

Biennial activity report from JCTLM Member organizations

Organization: TUBITAK National Metrology Institute, TUBITAK UME

JCTLM Member status: Member

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Period covered: 2015 – 2017

1. Major achievement(s) in support of standardization in laboratory medicine

➤ **The production of certified reference materials:**

- A certified reference material preparation project “25-Hydroxy Vitamin D2 and 25-Hydroxy Vitamin D3 in Lyophilized Serum” was completed and the CRM is on sale. The application was submitted for listing under JCTLM database.
- The project titled “Development and Production of Certified Reference Materials and Quality Control Materials for Newborn Screening and Measurements Routinely Performed by Clinical Biochemistry Labs for Amino Acids and Organic Acids” is a certified reference material production project that was financially supported by TUBITAK. Zivak Technologies, a LC-MS based test and instrument producer company, is also an industrial partner in the project. The project aims to produce and certify about 50 amino acids and 50 organic acids which are the metabolites for different genetic diseases in human plasma and urine, respectively. The certification of analytes was completed and the CRMs will be on sale in 2018.
- “HbA1c CRM Production” project, internally financed, started in 2016 and it is expected to be completed in 2018 and the CRM will be on sale in 2019.

➤ **Development of reference measurement methods**

- Primary ID-LC-MS reference measurement method for 25-Hydroxy Vitamin D2 and 25-Hydroxy Vitamin D3 in Lyophilized Serum was developed and a manuscript will be submitted to a peer reviewed journal.

➤ **Projects in the field of Laboratory Medicine:**

- **EMRP and EMPIR projects:**

Chemistry Group Laboratories have been collaborating in health related EMRP and EMPIR projects and contributes to the studies in the development of new measurements methods and certified reference materials in Laboratory Medicine. Our laboratories were partners in the following EMRP projects completed in the period of 2015-2017:

- EMRP-Metallomics: Metrology for Metalloproteins, 2012-2015.
- EMRP-INFECT-MET: Metrology for monitoring infectious diseases, antimicrobial resistance, and harmful micro-organisms, 2012-2015.
- EMRP-Bio-SITrace: Traceability for biologically relevant molecules and entities, 2013-2016.

Our laboratories are partners in the following health related EMPIR projects:

- EMPIR-AntiMicroResist - Novel methods and materials for the detection, traceable monitoring and evaluation of antimicrobial resistance, 2016-2019.
- EMPIR-ReMIND - Role of metals and metal containing biomolecules in neurodegenerative diseases such as Alzheimer’s disease, 2016-2019.

- **Dual Collaboration Project:**

A dual collaboration project, “Korea-Turkey collaboration project on the development of an international standard system for the measurement of gene methylation” started in 2015 and it will be completed at the end of 2017. A new measurement method for genomic DNA methylation measurement, which is a critical marker on the diagnosis of several diseases including cancer, will be suggested.

➤ **The establishment of calibration (reference) measurement services.**

The following reference measurement services were offered in 2017 and listed under TUBITAK UME services (Table 1).

Additionally, method verification was performed for a ProCalcitonin auto analyzer.

Table 1. CCQM and EURAMET international measurement comparison studies between 2015 and 2017.

Code	Measurement Service Name
G3OK-4600	Determination of 25-OH Vitamin D2, D3 and 3-epi-25-Hydroxyvitamin D3 in Serum
G3OK-4610	Determination of Vitamin 25-OH D2
G3OK-4620	Determination of Vitamin 25-OH D3 and 3-epi-25-OH Vitamin D3
G3OK-5600	Determination of Cholesterol in Serum
G3OK-5700	Determination of Glucose in Serum
G3OK-5800	Determination of Creatinine in Serum
G3IK-3301	Determination of As in Biological Materials
G3IK-3302	Determination of Pb in Biological Materials
G3IK-3303	Determination of Cr in Biological Materials
G3IK-3306	Determination of K in Human Serum
G3IK-3307	Determination of Ca in Human Serum
G3IK-3308	Determination of Mg in Human Serum
G3IK-3309	Determination of Fe in Human Serum
G3IK-3310	Determination of Selenomethionine in Human Serum
G3BA-2100	Determination of Relative Gene Expression Levels - Real Time PCR
G3BA-5100	Amino Acid Analysis For Pure Protein/Peptide - LC-ID-MS
G3BA-5200	Protein Identification - Peptide Mass Fingerprinting Method, LC-MS/MS
G3BA-5300	Quantification of c-peptide/angiotensin - LC-ID-MS(n)
G3BA-5400	Quantification of Protein Concentration - HPLC
G3BA-5500	Quantification of Protein Concentration - ELISA
G3BA-5600	Quantification of Protein Concentration - PICAA
G3BA-5700	Relative Quantification of HbA1c Protein - HPLC
G3BA-5800	Relative Quantification of HbA1c Protein- LC-ID-MS
G3BA-5900	Determination of Amylase Enzyme Activity

➤ **Outline of the measurement area(s)/measurands covered:**

- **CCQM and EURAMET Measurement Comparison Studies**

TUBITAK UME Chemistry Group Laboratories have participated to several international measurement comparison studies in the field of Laboratory Medicine between 2015 and 2017 (Table 2). More resources will be devoted to the development of new reference measurement systems in this field.

Table 2. Participated CCQM and EURAMET international measurement comparison studies by TUBITAK UME between 2015 and 2017.

Comparison #	Comparison Name
EURAMET 1185	Determination of Selenomethionine in Human Serum
CCQM-K6.2	Determination of Total Cholesterol in Human Serum
CCQM-K11.2	Determination of Glucose in Human Serum
CCQM-K12.2	Determination of Creatinine in Human Serum
CCQM-K55.c	Characterization of Organic Substances for Chemical Purity (Valine)
CCQM-K55.d	Organic Purity Assessment Folic Acid
CCQM-K78.a/P121.a	Polar Analytes in a Multicomponent Aqueous Solution: Mass Fraction of Amino Acids in Acidic Solution
CCQM-K132	Low Polarity Analytes in a Biological Matrix: Vitamin D Metabolites in Human Serum
CCQM-K107	Total Elements and Selenomethionine in Human Serum
CCQM-K109/P148	Urea and Uric Acid in Human Serum
CCQM-K115/P55.2	Peptide Purity Determination - Synthetic Human C Peptide (HCP)
CCQM-K139*	Elements in Human Serum
CCQM-P58.1	Fluorescence in ELISA (Stage 2) - cTnI Cardiac Troponin I Measurement
CCQM-P94.2	Quantification of DNA Methylation
CCQM-P117.d	Purity Assessment of High Purity Organic Materials: Folic Acid
CCQM-P137	Clinical Amylase Measurement
CCQM-P155	Multiple Cancer Cell Biomarker Measurement
CCQM-P164*	hGH Quantification in Serum
CCQM-P165	Quantification of CD34+ Cell Counts
CCQM-P184*	Copy Number Concentration and Fractional Abundance of a Mutation (SNV or INDEL) Mixed with Wild-type DNA

*In progress

➤ **Calibration and Measurement Capabilities:**

By using its expertise and providing services in clinical measurements, TUBITAK UME Chemistry Group Laboratories applied for calibration and measurement capabilities (CMC) tables and several measurement capabilities were obtained (Table 3).

Table 3. TUBITAK UME Calibration and Measurement Capabilities gained between 2015 and 2017.

Measurement Service Sub-Category	Matrix	Analyte or Component
Biological fluids and materials	Blood serum	25-hydroxyvitamin D2(25(OH)D2
Biological fluids and materials	Blood serum	25-hydroxyvitamin D3(25(OH)D3
Biological fluids and materials	Blood serum	Arsenic
Biological fluids and materials	Blood serum	Lead
Biological fluids and materials	Blood serum	Chromium
Biological fluids and materials	Blood serum	Calcium
Biological fluids and materials	Blood serum	Potassium
Biological fluids and materials	Blood serum	Magnesium
Biological fluids and materials	Blood serum	Selenomethionine
Biological fluids and materials	Blood serum	Sodium
Biological fluids and materials	Blood serum	Chlorine
Biological fluids and materials	Blood serum	Copper
Biological fluids and materials	Blood serum	Selenium
Biological fluids and materials	Blood serum	Phosphorus
Biological fluids and materials	Blood serum	Transferrin
Biological fluids and materials	Blood serum	Cholesterol
Biological fluids and materials	Blood serum	Glucose
Biological fluids and materials	Blood serum	Creatinine
Other	Pure DNA sequence in buffered matrix	Linearized plasmid DNA between 3 kbp and 6 kbp

➤ **Listing of the relevant technical/scientific publications:**

- Ralf D. Josephs, Norbert Stoppacher, Steven Westwood, Robert I. Wielgosz, Ming Li, Milena Quaglia, Jeremy Melanson, Gustavo Martos, Desiree Prevoo, Liqing Wu, Sandra Scapin, Merve Öztug Senal, Lingkai Wong, Ji-Seon Jeong, Kelly Wai Yi Chan, Cristian G. Arsene and Sang-Ryoul Park. Concept paper on SI value assignment of purity - Model for the classification of peptide/protein purity determinations, *J. Chem. Metrol.* 11:1 (2017) 1-8
- Josephs, R.D., Li, M., Song, D., Westwood, S., Stoppacher, N., Flatschart, R., Oliveira, R., Melanson, J.E., Ohlendorf, R., Henrion, A., Kinumi, T., Wong, L., Liu, Q., Oztug Senal, M., Vatansever, B., Un, I., Goren, A.C., Akgoz, M., Quaglia, M., Warren, J. Key Comparison Study on Peptide Purity—Synthetic Human C-Peptide. *Metrologia*, 54:Technical Supplement, 2017, 08007
- Pavšič J., Devonshire A., Blejec A., Foy C.A., Van Heuverswyn F., Jones G.M., Schimmel H., Žel J., Huggett J.F., Redshaw N., Karczmarczyk M., Mozioglu E., Akyürek S., Akgoz M., Milavec M. Inter-laboratory assessment of different digital PCR platforms for quantification of human cytomegalovirus DNA. *Anal Bioanal Chem.* 2017
- Devonshire, A., O'Sullivan, D., Honeyborne, I., Jones, G., Karczmarczyk, M., Pavsic, J., Gutteridge, A., Milavec, M., Mendoza, P., Schimmel, H., Van Heuverswyn, F., Gorton, R., Cirillo, D., Borroni, E., Harris, K., Barnard, M., Heydenrych, A., Ndusilo, N., Wallis, C., Pillay, K., Mozioglu, E., Akyurek, S., Yalcinkaya, B., Akgoz, M., Hugget, J. The Use of Digital PCR to Improve the Application of Quantitative Molecular Diagnostic Methods for Tuberculosis. *BMC Infectious Diseases*, 16:3, 2016, : 1-10
- Coskun, A., Baykal, A.T., Kazan, D., Akgoz, M., Oztug Senal, M., Berber, I., Titiz, I., Bilsel, G., Kilercik, H., Karaosmanoglu, K., Cicek, M., Yurtsever, i., Yazici, C. Proteomic Analysis of Kidney Preservation Solutions Prior to Renal Transplantation. *Plos One*, 11:12, 2016, 1-18
- Devonshire, A.S., Sanders, R., Whale, A.S., Nixon, G.J., Cowen, S., Ellison, S., Parkers, H., Pine, P.S., Salit, M., Mcdaniel, J., Munro, S., Lund, S., Matsukura, S., Sekiguchi, Y., Kawaharasaki, M., Granjeiro, J.M., Lotsch, P.F., Saraiva, A.M., Couto, P., Yang, I., Kwon, H., Park, S., Demsar, T., Akyurek, S., Akgoz, M. An International Comparability Study on Quantification of mRNA Gene Expression Ratios: CCQM-P103.1. *Biomolecular Detection and Quantification*, 8, 2016, 15-28
- Mozioglu E, Gokmen O, Tamerler C, Kocagoz ZT, Akgoz M. Selection of Nucleic Acid Aptamers Specific for Mycobacterium tuberculosis. *Appl Biochem Biotechnol.* 178, 4, 2016, 849-64
- Mozioglu, Erkan; Akgoz, Muslum; Kocagoz, Tanil; Tamerler, Candan, Detection of nuclease activity using a simple fluorescence based biosensor, *Analytical Methods*, 2016

2. Planned activity(ies) in support of standardization in laboratory medicine

➤ R&D project(s) and/or programme(s)

TUBITAK UME Chemistry Group Laboratories collaborate with public and private institutions to develop measurement procedures in the field of industrial, legal and scientific metrology.

In Bioanalysis Laboratory, the levels of biological molecules such as bacterial, viral or eukaryotic genomic DNA, RNA and peptides/proteins are measured in biological systems using reference measurement systems.

The measurements of small biological molecules such as amino acids, organic acids, hormones, lipids, antibiotics, pesticides and other metabolites are carried out by Organic Chemistry Laboratory which utilizes the measurement power of qNMR as a new reference measurement system in addition to the classical primary reference measurement methods.

Inorganic Chemistry Laboratory develops new measurement methods with the use of a new clean laboratory and new state-of-the-art instruments.

After establishing measurement capabilities, priority will be given to the preparation of the certified reference materials by Reference Materials Laboratory according to the needs of clinical laboratories in Turkey (Table 4). Proficiency testing schemes will be extended to the field of Laboratory Medicine starting with HbA1c.

Table 4. Certified Reference Materials Preparation Planning at TUBITAK UME

Date of Release	Description of CRM or RM
On sale	25-OH Vitamin D2 and D3 in serum (2012-2014)
Will be on sale 2018	Amino acids for neonatal screening (2014-2017)
Will be on sale 2018	Organic acids for neonatal screening (2014-2017)
Will be on sale 2019	HbA1c CRM (2016-2018)
	Elements in serum (2019-2022)
	Elements in whole blood (2019-2022)
	Elements in urine (2019-2022)
	Hemoglobin variants in blood (2019-2021)
	Hormones in serum (2019-2022)

➤ EMRP and EMPiR projects:

The following EMPiR projects will be completed as outlined in the project proposals and new measurement methods and CRMs/RMs will be produced.

- EMPiR-AntiMicroResist - Novel methods and materials for the detection, traceable monitoring and evaluation of antimicrobial resistance, 2016-2019.
- EMPiR-ReMIND - Role of metals and metal containing biomolecules in neurodegenerative diseases such as Alzheimer's disease, 2016-2019.

New EMPiR health call will be advertised in 2018, Chemistry Group Laboratories plan to be involved in these health related projects.

➤ **Korea-Turkey Dual Collaboration Project:**

“Korea-Turkey collaboration project on the development of an international standard system for the measurement of gene methylation” will be finalized in 2017 and “DNA Methylation measurement comparison study” will be proposed with collaboration with our project partner KRISS, Korea under CCQM in 2018. It has been planned to produce and distribute RM in the context of this measurement comparison study (Table 4).

➤ **Measurement Comparison Studies:**

TUBITAK UME Chemistry Group Laboratories have been planning to attend to the following measurement comparison studies (Table 5).

Table 5. CCQM international measurement comparison studies planned between 2017 and 2019.

Comparison #	Comparison Name
CCQM-K139	Elements in Human Serum
CCQM-KXX	Quantification of DNA Methylation (Planning to organize)
CCQM-P184*	Copy Number Concentration and Fractional Abundance of a Mutation (SNV or INDEL) Mixed with Wild-Type DNA
CCQM-KXX	Multiple Cancer Cell Biomarker Measurement
CCQM-XX	Total Hemoglobin in Serum
CCQM-XX	HbA1c Absolute Quantification
CCQM-XX	HbA1c Purity Assessment
CCQM-XX	ProCalcitonin in Serum
CCQM-XX	Oxytosin-Purity Assessment
CCQM-XX	Brain Natriuretic Peptide (BNP)
CCQM-XX	Quantification of CD34+ Cell Counts

* Study in progress, the result will be submitted in 2018.

➤ **Calibration and Measurement Capabilities:**

Participating to the new measurement comparisons in clinical area it is aimed to develop new measurement methods and new reference measurement materials, to broaden our services in the field of Laboratory Medicine (Table 6) leading to new CMC entries in the area.

Table 6. New Calibration and Measurement Capabilities applications planned between 2017 and 2019.

Measurement Service Sub-Category	Matrix	Analyte or Component
Other	cultured or purified mammalian cells or tissues	mRNA copy number concentration
Other	cultured or purified mammalian cells or tissues	reference gene normalized mRNA copy number
Other	cultured or purified mammalian cells or tissues	reference gene normalized mRNA copy number ratio
Peptides	Lyophilized or mQ-H ₂ O 20 mM Phosphate or other buffers with similar level of complexity	pure peptide primary reference material

3. Promoting traceability in laboratory medicine

➤ Congress Posters:

- Akyürek, S., Yang, I., Aşıcıoğlu, M., Park, S.R., Akgoz, M. DNA methylation measurement optimisation for APC gene. 28th Biochemistry Congress, 19-23 Sept 2017, Erzurum, Turkey
- Aşıcıoğlu, M., Akyürek, S., Akgoz, M., Öztürk, N. Circadian rhythm disruption effect on methylation profile of BRCA1 gene. 5th International Congress of the Molecular Biology Association of TURKEY, 8-10 September 2017, Boğaziçi University, Istanbul, Turkey
- Aşıcıoğlu, M., Akyürek, S., Akgoz, M. and Öztürk, M. Investigation of the Effect of Circadian Clock Disruption on DNA Methylation. Gebze Technical University, Graduate Studies Symposium and Introductory Days, Graduate School of Natural and Applied Sciences, 17-18 May 2017, Kocaeli, Turkey
- Yalcinkaya, B., Akyürek, S., Mozioglu E. and Akgöz, M. The aim of Biometrology (Tr: Biyometroloji ve Amacı). 3rd National Laboratory Accreditation and Safety Symposium and Exhibition. 11-13 May 2016, Istanbul, Turkey
- Akyürek, S., Yu, S., Akgoz, M., Park, S.R., Yang, I. Optimization strategy for DNA methylation measurements. The FEBS Journal 283 (Suppl. 1), 2016, p-05.02.2-036
- Yalcinkaya, B., Senal, M., Akgoz, M. Determination of nucleic acid purity by HPLC analysis. The FEBS Journal 283 (Suppl. 1), 2016, p:207, P-01.02.2-007
- Oztug Senal, M., Vatansever, B., Goren, A.C., Un, I., Akgoz, M. Development and validation of PICA method for the determination of the purity of synthetic human C-peptide. The FEBS Journal 283 (Suppl. 1), 2016, p:217, P-02.02.2-033
- Gunduz, S., Yilmaz, H., Bilsel, G., Goren, A.C. Determination of Folic Acid and Folic Acid Impurities by LC-MS Method (Tr: Folik Asit ve Folik Asit Safsızlıklarının LC-MS Yöntemi ile Belirlenmesi). 4th International Drug Chemistry Congress, 17-20 March 2016, Kuşadası, Muğla, Turkey
- Bilsel, G., Topal, K., Hatipoğlu, S., İşleyen, A., Vatansever, B., Goren, A. 25-OH-Vitamin D2/25-OH Vitamin D3 SRM Characterization Studies in Serum Used in Clinical Measurements (Tr: Klinik Ölçümlerde Kullanılan Serumda 25-OH-Vitamin D2/ 25-OH Vitamin D3 SRM'si Karakterizasyon Çalışmaları). 27th National Chemistry Congress, 23-28 August 2015, Çanakkale, Turkey
- Bilsel, M., Goren, A., Bilsel G., Yılmaz, H. Creatine Determination in Serum using LC-IDMS (Tr: LC-IDMS ile Serumda Kreatinin Tayini). 27th National Chemistry Congress, 23-28 August 2015, Çanakkale, Turkey
- Sahin, J., Oztug Senal, M., Akgoz, M. Metrological Traceability in Personalized Medicine. International Symposium on Advances in Predictive & Personalized Medicine, 02-03 April 2015. İstanbul, Turkey
- Hatipoğlu, S. D., Çelik, M., İşleyen, A., Ün, İ., Sadak, A. E., Gündüz, S., Zorlu, N., Gören, A. C. Two New Candidate Certified Reference Materials for Newborn Screening for the Diagnosis of Metabolic Disorders. The 14th International Symposium on Biological and Environmental Reference Materials, 11-15 October 2015, Washington, USA

➤ **Congress Presentations:**

- Gören, A. C. Ün, İ., Gündüz, S. Importance of Reference Materials for Traceable Measurements in Drug Industry, Drug Monitoring and Medical Instruments. 5th International Drug Chemistry Congress, 30 March-02 April 2017, Antalya, Turkey
- Öztuğ Şenal, M., Vatansever, B., Ün, İ., Gören, A.C. Akgoz, M. Impurity Determination of Peptide Based Pharmaceuticals (Tr: Peptit Yapılı İlaçların Safsızlık Analizleri). 5th International Drug Chemistry Congress, 30 March-02 April 2017, Antalya, Turkey

4. Reference laboratory networks /collaborations focusing on developing /implementing reference measurement systems

➤ JCTLM Activities

TUBITAK UME became a member of JCTLM in 2012 and has actively participated to the JCTLM Members' and Stakeholders' Meetings since then. Dr. Akgoz, TUBITAK UME representative, attended JCTLM Members' and Stakeholders' Meeting, on 30 Nov-1Dec 2015, Sevres and presented the Activities of TUBITAK UME Chemistry Group Laboratories in the Field of Laboratory Medicine. Biennial report was also submitted to JCTLM Executive Committee in 2015.

➤ CCQM Activities

As a member of CCQM, TUBITAK UME Chemistry Group scientists attended and actively contributed to OAWG, IAWG, NAWG, PAWG and CAWG meetings (except October 2017 OAWG, NAWG, PAWG and CAWG meetings).

Merve OZTUĞ is also a member of PAWG Focus Group 1 (SI Value Assignment of Purity) and PAWG Focus Group 2 (SI-traceable determination of peptides and proteins in complex matrices).

➤ 2016 and 2017 EurA1c European HbA1c Trail

In 2016 and 2017 several EQA organizers decided to participate in the "EurA1c" projects which are organized by The International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) Committee Education in the Use of Biomarkers in Diabetes (C-EUBD). TUBITAK UME was also involved in these trials as a partner and submitted the measurement results of Turkish clinical laboratories.