2024 C-Peptide Standardization Manufacturer Meeting
Minutes

Tuesday, October1 9:00-11:00 AM CST (Virtual Meeting)

Participants:

C-peptide Standardization Committee Members
Randie Little--Chair, Univ of Missouri
Kuanysh Kabytaev--Co-chair, Univ of Missouri
William Hagopian--Univ of Washington
Andy Hoofnagle--Univ of Washington
Paolo Pozzilli--Campus Bio-Med Univ of Rome
Salvatore Sechi--Liaison, NIH/NIDDK
Beena Akolkar--Liaison, NIH/NIDDK

Committee members not present
Robert Wielgosz--BIPM, France

Guests
Shawn Connolly--Univ of Missouri
Lutz Heinemann--dia team
Daniel Holmes--Providence Health
Amy Karger--Univ of Minnesota
Leslie Landree--FDA
Jieli Li--Ohio State Univ
Gustavo Martos--BIPM
Masami Murakami--Gunma Univ Japan
Curt Rohlfing--Univ of Missouri
David Sacks--NIH
Gwen Wark--NHS

Manufacturer Representatives
Yukie Arizono--Miraca Life Sciences
Ming Bangfa--Mindray
Paul Barto--Quidel Ortho
Jean-Sebastien Blanchet--Beckman
Stewart Cristan--Beckman Coulter Diag

Manufacturer Representatives (contd.)
Jamie Deeter--Roche Diagnostics
Benjamin Escher--Roche Diagnostics
Claudia Gonzalez--Tosoh Bioscience
Wictor Gustafsson--Mercodia
Yanlin Han--Autobio
Yuka Imai--Miraca Life Sciences
John Jaraczewski--Abbott
Michael Kjome--Beckman Coulter Diag
Takayuki Kosaka--Tosoh Bioscience
Carlos Martin--Tosoh Bioscience
Tiia Maxwell--Siemens Healthineers
Ross Molinaro--Siemens Healthineers
Asuka Monobe--Miraca Life Sciences
Shanti Narayanan--Raisin Bioscience
Godwin Ogbonna--Quidel Ortho
Christopher Reamer--Siemens Healthineers
Jeanne Rhea-McManus--Siemens Healthineers
Adriana Rycovska-Blume--Diasorin
Miguel Sainz--Mercodia
Hiroki Sato--Tosoh Bioscience
Ana Shulla-Mesi--Abbott
Matthew Stewart--Quidel Ortho
Joe Sun--Snibe
Yutaka Tahara--Tosoh Bioscience
H. Tsukamoto--Tosoh Bioscience
Edward Vachula--Abbott
Mie Wakabayashi--Miraca Life Sciences
Chris Wisherd--Alpco
Zeng Wu--Quest Diagnostics
Irene --Snibe

1) **Welcome and Introduction—Randie Little**
   R. Little welcomed those in attendance. Participants introduced themselves and the 2023 meeting minutes were approved.
2) **The clinical importance of C-peptide measurement for type 1 diabetes prevention—Paolo Pozzilli**
   - Clinical outlook of C-peptide in type 1 diabetes (T1D)
     - Measurement of C-peptide is useful to identify T1D patients who maintain residual beta-cell function. Detectable C-peptide characterizes long standing T1D patients who do not develop late complications (retinopathy, neuropathy, nephropathy).
     - The assessment of C-peptide during the prodromic phase of T1D (i.e. after islet cell autoantibody sero-conversion but before clinical onset) represents a marker to predict progression towards T1D development in high risk islet autoantibody positive subjects.
     - Maddaloni E.,…, Pozzilli P., Buzzetti R., Diabetes Obes Metab. 2022, Schatz D, et al., Pediatr Diabetes. 2004, Jones AG, Hattersley AT., Diabet Med. 2013
   - Stages 1 and 2 of T1D are pre-symptomatic despite the presence of 2 or more islet autoantibodies, and stage 3 is characterized by clinical diagnosis of T1D
     - Genetic Risk: Starting Point
     - Immune Activation
       1) Trigger event

2) Development of single antibody
3) C-peptide pathophysiology

- Stage 1
  1) Normal blood sugar
  2) ≥2 autoantibodies
  3) C-peptide may be normal
  4) 5 yr risk of clinical diagnosis of T1D: 44%
- Stage 2
  1) Abnormal blood sugar
  2) ≥2 autoantibodies + dysglycemia
  3) Dysglycemia may appear, C-peptide declining
  4) 5 yr risk of clinical diagnosis of T1D: 75%
- Stage 3
  1) Clinical diagnosis based on ADA criteria for diagnosis of diabetes
  2) ≥2 autoantibodies
  3) C-peptide still detectable
- Long standing T1D
  1) >Established T1D
  2) C-peptide continues to decline after diagnosis
- By the time people with T1D display symptoms, β cell mass is significantly reduced.
- Sims EK, et. al. Diabetes 2022; Besser RE, et al. Pediatr Diabetes 2022; ElSayed NA et al. Diabetes Care. 2023

- Individuals with stage 2 type 1 diabetes have a 74% chance of progressing to stage 3 within 5 years, and a near 100% lifetime probability of progression
- Pathophysiology: β cell function, defined based on dysglycemia, can be identified using provocative testing (e.g. oral glucose tolerance tests)
- Standardization of C-peptide measurement
  - Assessment of endogenous insulin secretion by measuring plasma C-peptide is widely accepted. However, considerable variation currently exists between different assay methods of C-peptide, giving variable results, despite their common traceability to the original World Health Organization standard (International Reference Reagent (IRR) 84/510).
  - Interest from manufacturers in moving forward with standardization could come from specific clinical recommendations from well-recognized clinical organizations with high promotion potentials (e.g. ADA, European Association for the Study of Diabetes [EASD], among others) that would increase awareness of the clinical utility of C-peptide testing.
  - Maddaloni E.,, Pozzilli P., Buzzetti R., Diabetes Obes Metab.2022
- Final remarks
  - A new paradigm of C-peptide measurement is represented by its measurement in subjects positive for islet cell antibodies and progressing to T1D onset.
  - Trials to prevent T1D are now ongoing in islet cell autoantibody positive (GAD-IA2, ZnT-8) subjects at stage 1 and 2 of the pre-T1D phase.
  - C-peptide represents the primary endpoint of trials aimed to prevent T1D (FDA approved).
  - Sample size are based on this marker; therefore, the use of well standardized C-peptide assays represents a key aspect for designing T1D preventing trials.

Discussion:

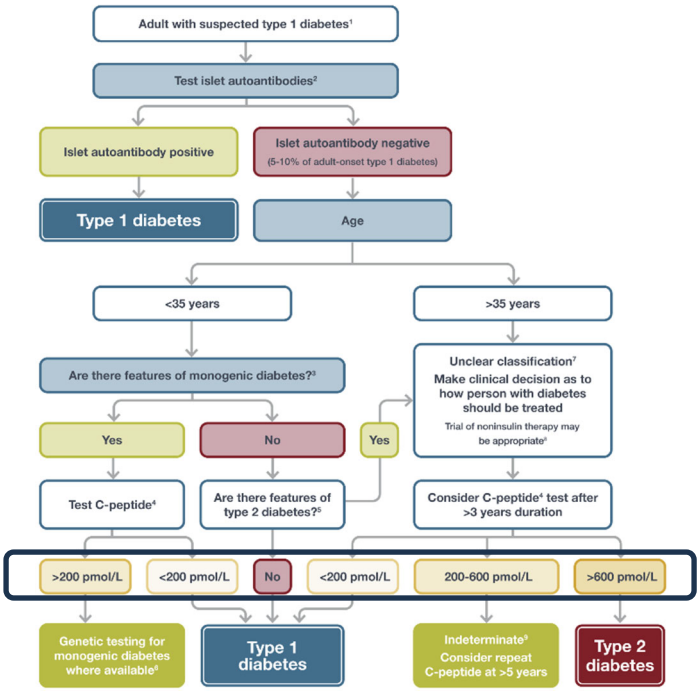
P. Pozzilli said that C-peptide will become more widely used in screening of subjects at risk for T1D in populations. A. Hoofnagle asked about cutoffs for detecting people that are C-peptide deficient, is there a limit of quantification that we need to be targeting in our assays? P. Pozzilli responded that the focus is on 3 groups: <0.2, 0.2-0.6 and >0.6 nmolar C-peptide. Of particular interest are subjects in the 0.2-0.4 range, because these are the subjects most likely to benefit from therapeutic interventions. J. Li asked if there are other tests besides C-peptide that should be used in determining which subjects are at risk for development of T1D, e.g. autoantibodies. P. Pozzilli said the C-peptide is generally going to be measured in subjects that are already autoantibody positive. Autoantibodies indicate that immune destruction of the beta cells has initiated, then the C-peptide level will indicate how extensive this beta cell destruction is. D. Holmes noted that C-peptide standardization becomes very important when absolute

thresholds are utilized, and asked P. Pozzilli if he anticipates the widespread adoption of the thresholds being discussed. P. Pozzilli responded that these intervention thresholds are already being recognized in terms of initiating insulin therapy along with other interventions, the recommendations are 0.2-0.4 or 0.3-0.5 nmolar depending upon whether the American or European guidelines are used. In T1D trials C-peptide is used as the endpoint in determining the effectiveness of the treatments used to delay or prevent diabetes, so the assay is very important. Standardization is important to this, and to convince regulatory organizations such as the FDA that C-peptide is the endpoint that should be used to assess the efficacy of these treatments. W. Hagopian said that in trials C-peptide is assessed as an area under the curve after a mixed-meal tolerance test. Random C-peptide is more practical, but they generally measure glucose as well to make sure it is not low which would indicate that the C-peptide is not measuring residual C-peptide function. P. Pozzilli said that random C-peptide is still a good test for subjects at risk, but also a oral glucose tolerance test or continuous glucose monitoring should be performed as well. There has been an explosion in the number of T1D intervention trials that are using different C-peptide assays, in this situation it is very difficult to use C-peptide as a primary endpoint, this is why standardization is extremely important. W. Hagopian asked about identifying dysglycemia in stage 2. There are different ways to do so, including OGTT, HbA1c and home glucose monitoring but they are not always consistent, would C-peptide be a better way of determining who should receive immunotherapeutic intervention? P. Pozzilli said yes, the problem is that the FDA is currently opposed to using C-peptide, they look at HbA1c. If you can convince them that measuring C-peptide and using CGM to assess time-in-range are the best way the FDA might come to recognize C-peptide, but no one is currently considering time-in-range as the best way to determine if the patient is doing well. R. Little asked about C-peptide in Type 2 patients. P. Pozzilli said that in T2 patients with poor metabolic control they measure C-peptide and if it is <0.2 nmol they can test for autoantibodies. If they are present they can conclude that the patient actually has LADA or T1 and requires insulin therapy.

3) C-Peptide Study Update—Kuanysh Kabytaev

- C-peptide Testing: Current State
  - Recommendation: Insulin and C-peptide assays should be standardized to facilitate measures of insulin secretion and sensitivity that will be comparable across research studies. GPP—Guidelines and Recommendations for Laboratory Analysis in the Diagnosis and Management of Diabetes Mellitus. Diabetes Care 2023;46(10):e151-e199.
  - Diagnosis and Classification of Diabetes: Standards of Care in Diabetes—2024, American Diabetes Association Professional Practice Committee, Diabetes Care 2024;47 (Supplement_1):S20-S42

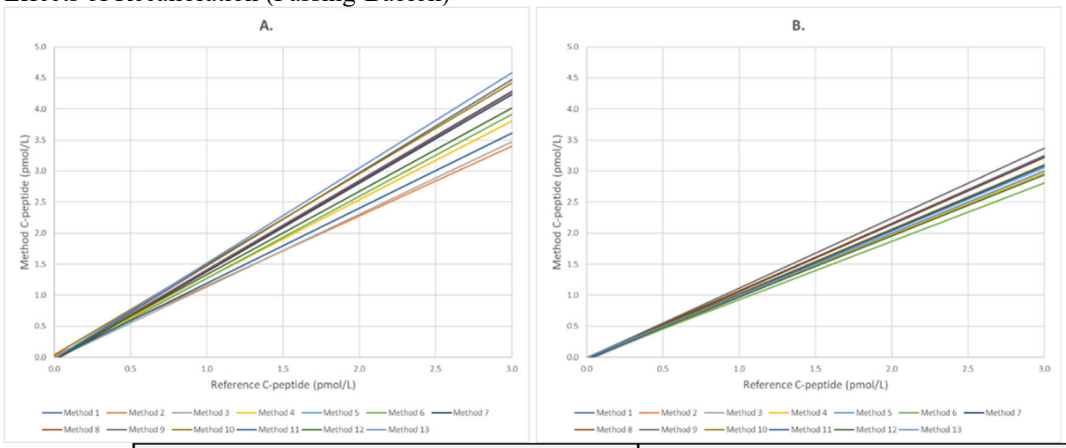
Flow chart for investigation of suspected type 1 diabetes in newly diagnosed adults, based on data from White European populations



- Current Study
  - C-peptide Methods

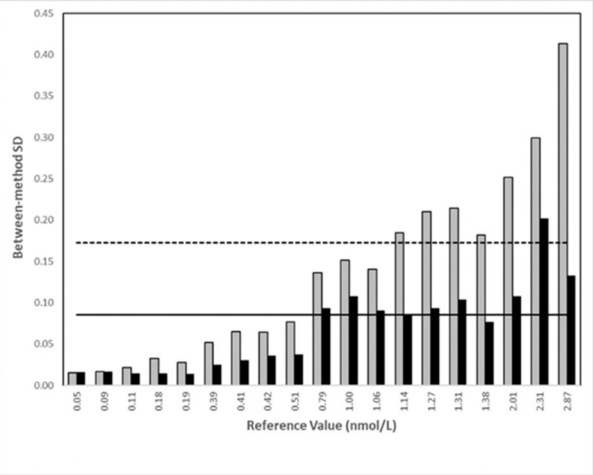
Vendor	Method/Platform	Primary Calibrator	Analytical Measurement Range, nmol/L	Assay Principle
Abbott Laboratories	Alinity i	WHO 84/510	0.01-10	Chemiluminescence immunoassay
Beckman Coulter, Inc	C-Peptide/Access2 Immunoassay System	WHO 13/146	0.03-10	Chemiluminescence immunoassay
DiaSorin Deutschland GmbH	LIAISON® C-Peptide / LIAISON® Analyzer family	WHO 84/510	0.003-10	Chemiluminescence immunoassay
QuidelOrtho	VITROS /(ECi/ECiQ, 3600 Immunodiagnostic Systems & XT 7600 Integrated Systems)	WHO 84/510	0.02-10	Chemiluminescence immunoassay
Fujirebio Inc	Lumipulse C-peptide / Lumipulse G1200	WHO 84/510	0.01-10	Chemiluminescence immunoassay
Siemens Healthineers	Atellica IM	WHO 13/146	0.02-10	Chemiluminescence immunoassay
Tosoh Corporation	CL AIA-PACK C-Peptide / AIA-CL2400	WHO 84/510	0.003-13.2	Chemiluminescence immunoassay
ALPCO	ELISA Chemiluminescent	WHO 84/510	0.001-4.3	Chemiluminescence immunoassay
Tosoh Corporation	ST AIA-PACK C-Peptide II / AIA-2000	WHO 84/510	0.01-10	Fluorescence enzyme immunoassay
Mercodia AB	Mercodia C-peptide ELISA	WHO 84/510	0.1-4	Sandwich ELISA
Roche Diagnostics GmbH	Elecsys C-Peptide	WHO 84/510	0.01-13.2	Electrochemiluminescence immunoassay

  - Effects of Recalibration (Passing-Bablok)

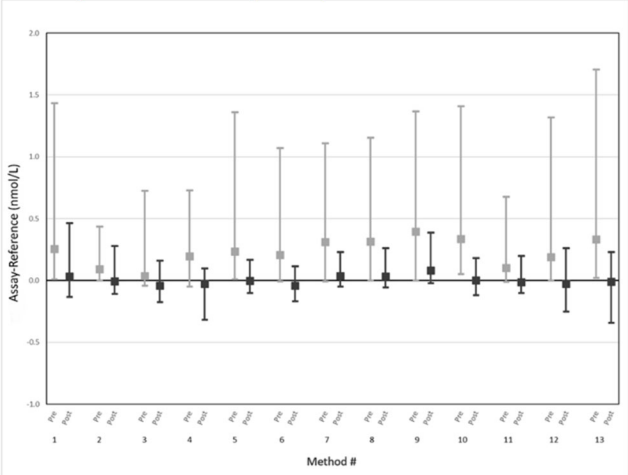


	Pre-Calibration				Post-Calibration			
	Int (95% CI)	*Slope (95% CI)*	*Int=0*	*Slope=1*	*Int (95% CI)*	*Slope (95% CI)*	*Int=0*	*Slope=1*
Method 1	-0.029 (-0.088-0.020)	1.438 (1.351-1.567)	Y	N	-0.028 (-0.050-0.018)	1.094 (1.029-1.189)	Y	N
Method 2	0.026 (-0.019-0.061)	1.123 (1.048-1.207)	Y	N	-0.026 (-0.071-0.000)	1.041 (0.973-1.120)	Y	Y
Method 3	-0.035 (-0.088--0.005)	1.168 (1.092-1.265)	N	N	-0.047 (-0.074--0.010)	1.016 (0.942-1.081)	N	Y
Method 4	0.007 (-0.041-0.028)	1.265 (1.200-1.387)	Y	N	-0.025 (-0.059--0.009)	0.998 (0.946-1.074)	N	Y
Method 5	-0.015 (-0.065-0.013)	1.416 (1.317-1.500)	Y	N	-0.001 (-0.044-0.015)	1.020 (0.955-1.072)	Y	Y
Method 6	-0.052 (-0.090--0.005)	1.322 (1.258-1.409)	N	N	-0.013 (-0.062-0.019)	0.939 (0.900-1.045)	Y	Y
Method 7	-0.048 (-0.084--0.001)	1.426 (1.370-1.495)	N	N	-0.017 (-0.051-0.018)	1.079 (1.021-1.111)	Y	N
Method 8	-0.039 (-0.085--0.008)	1.437 (1.376-1.500)	N	N	-0.004 (-0.054-0.017)	1.078 (1.030-1.125)	Y	N
Method 9	-0.030 (-0.091--0.010)	1.500 (1.444-1.571)	N	N	-0.008 (-0.057-0.014)	1.126 (1.074-1.177)	Y	N
Method 10	0.036 (-0.034-0.061)	1.462 (1.354-1.538)	Y	N	-0.011 (-0.049-0.015)	1.039 (0.954-1.086)	Y	Y
Method 11	-0.019 (-0.043-0.018)	1.211 (1.113-1.262)	Y	N	-0.034 (-0.068--0.006)	1.041 (0.967-1.097)	N	Y
Method 12	-0.013 (-0.087-0.012)	1.341 (1.266-1.457)	Y	N	-0.013 (-0.062-0.011)	0.984 (0.925-1.063)	Y	Y
Method 13	-0.018 (-0.074-0.023)	1.533 (1.429-1.653)	Y	N	0.000 (-0.048-0.020)	1.000 (0.943-1.094)	Y	Y

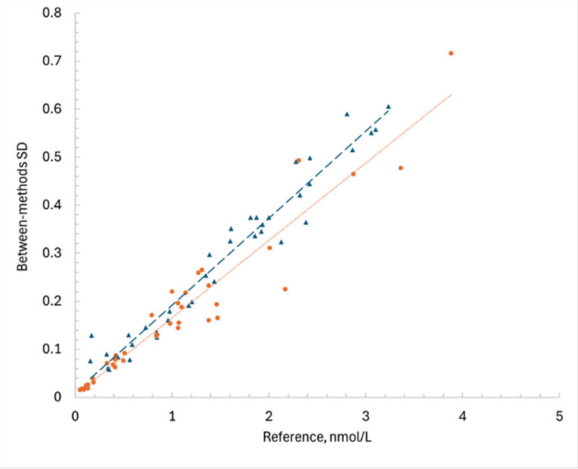
  - Variation Among Methods for Different C-peptide Levels



- Limits of Agreement Among Assays



- Study 2008 vs. Current Study (Pre-calibration)



Discussion:

K. Kabytaev noted that despite intervening years of technical improvement and replacement of older methods with newer ones, there has been little improvement in the comparability among assays since 2008. The explanation for this is likely the lack of commutability of the WHO material, and it is possible that the new WHO material may be

even less commutable as it does not contain albumin. G. Marcos said the post-calibration data look promising, as noted in the previous presentation the lower nmolar levels will be especially important given that there are now diagnostic guidelines with cutoff points in that range. R. Little said it is not surprising that the pre-calibration data look similar between 2008 and the current study despite technological improvements, as the methods are still calibrated with non-commutable material. K. Kabytaev said that going forward the UM group will be collecting more samples and will try to obtain a better distribution of values so as to examine different ranges more closely. The lower end will be the most important. G. Ogbonna asked about bias at the low end of the range, noting the slopes all indicate systematic positive bias prior to re-calibration but after calibration the slopes are all close to one. Should we focus on the slopes in the lower range, and what is acceptable? What is clinically relevant? R. Little responded that the low end of the range is important in terms of Type 1 diabetes, but C-peptide is also used in subjects with Type 2 diabetes when looking at insulin resistance, there are equations that include C-peptide in determining the degree of IR. Standardization of C-peptide would make these equations more useful, she was not sure what the clinically relevant range is for this purpose. Once standardization has been implemented we can look more closely at the clinically relevant ranges, right now we are just focused on overall standardization. K. Kabytaev added that in the current study most values were in the middle range, there were only a few samples in the low range. In the future there will be a focus on obtaining a more even distribution of values including more samples in the low range. A. Hoofnagle noted that all of the intercepts are negative in the current study and asked what might be driving this. K. Kabytaev replied that this may be a function of the distribution of the values in the dataset where samples at the higher end are affecting the overall equations such that they impacting the low end. This is why more samples with a good range distribution are needed, then we can divide the ranges and look at subranges more closely, especially the low end. Also many manufacturers are asking for samples that cover their analytical range including unphysiological high levels, which would impact these equations when looking at the entire range. A. Hoofnagle asked about the reproducibility of the samples at the low end. K. Kabytaev said that some vendors provided duplicate values while others provided single values, and there were only a few samples at the low end so it was not possible to obtain very good estimates. It is also important to run more replicates of samples at the low end on the mass spec reference method.

4) C-peptide Standardization Project using the SRM of DDL in Japan: Masami Murakami

- Collaboration involving six universities in Japan
- SRM and serum and urine samples collected at multiple institutions will be distributed to participating manufacturers to conduct measurements
  - Dr. Little and Dr. Kabytaev of University of Missouri have kindly provided 8 sets of 8 concentrations of SRM.
  - Samples to be delivered
    - **1)** SRM from DDL network
    - **2)** Patient serum (299 samples)
    - **3)** Patient urine (121 samples)
- Abbott Japan, Beckman Coulter, FUJIFILM Wako Pure Chemical Corporation, FUJIREBIO, Ortho Clinical Diagnostics, Roche Diagnostics, Siemens HealthCare Diagnostics, and Tosoh Corporation have agreed to participate in this project.
- Number of serum and urine samples collected at multiple institutions

Serum Samples of Patients for C-peptide Standardization Project

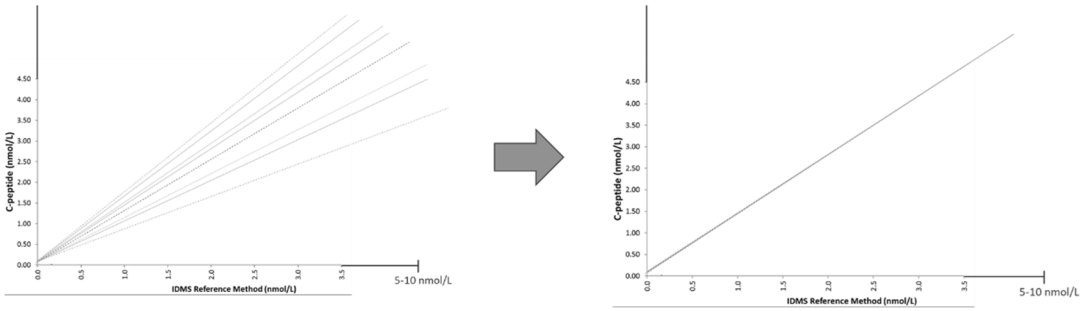
C-peptide Range of Serum Samples		Number of Samples Collected at Multiple Institutions
ng/mL	nmol/L	
<0.01	<0.003	4
0.01～1.0	0.003-0.3	64
1.0～3.0	0.3-1.0	113
3.0～5.0	1.0-1.7	60
>5.0	>1.7	58
Total		299

Urine* Samples of Patients for C-peptide Standardization Project

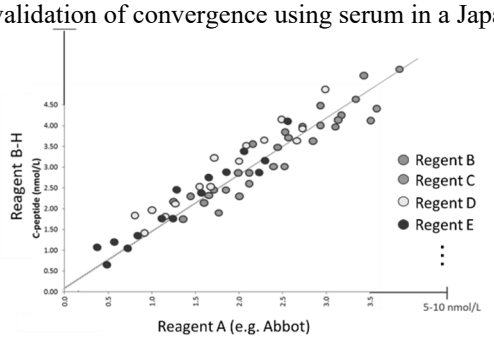
C-peptide Range of Urine Samples		Number of Samples Collected at Multiple Institutions
ng/mL	nmol/L	
0.1～10	0.03～0.3	30
10～50	3～17	49
50～100	17～33	27
>100	>33	15
Total		121

*Mainly 24-hour urine collection, with preservatives added.

- SRM will be measured using the reagent of each manufacturer, and a correction coefficient will be determined from the results of reference method of the University of Missouri DDL Network



- Anticipated findings and outcomes of this study
  - Correction coefficients of eight Japanese reagent manufacturers will be identified.
  - Because all Japanese C-peptide reagent manufacturers participate in this study, it will support C-peptide standardization in Japan.
- Re-validation of convergence using serum in a Japanese cohort (CV% evaluation)



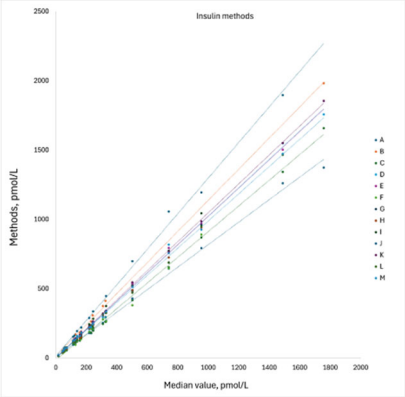
- In the future, we hope to verify applicability of standardized C-peptide measurement to urine samples.
- Tentative Plan for the Future
  - Based on the results of the present study, we hope to obtain agreement of academic societies such as Japan Diabetes Society (JDS) on standardization of C-peptide measurement.
  - We would like to ask eight participating manufacturers to provide standardized values with correction coefficient in accordance with the DDL network in Japan.
  - We hope to conduct EQA/PT of C-peptide measurement in Japan.

5) Insulin Update — Kuanysh Kabytaev

- Insulin methods

Vendor	Method/Platform	Primary Calibrator	Reported Units	Lowest Detection Limit	Assay Range Without Dilution
Abbott	Alinity i	WHO 66/304	µU/mL	0.4 µU/mL	1.6 – 300.0 µU/mL
ALPCO	ELISA Chemiluminescent	WHO 11/212	pg/mL	2 pg/mL	5 – 30000 pg/mL
Fujirebio Inc	Lumipulse G Insulin-N / Lumipulse G1200	WHO 66/304	µIU/mL	0.0197 µIU/mL	0.6 – 400 µIU/mL
Mercodia	Mercodia Insulin ELISA	WHO 66/304	mU/L	1 mU/L	3 – 200 mU/L
QuidelOrtho	VITROS /(ECi/ECiQ, 3600 Immunodiagnostic Systems & XT 7600 Integrated Systems)	WHO 66/304	µIU/mL or pmol/L	0.077 µIU/mL	1.00 – 300 µIU/mL
Roche Diagnostics Siemens	Elecsys Insulin	WHO 66/304	µU/mL or pmol/L	≤ 0.4 µU/mL	0.4 – 1000 µU/mL
Healthineers	Atellica IM	WHO 66/304	mU/L	0.8 mU/L	0.5 – 300.0 mU/L
Tosoh (AIA)	ST AIA-PACK IRI / AIA-2000	WHO 66/304	µU/mL	0.5 µU/mL	0.5 – 320 µU/mL
Tosoh (CL)	CL AIA-PACK IRI / AIA-CL2400	WHO 66/304	µU/mL	0.033 µU/mL	0.2 – 1000 µU/mL

- Discordance Among Methods



Discussion:

K. Kabytaev said the UM insulin reference method is still being developed. One of the major issues is that the current chemical enrichment protocol is not providing adequate sensitivity in the low end of the measurement range. One of the vendors has provided an antibody enrichment system that should provide improved sensitivity. Regarding the comparison among methods, there were some samples where the subjects were using exogenous insulin and large discrepancies were observed, these samples were therefore excluded from the final comparison. There are several insulin standards available for the reference method that are being considered. R. Little said that once the insulin reference method is established the plan is to use the same recalibration scheme that has been developed for C-peptide.

Feedback from Manufacturers

R. Little asked for feedback from manufacturers. There is now data from two studies and the reference method has been improved to the point where it is stable. The latest study results are about to be published, and data from these studies has clearly shown that the traceability scheme using serum samples with reference method-assigned values greatly reduces variability among the assays. She asked manufacturers to consider publishing white papers for their customers introducing them to the reference method and showing how their methods compare to the reference method. She has received some feedback, so far there have been requests for more samples and that the samples need to span the measurement range. The ranges for some assays go up to 10 nmol/L or higher, which is not physiological so it will not be possible to obtain samples in this range. For the next study samples spiked with certified reference material could be included in order to obtain these high levels. She asked the manufacturers what is needed in order for them to publish white papers. B. Escher said he appreciates the efforts to standardize these analytes, from their perspective they have to follow CLSI guideline meaning more samples are needed. Also, in order to publish a white paper they need the samples to cover the measurement range even if the higher levels are not physiological. R. Little asked what number of samples are needed, B. Escher responded it is typically over 100, i.e. 120. R. Little said we now have 40 samples and are in the process of collecting another set, but did not think more than that could be collected in the near term, maybe 80 samples would be adequate? G. Ogbonna agreed with B. Escher, for CLSI above 100 is considered the minimum. Calibration is very important, when re-calibration occurs there can be a shift in controls that has a huge impact. Before this is done we have to be really confident that the samples span the measurement range, also a large number of sample in the clinically relevant range is very important. He suggested sending the distribution of samples to manufacturers, then they can provide feedback. R. Little said they can share their goals as far as the levels are concerned with manufacturers. They can be more specific for the samples that will be spiked, and subjects are screened before the actual draw, but in the low and middle ranges they cannot be sure of what the levels are until the samples have been drawn. She asked about CLSI, it seems the guideline is for method comparison but she was not aware of a guideline for re-calibration of assays. G. Ogbonna said that the CLSI guideline is for method comparisons, and for that purpose 100 samples is a minimum. When they assign values to calibrators they actually run hundreds of samples. The value assignments to calibrators is extremely important in terms of customers and the manufacturing process, so they have to make sure they are comfortable with the results. R. Little asked if for the purpose of a white paper 80 samples would be acceptable, then additional samples could be provided later for a total of 120 samples prior to the actual re-calibration. K. Kabytaev added that the white paper is just to provide customers with initial study results to let them know about the standardization and provide an initial view of the relationship between the assay and reference methods. For this

purpose, as opposed to the formal recalibration, is 40 samples adequate? G. Ogbonna responded that statistically 40 samples just gives an idea of the relationship between two methods. Above 100 samples gives more confidence, but for things their company does internally they analyze hundreds of specimens. Regarding a white paper, what would the benefit be? K. Kabytaev said the purpose of the white paper would be to simply inform customers of the standardization and give an idea of how the method relates to the reference method. R. Little added that the white paper would inform customers prior to actual recalibration. S. Narayanan agreed with G. Ogbonna and B. Escher regarding the CLSI guideline requirements for sample sizes in terms of their requirements in their SOPs. However, she noted that the white paper is to establish the need for recalibration and how the methods will be brought closer together. The purpose is to make not only customers but also regulatory bodies that will be reviewing the manufacturers' methods aware of the recalibration process. For C-peptide a new 510k will not be required since it is a Class I exempt analyte, but data will still need to be kept on file. R. Little asked L. Landree to comment, she said that S. Narayanan is correct but added that manufacturers should consider intended use. If the intended use changes that could change the classification. There are still other requirements to register and list, and they recommend submission of a CLIA categorization request if recalibration is performed and the assay performance changes. S. Narayanan asked about long-term clinical studies, there will likely be shifts after recalibration and this is a concern for both manufacturers and study sponsors. How do we address this? R. Little said the relationship between the old and new calibrations will be known, so a given central lab could use the old calibration for existing studies and the post-recalibration results for new studies. S. Narayanan said this would effectively require manufacturers to maintain two assays, one pre and one post-calibration. R. Little did not know the rules involved with that but said at least the relationship would be known and could be introduced by a white paper. S. Narayanan said in that sense the white paper could be beneficial to manufacturers, in that there are many manufacturers involved and knowing the relationships would give confidence to sponsors of clinical studies. They would still need crossover studies performed prior to the change to the new calibration, but at least it would give them an idea of where they stand. G. Ogbonna noted that the reference intervals would change asked about the crossover studies, would these be performed by the manufacturers or the individual labs? R. Little asked what manufacturers normally do, for example if a switch to the new WHO material results in a shift in the reference range. G. Ogbonna responded that the manufacturers do this internally. S. Narayanan agreed, saying manufacturers would need to do their own reference range studies if there is a significant shift, but in the case of long-term clinical studies the labs involved would also likely need to do their own validation studies based on the CLSI protocol for verifying reference intervals. She asked L. Landree for comments, L. Landree replied that she does not have experience with these types of trials and suggested checking with CDER or whatever organization(s) are responsible for reviewing these clinical studies for advice prior to implementing changes. G. Martos asked about the study showing the improvement of comparability when serum samples are used for calibration: is there a reason that the current reference materials were not included as part of the study? R. Little said one of the early studies the WHO material was sent to manufacturers, then when the results were used to re-calibrate the different assays it did not bring the results closer together. This is why the traceability chain was developed where a primary reference material is used to calibrate the reference method, which then assigns values to commutable serum materials. K. Kabytaev added that internal studies at UM showed that the NMIJ material is not commutable for commercial assays, but it is useful to calibrate the reference method because it is better characterized and well-defined. R. Little then outlined the next steps. UM is in the process of collecting another set of serum samples, as discussed earlier a proposal for the target ranges will be provided to manufacturers. It will need to be decided whether the 40 samples will again be divided into two groups or just send as one group. Values will be assigned by the reference method, then the samples will be sent to each manufacturer. She then asked for additional comments from manufacturers regarding the development of a white paper on the part of each manufacturer, using the data already provided. M. Sainz said they have requested the final draft, once they have been able to review it further internal discussions will take place. R. Little said the final draft will not be significantly different from the draft already provided. The final draft will be sent to the manufacturers, but the data will be the same. Now is the time for the discussions to take place while the new set of samples is being collected. Someone from Mercodia actually brought up the idea of a white paper at a previous meeting, would they be open to that? M. Sainz responded that there have been some personnel changes and more internal discussions involving management will need to take place. R. Little asked for input from Tosoh, C. Martin said he needs to consult with R&D about whether they have received the samples and their status. R. Little asked for someone from Abbott. J. Jaraczewski said the problem with the white paper is that it implies they are making a claim and they cannot use white papers to make a claim that you can just use a conversion to make a switch. To standardize would have to be full bore, involving resubmissions or updates by country. R. Little said the purpose of a white paper is not to make a claim, but just to provide information for customers regarding the reference method and presenting the preliminary relationship with the reference method. J. Jaraczewski replied

that it is a very fine line. L. Heinemann encourage the use of a white paper, as there is a worldwide lack of knowledge among diabetologists. R. Little reiterated that the purpose is that it is a way to inform customers prior to actual recalibration. She looked into the definition of a white paper and did not get the impression that it is for the purpose of making a claim. S. Narayanan asked about authorship, who would actually author the white paper. R. Little responded that each manufacturer would author their own white paper to present to their customers. She noted that the clinical importance of standardization has been presented, and that standardization is better when performed before there is a pressing clinical need. This way the change is not made in the middle of patient treatment/surveillance, it can start with a white paper then the recalibration can occur as more physicians start to use C-peptide. G. Ogbonna agreed that standardization is important and stated that he will need to have internal discussions with the various departments involved and get back to R. Little. R. Little said they are happy to help if any manufacturers need further information. A. Hoofnagle asked about other standardization programs such as HbA1c, testosterone, estradiol, etc., he did not recall any of them using more than 40 samples and asked R. Little for her experience with HbA1c. R. Little noted that the NGSP began before there was a true reference method for HbA1c or a traceability chain. The drive for it was the results of the DCCT study, from which data are still being obtained. Over the years that they have been working on C-peptide standardization they have received a number of requests in terms of what is needed for standardization, including listing of the reference method on JCTLM, use of a certified reference material and a traceability chain. These have been achieved, she was not sure about previous standardization schemes for other analytes and did not think they are truly standardized even if there is a program for standardization. For example, with TSH there was a feeling that manufacturers had gotten closer to the reference method and there was not a point where manufacturers formally recalibrated. In the case of the AACC or IFCC standardization groups, it seems that once they have a list of methods and descriptions and have a complete traceability chain they consider the analyte standardized. However, if you look at proficiency testing results it does not necessarily show that manufacturers have adjusted their methods so they match. There is now an accuracy-based CAP PT survey that includes C-peptide and they are trying to get more labs to participate in the survey. That is the only well to tell if an analyte is truly standardized. With HbA1c there is accuracy-based PT and a certification program, at some point this could be done with C-peptide, right now the goal is to get the different assays aligned and then actual criteria for what constitutes acceptable performance can be determined. A. Hoofnagle said that the example of the DCCT driving the need for standardization of HbA1c is a good one and that there will eventually be a similar need with C-peptide. He supported the idea of a white paper and achieving standardization prior to the need becoming apparent. D. Holmes asked about the white paper, and asked is the proposal that the white paper which is a document normally used within a given organization be issued by each manufacturer with similar verbiage? That would seem to be a lot of work, or is the proposal to have this group prepare a white paper that manufacturers can agree to then that being passed along? R. Little said years ago it was proposed that the group could prepare a document that could assist manufacturers with providing the information. This was done, and could be done again if it would be helpful. D. Holmes said it would seem to be time-consuming to have each manufacturer prepare their own white paper, and it would likely be done over a dispersed period of time. He wondered if it would be more efficient to get all or at least some of the manufacturers to participate in publishing a paper with the IFCC or ADLM, or in a journal. R. Little, said she could ask in an e-mail if it would help the manufacturers if the group wrote something, but in the end they would have to be the ones to present the information to their customers. She and Dr. Kabytaev will be attending the upcoming IFCC meeting, they are hoping to get more information from the IFCC regarding how to proceed in these efforts. All of this originally came out of the IFCC and the EU Directive, but they do not seem to be backing up their request for everyone to standardize measurements. This group has been working toward C-peptide standardization for years but thus far has made little progress. She asked manufacturer representatives present if it would help for the group to prepare an example of a white paper. K. Kabytaev suggested that the manufacturer white papers would likely be different for different manufacturers. Some methods are already close to the reference method and those manufacturers might be more motivated to share white papers. For those manufacturers there may be less involved in terms of regulatory requirements, paperwork, etc. R. Little agreed, noting that they may just have to show the data demonstrating that they are already well aligned.

R. Little said that she would like to obtain more comments via e-mail, and thanked everyone present for their attendance, the meeting was adjourned at 11:00 AM CST.

Minutes prepared by Curt Rohlfing 11/22/2024.