Mini Review

Aasne K. Aarsand*, Thomas Røraas, William A. Bartlett, Abdurrahman Coşkun, Anna Carobene, Pilar Fernandez-Calle, Niels Jonker, Jorge Díaz-Garzón, Federica Braga and Sverre Sandberg, on behalf of the European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) Working Group on Biological Variation

Harmonization initiatives in the generation, reporting and application of biological variation data

https://doi.org/10.1515/cclm-2018-0058
Received January 15, 2018; accepted February 12, 2018

Abstract: Biological variation (BV) data have many applications in laboratory medicine. However, concern has been raised that some BV estimates in use today may be irrelevant or of unacceptable quality. A number of initiatives have been launched by the European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) and other parties to deliver a more harmonized practice in the generation, reporting and application of BV data. Resulting from a necessary focus upon the veracity of historical BV studies, critical appraisal and meta-analysis of published BV studies is possible through application of the Biological Variation Data Critical Appraisal Checklist (BIVAC), published in 2017. The BIVAC compliant large-scale European Biological Variation Study delivers updated high-quality BV data for a wide range of measurands. Other significant developments include the publication of a Medical Subject Heading term for BV and recommendations for common terminology for reporting of BV data. In the near future, global BV estimates derived from meta-analysis of BIVAC appraised publications will be accessible in a Biological Variation Database at the EFLM website. The availability of these high-quality data, which have many applications that impact on the quality and interpretation of clinical laboratory results, will afford improved patient care.

Keywords: analytical performance specification; biological variation; evidence-based medicine; meta-analysis; reference change values.

Introduction

Many different sources of variation may impact upon laboratory test results as an individual is monitored over time. Natural biological variation (BV) describes the variation in constituents regulated by homeostatic processes in the body [1]. Many measurands of clinical interest are characterized by random variation around a stable homeostatic set-point (e.g. serum sodium concentration), whereas

***Corresponding author: Aasne K. Aarsand,** Norwegian Porphyria Centre, Laboratory of Clinical Biochemistry, Haukeland University Hospital, NO-5021 Bergen, Norway, Phone: +47 55 97 31 27/ +47 55 97 31 70, Fax: +47 55 97 31 15,
E-mail: aasne.aarsand@helse-bergen.no; and Norwegian Quality Improvement of Laboratory Examinations (Noklus), Haraldsplass Deaconess Hospital, Bergen, Norway
Thomas Røraas: Norwegian Quality Improvement of Laboratory Examinations (Noklus), Haraldsplass Deaconess Hospital, Bergen, Norway
William A. Bartlett: Blood Sciences, Ninewells Hospital and Medical School, Scotland, UK
Abdurrahman Coşkun: Acibadem University, School of Medicine, Atasehir, Istanbul, Turkey
Anna Carobene: Servizio di Medicina di Laboratorio, Ospedale San Raffaele, Milan, Italy
Pilar Fernandez-Calle and Jorge Díaz-Garzón: Hospital Universitario La Paz, Madrid, Spain; and Spanish Society of Laboratory Medicine (SEQC^{ML}), Barcelona, Spain
Niels Jonker: Certe, Wilhelmina Ziekenhuis Assen, Assen, The Netherlands
Federica Braga: Research Centre for Metrological Traceability in Laboratory Medicine (CIRME), University of Milan, Milan, Italy
Sverre Sandberg: Norwegian Porphyria Centre, Laboratory of Clinical Biochemistry, Haukeland University Hospital, Bergen, Norway; Norwegian Quality Improvement of Laboratory Examinations (Noklus), Haraldsplass Deaconess Hospital, Bergen, Norway; and Department of Global Health and Primary Care, Faculty of Medicine and Dentistry, University of Bergen, Bergen, Norway

others display change during the lifetime of an individual or predictable cyclical variation (e.g. pituitary gonadotropins in females). An understanding of this variation is required to enable appropriate application of clinical laboratory measurements. The magnitude of within-subject BV (CV_I) describes the variation observed within an individual and the between-subject BV (CV_G) the variation between individuals. BV data can be quantified to these ends; the resultant data enabling (1) setting analytical performance specifications (APS), (2) determination of reference change values (RCV) to assess the significance of change in serial measurements within a subject and (3) the derivation of index of individuality to assess the applicability of population based reference intervals [2]. The utility and validity of APS and other applications for diagnosis and monitoring based on BV data deliver a requirement that the applied BV data must be of high quality and relevant to the population served by the laboratory. This raises an issue in that BV data are reference data, but they are often applied without this understanding. This leads to indiscriminate application of the data to populations across health care systems that may have characteristics that are different to the reference population. Thus, this delivers an imperative for those who use published BV data to apply the sort of scrutiny that they might apply when adopting population based reference intervals. Furthermore, BV estimates for the same measurand obtained from independent studies may deliver estimates that vary substantially. The reason for this variation may be multi-factorial with lack of harmonization in study design, applied methodology and data handling for BV studies representing a major component source. There are a considerable number of published BV studies, with varying quality, stretching back over 40 years. Access to a compilation of these data has been enabled via an online database which presents BV data from studies identified through literature searches. The BV data are presented for a range of measurands alongside APS for bias, imprecision and total error derived from those data [3]. This is the work of the Analytical Quality Commission of the Spanish Society of Laboratory Medicine ($SEQC^{ML}$) [4], first presented at the Stockholm Conference in 1999 [5]. The database was updated every 2 years until 2014 [6] and has proven a useful source of BV data for the laboratory community. However, a number of questions have been raised regarding the accuracy of the estimates presented in this database [7, 8]. Prior to inclusion of the data into the pool from which the online 2014 BV database is compiled, publications on BV have been reviewed with regard to a set of criteria, such as the applied statistical model and ratio between analytical variation (CV_A) and CV_I estimates [6]. However, a rigorous appraisal with regard to study design or statistical analysis has not been performed. Furthermore, the validity of the methods used for generation of the global estimates of BV has been questioned and the estimates provided in the database have been unaccompanied by measures of uncertainty. That estimates from historical studies employing obsolete analytical methods may be included delivers a further degree of complexity to be considered by users. It is thus a concern that the headline BV estimates from this compiled database may not be fit for purpose and not the product of a harmonized approach to delivery. The data are applied by the laboratories and clinicians in their everyday practice and this may both lead to adoption of inappropriate APS and also consequences for patients' diagnosis and monitoring.

Several initiatives have been established to provide a more harmonized practice and consequent delivery of fit for purpose and appropriately characterized BV estimates; they include those driven by the European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) Working Group on Biological Variation (WG-BV) [9] and the Task and Finish Group for the Biological Variation Database (TFG-BVD) [10]. This article aims to identify challenges in the generation and application of BV data and describes EFLM initiatives and other important harmonization contributions that aim to provide solutions to the many challenges identified to assure the delivery of BV estimates that are fit for purpose to enable safe and effective clinical applications.

Generation and estimation of components of BV

Study design and reliability

Numerous studies on BV components have been published during the last four decades. There are variations between these publications in study design, methodology and applied statistical approach that may affect the BV estimates they present. To generate high-quality BV data, adherence to strict experimental and statistical protocols is required. Furthermore, in order for estimates to be applicable in clinical practice today, the analytical methods used to derive the BV data must be comparable with contemporary methods in terms of analytical specificity, as obsolete methods using different principles may in fact deliver different BV estimates (e.g. switching from competitive immunoassay to immunometric assays for parathyroid hormone). Typically, when generating BV

data, a group of reference individuals, in most cases cited as apparently healthy individuals, are studied. From each of these individuals, specimens are drawn at regular and appropriate time intervals while minimizing all sources of pre-analytical variation. All specimens must be drawn, processed, transported and stored under controlled and standardized conditions [2]. Ideally, analysis should be performed in duplicate, with all samples from the same individual being processed in the same analytical run if possible, while minimizing analytical sources of variation. Thereafter, appropriate statistical analysis including assessment of outliers and homogeneity of variances must be applied, followed by dissection of the CV_A, CV_I, and CV_G components by a nested analysis of variance (ANOVA), general/generalized linear model or a similar approach [2].

To produce robust, well-characterized BV data and to ensure that data are transferable across health care systems and different populations, BV studies must conform to an ideal standardized approach, be adequately powered and properly documented at the reporting stage. This is, unfortunately, not always the case. The number of replicates, number of samples and number of individuals included in the study influence the reliability and confidence intervals (CI) of the BV estimates, and the effect of these variables varies with the ratio of analytical standard deviation (SD_A) to within-subject SD (SD_I) [11]. Generally, the lower the SD_A/SD_I ratio, the narrower the CIs. If subgroups are to be explored, a balanced number of subjects in each planned sub-group is preferable. When published, BV estimates must be accompanied by their corresponding CIs to allow for the assessment of their reliability, comparison between subgroups and the comparability with other studies.

The European Biological Variation Study (EuBIVAS)

Responding to the need for reliable BV estimates delivered from big high-quality studies, the EFLM WG-BV has designed and implemented the European Biological Variation Study (EuBIVAS) [12]. In the EuBIVAS, six European clinical laboratories in five different countries followed a strict, detailed protocol for the recruitment of subjects and for the pre-analytical phase [13]. The study population included 91 healthy volunteers (38 males and 53 females, age 21–69 years). Fasting blood samples were drawn for 10 consecutive weeks (April–June 2015) for all individuals. All samples were processed and prepared in the same way as detailed [13] and stored at −80 °C prior to shipment on dry-ice to the coordinating center in Milan. All samples from each of the participants were analyzed in duplicate on platforms within the Milan laboratory under standardized conditions. The resultant data sets underwent rigorous scrutiny and appropriate statistical analysis to enable delivery of BV estimates accompanied by CIs. So far, estimates for liver enzymes [14] and creatinine using enzymatic and alkaline picrate methods [15] have been published. For these measurands, EuBIVAS estimates are lower than the corresponding estimates available in the online 2014 BV database (Table 1) [3]. Though the reasons for this may be multi-factorial, our data so far indicate that a large and strictly performed study such as the EuBIVAS delivers lower estimates than many other BV studies; these differences have implications for the setting of APS, diagnosis and monitoring and other applications of BV data. In addition, the EuBIVAS study has enabled comparison of measurements using different analytical approaches; in the case of serum creatinine the EuBIVAS results indicated that current alkaline picrate methods may not meet BV-based APS for imprecision [15]. The next phase of the EuBIVAS, providing BV estimates for electrolytes, lipids, hormones and specific proteins, is under way [12]. Thus, the EuBIVAS provides a valuable resource and infra-structure to enable delivery of high-quality and well characterized, and therefore transportable, BV data for a large number of measurands, using a protocol that is fully compliant with the newly developed Biological Variation Data Critical Appraisal Checklist (BIVAC) [16].

Critical appraisal of BV publications and the delivery of global estimates

Following the 1st Strategic Conference of the EFLM defining APS in November 2014 [17], the EFLM TFG-BVD was established [10], with the objective to appraise the quality of BV data that is publicly available. Its terms of reference were to develop a critical appraisal list for the evaluation of BV studies, to use this to assess the existing literature on BV and to extract essential information from those papers and to summarize the results. The remit has expanded to the development of a new database containing measures of BV with indications of their quality and associated APS. The TFG-BVD was made up by members from the EFLM WG-BV [9], the Analytical Quality Commission of the SEQC^{ML} [4], and other key persons with experience in theoretical and practical aspects of generating BV data. The results of the work of this TFG are three-fold. Firstly, the TFG-BVD in collaboration with the WG-BV has developed

Table 1: Biological variation within-subject (CV_I) and between-subject (CV_G) estimates from the European Biological Variation Study (EuBIVAS) with 95% confidence intervals (CI) for serum creatinine concentrations and enzyme activities and corresponding estimates available in the online 2014 BV database.

Measurand	Study population	EuBIVAS[a]		Online 2014 BV database	
		CV_I % (95% CI)	CV_G % (95% CI)	CV_I, %	CV_G, %
Creatinine (enzymatic method)	All subjects	4.4 (4.2–4.7)	14.8 (12.8–17.8)	6.0	14.7
	Males	4.2 (4.0–4.7)	12.8 (10.1–17.0)		
	Females	4.6 (4.3–4.9)	10.2 (8.4–13.0)		
Alanine amino transferase (ALT)	All subjects	9.3 (8.7–10.0)	28.0 (24.7–33.9)	19.4	41.6
	Males	10.1 (9.2–11.2)	28.2 (22.6–37.4)		
	Females	9.6 (8.8–10.5)	25.2 (21.3–32.1)		
Aspartate aminotransferase (AST)	All subjects	9.5 (9.0–10.2)	20.3 (17.7–24.2)	12.3	23.1
	Males	10.3 (9.5–11.3)	14.7 (12.4–20.3)		
	Females	8.9 (8.3–9.8)	22.4 (18.7–28.3)		
Gamma-glutamyl transferase (GGT)	All subjects	8.9 (8.1–9.7)	45.1 (38.9–54.2)	13.4	42.2
	Males	8.3 (7.1–9.5)	41.7 (33.5–57.4)		
	Females	8.3 (7.4–9.5)	34.2 (28.1–43.4)		
Alkaline phosphatase (ALP)	All subjects	5.3 (5.0–5.7)	24.9 (21.4–29.3)	6.5	26.1
	Males	5.0 (4.6–5.5)	25.4 (20.3–34.1)		
	Females	5.4 (5.1–5.9)	24.2 (20.6–30.9)		
Lactate dehydrogenase (LDH)	All subjects	5.2 (5.0–5.5)	12.6 (10.8–14.7)	8.6	14.7
	Males	5.5 (5.1–6.0)	10.2 (8.1–13.3)		
	Females	5.2 (4.9–5.6)	14.0 (11.6–17.3)		
Creatine kinase (CK)	All subjects	14.5 (13.8–15.4)	37.9 (32.8–45.8)	22.8	40
	Males	16.0 (14.8–17.5)	31.5 (25.2–42.5)		
	Females	15.7 (14.6–16.8)	30.5 (24.8–38.4)		
Amylase	All subjects	6.8 (6.5–7.2)	30.4 (26.5–36.3)	8.7	28.3
	Males	6.3 (5.9–6.9)	30.0 (23.7–39.1)		
	Females	7.1 (6.7–7.6)	31.4 (26.6–40.0)		
Pancreatic amylase	All subjects	6.3 (6.0–6.7)	24.9 (21.9–30.1)	11.7	29.9
	Males	5.9 (5.5–6.5)	23.3 (18.8–30.8)		
	Females	6.8 (6.3–7.3)	26.0 (22.3–34.2)		
Lipase	All subjects	7.7 (7.2–8.3)	23.8 (20.6–28.2)	32.2	31.8
	Males	6.4 (5.5–7.2)	22.2 (18.1–29.2)		
	Females	8.9 (8.3–9.8)	23.9 (19.9–30.2)		

[a]EuBIVAS estimates adapted from Carobene et al. [14, 15].

the recently published BIVAC [16]. Secondly, as part of this work, a meta-analysis approach for pooling BIVAC compliant estimates to provide global BV estimate has been developed. And thirdly, using the results of this work, the TFG, now transformed to a more permanent Task Group, is in the process of establishing a new Biological Variation Database, to be available at the EFLM website.

The Biological Variation Data Critical Appraisal Checklist (BIVAC)

The BIVAC is designed to assess the quality of BV publications by verifying whether all essential elements that may impact upon veracity and utility of the associated BV estimates are present [16]. Its main focuses are the effect of study design, the measurement procedure and statistical handling of data on CV_I estimates. The BIVAC consists of 14 quality items (QI), which can be awarded scores A, B, C or D (Table 2) [16]. Based on the individual scores for each of the QI, an overall grade is set for the publication under review. The grade A is achieved if the publication shows full compliance with all BIVAC QI. If the lowest score for any QI is a B, then the overall grade is a B and similarly C or D if the lowest QI score is a C or D, respectively. In the BIVAC scoring system, the QI related to the overall grade are shown as a subscript, as exemplified by $C_{4,8,13}$ and $B_{6,7,8,9,11,13}$. BV estimates derived from studies that receive one or more D scores are considered unsuitable for use in clinical practice. The BIVAC QI relating to

Table 2: The quality items of the Biological Variation Data Critical Appraisal Checklist with achievable scores.[a]

Quality item number	Quality item	Achievable scores			
1	Scale	A	B	–	–
2	Subjects	A	B	C	D
3	Samples	A	B	C	D
4	Measurand	A	B	C	D
5	Pre-analytical procedures	A	B	C	–
6	Estimates of analytical variation	A	B	C	–
7	Steady state	A	B	C	–
8	Outliers	A	B	C	–
9	Normally distributed data	A	B	–	–
10	Variance homogeneity	A	–	C	–
11	Statistical method	A	B	C	–
12	Confidence limits	A	–	C	–
13	Number of included results	A	B	C	–
14	Concentrations	A	B	–	–

[a]Adapted from Aarsand et al. [16].

subjects (QI 2), samples (QI 3) and the measurement procedure (QI 4) are considered critical for the reliability and applicability of the associated measures of BV and thus can be scored as D (Table 2). It is an absolute requirement that populations from which BV data have been derived are adequately characterized. Furthermore, details on the study population, samples, sample material and timing of samples are necessary to compare with other studies, to deliver CIs and to generate global BV estimates. Additionally, it is essential that historical publications assess the same measurand as contemporary methods for the reported BV estimates to be relevant. QI 5–7 refer to pre-analytical procedures, estimates of CV_A and demonstration of steady state, all of which are required to obtain high-quality BV estimates. Statistical elements QI 8–12 relate to analysis of outliers, normality, homogeneity of variances, the applied statistical model and CI which are central for the quality of the associated BV estimates and their applicability. QI 1 (scale), 13 (number of included results) and 14 (concentrations) do not reflect the reliability of the BV estimates in themselves, but are necessary elements for interpretation and application of the data. Applying the BIVAC to 128 BV publications for 28 different measurands revealed that the QI for outlier analysis and variance homogeneity testing were most often scored as C, meaning that appropriate analysis of these elements were missing in more than 60% of publications [16]. Although many of the historical papers on BV were performed according to the standards existing at the time of publication, it is, based on our review, evident that many BV publications omit, or fail to address, essential elements, in particular for statistical analysis, which may affect the reliability and applicability of the reported BV estimates.

Meta-analysis of BV studies

For commonly requested measurands, many studies on BV have been published, as exemplified for total cholesterol, where 46 publications are included in the online 2014 BV database [3]. The common estimate that is presented in this database represents the median of estimates from studies fulfilling its inclusion criteria [6]. We have recently developed a meta-analysis approach for the delivery of global estimates [16]. With this approach, BV publications for the measurand in question are identified by systematic searches and relevant publications are thereafter appraised by the BIVAC. Only studies receiving an overall BIVAC grade A, B or C are considered fit to be considered for the meta-analysis. Thereafter, the study design is reviewed so that only studies with similar characteristics are included in the meta-analysis; as a first step studies performed in healthy adults where sampling is weekly. For the meta-analysis, the associated BIVAC grade and the width of the CI are used as weight, with the global estimate being delivered by a weighted median approach [16]. If a sufficient number of higher quality studies, i.e. BIVAC grades A and B, are available, the global estimate may be based on these alone.

The EFLM Biological Variation Database

As part of our initial BIVAC review of BV publications for enzymes, lipids, kidney and diabetes related measurands [16], work is in progress in providing updated global estimates for all these 28 measurands. These, and other measurands that will be critically appraised in the near future, will be published in a new Biological Variation Database, available on the EFLM website. Here global BV estimates with the evidence behind them and related APS will be accessible, with separate estimates being delivered for different populations and sampling intervals when relevant. This will be an important initiative in providing updated evidence-based global estimates easily accessible to users. Once this database covers the majority of relevant measurands and gains momentum in reaching potential users, it may contribute to a harmonized approach so that where appropriate, the same global BV estimates and APS will be applied in laboratories worldwide.

Reporting of BV data and BV terminology

The Biological Variation Data Reporting Checklist

There have historically been no internationally recognized standards for production, reporting and transmission of BV data. The WG-BV published in 2015 the Biological Variation Data Reporting Checklist [18], which identifies key elements required in studies to enable accurate and effective use of BV data across population groups and healthcare systems. It is based on the same structure as the STARD [19] and identifies six main minimum data set domains including (1) title/abstract/keywords, (2) introduction, (3) methods, (4) data analysis, (5) results and (6) discussion. Compliance with this checklist indicates that studies are fit for purpose, that essential statistical analyses such as outlier and variance homogeneity testing have been reported, that common terminology has been used and that BV estimates are accompanied by key metadata. Additionally, the BIVAC, although designed for critical appraisal of already published studies (16), provides a detailed guide for those designing and delivering future studies and an aide for reviewers of manuscripts reporting on those to assure publication of the required high-quality BV publications.

Terminology for components of BV

There is a wide range of terms and symbols used for the definition of components of BV in the published literature. A recent study reviewed the 13 highest ranked clinical chemistry journals and identified 68 terms and 25 symbols for components related to individuals and 47 terms and 18 symbols for components related to groups of individuals [20]. There is a clear need for a harmonized use of terminology, and the EFLM WG-BV supports the terms and symbols for components of BV suggested by Simundic et al. [20], as outlined in Table 3. This terminology has also been supported by the Clinical Chemistry and Laboratory Medicine [21]. Additionally, up until now, there has not been a Medical Subject Heading (MeSH) heading for BV, which both adds to the varied use of BV terms as well as complicating literature-searching strategies [16]. The WG-BV suggested in 2016 the establishment of a new MeSH term for "Biological Variation" to the National Library of Medicine. This was accepted and the MeSH term has been available from December 2017, thus facilitating systematic searches for BV publications. However, the National Library of Medicine usually does not re-index previously indexed articles when new headings are added.

Table 3: Terminology and abbreviations for biological variation related components and applications.

Abbreviation/symbol	Term
CV_I	Within-subject biological variation
CV_G	Between-subject biological variation
CV_A	Analytical variation
RCV	Reference change value
II	Index of individuality

Applications of BV data

There are many applications for BV data in the laboratory and in clinical practice, most central are the setting of APS and RCV. It has recently been suggested that laboratory tests can be allocated to three different models for the determination of APS; (1) the effect on the clinical outcome, (2) BV data or, (3) the state-of-the-art of measurement [22]. It has been suggested that most measurands should have APS assigned using the BV model [22, 23], further emphasizing the central role of BV data in laboratory medicine. Additionally, for many measurands where the outcome model is recommended, studies are not currently available to support this approach. In such cases application of the BV model may represent a pragmatic interim approach to setting of APS if appropriate data are available. For RCV, they are at any given level of probability a function of the CV_A and CV_I. For local use, local RCV must be calculated based on appropriate CV_A from the laboratory's own quality control for the relevant time interval and the relevant z-value chosen, depending on the clinical question in mind and the wanted level of probability [2]. A log-normal approach as described below is preferable for calculating RCV when using estimates of CV_A and CV_I in % [24], resulting in asymmetric limits, i.e. differently sized RCV for a decrease and an increase in the measurand concentration:

$$SD^2_{A,\log} = \log_e(CV^2_A + 1)$$

$$SD^2_{I,\log} = \log_e(CV^2_I + 1)$$

$$SD_{\text{combined},\log} = \sqrt{SD^2_{A,\log} + SD^2_{I,\log}}$$

$$RCV\% = 100\% \times e^{((\pm Z_\alpha \times \sqrt{2} \times SD_{\text{combined},\log}) - 1)}.$$

This approach is also superior to classical RCV calculation in that it is not possible to achieve paradoxical decreases greater than 100% [24]. For the best performance, this approach assumes log-normal distributed data or estimates of CV_I <12% [25, 26]. A rearrangement of the RCV equation can make z the unknown so that, for any change in serial results detected, using the estimates of CV_I and CV_A, the probability that the seen change is significant can be calculated [2]. Additionally, a model has been developed to set limits for significant bidirectional changes when more than two serial results are available [27].

Conclusions

Over time, concern has been raised regarding the quality of available BV estimates and the effect of their application in clinical practice. With this in mind, a number of initiatives have been launched aiming to deliver robust and reliable BV estimates. Harmonized approaches to the future delivery and application of these important data will drive up their quality and assure safe use. An understanding of the importance of study design and power to deliver robust data with narrow CI has been published. The need to derive new BV estimates for existing and new analytical systems has led to the implementation of the EuBIVAS. This is a large-scale, highly powered and BIVAC compliant BV study providing updated and well-characterized BV estimates for many clinically important measurands. EFLM groups have developed both the BIVAC for critical appraisal of existing BV studies and an approach to meta-analysis for BV studies that enables more robust point estimates of BV from multiple quality assessed studies. The establishment of a MeSH term, common terminology and a standard for reporting of BV studies will provide accessibility and understanding of and transportability of the published BV data. The results of these initiatives will enable delivery of a new EFLM Biological Variation Database, where global BV estimates resulting from meta-analysis of critically appraised publications will be made publicly available. The use of this evidence-based database will provide laboratories the opportunity to implement global, quality assured BV estimates and APS and thus contribute to a harmonized use of these data and potentially better quality of patient care.

Acknowledgments: We thank the members of the EFLM TFG-BV for their continued efforts in contributing to the delivery of high-quality estimates of BV; Carmen Ricos, Joana Minchinela, Margarita Simón, Virtudes Álvarez, Pilar Fernández-Fernández, Beatriz Boned, Zoraida Corte and Fernando Marqués García from the Analytical Quality Commission of the SEQC^{ML}; Berna Aslan, the Institute for Quality Management in Healthcare, Toronto and Craig Webster, Birmingham Heartlands Hospital, Birmingham.
Author contributions: All the authors have accepted responsibility for the entire content of this submitted manuscript and approved submission.
Conflict of interest: None declared
Research funding: None declared.
Employment or leadership: None declared.
Honorarium: None declared.
Competing interests: The funding organization(s) played no role in the study design; in the collection, analysis, and interpretation of data; in the writing of the report; or in the decision to submit the report for publication.

References

1. Fraser CG. Biological variation: from principles to practice. Washington: AACC Press, 2001.
2. Fraser CG, Sandberg S. Biological variation. In: Rifai N, Horvath AR, Wittwer CT, editors. Tietz textbook of clinical chemistry and molecular biology, 6th ed. St. Louis: Elsevier, 2017:157–70.
3. Minchinela J, Ricós C, Perich C, Fernández-Calle P, Álvarez V, Doménech MV, et al. The online 2014 biological variation database. http://www.westgard.com/biodatabase1.htm#1. Accessed: 12 Jan 2018.
4. Analytical Quality Commission of the Spanish Society of Laboratory Medicine. http://www.seqc.es/es/comisiones/comision-de-calidad-analitica/_id:4/. Accessed: 12 Jan 2018.
5. Ricos C, Alvarez V, Cava F, Garcia-Lario JV, Hernandez A, Jimenez CV, et al. Current databases on biological variation: Pros, cons and progress. Scand J Clin Lab Invest 1999;59:491–500.
6. Perich C, Minchinela J, Ricos C, Fernandez-Calle P, Alvarez V, Domenech MV, et al. Biological variation database: structure and criteria used for generation and update. Clin Chem Lab Med 2015;53:299–305.
7. Aarsand AK, Roraas T, Sandberg S. Biological variation – reliable data is essential. [Editorial]. Clin Chem Lab Med 2015;53:153–4.
8. Carobene A. Reliability of biological variation data available in an online database: need for improvement. Clin Chem Lab Med 2015;53:871–7.
9. European Federation of Clinical Chemistry and Laboratory Medicine Working Group on Biological Variation. www.eflm.eu/site/page/a/1148. Accessed: 12 Jan 2018.
10. European Federation of Clinical Chemistry and Laboratory Medicine Task and Finish Group Biological Variation Database. https://www.eflm.eu/site/page/a/1084. Accessed: 12 Jan 2018.
11. Roraas T, Petersen PH, Sandberg S. Confidence intervals and power calculations for within-person biological variation: effect of analytical imprecision, number of replicates, number of samples, and number of individuals. Clin Chem 2012;58:1306–13.
12. Carobene A. The European Biological Variation Study (EuBIVAS): delivery of updated biological variation estimates, a project by the Working Group on Biological Variation in the European

Federation of Clinical Chemistry and Laboratory Medicine. J Lab Precis Med 2017;2:70.
13. Carobene A, Strollo M, Jonker N, Barla G, Bartlett WA, Sandberg S, et al. Sample collections from healthy volunteers for biological variation estimates' update: a new project undertaken by the Working Group on Biological Variation established by the European Federation of Clinical Chemistry and Laboratory Medicine. Clin Chem Lab Med 2016;54:1599–608.
14. Carobene A, Roraas T, Solvik UO, Sylte MS, Sandberg S, Guerra E, et al. Biological variation estimates obtained from 91 healthy study participants for 9 enzymes in serum. Clin Chem 2017;63:1141–50.
15. Carobene A, Marino I, Coskun A, Serteser M, Unsal I, Guerra E, et al. The EuBIVAS project: within- and between-subject biological variation data for serum creatinine using enzymatic and alkaline picrate methods and implications for monitoring. Clin Chem 2017;63:1527–36.
16. Aarsand AK, Roraas T, Fernandez-Calle P, Ricos C, Diaz-Garzon J, Jonker N, et al. The Biological Variation Data Critical Appraisal Checklist: a standard for evaluating studies on biological variation. Clin Chem 2018;64:501–14.
17. Panteghini M, Sandberg S. Defining analytical performance specifications 15 years after the Stockholm conference [editorial]. Clin Chem Lab Med 2015;53:829–32.
18. Bartlett WA, Braga F, Carobene A, Coskun A, Prusa R, Fernandez-Calle P, et al. A checklist for critical appraisal of studies of biological variation. Clin Chem Lab Med 2015;53:879–85.
19. Bossuyt PM, Reitsma JB, Bruns DE, Gatsonis CA, Glasziou PP, Irwig L, et al. STARD 2015: an updated list of essential items for reporting diagnostic accuracy studies. Clin Chem 2015;61:1446–52.
20. Simundic AM, Kackov S, Miler M, Fraser CG, Petersen PH. Terms and symbols used in studies on biological variation: the need for harmonization. Clin Chem 2015;61:438–9.
21. Plebani M, Padoan A, Lippi G. Biological variation: back to basics [editorial]. Clin Chem Lab Med 2015;53:155–6.
22. Sandberg S, Fraser CG, Horvath AR, Jansen R, Jones G, Oosterhuis W, et al. Defining analytical performance specifications: consensus statement from the 1st strategic conference of the European Federation of Clinical Chemistry and Laboratory Medicine. Clin Chem Lab Med 2015;53:833–5.
23. Ceriotti F, Fernandez-Calle P, Klee GG, Nordin G, Sandberg S, Streichert T, et al. Criteria for assigning laboratory measurands to models for analytical performance specifications defined in the 1st EFLM strategic conference. Clin Chem Lab Med 2017;55:189–94.
24. Fokkema MR, Herrmann Z, Muskiet FA, Moecks J. Reference change values for brain natriuretic peptides revisited. Clin Chem 2006;52:1602–3.
25. Braga F, Ferraro S, Ieva F, Paganoni A, Panteghini M. A new robust statistical model for interpretation of differences in serial test results from an individual. Clin Chem Lab Med 2015;53:815–22.
26. Roraas T, Stove B, Petersen PH, Sandberg S. Biological variation: the effect of different distributions on estimated within-person variation and reference change values. Clin Chem 2016;62:725–36.
27. Lund F, Petersen PH, Fraser CG, Soletormos G. Calculation of limits for significant bidirectional changes in two or more serial results of a biomarker based on a computer simulation model. Ann Clin Biochem 2015;52:434–40.