European Partnership on Metrology
Call 2022 – Digital Transformation, Health, Integrated European Metrology, Normative and Research Potential

Selected Research Topic number: **SRT-h13**
Version: 1.0

Title: Metrology for Metal Metabolism Disorders

Abstract

Deficiency or excess of essential metals in the body is associated with sever health problems. Metal Metabolism Disorders (MMD) with genetic origin and, for which the functions and levels of physiologically relevant metals in the blood is controlled by the specific proteins which can result in protein malfunction resulting in toxic accumulation of metals in the body. Therefore, the need to underpin current bio-chemical tests for such disease. This proposal aims to develop reference methods for already established biomarkers of MMDs (e.g., Wilson's disease, hemochromatosis), as well as developing integrated methodology and data platforms for emerging biomarkers (e.g., metal isotopic compositions and bio-distribution markers) enabling diagnosis and guiding therapeutics through engagement with clinicians, PT providers (Proficiency Testing) and pharmaceutical industry to support quality control/quality assurance to improve disease diagnosis and therapy.

Keywords

Wilson's disease, exchangeable copper, Ferritin, hemochromatosis, Fe imbalance, Cu imbalance, traceable methods, clinical biomarkers, disease diagnosis and treatment, quality assurance.

Background to the Metrological Challenges

The global metabolic disorders drug market is expected to reach $198 billion in 2025 at a compound annual growth rate of 8 %. There is presently no known cure for such MMD disease and therefore, reliable treatment monitoring methods is needed to support patient wellbeing and the development of more efficacious personalised therapies. Currently, the diagnosis involves gene mutation testing, clinical observation, and bio-chemical testing. In Wilson's disease, exchangeable Cu is measured by nephelometry as the amount of total Cu minus that of CER-bound Cu. The main limitation of this test lies in the inaccuracy of measuring CER (ceruloplasmin) by immunological methods not able to distinguish between the apo-CER and the active holo-CER, thus leading to biased results.

Moreover, there is no SI traceable methods for exchangeable Cu (e.g., *via* traceable determination of Cu-CER). There is no available validated methodology to use to determine exchangeable Cu in a clinical trial for a large number of samples to measure stability and no traceable methods is available to achieve relative limits of quantification at the low µg kg^{-1} level for bio-species in micro-sections of complex biological materials.

Furthermore, due to the low concentrations, small sample volumes, labile nature of the measurands and difficult matrices, the use of a wide range of expertise and instrumentation is required, which is rarely available in a single institution. Also, the lack of SI traceable methodology for ferritin in human serum, for which the complexity of the matric combined with the low ferritin levels (µg kg^{-1}) represent challenges to both accurate quantification and species structural confirmation.

Previously developed WHO Standard for Ferritin using immunoassays methods indicated results is hardly comparable and is not well understood and no traceability chain has been established for the available ferritin standards. From the foregoing account, there is an urgent need for the reference methods for the quantification and identification of biochemical markers for the early diagnosis, treatment and monitoring of MMD.

The International Organisation for Standardisation (ISO) documents including ISO 17511 and ISO 18153 has recognised and accepted reference materials and measurements is considered the key element in assuring the accuracy and comparability of clinical laboratory measurements. Also, the development of methods for the traceable determination of both established and emerging biomarkers of MMD can improve the quality of measurement science of European NMIs and support quality assurance of the measurements performed in clinical and less experienced laboratories. To conclude, more efficacious MMD therapies can benefit economic

in terms of pharmaceutical industry drug pipelines reducing treatment costs and have a positive impact on the quality of life of those affected by MMD.

Objectives

Proposers should address the objectives stated below, which are based on the PRT submissions. Proposers may identify amendments to the objectives or choose to address a subset of them in order to maximise the overall impact, or address budgetary or scientific / technical constraints, but the reasons for this should be clearly stated in the protocol.

The JRP shall focus on the traceable measurement and characterisation of biochemical markers used for the early on diagnosis of metal metabolism disorders (MMD).

The specific objectives are

1. To develop and validate the protocols for selection, preparation, fractionation, mounting and pre-concentration of samples (e.g. tissues, cells, plasma/serum) with preservation of species integrity. This includes the strategies for preparation and characterisation of calibrants, labelled materials, model samples and reference materials as well as stability studies for labile species.
2. To develop and validate the SI-traceable methods for the quantification and identification of established bio-chemical markers of MMD at $\mu g\ kg^{-1}$ levels in biological samples (e.g. Cu-Ceruloplasmin in plasma/serum for Wilson's disease; Ferritin light chain in serum for Fe-overload states like hemochromatosis). In addition, the comparison of data obtained with that of routinely used bio-chemical tests should be undertaken.
3. To develop methodology and data platforms for emerging bio-chemical markers (e.g. metal isotopic compositions, metal:sulphur ratios and bio-distribution markers) in biological samples (e.g. fluids, tissues and cells) enabling diagnosis and guiding therapeutics. Quantitative strategies for elemental spatial distribution in tissues and cells (to assess drug penetration, effect and bio-distribution) should be linked to histopathology data.
4. To assess of the methodology performance and definition of requirements for quality control materials/reference materials by organisation of an inter-laboratory comparison. In addition, to develop a best practice guide on the traceable measurement and characterisation of key biomarkers of MMD in complex biological matrices.
5. To facilitate the take up of the technology and measurement infrastructure developed in the project by the measurement supply chain (in coordination with the European Metrology Network "Traceability in Laboratory Medicine" (EMN TraceLabMed) and through engagement with metrology institutes, accredited laboratories), standard development organisations (CEN/TC 140, ISO/TC 212, VAMAS TWA 40) and end users (e.g. clinicians, pharmaceutical industry, PT providers, healthcare products)

These objectives will require large-scale approaches that are beyond the capabilities of single National Metrology Institutes and Designated Institutes, and it is expected that multidisciplinary teams will be required. To enhance the impact of the research, the involvement of the appropriate user community such as medical practitioners, medical (academic) hospitals and industry is strongly recommended, both prior to and during methodology development.

Proposers should establish the current state of the art and explain how their proposed project goes beyond this. In particular, proposers should outline the achievements of the EMPIR project 15HLT02 ReMiND and how their proposal will build on those.

EURAMET expects the average EU Contribution for the selected JRPs in this TP to be 1.9 M€ and has defined an upper limit of 2.6 M€ for this project.

EURAMET also expects the EU Contribution to the external funded beneficiaries to not exceed 35 % of the total EU Contribution across all selected projects in this TP.

Any industrial beneficiaries that will receive significant benefit from the results of the proposed project are expected to be beneficiaries without receiving funding or associated partners.

Potential Impact

Proposals must demonstrate adequate and appropriate participation/links to the 'end user' community, describing how the project partners will engage with relevant communities during the project to facilitate

knowledge transfer and accelerate the uptake of project outputs. Evidence of support from the 'end user' community (e.g. letters of support) is also encouraged.

You should detail how your JRP results are going to:

- Address the SRT objectives and deliver solutions to the documented needs,
- Feed into the development of urgent documentary standards through appropriate standards bodies,
- Transfer knowledge to the Healthcare sector.

You should detail other impacts of your proposed JRP as specified in the document "Guide 4: Writing Joint Research Projects (JRPs)"

You should also detail how your approach to realising the objectives will further the aim of the Partnership to develop a coherent approach at the European level in the field of metrology and include the best available contributions from across the metrology community. Specifically, the opportunities for:

- improvement of the efficiency of use of available resources to better meet metrological needs and to assure the traceability of national standards
- the metrology capacity of EURAMET Member States whose metrology programmes are at an early stage of development to be increased
- organisations other than NMIs and DIs to be involved in the work.

Time-scale

The project should be of up to 3 years duration.