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Introducing the Lipidomics Minimal Reporting Checklist

The rapid increase in lipidomic data has triggered a community-based movement to develop guidelines and minimum requirements for generating, reporting and publishing lipidomic data. The creation of a dynamic checklist summarizing key details of lipidomic analyses using a common language has the potential to harmonize the field by improving both traceability and reproducibility.

Jeffrey G. McDonald, Christer S. Ejsing, Dominik Kopczynski, Michal Holčapek, Junken Aoki, Makoto Arita, Masanori Arita, Erin S. Baker, Justine Bertrand-Michel, John A. Bowden, Britta Brügger, Shane R. Ellis, Maria Fedorova, William J. Griffiths, Xianlin Han, Jürgen Hartler, Nils Hoffmann, Jeremy P. Koelmel, Harald C. Köfeler, Todd W. Mitchell, Valerie B. O'Donnell, Daisuke Saigusa, Dominik Schwudke, Andrej Shevchenko, Candice Z. Ulmer, Markus R. Wenk, Michael Witting, Denise Wolrab, Yu Xia, Robert Ahrends, Gerhard Liebisch and Kim Ekroos

Since the early 2000s, lipid analysis by mass spectrometry (MS) has undergone substantial growth. This growth has benefited many scientific fields, and now lipid measurements are increasingly common in a diverse array of scientific fields and journals.

Problems in lipidomics reporting

This expansion has led to confusion and uncertainty regarding published data when analyses are performed. Methods sections are frequently minimized, and methods, when reported, are relegated to supplementary material where they are often truncated and not carefully reviewed. Submissions to non-lipid-focused journals do not always receive technical input from peer reviewers with lipidomic expertise to properly assess the quality of the methods. Furthermore, the expanded use of commercial services that provide lipidomic measurements leads to data being published with little or no description of the methods due to claims of proprietary methodology. A further uncertainty appears when researchers annotate MS data using databases and search algorithms without sufficient knowledge of how the curation is applied or without considering the level of accuracy that is ascribed. Not all of these databases or algorithms have been thoroughly vetted by the community, and many have been developed for a different purpose to lipid annotation. With few exceptions, lipids are measured with commercially available instrumentation (MS and liquid chromatography) using variations on several over-arching methods. These methods must be described sufficiently for a reader to judge the quality and validity of reported lipidomic data. When researchers who lack lipid expertise receive data from colleagues, databases or proprietary commercial services, it is often difficult to judge its quality and completeness. This difficulty can lead to the publication of papers with inadequate or incorrect lipid data[1,2].

A move towards standardization

In a previous Comment, we called for the lipidomics community to work together towards standardization in the field by establishing guidelines and minimum requirements for the publication of lipidomics data[3]. Established in 2019, the Lipidomics Standard Initiative (LSI) began coordinating these efforts through a series of public web-based workshops over the summer of 2020, which attracted approximately 150 international researchers from both the lipidomic and metabolomic fields. LSI integrated feedback from these meetings to prepare guidelines and minimum reporting standards. These guidelines are also published on the LSI website that is affiliated as interest group to the International Lipidomics Society (ILS). Although consensus-driven guidelines for lipidomics are now in place, actionable use of these guidelines remains absent. Here, we propose a checklist concept that leverages and expands these guidelines into a freely available, virtual document to accelerate standardization in our field.

The checklist concept

We have compiled the guidelines and minimum requirements into a dynamic, interactive, virtual checklist accessible to everyone. The checklist is composed of nine sections.

The 'Pre-Analytics' section covers aspects of the samples before processing, extraction or analysis. This section includes sample type, origin, storage conditions, freeze-thaw cycles and other information pertinent to the quality and integrity of the samples. While researchers cannot always control every aspect of pre-analytics (for example, biobank samples), this section allows interested parties to rapidly assess issues pertaining to the history of the samples before analysis.

'Overall Study Design' gives a snapshot of the general workflow and types of analytical methods used. Mode of sample introduction (for example, direct infusion and chromatography) and ionization method (for example, electrospray ionization and matrix-assisted laser desorption/ionization) and whether the analysis is qualitative or quantitative are covered in this section.

'Lipid Extraction' includes all aspects of extracting and isolating lipids from biological samples. The extraction method, the solvents used, the internal standards and the details on additional processing is covered here. This section is particularly important because suboptimal extraction of lipids has a profound negative impact on all downstream analysis steps.

'Analytical Platform' provides details on the type of MS approach used and details on liquid chromatography. Instrument type and vendor, sample introduction and any orthogonal dimensions of analysis are described here. Key parameters including instrument resolution, mass accuracy and acquisition mode will also be included.

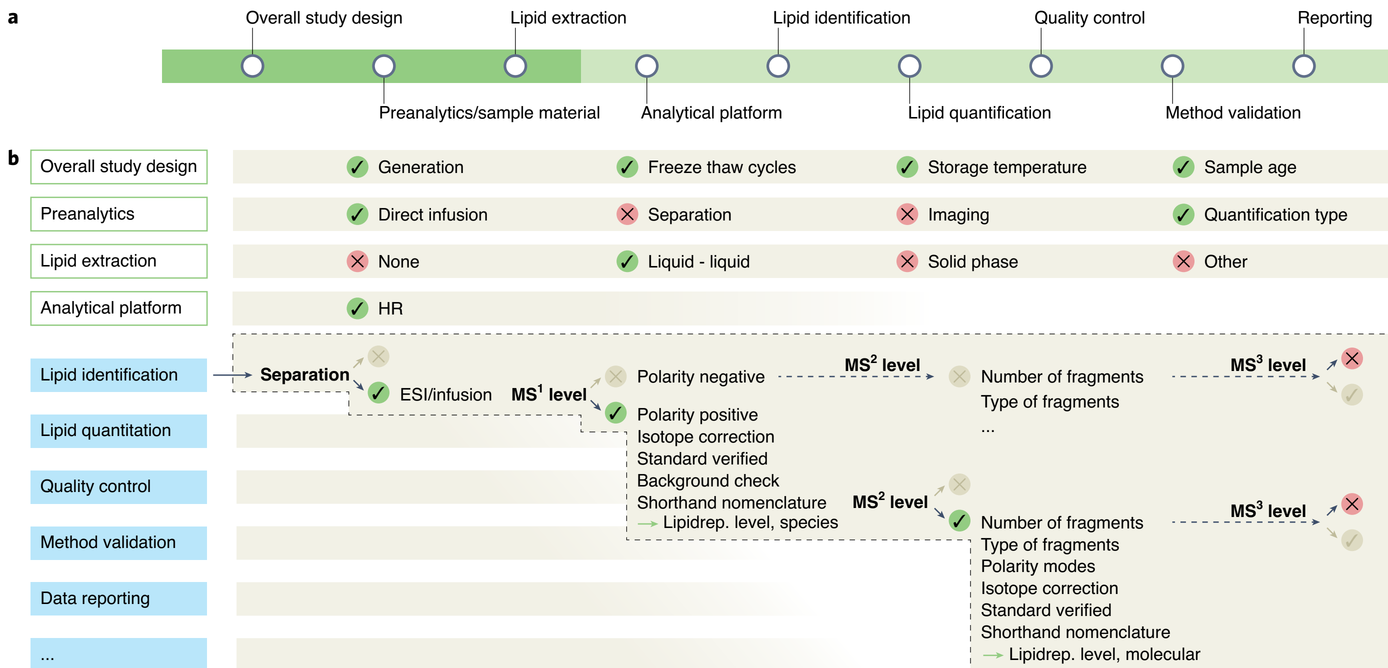

Fig. 1 | Graphical overview of the Lipidomics Minimal Reporting Checklist concept. **a**, A progress bar illustrates the various sections of the lipidomics checklist. **b**, The nine major categories that are necessary to conduct lipidomic research are shown on the left side. An expansion of the 'Separation' checkbox within the 'Lipid Identification' section shows a decision tree with subcategories and dropdown menu options for this section that are available on the online platform. After completion of the workbook, a summary report is generated that we recommend be included with a manuscript submission to a journal and added to supplementary material when published. This checklist will allow an editor, reviewer or reader to determine if minimum guidelines were met for lipidomic data presented in a manuscript. This workbook will be available on the LSI website where it will undergo regular updates and revisions based on the needs of the lipidomics field.

'Lipid Identification' defines how this instrumentation was applied to identify a lipid molecule from chromatographic and/or mass spectral data. MS level (MS^1, MS^2), ionization polarity, isotope correction, retention time and use of authentic standards are included here. This section of the checklist is unique in that it links to an expanded table where details on the respective lipid classes can be reported. Data such as precursor, fragment ions and data manipulation steps such as smoothing, check of background ions and signal to noise level are reported. Like the section on lipid extraction, this section is also crucial due to the highly complex and often isomeric and isobaric nature of lipids.

'Lipid Quantitation' designates how mass spectral data was transformed into quantitative values. Use of calibration curves or relative response, number of standards per lipid class and use of internal standards are among the checklist items covered.

Finally, 'Quality Control', 'Method Validation' and 'Reporting' cover aspects of data and method quality, and how the data were reported. The use of blank and quality control samples is included here, as well as depth of method validation including dynamic ranges and limits of detection and quantitation. Availability of (raw) is also reported.

An example of the checklist is shown in Fig. 1. A glossary of terms for each entry accompanies the checklist.

Intent and implementation

The Lipidomics Minimal Reporting Checklist has broad implications and can serve as a pillar for the field, setting standards for ongoing and future work. The checklist has multiple uses, but we first and foremost recommend that it should be included as supplementary material in publications containing lipidomic data. Modern MS-based lipidomic analysis comes in many flavours, and due to the continued growth of the field, few scientists have sufficient training and expertise to fully evaluate manuscripts with lipidomic data. This skill gap poses an essential problem for journal editors and reviewers, mainly when manuscripts with lipidomic data are submitted to multidisciplinary journals or journals focusing on biomedical research. The proposed checklist will give editors and reviewers feedback regarding the quality and completeness of lipidomic data and will, in a planned future update, introduce a numeric and/or colour-coded scoring system. Deficiencies in critical areas are flagged so that editors and reviewers know that further guidance from experts in lipidomics might be warranted. We expect this checklist system to improve the review process for journals at no additional cost and minimal effort on their part, as the LSI is committed to maintaining the checklist as a freely available resource.

The checklist also has orthogonal benefits. It serves as an excellent resource for the design of experiments as it encompasses best practices in lipidomics. An experiment designed with the help of the Lipidomics Minimal Reporting Checklist should be of high quality and yield data that are both consistent and interpretable. The checklist can also be used to prepare a manuscript to ensure that lipidomic data reported are at the highest level of rigor. Finally, the checklist represents a valuable educational tool for both the inexperienced and expert lipid mass spectrometrist, serving as a continually updated, central repository of best practices that can be used as the foundation for the education of students, fellows and colleagues.

We developed the checklist to be informative yet require only a comparatively short amount of time to complete. The

Lipidomics Minimal Reporting Checklist is not meant to recapitulate or replace the methods section in a manuscript but instead offers an easily understandable summary as a guide for readers. The checklist can be completed in 30–60 min, depending on the complexity of the experiments and data. If researchers use the checklist to design the experiments and organize their manuscripts, then completion of the checklist will take even less time.

As a virtual resource, the checklist is flexible and will be revised based on feedback from users, evolving in parallel with the lipidomics field. As recently emphasized[2], a myriad of methods exists to analyse lipids, each with its own purpose and merits. We do not intend to dictate how to measure lipids. As a diverse group of scientists with extensive knowledge and expertise in all aspects of lipidomics, we define the minimum requirements necessary to measure lipids and report lipidomic data. This checklist thus constitutes a consensus-driven tool for researchers to navigate this evolving scientific field successfully. By adopting the Lipidomics Minimal Reporting Checklist, the lipidomics field will converge into a stronger, more robust and more harmonious area. ❐

Jeffrey G. McDonald[1,37], Christer S. Ejsing[2,3,37], Dominik Kopczynski[4,37], Michal Holčapek[5,37], Junken Aoki[6,7], Makoto Arita[8], Masanori Arita[9], Erin S. Baker[10], Justine Bertrand-Michel[11], John A. Bowden[12], Britta Brügger[13], Shane R. Ellis[14,15], Maria Fedorova[16], William J. Griffiths[17], Xianlin Han[18,19], Jürgen Hartler[20,21], Nils Hoffmann[22], Jeremy P. Koelmel[23], Harald C. Köfeler[24], Todd W. Mitchell[15], Valerie B. O'Donnell[25], Daisuke Saigusa[26], Dominik Schwudke[27,28,29], Andrej Shevchenko[30], Candice Z. Ulmer[31], Markus R. Wenk[32], Michael Witting[33], Denise Wolrab[5], Yu Xia[34], Robert Ahrends[4]✉, Gerhard Liebisch[35]✉ and Kim Ekroos[36]✉

[1]Center for Human Nutrition and Department of Molecular Genetics, UT Southwestern Medical Center, Dallas, TX, USA. [2]Department of Biochemistry and Molecular Biology, VILLUM Center for Bioanalytical Sciences, University of Southern Denmark, Odense, Denmark. [3]Cell Biology and Biophysics Unit, European Molecular Biology Laboratory, Heidelberg, Germany. [4]Department of Analytical Chemistry, Faculty of Chemistry, University of Vienna, Vienna, Austria. [5]Department of Analytical Chemistry, Faculty of Chemical Technology, University of Pardubice, Pardubice, Czech Republic. [6]Graduate School of Pharmaceutical Sciences, Tohoku University, Sendai, Japan. [7]Graduate School of Pharmaceutical Sciences, University of Tokyo, Tokyo, Japan. [8]RIKEN, Center for Integrative Medical Sciences, Yokohama, Japan. [9]National Institute of Genetics, Mishima, Japan. [10]Department of Chemistry, North Carolina State University, Raleigh, NC, USA. [11]MetaboHUB-Metatoul, National Infrastructure of Metabolomics and Fluxomics, Inserm I2MC, Toulouse, France. [12]Center for Environmental and Human Toxicology, Department of Physiological Sciences, College of Veterinary Medicine, University of Florida, Gainesville, FL, USA. [13]Heidelberg University Biochemistry Center (BZH), University of Heidelberg, Heidelberg, Germany. [14]Molecular Horizons and School of Chemistry and Molecular Bioscience, University of Wollongong, Wollongong, New South Wales, Australia. [15]Illawarra Heath and Medical Research Institute, Wollongong, New South Wales, Australia. [16]Center for Membrane Biochemistry and Lipid Research, Faculty of Medicine Carl Gustav Carus of TU Dresden, Dresden, Germany. [17]Swansea University Medical School, Swansea, UK. [18]Barshop Institute for Longevity and Aging Studies, University of Texas Health Science Center at San Antonio, San Antonio, TX, USA. [19]Department of Medicine - Diabetes, University of Texas Health Science Center at San Antonio, San Antonio, TX, USA. [20]Institute of Pharmaceutical Sciences, University of Graz, Graz, Austria. [21]Field of Excellence BioHealth – University of Graz, Graz, Austria. [22]Center for Biotechnology (CeBiTec), Bielefeld University, Bielefeld, Germany. [23]Department of Environmental Health Sciences, Yale School of Public Health, New Haven, CT, USA. [24]Core Facility Mass Spectrometry and Lipidomics, ZMF, Medical University of Graz, Graz, Austria. [25]Systems Immunity Research Institute, School of Medicine, Cardiff University, Cardiff, UK. [26]Department of Integrative Genomics, Tohoku Medical Megabank Organization, Tohoku University, Sendai, Japan. [27]Research Center Borstel - Leibniz Lung Center, Borstel, Germany. [28]German Center for Infection Research, Thematic Translational Unit Tuberculosis, Partner Site Hamburg-Lübeck-Borstel-Riems, Borstel, Germany. [29]German Center for Lung Research (DZL), Airway Research Center North (ARCN), Research Center Borstel, Leibniz Lung Center, Borstel, Germany. [30]Max-Planck-Institute of Molecular Cell Biology and Genetics, Dresden, Germany. [31]Office of Public Health Science, Food Safety and Inspection Service, US Department of Agriculture, Athens, GA, USA. [32]Singapore Lipidomics Incubator (SLING), Department of Biochemistry, YLL School of Medicine, National University of Singapore, Singapore, Singapore. [33]Metabolomics and Proteomics Core, Helmholtz Zentrum München, Neuherberg, Germany. [34]MOE Key Laboratory of Bioorganic Phosphorus Chemistry & Chemical Biology, Department of Chemistry, Tsinghua University, Beijing, China. [35]Institute of Clinical Chemistry and Laboratory Medicine, University of Regensburg, Regensburg, Germany. [36]Lipidomics Consulting Ltd., Esbo, Finland. [37]These authors contributed equally: Jeffrey G. McDonald, Christer S. Ejsing, Dominik Kopczynski, Michal Holčapek.
✉*e-mail: robert.ahrends@lipidomics.at; Gerhard.Liebisch@klinik.uni-regensburg.de; kim@lipidomicsconsulting.com*

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Author contributions

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Competing interests

Kim Ekroos is the owner of Lipidomics Consulting Ltd.