



Research Paper

Noninvasive skin fluorescence spectroscopy for detection of abnormal glucose tolerance[☆]

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ABSTRACT

The ENGINE study evaluated noninvasive skin fluorescence spectroscopy (SFS) for detection of abnormal glucose tolerance (AGT). The AGT detection performance of SFS was compared to fasting plasma glucose (FPG) and hemoglobin A_{1c} (A1C). The study was a head-to-head comparison of SFS to FPG and A1C in an at-risk population of 507 subjects, with no prior diagnosis of diabetes, each of whom received a 75 g, two-hour oral glucose tolerance test (OGTT). Subjects were measured by SFS on multiple days in fasting and non-fasting states. SFS data were acquired and analyzed with the SCOUT DS[®] device (Veralight, Albuquerque, NM, USA). Disease truth was AGT, defined as OGTT \geq 7.8 mmol/L. Sensitivity, false positive rate (FPR), ROC area, and equal error rate (EER) for detection of AGT were computed. The reproducibility of SFS and FPG was assessed. The AGT sensitivity of SFS at the device's recommended screening threshold of 50 was 75.2%, higher than that of FPG (thresholds of 5.6 mmol/L or 6.1 mmol/L) and A1C (thresholds of 5.7% or 6.0%). The SFS FPR was 42.1%, comparable to an A1C threshold of 5.7% (FPR = 43.5%). The EERs of SFS, FPG and A1C were similar, as were the partial ROC areas for FPRs of 20–50%. The reproducibility of SFS was 7.7% versus 8.1% for FPG. SFS had similar AGT detection performance to FPG and A1C and is a viable alternative to screening individuals for AGT.

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Introduction

Diabetes mellitus is a growing, global health concern. The International Diabetes Federation estimates that in 2011, 366 million people ages 20–79 around the globe had diabetes. It is estimated that 50% of diseased individuals are undiagnosed, with type 2 diabetes representing 90–95% of cases [1]. Another 280 million people had impaired glucose tolerance (IGT), a significant risk factor for developing type 2 diabetes within 10 years [1]. By the year 2030 the prevalence of diabetes and IGT are projected to grow to 552 and 398 million people ages 20–79, respectively [2]. Persons with diabetes are at increased risk for nephropathy, retinopathy, autonomic neuropathy, peripheral neuropathies that may lead to lower

limb amputation, cardiovascular disease and stroke. Diabetes and its attendant complications place a significant burden on the health care systems of the world, with an aggregate cost estimated at \$465 billion (USD) in 2011, representing 11% of health expenditures in adults 20–79 years of age [1]. Caring for people with diabetes may become economically unsustainable given the projected growth of the disease.

Early identification and intervention have been identified as critical components of programs seeking to prevent or delay the onset of type 2 diabetes. The Diabetes Prevention Program (DPP) and Finnish Diabetes Prevention Study (DPS) both demonstrated that a 4–7% decrease in body weight coupled with 150 minutes a week of moderate exercise could reduce the conversion from IGT to type 2 diabetes by 58% over approximately 3 years [3,4] and by 34% over 10 years, according to the DPP Observational Study (DPPOS) [5]. Furthermore, DPP/DPPOS showed that pharmacological therapy with metformin reduced conversion to type 2 diabetes by 31% over 3 years and 18% over 10 years [3,5].

Screening at-risk individuals is a critical component of primary diabetes prevention. Traditional blood-based measurements of glycemia are commonly used to screen for type 2

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diabetes, but these methods have drawbacks that limit the percentage of the at-risk population that is tested [6]. Requirements for overnight fasting and the lag time of laboratory processing of the blood samples represent barriers to implementing effective diabetes screening programs [7]. The American Diabetes Association (ADA) has recently recommended lab-based A1C for diabetes screening and diagnosis to eliminate fasting requirements and reduce potential pre-analytical errors such as fasting compliance, acute illness and *in vitro* glycolysis [8]. However, laboratory-based A1C still requires drawing venous blood and waiting for results to be reported to the physician. In contrast, various researchers have found that risk stratification using a noninvasive screening measure increases participation in confirmatory blood-based testing [9,10], and that such screening approaches are most cost-effective with respect to the cost per identified case of disease [11].

Recently, noninvasive diabetes screening based on skin fluorescence spectroscopy (SFS) has been proposed [12–16]. The SCOUT DS device (VeraLight, USA) uses fluorescence spectroscopy to noninvasively measure biomarkers of diabetes in the skin, including fluorescent advanced glycation endproducts (AGEs) like pentosidine and cross-lines as well as indicators for cell metabolism and oxidative stress such as reduced nicotinamide adenine dinucleotide (NADH) and flavin adenine dinucleotide (FAD) [17,18]. As shown in Figure 1, the tabletop device illuminates the left volar forearm skin with low-intensity light at multiple near-ultraviolet and visible wavelengths using light emitting diodes (LED). A specially designed fiber-optic probe couples the excitation light to the subject's forearm and relays the resulting skin fluorescence and reflectance to a spectrograph and camera. The SCOUT device measures the reflectance of each excitation LED and automatically adjusts the LED brightness and camera exposure time to compensate for subject specific variation in melanin content, hemoglobin and light scattering, facilitating measurement of all but the darkest skin tones. The optical signals are then processed to produce a score that is related to the presence of prediabetes or diabetes. The score is reported on a scale of 0–100, with higher values indicating higher disease probability. Subjects with a score ≥ 50 are typically considered to have screened positively and are referred for a follow-up blood test to make a diagnosis of prediabetes or type 2 diabetes.

Methods

Objectives

The ENGINE (Evaluation of a Noninvasive Diabetes Screening Device in Subjects at Risk for Diabetes) trial was a prospective,

multi-center validation of SFS detection of abnormal glucose tolerance (AGT) in the subjects at-risk for but without an established diagnosis of type 2 diabetes (clinicaltrials.gov, NCT01080157). FPG and A1C were used as comparative screening methods. Disease truth was abnormal glucose tolerance, defined as a post-challenge plasma glucose of at least 7.8 mmol/L after a 75 gram, two-hour oral glucose tolerance test (OGTT). A secondary study objective was to confirm that SFS had a within-patient, inter-day coefficient of variation (CV) less than 10%. The results reported here are fully prospective: all aspects of data processing, quality assurance metrics, metric thresholds, and the prediction algorithm were derived from a previous calibration data set and fixed prior to data unblinding. Therefore, these results constitute the most rigorous test to date of the SFS system under realistic operating conditions.

Study design

Subjects at risk for type 2 diabetes were recruited using clinical databases and advertising from 12 research sites distributed across the United States. The study protocol was approved by the Schulman Associates institutional review board and encompassed all sites. Subjects were scheduled for three visits to the center with a minimum of 1 day and a maximum of two weeks separating each visit. Each site was supplied with a single SCOUT DS instrument for measurement of skin fluorescence and reflectance. Site staff were trained to explain to study participants how to seat themselves at the device and to place their arms on the optical sensor. The SFS devices perform automated quality control measurements as needed when subject measurements are not being performed.

Inclusion criteria were (i) age greater than or equal to 45 years or (ii) age 18–44 years with a BMI >25 kg/m² and one or more additional ADA-defined risk factors for type 2 diabetes, including physical inactivity, first-degree relative with diabetes, high risk race/ethnicity, women who delivered a baby weighing >9 lbs or diagnosed with gestational diabetes, hypertension, dyslipidemia, women with PCOS, prediabetes, other clinical conditions associated with insulin resistance or history of CVD [19]. Exclusion criteria were participation in previous studies of the SCOUT DS device, receiving chemotherapy in last 12 months, receiving dialysis or compromised renal function, receiving investigational treatments within the past two weeks, a prior diagnosis of diabetes, scars or tattoos on the left volar forearm, or a known skin photosensitivity condition.

On Visit 1, subjects reported to the study site in the morning after an overnight fast of at least eight hours. Informed consent was obtained for all subjects. Each subject completed a short health



Figure 1. Pictures of SCOUT DS device with and without subject forearm.

history and had physical measurements of height, weight, waist circumference (at the midpoint between the top of the iliac crest and inferior margin of the last rib) and blood pressure. The subject placed his/her forearm on the SFS device and the spectral data were analyzed by the system to produce a SFS score for that subject (SFS (Visit 1)). Venipuncture was performed to collect FPG (FPG (Visit 1)) and A1C (A1C (Visit 1)) specimens. The subject then consumed a 75 gm oral glucose load (glucola, 10 fl oz) within 5 minutes. Two hours \pm 10 minutes after consumption, a venipuncture was performed to collect the two hour post-challenge plasma glucose specimen.

On Visit 2, subjects reported any time of day in a non-fasting state to test SFS in its intended-use environment and assess the inter-day coefficient of variation (CV).

On Visit 3, subjects reported to the study site in the morning after an overnight fast of at least 8 hours. A venipuncture was performed for a second FPG (FPG (Visit 3)) in order to assess the inter-day CV of FPG.

Analytical methods

Plasma glucose and A1C assays were performed at local reference laboratories accredited by the College of American Pathology (CAP) and certified in compliance with the Clinical Laboratory Improvements Amendments (CLIA). Each laboratory participated in CAP proficiency testing and provided the results of the proficiency testing for the laboratory tests being performed for the study. All assays had to be judged as acceptable by CAP.

All blood assays were acquired by venipuncture. A1C was collected in EDTA vacutainer tubes, mixed immediately by repeated gentle inversion, and then refrigerated. The A1C assay was traceable to the National Glycohemoglobin Standardization Program (NGSP).

Plasma glucose blood samples were drawn in lithium heparin plasma-separator vacutainer tubes, mixed by gentle inversion, and immediately centrifuged at 2000g for 15–20 minutes. Aliquots of the plasma specimens were placed in transfer tubes and refrigerated. Glucose assays utilized either the glucose oxidase or hexokinase method.

Calculation

Point estimates and 95% confidence intervals on the sensitivity and false positive rate (FPR) for the detection of AGT were calculated for FPG, A1C, and SFS. Receiver operating characteristic (ROC) curves were generated for each test, and the equal error rate (EER, i.e., the point on the ROC where sensitivity and specificity are equal) was computed. The area under the ROC curve (AUC) and partial area (pAUC) for FPRs in the range of 0.2–0.5 (typical of FPG) were also computed for each test. The FPR range for the pAUC calculations was selected to match the typical behavior of the FPG test; refer to the Discussion section for additional details. Partial ROC areas were computed per Dodd and Pepe [20]. Confidence intervals on the AUC and pAUC were determined as described by Qin et al. [21]. The sensitivities of FPG, A1C and SFS for detection of frank diabetes (DM, defined as OGTT \geq 11.1 mmol/L) and impaired glucose tolerance (IGT, defined as 7.8 mmol/L \leq OGTT < 11.1 mmol/L) were also computed.

SFS and FPG measurements were recorded for each subject at each of two visits. By collecting measurements on separate days, the inter-day reproducibility of SFS and FPG could be assessed. Reproducibility was assessed via the intra-individual coefficient of variation from the Hoorn study (Hoorn CV) [22], given by

$$\text{Hoorn CV} = \frac{SD_{\text{dif}} / \sqrt{2}}{\text{median}(W_j)} \times 100,$$

where SD_{dif} is standard deviation of the difference between each subject's first and second measurements, and M_{mean} is the median of the mean of each subject's first and second measurements.

Statistical analyses were performed with MATLAB 7.5 (R2007b).

Results

Data description

Refer to Figure 2 for a trial flow summary. A total of 537 subjects enrolled in the ENGINE trial. Of the enrolled subjects, 15 subjects were screen failures due to inability to collect an OGTT blood specimen, inability to complete the 75 g glucose challenge, or laboratory errors. An additional 4 subjects received two-hour OGTT blood draws that were outside the allowable 2 hours \pm 10 minutes window, leaving a total of 518 subjects with complete OGTT reference data. Two subjects were not measurable on the SFS device due to extremely low signal (<1% of expected reflectance at 375 nm excitation wavelength). In addition, 9 subjects were lost to follow-up and did not complete the 3-visit sequence of the ENGINE trial, leaving a total of 507 subjects who completed the entire protocol.

Valid first attempt SFS (Visit 2) values were obtained from 408 of the 507 (80%) subjects who completed the ENGINE study and from 482 of 507 subjects (95%) if a single retry was allowed in the case that the first attempt failed. Invalid SFS measurements most commonly resulted from (i) failure to collect a complete set of SFS data ($n = 36$, due to failure of one or more automated data quality

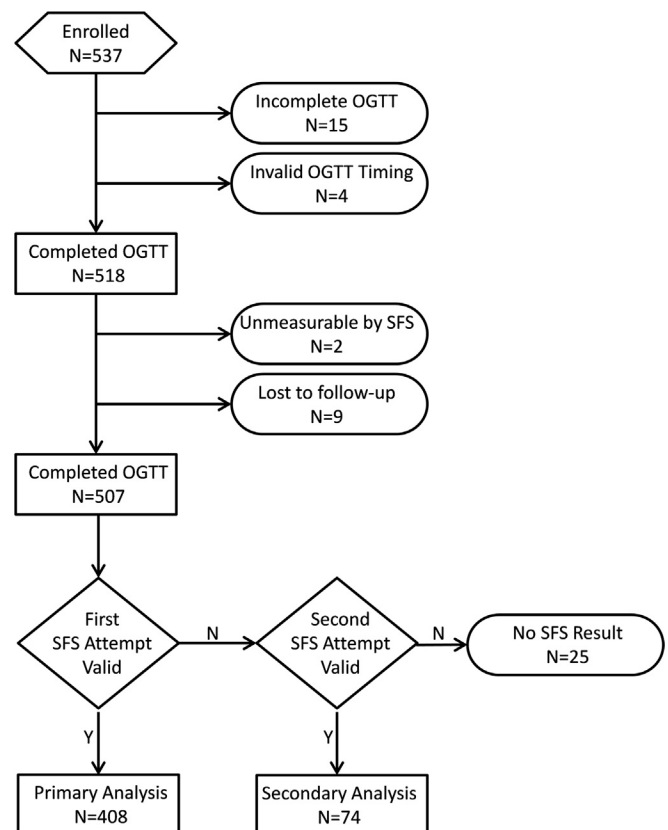


Figure 2. Trial flow summary for the ENGINE study.

metrics, most commonly originating with the presence of room lights in the fluorescence data) or (ii) identification of the acquired spectral data as an outlier ($n = 63$, most commonly due to successive forearm measurements resulting in dissimilar spectra due to inconsistent contact with the optical sensor). All outlier metrics and thresholds were pre-specified and were developed exclusively from a separate and previously collected algorithm training data set. The study protocol did not provide time for subjects who failed to obtain a valid SFS score to make an additional measurement attempt during Visit 2. The primary analysis looked at the 408 subjects with valid first attempt SFS measurements and a secondary analysis looked at the 482 subjects who had valid SFS measurements on the first or second attempts.

Cohort description

The demographic characteristics of the 408 subjects in the primary analysis set are summarized in Table 1. The AGT prevalence was 26.7%. The AGT and normal glucose tolerance (NGT) groups had similar distributions of gender and family history of diabetes. Subject age, BMI, hypertension, and waist circumference were all significantly higher in subjects with AGT versus NGT. The ethnicities of the AGT and NGT groups were different ($p = 0.002$). The AGT group had 65% Caucasian, 22% Latino and 6% African American subjects, versus 61% Caucasian, 17% Latino, and 19% African American in the NGT group.

The demographic characteristics of the 408-member primary analysis set were also compared to those subjects who completed the ENGINE protocol but failed to obtain a valid SFS (Visit 2) score on the first attempt for the reasons described above (Table 2). None of the above-mentioned demographic properties were significantly different between the two groups. The standard deviations of the FPG and A1C values for subjects in the primary analysis set were slightly larger than those for subjects without a valid first attempt SFS (Visit 2) score (1.2 vs 0.7 mmol/L for FPG and 0.7 vs 0.5% for A1C). However, the mean FPG and A1C values of subjects in the primary

Table 2

Demographic characteristics of the ENGINE analysis set versus study completers with a valid OGTT result who were not in the primary analysis set.

	Primary analysis set $n = 408$ (80%)	Completers, not in primary analysis set $n = 99$ (20%)	p value
Gender			0.202 ^a
Male	173 (42%)	49 (49%)	
Female	235 (58%)	50 (51%)	
Ethnicity			0.576 ^a
White	254 (62%)	55 (56%)	
Latino	76 (19%)	20 (20%)	
Af.Amer.	65 (16%)	19 (19%)	
Other	13 (3%)	5 (5%)	
Parent with diabetes	137 (34%)	37 (37%)	0.476 ^a
Sibling with diabetes	59 (14%)	15 (15%)	0.861 ^a
Hypertensive	96 (24%)	26 (26%)	0.568 ^a
Age (years)	51.6 ± 13.6	53.0 ± 12.9	0.494 ^b
BMI (kg/m ²)	31.0 ± 6.9	31.4 ± 7.2	0.661 ^b
Waist, male (in)	40.8 ± 5.5	41.9 ± 6.8	0.344 ^b
Waist, female (in)	38.0 ± 6.7	39.5 ± 6.0	0.107 ^b
2-hour glucose on OGTT (mg/dL)	7.1 ± 3.2	7.1 ± 2.6	0.675 ^b
FPG (mg/dL)	5.5 ± 1.2	5.6 ± 0.7	0.011 ^b
A1C (%)	5.8 ± 0.7	5.8 ± 0.5	0.041 ^b
AGT	109 (27%)	34 (34%)	0.130 ^a

AGT = abnormal glucose tolerance.

^a Pearson's χ^2 test.

^b Wilcoxon rank sum test.

Table 1
Demographic characteristics of the ENGINE cohort expressed as either number (%) or mean ± standard deviation.

	Normal glucose tolerance (NGT) $n = 299$	Abnormal glucose tolerance (AGT) $n = 109$	p value
Gender			0.96 ^a
Male	127 (42%)	46 (42%)	
Female	172 (58%)	63 (58%)	
Ethnicity			0.002 ^a
White	183 (61%)	71 (65%)	
Latino	52 (17%)	24 (22%)	
Af.Amer.	58 (19%)	7 (6%)	
Other	6 (2%)	7 (6%)	
Parent with diabetes	96 (32%)	41 (38%)	0.30 ^a
Sibling with diabetes	40 (13%)	19 (17%)	0.30 ^a
Hypertensive	59 (20%)	37 (34%)	0.003 ^a
Age (years)	50.2 ± 13.7	55.5 ± 12.4	<0.001 ^b
BMI (kg/m ²)	30.0 ± 6.5	33.8 ± 7.1	<0.001 ^b
Waist, male (in)	40.1 ± 5.1	42.8 ± 6.2	0.01 ^b
Waist, female (in)	36.8 ± 6.3	41.3 ± 6.8	<0.001 ^b
2-hour glucose on OGTT (mmol/L)	5.8 ± 1.1	10.8 ± 4.0	N/A ^c
FPG (mg/dL)	5.2 ± 0.5	6.1 ± 2.0	<0.001 ^b
A1C (%)	5.6 ± 0.3	6.2 ± 1.1	<0.001 ^b
SFS Score	49.1 ± 9.3	55.9 ± 9.7	<0.001 ^b

^a Pearson's χ^2 test.

^b Wilcoxon rank sum test.

^c AGT and NGT groups were stratified on the basis of an OGTT threshold of 7.8 mmol/L.

analysis set were very similar to those of subjects without a valid SFS (Visit 2) score (5.5 vs 5.6 mmol/L for FPG, 5.8% vs 5.8% for A1C). In addition, neither the AGT prevalence nor the distributions of OGTT values were significantly different between the two groups. Thus, there is no evidence of selection bias in the individuals who were able to obtain a valid SFS (Visit 2) score on the first attempt versus those who were not.

ROC Curves for AGT Detection

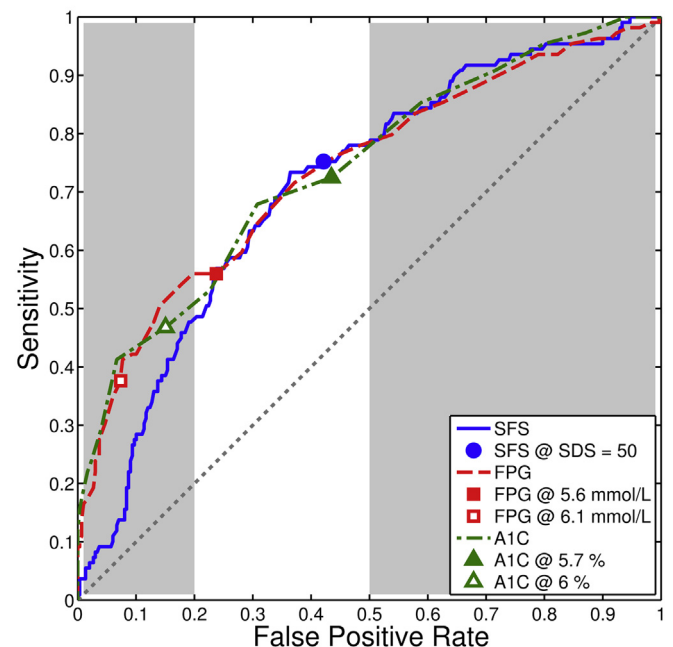


Figure 3. ENGINE study ROC curves for detection of abnormal glucose tolerance (AGT) for SFS DS (Visit 2), FPG (Visit 1) and A1C (Visit 1). The white region denotes $0.2 \leq \text{FPR} \leq 0.5$, which is the range used to compute the partial ROC areas in Table 2.

Table 3
Summary of screening test performance metrics as compared with OGTT.

Test	AGT AUC [95% CI]	AGT equal error rate (%)	AGT pAUC 0.2 ≤ FPR ≤ 0.5 [95% CI]	Threshold	AGT false positive rate (%) [95% CI]	AGT sensitivity (%) [95% CI]
SFS (Visit 2, No Retry)	0.711 [0.655–0.767]	33.0	0.203 [0.174–0.231]	50	42.1 [36.7–47.8]	75.2 [66.4–82.4]
SFS (Up to One Retry)	0.701 [0.650–0.753]	33.4	0.199 [0.172–0.225]	50	42.5 [37.4–47.7]	75.0 [67.0–81.6]
FPG (Visit 1)	0.735 [0.679–0.792]	33.0	0.204 [0.176–0.233]	5.6 mmol/L	23.7 [19.3–28.9]	56.0 [46.6–64.9]
A1C (Visit 1)	0.740 [0.688–0.791]	31.8	0.204 [0.166–0.242]	6.1 mmol/L	7.4 [4.9–10.9]	37.6 [29.1–47.0]
				5.7%	43.5 [38.0–49.1]	72.5 [63.4–80.0]
				6.0%	15.1 [11.4–19.5]	46.8 [37.7–56.1]

Comparative performance

Because skin fluorescence spectroscopy is not influenced by fasting status and is intended to be performed in a non-fasting state, the primary analysis compared SFS detection of AGT from the non-fasting visit (Visit 2) to that of FPG and A1C. The AGT ROC curves for SFS (Visit 2) (blue/solid line), FPG (Visit 1) (red/dashed line) and A1C (Visit 1) (green/dash-dotted line) are shown in Figure 3. Points indicate performance of the tests at their respective screening thresholds of 50 AU (SFS, blue circle), 5.6 mmol/L (FPG, solid red square), 6.1 mmol/L (FPG, open red square), 5.7% (A1C, solid green triangle), 6.0% (A1C, open green triangle). The ROC curves for SFS, FPG, and A1C are very similar for $FPR > 0.2$, with nearly identical pAUCs for $0.2 \leq FPR \leq 0.5$.

Table 3 is a summary of the AUC, pAUC, equal error rate, sensitivity, and FPR for SFS and the FPG and A1C tests using the screening thresholds depicted in Figure 3 as well as SFS if a single measurement retry was allowed. The AGT sensitivity of SFS at its decision threshold of 50 AU was higher than that of FPG at the 5.6 and 6.1 mmol/L thresholds and of A1C at the 5.7% and 6.0% thresholds. The AGT FPR of SFS was higher than that of FPG but comparable to A1C at a 5.7% threshold. The SFS threshold favors test sensitivity at the expense of a moderate FPR, while the A1C and FPG thresholds of 6.0% and 6.1 mmol/L, respectively, favor lower FPR at the expense of sensitivity. SFS and FPG had equal error rates of 33%; the equal error rate of A1C was 32%. The SFS pAUC for detection of AGT was 0.203 (0.199 with one retry), which is comparable to and not significantly different from the FPG and A1C pAUCs, both of which were 0.204. The sensitivity of SFS for undiagnosed diabetes ($OGTT \geq 11.1$ mmol/L) was 79.3%, equal to that of FPG at a threshold of 5.6 mmol/L (79.3%) and comparable to that of A1C at a threshold of 5.7% (sensitivity = 86.2%). The sensitivity of SFS (73.8%) for detection of IGT was higher than that of FPG (47.5%, 27.5%) or A1C (67.5%, 36.3%) at either of the thresholds evaluated for these tests.

The inter-day Hoorn coefficients of variation were 7.7% for SFS (SFS (Visit 1) vs SFS (Visit 2)) and 8.1% for FPG (FPG (Visit 1) vs FPG (Visit 3)).

Although the primary analysis considered SFS performance from Visit 2 (non-fasting), additional analyses found that SFS ROC metrics (AUC = 0.701, pAUC = 0.206) and sensitivity (70.5%) at SFS = 50 from fasting Visit 1 measurements were not significantly different from those of Visit 2. The fasting Visit 1 FPR of SFS was statistically significantly lower than the non-fasting Visit 2 FPR (33.3% vs 42.1%).

Discussion

The ENGINE study results indicate that SFS is effective for screening individuals at risk for pre-diabetes and type 2 diabetes. Specifically, when screening for abnormal glucose tolerance, the ROC curve of SFS was comparable to that of FPG and A1C for $0.2 \leq FPR \leq 0.5$. In a review of FPG AGT screening performance in large cohorts of at-risk, previously-undiagnosed subjects, the FPR of

the FPG test at the ADA-recommended impaired fasting glucose cutpoint of 5.6 mmol/L was found to be constrained to this range ([23–26], Supplementary Table 1). The inter-day Hoorn coefficient of variation of SCOUT DS (7.7%) was comparable to that of the FPG test (8.1%).

Detection of undiagnosed diabetes is also a critical aspect of primary diabetes screening: the United Kingdom Prospective Diabetes Study (UKPDS) established the importance of glycemic control for reducing the risk of developing the diabetes-related complications [27,28]. In addition, tools that facilitate effective AGT screening are important because impaired glucose tolerance is recognized as an early risk indicator for development of type 2 diabetes as well as cardiovascular disease. For example, Tominga et al found that the hazard ratio for death from cardiovascular disease was significantly elevated for individuals with IGT versus NGT (ratio = 2.2), but not for individuals with impaired versus normal fasting glucose (ratio = 1.1) [29]. Other studies have also established a strong association between cardiovascular complications and compromised glucose tolerance [30,31], and primary diabetes prevention studies such as the DPP/DDPOS and DPS have traditionally focused on altered glucose tolerance as the most reliable early indicator of glycemic dysfunction in type 2 diabetes [3–5].

It is therefore desirable that a primary diabetes screening test be as sensitive as possible in identifying individuals with abnormal glucose tolerance. The balance between a test's sensitivity and false positive rate is described by the ROC curve. As seen in Figure 3/ Table 3, the SFS threshold of 50 AU exhibited the highest AGT sensitivity (75.2%) of all screening tests involved in the ENGINE trial. The FPR of SFS was 42.1%, which was slightly lower than that of the A1C test (43.5%) at the ADA-recommended screening threshold of 5.7% [8]. The FPR of the FPG test was lower (23.7% at the ADA-recommended threshold of 5.6 mmol/L), but at the cost of decreased AGT sensitivity (56.0%).

While a low FPR is highly desirable for a diagnostic test, where false positives lead to costly and unnecessary treatment, selecting a moderate FPR in order to maximize AGT sensitivity in primary screening is well justified if ruling out disease in the false positive cases is not excessively inconvenient and/or costly relative to the benefits of early detection and intervention. In a recent modeling study, Chatterjee et al found that widespread opportunistic screening for type 2 diabetes was economically superior to the case of no or limited screening, after accounting for the costs of screening and therapeutic intervention, even for moderate false positive rates such as those reported here [32]. Similarly, a detailed model by Herman et al found that widespread opportunistic screening and implementation of the DPP interventions were cost-effective or marginally cost-saving, even if metformin therapy was extended to individuals as old as 65 years of age [33].

There is ongoing debate in the public health community as to the economic benefit of widespread diabetes screening. For example, citing a study that showed no effect of diabetes screening on mortality, the Canadian Task Force on Prevention Periodic Health

Examination recently issued a guideline that screening for IGT and/or DM is not indicated [34]. Nevertheless, in an effort to reduce the economic burden of caring for those with the disease and to improve workforce productivity, national diabetes screening efforts are being implemented by governments throughout the world. Examples include the National Diabetes Prevention Program (<http://www.cdc.gov/diabetes/prevention/about.htm>) in the United States and the National Programme for Prevention and Control of Diabetes, Cardio-Vascular Diseases and Stroke in India [35]. In addition, several provisions in the United States Patient Protection and Affordable Care Act (2010) directly address gaps in diabetes screening, prevention, and therapy.

While the need for effective primary screening methods for those at risk is generally accepted, multiple obstacles limit the effectiveness of existing blood-based screening modalities. Patient convenience and compliance are barriers to the effectiveness of FPG and OGTT, which require overnight fasting [7,9]. As shown above, FPG suffers from poor sensitivity at its typical screening thresholds. The need to properly handle and dispose of biohazardous waste generated by blood-based methods may be an additional obstacle in certain instances such as employee wellness clinics or community health fairs.

A limitation of the study is the fact that was cross-sectional and only shows the association of SFS with current impaired glucose tolerance or undiagnosed type 2 diabetes. A longitudinal study would be more powerful in elucidating an SFS threshold that predicts the development of type 2 diabetes.

Strengths of the study include that all members of the study cohort were at risk for type 2 diabetes by ADA guidelines and therefore members of the intended-use population. In addition, head-to-head SFS, FPG, A1C, and OGTT data were acquired. The cohort also had a representative mixture of patient age, gender, ethnicity, and BMI. The clinically-realistic, multi-center, multi-device nature of the ENGINE trial represents an additional strength.

Conclusion

The elimination of overnight fasting, the absence of blood, and the rapid, real-time communication of screening results are aspects of noninvasive skin fluorescence spectroscopy that facilitate opportunistic screening of individuals at risk for type 2 diabetes while delivering performance that is comparable to FPG and A1C for detection of abnormal glucose tolerance.

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Author contributions: ELH co-wrote and edited the manuscript and analyzed data. NIM, MNE, BPO, and AJM analyzed data and

reviewed/edited the manuscript. KEV and JDM co-wrote and edited the manuscript and contributed to discussion. JFW was responsible for overseeing all data collection and contributed to discussion.

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Supplementary Table 1

Performance of FPG when screening for abnormal glucose tolerance at a test threshold of 5.6 mmol/L. Disease truth is defined as a two-hour OGTT result of at least 7.8 mmol/L.

Data set	Number of subjects	Disease prevalence	FPG FPR	FPG sensitivity
NHANES III, Phase 1 (1988-91) ^a [23]	1460	28.0%	36.7%	66.1%
NHANES III, Phase 2 (1991-94) ^a [23]	1566	28.9%	26.9%	62.5%
NHANES 05-06 ^a [24]	1293	26.6%	34.9%	74.7%
NHANES 07-08 ^a [25]	1512	29.6%	49.0%	75.9%
Robles-Osorio et al. [26]	1239	36.7%	18.9%	54.9%

^a At-risk population (per ADA guidelines) only, weighted per NHANES guidelines to reflect overall US demographics for age ≥ 18 .