrimination of TCM-based T2D syndrome subtypes in the previous urine metabolomics study.[15] Fig. 3 illustrates the correlation network which has a total of 26 pairwise correlations between 10 UPE parameters and 13 urine metabolites including amino acids (e.g., L-cysteine and L-serine), organic acids (e.g., Citric acid and Pyruvic acid) and sugars (e.g., D-Glucose and D-Ribose). These results indicate that the biological interpretation of the UPE parameters may be correlated to the biological pathways reflected by these specific urine metabolites in the T2D subjects, and thereby UPE parameters may provide more biological information to the TCM-based diagnostics. However, further validation of this strategy is required. Given that additional metabolomics platforms (e.g., amino acid and oxylipin platforms in plasma sample) are used to identify more metabolites such as oxidative stress/ reactive oxygen species (ROS) products etc., as well as their biological pathways in the disease state,[21,53–55] it may be possible to establish a comprehensive correlation between UPE parameters and multi-metabolic biomarkers. Therefore, UPE may serve to complement metabolomics, providing an integrated strategy for interpreting both UPE properties and TCM-based diagnostics, ultimately contribute to personalized medicine.[17,21]

4. Conclusions

Phenotyping is believed to play an essential role in realizing the goal of personalized

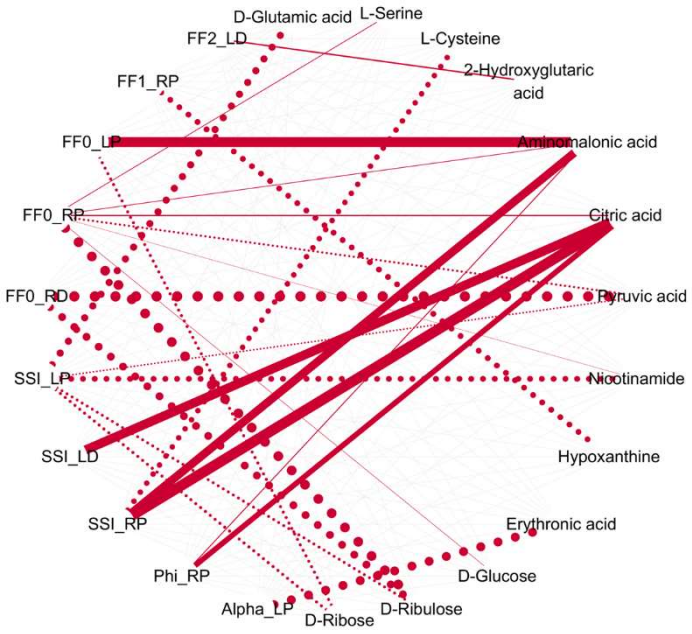

Fig. 3 Correlation network of the UPE parameters and the urine metabolites for pre-diabetic subjects. Visualization of the data is concentrated on the correlations between UPE parameters and urine metabolites using Cytoscape. Linear relationships are drawn for correlations in which Spearman's $|\rho|$ was >0.30. Negative correlations are indicated with dotted lines, and positive correlations are indicated with solid lines; thicker lines indicate a higher correlation (i.e., a bigger Spearman's $|\rho|$). The length of each line has no meaning.

medicine,[10] and traditional Chinese medicine–based diagnostics provides important information (e.g., syndrome subtype) regarding personalized phenotypes.[10] Given that TCM-based diagnostics is a phenomenological and descriptive approach used to reflect regulation at a systems level—including dynamics—it can be used to monitor changes in lifestyle and system-based treatments.[12,21] The key challenge is to both distinguish between and standardize TCM-based syndrome subtypes using modern technology.[12] Here, we present the first study combining UPE parameters with TCM-based diagnostics in order to investigate syndrome subtypes among pre-diabetic subjects. Several main conclusions can be drawn from our study.

First, based on the characteristic nature of the UPE signal, ten properties were

extracted from four anatomical sites, resulting in 40 separate UPE parameters measured in 44 subjects with pre-T2D; 16 of these 40 parameters were able to discriminate between the three pre-diabetes subtypes. Second, our correlation network between UPE parameters and TCM-based diagnostics provides key insight into the identification of three pre-T2D syndrome subtypes. These results indicate that UPE profiles may reflect personalized TCM-based diagnostics. A future strategy to reveal powerful UPE parameters is combing TCM-based diagnostics, UPE measurements and metabolomics analysis. The correlation between UPE parameters and metabolomics data for a given subtype as determined by TCM-based diagnostics, could lead to a better understanding of the underlying biochemistry and regulatory processes on which TCM diagnosis is based. The previously published urine metabolomics study reported only limited differentiation between three TCM-based syndrome subtypes in the pre-diabetic subjects.[15] In the present study it seems that a better segregation is obtained in the personalized diagnosis using 44 subjects, when a combination of UPE parameters and urine metabolites data is used. Further study should focus on the biological interpretation of UPE parameters.

Finally, the results of our study create new avenues for investigation. Future studies should include larger patient cohorts. In addition, similar analyses can be performed with other disease cohorts. In conclusion, UPE is a highly sensitive, non-invasive, robust technique that represents a new technological platform for diagnosing disease and disease subtypes, and for studying the efficacy of various therapies and healthcare approaches.

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