

Transdermal Blood Sampling for C-Peptide Is a Minimally Invasive, Reliable Alternative to Venous Sampling in Children and Adults With Type 1 Diabetes

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Transdermal blood sampling for C-peptide is a minimally invasive, reliable alternative to venous sampling in children with type 1 diabetes

BACKGROUND AND AIMS

C-peptide and islet autoantibodies are key type 1 diabetes biomarkers, typically requiring venous sampling, which limits their utility. We assessed transdermal capillary blood plasma (TCB) collection as a practical alternative.

Figure 1. TCB device



METHODS

Ninety-one individuals (71 type 1 diabetes, 20 controls) underwent a contemporaneous venous and TCB sample for measurement of plasma C-peptide; see Table 1. Type 1 diabetes participants also provided venous serum and plasma, and TCB plasma for measurement of autoantibodies to glutamate decarboxylase (GADA), islet antigen-2 (IA-2A), and zinc transporter 8 (ZnT8RA/WA). The ability of TCB plasma to detect significant endogenous insulin secretion (venous plasma C-peptide ≥ 200 pmol/L) was compared along with agreement in levels using Bland-Altman. Venous serum was compared with venous and TCB plasma for detection of autoantibodies using established thresholds. Acceptability was assessed by age-appropriate questionnaire.

Table 1. Participant demographics

	Type 1 diabetes N=71	Control N=20
Age median (IQR) yr	14.8 (9.1–17.7)	42.2 (38.0–52.1)
Diabetes duration median (IQR) yr	4.0 (1.5–7.7)	-
N(%) females	38/71 (54%)	19/20 (95%)
Ethnicity		
Asian/Asian British	2 (3%)	0
Black/African/Caribbean/Black British	3 (4%)	1 (5%)
Mixed/Multiple Ethnic Groups	7 (10%)	1 (5%)
White	60 (83%)	18 (90%)

RESULTS

Median sample volume was 50 μ L (interquartile range (IQR) 40–50) with 3 of 91 (3.3%) failures, and 13 of 88 (14.7%) < 35 μ L.

C-peptide There was good agreement between TCB and venous C-peptide (mean venous ln(C-peptide) – TCB ln(C-peptide) = 0.008, 95% CI (-0.23, 0.29).

Figure 2. Bland Altman plot comparing the TCB and venous C-peptide measurements on 48 participants.

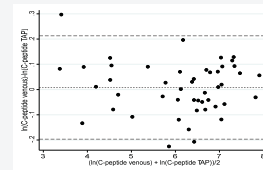
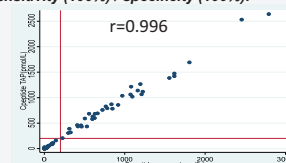


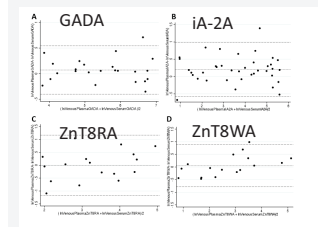
Figure 3. Scatter diagram between random venous and TCB C-peptide in controls and type 1 diabetes participants. Cutoff of C-peptide ≥ 200 pmol/L with sensitivity (100%) / specificity (100%).



Islet autoantibodies

Venous serum in multiple autoantibody positive TCB plasma agreed in 22 of 32 (sensitivity 69%), comparative specificity was 35 of 36 (97%).

Figure 4. Bland-Altman plot comparing the venous plasma and venous serum, log transformed. The upper and lower horizontal lines show the limits of agreement, and the middle line the mean difference of the measurements.



Discreet choice

TCB was preferred to venous sampling (type 1 diabetes: 63% vs 7%; 30% undecided).

CONCLUSIONS Transdermal testing for C-peptide is a sensitive, specific, acceptable and practical alternative to venous sampling. TCB sampling for islet autoantibodies needs further assessment.

ARTICLE HIGHLIGHTS

- Why did we undertake this study?**
 We aimed to determine the precision and acceptability of transdermal capillary blood (TCB) for measurement of C-peptide.
- What is the specific question(s) we wanted to answer?**
 We investigated whether TCB is a reliable and acceptable alternative to venous C-peptide measurement.
- What did we find?**
 From a study of 91 individuals (71 children and adults with type 1 diabetes, and 20 adult controls), TCB was found to be a sensitive, specific, and acceptable alternative to venous sampling.
- What are the implications of our findings?**
 TCB could be used as a reliable and practical alternative for C-peptide sampling.



Transdermal Blood Sampling for C-Peptide Is a Minimally Invasive, Reliable Alternative to Venous Sampling in Children and Adults With Type 1 Diabetes

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OBJECTIVE

C-peptide and islet autoantibodies are key type 1 diabetes biomarkers, typically requiring venous sampling, which limits their utility. We assessed transdermal capillary blood (TCB) collection as a practical alternative.

RESEARCH DESIGN AND METHODS

Ninety-one individuals (71 with type 1 diabetes, 20 control; individuals with type 1 diabetes: aged median 14.8 years [interquartile range (IQR) 9.1–17.1], diabetes duration 4.0 years [1.5–7.7]; control individuals: 42.2 years [38.0–52.1]) underwent contemporaneous venous and TCB sampling for measurement of plasma C-peptide. Participants with type 1 diabetes also provided venous serum and plasma, and TCB plasma for measurement of autoantibodies to glutamate decarboxylase, islet antigen-2, and zinc transporter 8. The ability of TCB plasma to detect significant endogenous insulin secretion (venous C-peptide ≥ 200 pmol/L) was compared along with agreement in levels, using Bland-Altman. Venous serum was compared with venous and TCB plasma for detection of autoantibodies, using established thresholds. Acceptability was assessed by age-appropriate questionnaire.

RESULTS

Transdermal sampling took a mean of 2.35 min (SD 1.49). Median sample volume was 50 μ L (IQR 40–50) with 3 of 91 (3.3%) failures, and 13 of 88 (14.7%) < 35 μ L. TCB C-peptide showed good agreement with venous plasma (mean venous \ln [C-peptide] – TCB \ln [C-peptide] = 0.008, 95% CI [–0.23, 0.29], with 100% [36 of 36] sensitivity/100% [50 of 50] specificity to detect venous C-peptide ≥ 200 pmol/L). Where venous serum in multiple autoantibody positive TCB plasma agreed in 22 of 32 (sensitivity 69%), comparative specificity was 35 of 36 (97%). TCB was preferred to venous sampling (type 1 diabetes: 63% vs. 7%; 30% undecided).

CONCLUSIONS

Transdermal capillary testing for C-peptide is a sensitive, specific, and acceptable alternative to venous sampling; TCB sampling for islet autoantibodies needs further assessment.

C-peptide and pancreatic islet autoantibodies are key biomarkers used in type 1 diabetes. C-peptide reflects endogenous β -cell function and is used in clinical care to aid in

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the correct classification of diabetes subtype (1–4). In research, C-peptide is the primary outcome following interventions aiming to preserve β -cell function (5). Islet autoantibody testing may be needed to confirm diabetes etiology in clinical care and in research trials (4,6), and can be used to identify children at risk for future clinically diagnosed disease (7). The ability to accurately measure type 1 diabetes biomarkers, that is effective and painless as well as acceptable, would be highly valuable, particularly in children.

C-peptide is typically collected using a venous blood draw, which is invasive, involving a needle, which can be a challenge for young children, and restricts testing to the health care setting. Recently, urine C-peptide-to-creatinine ratio (UCPCR) (8,9), dried blood spot (DBS) C-peptide (10,11), and, more recently, the volumetric absorptive microsampling device (12) have been shown to be practical alternatives to venous sampling. However, UCPCR requires an individual to void on demand, typically not possible in very young children. DBS C-peptide and the volumetric absorptive microsampling device method are typically collected from a fingerstick blood spot using a lancet blade or needle, to produce a sample that is a mixture of arteriolar, venous, and capillary blood. DBS C-peptide requires careful sample handling and processing, making it expensive, time consuming, and difficult for measuring C-peptide levels at very low concentrations that are still clinically meaningful (13). Islet autoantibodies can be measured from serum taken from a venous or a capillary fingerstick or bloodspot sample, and which can be posted for analysis (14).

The Touch Activated Phlebotomy (TAP I) device has been developed for painless and minimally invasive blood collection (~100 μ L), recently upgraded to collect a larger blood volume of ~300 μ L, and has a Conformité Européenne (CE) mark for measurement of glycosylated hemoglobin (HbA_{1c}) in adults >21 years (15). The “CE” mark status indicates that the device has been sold in the European Economic Area, has been assessed to meet high standards, and complies with European Union legislation. The TAP I/II and a different microsampling device (Tasso+) have now been tested successfully for use in a number of clinical and laboratory settings (16–23). However, the accuracy of using this method for measuring

C-peptide or islet autoantibodies in type 1 diabetes has not been tested, and there has been no assessment of acceptability of using this method in children. We therefore aimed to assess whether C-peptide and autoantibodies collected from transdermal capillary blood (TCB) from the TAP I device was a reliable and acceptable alternative to venous sampling.

RESEARCH DESIGN AND METHODS

Population

We studied 71 individuals with type 1 diabetes (defined by clinical diagnosis) and 20 adult control individuals (Table 1). Individuals with type 1 diabetes were recruited from pediatric and adult diabetes clinics at the John Radcliffe Hospital, and the Oxford Centre for Diabetes Endocrinology and Metabolism, Oxford, U.K. Adult control participants without diabetes were recruited through poster advertisement at the John Radcliffe Hospital and the Oxford Centre for Diabetes Endocrinology and Metabolism, as well as parents without diabetes identified through patients attending the diabetes clinics involved in the study.

Recruitment was enriched for individuals with type 1 diabetes who had measurable C-peptide to allow assessment across a spread of C-peptide values, and we aimed to recruit at least 50% people with type 1 diabetes within 5 years of diagnosis. In order to assess acceptability, we aimed to recruit equal numbers of participants by age group, split between under 10 years, 10–16 years, and adults aged >16 years. Participants were excluded if they had known renal impairment (estimated glomerular filtration rate <60 mL/min/1.73 m²), were pregnant, had a known coagulopathy, were on medication interfering with renal excretion, or were non-English speakers. Parental consent along with assent was gained for children and

young people aged under 16 years, and consent was gained for participants aged over 16 years.

Study Design

Initially, we aimed to recruit 50 individuals with type 1 diabetes and 20 adult control individuals. Further individuals were recruited to replace the participants where sample collection was unsuccessful, defined as complete sample failure or collecting <35 μ L plasma, which was anticipated to be the minimum viable volume of plasma needed for C-peptide measurement. Measurement of C-peptide was the primary aim of our study. Measurement of islet autoantibodies was a secondary aim, to explore how much information a TCB sample could yield, and therefore was conducted after C-peptide analysis.

To minimize the impact of blood sampling for individuals with type 1 diabetes, sample collection was offered as part of their routine annual review visit, which would normally include a venous blood draw.

Topical anesthetic was offered according to local policy for venous sampling. Prior to sampling, individuals with type 1 diabetes performed a self-monitoring blood glucose test, and sampling was delayed if blood glucose was <4 mmol/L, until resolution, to avoid C-peptide suppression.

Participants with type 1 diabetes had a 5-mL Li heparin plasma sample (for C-peptide measurement), and 1.3-mL serum-separating-tube sample (for autoantibody measurement), and a concomitant TCB sample collected by a researcher from the participants' opposite upper arm. Control participants had only a 5-mL Li heparin plasma sample collected (for C-peptide measurement) with the concomitant TCB sample.

We recorded the time taken to collect the TCB sample, using the color indicator

Table 1—Participant characteristics

	Type 1 diabetes, <i>n</i> = 71	Control, <i>n</i> = 20
Age, years	14.8 (9.1–17.7)	42.2 (38.0–52.1)
Diabetes duration, years	4.0 (1.5–7.7)	—
<i>N</i> (%) females	38 of 71 (54%)	19 of 20 (95%)
Ethnicity		
Asian/Asian British	2 (3%)	0
Black/African/Caribbean/Black British	3 (4%)	1 (5%)
Mixed/multiple ethnic groups	7 (10%)	1 (5%)
White	60 (83%)	18 (90%)

Data are presented as median (interquartile range), unless otherwise stated.

turning from green to red on the TAP device. At the end of sample collection, the TAP device was removed, and the samples were centrifuged to allow plasma to be extracted and stored at -80°C at the Juvenile Diabetes Research Foundation (JDRF)/Wellcome Diabetes and Inflammation Laboratory at the Wellcome Centre for Human Genetics.

TAP I Device

The TAP I Blood Collection Device (manufacturer YourBio Health, Inc., previously known as Seventh Sense Biosystems) combines capillary action with the use of 1-mm-long solid microneedles and vacuum extraction through the skin, to obtain 100 μL capillary whole blood in LiHeparin anticoagulant (Supplementary Method 1). A visual marker indicates when the device reservoir is filled (green to red). Following removal of the device from the skin, the blood sample is extracted via pipette method.

Laboratory Methods

C-Peptide

C-peptide samples were analyzed centrally. Plasma C-peptide was measured by electrochemiluminescence immunoassay (intraassay coefficient of variation [CV] $<3.3\%$; interassay CV $<4.5\%$, assay limit 3.3 pmol/L) on a Roche Diagnostics (Mannheim, Germany) E170 analyzer by the Academic Department of Blood Sciences at the Royal Devon University National Health Service Healthcare Foundation Trust, Exeter, U.K.

All samples underwent a minimum dilution with an equine proteinaceous diluent (Diluent Multi Analyte; Roche Diagnostics, Mannheim, Germany) to achieve a minimum volume of 100 μL , required for analysis of C-peptide. Details of the dilutions performed can be found in Supplementary Table 1.

Islet Autoantibodies

Plasma remaining after C-peptide measurement and undiluted venous serum samples, from participants with diabetes, were refrozen and sent to the Learning and Research Centre at Southmead Hospital, Bristol, U.K. Islet autoantibodies to GAD, IA-2, and ZnT8R/W were measured using established standardized radioimmunoassays with ^{125}I - or ^{35}S -labeled antigens (intraassay CV 4%, 6%, and 6% and interassay CV for a positive sample 20%, 19%, and 21% measuring antibodies for GAD, IA-2, and ZnT8R/W, respectively)

(24,25). The sensitivity and specificity for these tests in Islet Autoantibody Standardization Program 2020 was 64% and 97.8% for GADA, 72% and 100% for IA-2A, 60% and 100% for ZnT8RA, and 56% and 100% for ZnT8WA, respectively.

Acceptability Assessment

Usability of the TAP I device was assessed using a questionnaire, adapted from Liu et al. (14). This included a traditional Likert scale, and a visual pain score (Wong-Baker Faces scale) (26). For participants aged under 16 years, the Likert scale was graded from 0 to 10, where 0 is “no hurt” and 10 is “hurts worst.” For adult participants, the Likert scale was graded from 0 to 7, where 0 is “no pain,” 4 is “moderate pain,” and 7 is “very painful” (Supplementary Method 2). Participants aged over 16 years completed the questionnaire independently, and, for those aged under 16 years, it was completed by both participant and guardian. We further recorded the choice of having a future test with either a TAP I device or venous sample (“TAP/Venous/don’t mind”).

Adverse Events

Adverse events were recorded following venous and TAP device sampling over 7 days. Data were recorded for redness, swelling, and bruising (yes/no), and pain was recorded on a scale of 0 to 4, where 0 was no pain, and 4 was spontaneously painful and prevents normal daily activities.

Statistical Analyses

We assessed the time taken (minutes, seconds) to complete sample collection from the TAP device. We recorded the volume (μL) of whole blood collected and the plasma extracted.

C-Peptide

We compared the C-peptide of the venous sample with the TCB plasma sample using paired samples, where both measurements were available for each participant, and excluded those where dilution resulted in raising the limit of detection so values could not be compared. Each venous sample had been divided into four or five aliquots, analyzed separately for C-peptide, and the mean value was calculated for each participant; the SD and range within each participant’s measurements were calculated. The C-peptide value was log transformed. Using the paired values, the mean of the venous and

TCB C-peptide and the difference between the two were plotted. A Bland-Altman plot was used to assess the bias and the limits of agreement between the two methods.

We assessed the association between venous and TCB C-peptide (Pearson correlation coefficient). We further assessed the ability of venous and TCB plasma C-peptide to correctly classify individuals with clinically significant endogenous insulin production (defined as C-peptide ≥ 200 pmol/L), with corresponding specificities and sensitivities.

Islet Autoantibodies

Thresholds for islet autoantibody positivity have been established using populations of people without diabetes as previously described and were 33 diabetic kidney (DK) units/mL for GADA, 1.4 DK units/mL for IA-2A, and 1.8 units for ZnT8R/WA (24,25).

Islet autoantibody levels were compared between venous serum with venous plasma, and separately with diluted TCB sample values.

Venous Serum Versus Venous Plasma. Venous serum samples (the gold standard) were compared with venous plasma samples using a Bland-Altman plot to assess the bias and the limits of agreement. Only observations where the level was great enough to be detected were included.

Venous Serum Versus TCB Plasma. Results for venous serum and TCB plasma were log transformed and plotted to determine the impact of dilution on the precision of positive values. Sensitivity and specificity of plasma was compared with detection in serum; that is, serum antibody positives were considered true positives, and serum antibody negatives were considered true negatives. We assessed the ability of the TCB plasma to detect two or more islet autoantibodies, as is used in type 1 diabetes screening studies.

Acceptability. For participants aged under 16 years, pain scores were grouped as follows: 0, no pain; 2, mild pain; 4–6, moderate pain; 8–10, severe pain. For adults aged 16 years and over, scores were reported as follows: 1, no pain; 2–3, mild pain; 4–6, moderate pain; 7, severe pain.

Ethical Considerations

This study was approved by the West Midlands–Edgbaston Research Ethics Committee, Nottingham, U.K. All subjects, and, for those <16 years, also their parents, gave informed consent.

RESULTS

Ninety-one individuals were recruited (71 with type 1 diabetes, 20 adult control) (Table 1). There were 20 participants aged under 10 years, 22 aged 10–16 years, and 29 aged >16 years. Participants with type 1 diabetes were aged median 14.8 years (interquartile range [IQR] 9.1–17.7) and range 1.2–41.0 years, with a diabetes duration of median 4.0 (1.5–7.7) years and range 0.1–23.0 years.

TCB Sample Collection

There were 3 of 91 (3.3%) absolute sample failures, and 13 of 88 (14.7%) with low plasma volume (<35 μ L). The absolute sample failures were all children (aged 11 months, 12 years, and 13 years); two were female, and one was male. The samples yielding low plasma volume were also all from children (median age 3.9 years, range 9 months to 9 years); five were female, and eight were male. A median of 50 μ L plasma (IQR 40–50 μ L), range 10–65 μ L, was collected ($n = 88$). Sample collection from 91 participants took a mean of 2.35 min (median 2.35), SD 1.49, range 0.37–7 min.

Relationship Between TCB and Venous Samples

C-Peptide

Bland Altman Agreement. There were 372 venous samples in total, separated from multiple aliquots from the 91 participants: 81 samples from 20 healthy control participants and 301 from 71 participants with type 1 diabetes. One hundred and thirty-nine samples had C-peptide <3 pmol/L, and all were from 34 of 71 participants with type 1 diabetes. The median value of the patient mean values was 45.5 pmol/L (IQR <3, 626) and range <3 to 2,792 pmol/L.

There were 48 participants with both detectable C-peptide (>3 pmol/L) from paired TCB and venous plasma samples. Fig. 1 shows the Bland Altman plot of these 48 paired examples, using the log transformed values. There is no statistically significant bias, the mean venous $\ln(\text{C-peptide}) - \text{TCB } \ln(\text{C-peptide}) = 0.008$, 95% CI (–0.23, 0.29). The limits of agreement are –0.197, 0.213.

Transforming the values back to the original scale, we can report the ratio of venous to TAP:

mean = 1.008, 95% CI 0.79 to 1.34

limits of agreement 0.82 to 1.24.

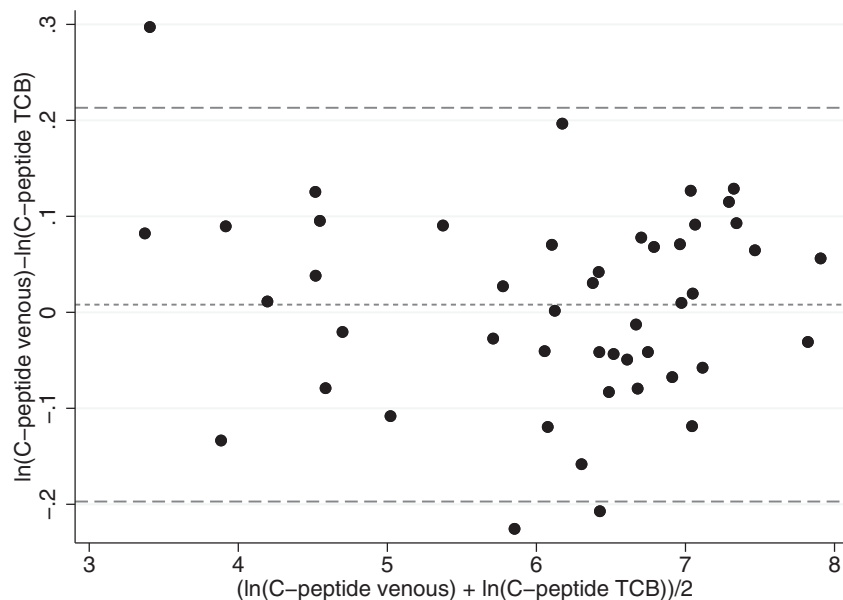


Figure 1—Bland-Altman plot comparing the venous and TCB C-peptide measurements on 48 participants. The upper and lower horizontal dashed lines show the limits of agreement.

Classification for C-Peptide Positivity.

C-peptide in TCB plasma was highly correlated to venous serum (Fig. 2): Pearson correlation coefficient 0.996. There was 100% (36 of 36) sensitivity and 100% specificity (50 of 50) for significant endogenous β -cell function (≥ 200 pmol/L) for the TCB compared with the venous sample. Table 2 reports the results of dichotomizing C-peptide at 200 pmol/L for 86 participants.

Islet Autoantibodies

Venous Serum Versus Venous Plasma

First, serum measurement (gold standard) was compared with venous plasma by Bland-Altman (Supplementary Fig. 1A). Using serum measurement to determine true positive and negative, sensitivity was >89% and specificity was >86% for all autoantibodies measured in venous plasma. Bland-Altman analysis identified mean biases of <1.3 (DK) units.

Venous Serum Versus TCB Plasma (Diluted)

Supplementary Fig. 2 shows the precision of TCB on islet autoantibody sampling. Sensitivity was 24 of 25 (96%) for GADA, 32 of 39 (82%) for IA-2A, 12 of 19 (63%) for ZnT8RA, and 10 of 20 (50%) for ZnT8WA; specificity was 41 of 43 (95%), 27 of 29 (93%), 48 of 49 (98%), and 48 of 48 (100%), respectively.

Classification for Islet Autoantibody Positivity Using TCB Plasma

Islet autoantibody positivity is considered commonly in research studies, and

the presence of two or more islet autoantibodies is considered a marker of early-stage type 1 diabetes, rather than the actual titer of the responses (27). The sensitivity and specificity for detecting two or more islet autoantibodies using TCB plasma was 68.8% (22 of 32) and 97.2% (35 of 36), respectively (Supplementary Table 3).

Acceptability

Usability

Likert Scale. Of the 71 participants, 15 (21%) with type 1 diabetes reported no difference between venous and TAP sampling, 48 of 71 (68%) scored venous more painful than TAP, and 8 of 71 (11%) scored TAP more painful than venous (all by children and young people aged <16 years).

Type 1 Diabetes: Under 10 Years

For children aged <10 years, 60% ($n = 12$) reported no pain, 35% ($n = 7$) reported mild/moderate pain, and 5% ($n = 1$) reported worst pain using the TAP device versus 50% ($n = 10$), 40% ($n = 8$), and 10% ($n = 2$), respectively, after venous sampling (Supplementary Table 4A).

Type 1 Diabetes: 10–16 Years

For children aged 10–16 years, 54% ($n = 12$) reported no pain, 36% ($n = 8$) reported mild/moderate pain, and 9% ($n = 2$) reported worst pain using the TAP device versus 27% ($n = 6$), 59% ($n = 13$), and 14%

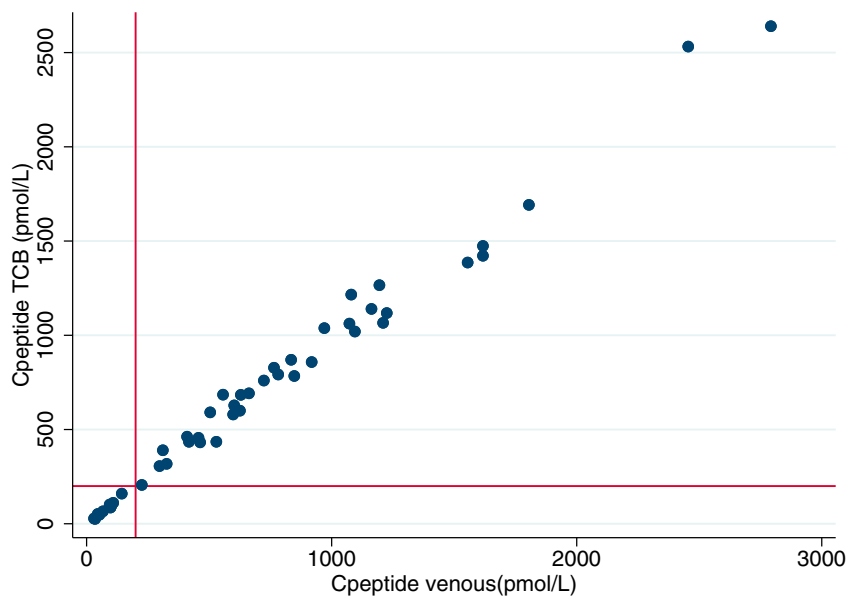


Figure 2—Scatter diagram showing the relationship between random venous and TCB C-peptide in 20 control participants and 71 participants with type 1 diabetes (Pearson correlation, $r = 0.996$). Cutoff of C-peptide ≥ 200 pmol/L is shown with corresponding sensitivity and specificity.

($n = 3$), respectively, after venous sampling (Supplementary Table 4B).

Adults

For adults with type 1 diabetes, 79% ($n = 23$) reported no pain, and 21% ($n = 6$) reported mild pain using the TAP device versus 10% ($n = 3$) no pain, 55% ($n = 16$) mild, 31% ($n = 9$) moderate, and 3% ($n = 1$) severe pain with venous sampling (Supplementary Table 4C). In adult control participants ($n = 20$), 100% reported mild pain versus 70% ($n = 14$) mild pain and 30% ($n = 4$) moderate pain on venous sampling (Supplementary Table 4D).

Patient Preference

When asked to choose their preferred method for future sampling, the majority (63%, 44 of 70) stated they would prefer the TAP device versus 7% (5 of 70) venous sampling, and 30% (21 of 70) were undecided. This was similar across all age groups, with the highest preference to TAP seen in adults with type 1 diabetes (19 of 29, 65%) (Supplementary Table 5).

Adverse Event Diaries

Redness on day 1 was reported in 44% (32 of 73) for the TAP device and 30% (21 of 69) for venous sampling (Supplementary Table 6). No analgesia was needed for either TAP or venous sample in the 7 days following sampling.

CONCLUSIONS

We show that transdermal plasma C-peptide shows good agreement with venous plasma, and is a sensitive, specific, and acceptable method to detect endogenous insulin secretion. Transdermal collection for islet antibodies needs further assessment.

Transdermal Blood Collection for C-Peptide

Our results showed a strong agreement between paired venous and transdermal C-peptide. The precision of the device in identifying significant endogenous insulin production (≥ 200 pmol/L) makes it an attractive alternative to venous sampling.

Although the transdermal method is not superior to venous sampling, it is more practical, since it does not involve a venous blood draw. While transdermal sampling was more favorable in our study, it was not uniformly chosen by children. This can be explained by the standard method for collecting a venous blood draw in a children’s hospital setting, which includes use of topical anesthetic and access to play specialists. Allowing a reliable method to be undertaken without a venous blood draw may mean it could avoid bringing children into the hospital unnecessarily, with an obvious cost saving. The main barrier to home testing would be the need to process the samples shortly after collection. This may be overcome by use of the next generation device (TAP II) that collects a greater blood volume directly into a blood collection tube containing an appropriate additive, which means it does not require immediate processing, a method recently tested in adults with rheumatic diseases with high acceptability (19). Since C-peptide is stable for at least 24 h in plasma, home samples could be collected in EDTA plasma and brought to a community setting for transport to the local hospital laboratory for processing, but would need further assessment.

Transdermal Blood Measurement for Islet Autoantibodies

The need to dilute the TCB samples in our study made the analysis of lower-level autoantibody responses challenging; however, the newer generation TAP II device would address this limitation.

We identified some differences between measurement in venous serum and TCB plasma samples. The lower specificity of IA-2A measured in TCB plasma is partially explained by a genuine difference between plasma and serum measurement in two participants; where IA-2A levels >15 DK units (more than 10 times the threshold) in venous or TCB plasma compared with undetectable levels in serum, other differences were more subtle. This contrasts with previous work suggesting EDTA plasma and serum showed very high correlation for GADA and IA-2A measured by radioimmunoassay (28), but, in that study, fewer IA-2A were detected close to the threshold. The small difference between venous plasma and serum does not fully explain the lower sensitivity in detecting individual islet autoantibodies using diluted TCB plasma. Even after a post hoc

Table 2—Categorization of positive values, defined as C-peptide ≥ 200 pmol/L, in TCB plasma and venous serum samples

	Venous C-peptide ≥ 200 pmol/L	Venous C-peptide < 200 pmol/L	Total
TCB C-peptide ≥ 200 pmol/L	36	0	36
TCB C-peptide < 200 pmol/L	0	50	50
Total	36	50	86

adjustment for dilution (data not shown), levels did not agree. We hypothesize that the impact of transdermal sampling, with a high level of interstitial fluid and unknown matrix effect, may have affected the results. Overall sensitivity for detecting two or more islet autoantibodies was relatively low (69%), with high 97% specificity, suggesting further work is needed before using the transdermal methods to measure islet autoantibodies.

Alternative Measures of Capillary C-Peptide

The strong relationship between transdermal blood collection for C-peptide and serum C-peptide is supported by previous studies assessing DBS compared with venous C-peptide during a mixed-meal tolerance test (11). Both DBS and TCB C-peptide use capillary sampling. The method of extraction and processing is, however, different, with DBS requiring extraction of very small volumes that are not measurable at low levels. In contrast, transdermal collection allows a larger volume to be processed and lower concentrations to be measured, although using this generation device with relatively low volumes dilution that will decrease the limit of detection, and that has already been addressed using the TAP II device (19). Compared with a timed urine collection for measurement of UCPCR that requires an individual to void on demand (9), transdermal blood can be collected at any age and has been tested in infants as young as 2 months of age (18).

Strengths

The study included a large range of C-peptide and age ranges, making the results translatable to pediatric and adult type 1 diabetes settings.

Study Limitations

Acceptability and pain were assessed; however, no topical analgesic was used for TCB sample collection, so it may be argued that this was not a fair comparison. Despite this, participants favored use of the TAP device. In adults, who are not routinely offered topical analgesia for venous sampling, 63% ($n = 19$) of adults with type 1 diabetes favored the TAP device, 7% ($n = 2$) favored the venous sample, and 27% ($n = 8$) were undecided.

Insulin autoantibodies were not measured in this study, owing to the relatively

large volume of sample required, and, after 2 weeks, exogenous insulin injection can stimulate insulin antibody production. For use in research, particularly young children at risk for type 1 diabetes, including measurement of insulin autoantibodies would be preferable, and tested on undiluted samples.

The absolute failure rate of sampling using the TAP device was only 3.3%, and 14.7% yielded a low plasma volume ($<35 \mu\text{L}$). Cost was not assessed as part of this study, but device failure and the volume of sampling required would need to be accounted for in further routine analysis and before health care integration and home testing.

Implications

The TAP I device may have a role in home collection of C-peptide in the research setting in prospective studies assessing β -cell function and following interventions of disease-modifying agents in both newly diagnosed (stage 3) as well as early-stage type 1 diabetes (stages 1 and 2) (29). Since the gold standard measure of endogenous insulin secretion in type 1 diabetes, the mixed-meal tolerance test, is costly and impractical, it is usually measured only 3–12 monthly following interventions in type 1 diabetes trials. Interim samples provide useful information, an approach adopted in our clinical trial in children with newly diagnosed type 1 diabetes (30), with home collection of DBS C-peptide after a standardized meal. The transdermal approach has advantages over DBS C-peptide, mostly related to the method and volume of blood extracted, and may therefore offer the ability to measure C-peptide less invasively, more frequently, and at lower assay limits. The current method is not sufficiently accurate for islet autoantibody testing.

Future Work

The feasibility of collecting a home TAP I sample for measurement of C-peptide needs investigating. It may offer a potential practical alternative to hospital testing. The feasibility and acceptability of collecting undiluted TAP samples for measurement of islet autoantibodies may have a role in screening for type 1 diabetes in the general population and first-degree relatives, in particular using the TAP II or Tasso+ device, and needs further assessment, since antibodies to rheumatic

diseases have been successfully measured (19). The validation, feasibility, and acceptability of home testing in other settings, in particular chronic diseases such as thyroid disease, would be worthwhile.

Transdermal blood collection may offer a precise and acceptable alternative to venous sampling for C-peptide in children and adults with type 1 diabetes. Further assessment is needed for home collection and for assessment of islet autoantibody measurement.

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