Brief Communication

We engineered a machine learning approach, MSHub, to enable auto-deconvolution of gas chromatography–mass spectrometry (GC–MS) data. We then designed workflows to enable the community to store, process, share, annotate, compare and perform molecular networking of GC–MS data within the Global Natural Product Social (GNPS) Molecular Networking analysis platform. MSHub/GNPS performs auto-deconvolution of compound fragmentation patterns via unsupervised non-negative matrix factorization and quantifies the reproducibility of fragmentation patterns across samples.

Given its ease of use and low operational cost, GC–MS has applications with broad societal effect, such as detection of metabolic disease in newborns, toxicology, doping, forensics, food science and clinical testing. The predominant ionization technique in GC–MS is electron ionization (EI), in which all compounds are ionized by high-energy (70-eV) electrons. Because fragmentation occurs with ionization, EI GC–MS data are subjected to spectral deconvolution, a process that separates fragmentation ion patterns for each eluting molecule into a composite mass spectrum.

The 70 eV for ionizing electrons in GC–MS has been the standard, making it possible to use decades-old EI reference spectra for annotation. There are ~1.2 million reference spectra that have been accumulated and curated over a period of more than 50 years. Many tools and repositories for GC–MS data have been introduced; however, much of GC–MS data processing is restricted to vendor-specific formats and software. Currently, deconvolution requires setting multiple parameters manually or possessing computational skills to run the software. Also, the lack of data sharing in a uniform format precludes data comparison between laboratories and prevents taking advantage of repository-scale information and community knowledge, resulting in infrequent reuse of GC–MS data.

Although batch modes exist, deconvolution quality is currently not enhanced by using information from all other files. To leverage across-file information, improve scalability of spectral deconvolution and eliminate the need for manually setting the deconvolution parameters (m/z error correction of the ions and peak shape—slopes of raising and trailing edges, peak RT shifts and noise/intensity thresholds), we developed an algorithmic learning strategy for auto-deconvolution (Fig. 1a–f). We deployed this functionality within GNPS/MassIVE (https://gnps.ucsd.edu) (Fig. 1f–i). To promote analysis reproducibility, all GNPS jobs performed are retained in the ‘My User’ space and can be shared as hyperlinks.

This user-independent ‘automatic’ parameter optimization is accomplished via fast Fourier transform (FFT), multiplication and inverse Fourier transform for each ion across an entire data set, followed by an unsupervised non-negative matrix factorization (one-layer neural network). Then, the compositional consistency of spectral patterns for each spectral feature deconvoluted across the entire data set can be summarized as a ‘balance score’. The balance score (mathematical definition in the Methods) quantifies reproducibility of the deconvoluted fragmentation patterns across the data, which, in turn, gives insight into how well the spectral
Fig. 1 | The processing pipeline and performance. a, Spectra are aligned and binned; noise is filtered and (b) baseline corrected. c, Common profile across the data set and peaks in RT dimension are aligned using FFT-accelerated correlation. d, Generation of both peak integrals for all samples and their common fragmentation patterns. e, Separation of overlapping peaks with patterns across samples using NMF. f, Peak integrals for all samples and canonical fragmentation patterns. NIST, National Institute of Standards and Technology. g, Annotation with public or private libraries. rT, retention time index. h, Molecular networks. i, Data and results are shared between users. j, Linear dependence of the MSHub processing time. k, Distributions of library matching scores with an increased volume of data (data sets with known spiked compounds, Test1–Test11; Supplementary Table 1) for all matches and (l) for the spiked compounds only. m, FDR for annotations (Test11) of the top match and (n) top ten matches. o, Number of library matches for spiked compounds. p–q, Cosine improves as higher volume of the data enhances deconvolution quality for the top match of biological samples: breath (non-derivatized, ICL1–ICL11; Supplementary Table 1) (p) and human and mouse blood serum, adipose tissue and cerebrospinal fluid (silylated, data sets UCD1–UCD16; Supplementary Table 1) (q). r, The unique annotations across data sets ICL1–ICL11 and (s) data sets UCD1–UCD16; no balance score filtering applied. t–u, Quantitative comparison of XCMS (t) and MSHub (u). RT, retention time.
Fig. 2 | Analysis and molecular networking of GC–MS data. Annotated spectra (a) without filtering and (b) with a 65% balance score filtering. c, Global network containing 35,544 nodes from 8,489 files in 38 GNPS data sets. The size of the node is proportional to the number of nodes that connect, and the edge thickness is proportional to the cosine score (Supplementary Fig. 6). The annotation is the top match with cosine above 0.65. d, Zoomed-in region. e, Cluster of compounds from dart frog skin samples—all nodes are alkaloids. f, Human surface volatilome visualized with ‘ili’. Molecular distributions for squalene; (g) hexanoic acid, a malodor molecule; (h) globulol, common in perfume; and (i) phenylene dibenzoate, common in skincare products.
All MSHub algorithms use efficient HDF5 technologies. The Fourier transform with multiplication improves MSHub's efficiency, resulting in deconvolution times that scale linearly with the number of files (Fig. 1) and Supplementary Figs. 1a, 2 and 4. We achieved this performance using out-of-core processing, a technique used to process data that are too large to fit in a computer's main memory (RAM); MSHub uploads files one at a time into the RAM module; data are then processed and deleted from memory, iteratively. Because only one sample is stored in the memory, the load is constant (Supplementary Fig. 2a–f). As machine learning approaches gain improved performance with increased volumes of information, including more data into analysis leads to better scores of spectral matches (Fig. 1k,l and Supplementary Fig. 1b). The spectral library match scores increase, and their distributions become narrower, indicating better quality of results (Fig. 1p,q). More files deconvoluted in MSHub leads to fewer chimeric spectra, resulting in higher-quality spectral features, and an increase in the number of annotations with improved scores (Fig. 1r,s). MSHub performs as well or better as other deconvolution tools (Fig. 1t,u and Supplementary Figs. 3–5). Linear scaling for MSHub makes it the only tool amenable to repository-scale operation in its present form (Supplementary Table 2). GNPS saves deconvoluted data as a summary file, so the deconvolution step does not need to be re-performed for any future analyses.

Once the summary file is generated by GNPS-MSHub or imported from another deconvolution tool, the spectra can be searched against public, private or commercial libraries. Matches are narrowed down based on user-defined filtering criteria, such as number of matched ions, Kovats index, balance score, cosine score and abundance. We provide freely available reference data of 19,808 spectra for 19,708 standards, a ~29% increase of free public libraries. All annotations should be considered level 3 (a molecular family) annotation. When multiple annotations can be assigned, GNPS provides all candidate matches within the user's filtering criteria.

One of the developments that enabled finding structural relationships within mass spectrometry data is spectral alignment, which forms the basis for molecular networking. GNPS has now expanded to include GC–MS-specific molecular networking. GNPS-based GC–MS analysis enables data co- and re-analysis, as the processing is agnostic to the data origin. To showcase this ability, we built a global network of various public GC–MS data sets and applied a balance score of 65% (Fig. 2a,b and Supplementary Fig. 6) to ensure that only good-quality deconvoluted spectra are matched against the reference library (Fig. 2c–e and Supplementary Figs. 9 and 10). Molecular networking can further guide the annotation at the molecular family level by using information from connected nodes rather than focusing on individual annotations (Supplementary Figs. 7 and 8). One can visualize aspects such as derivatized versus non-derivatized, candidate compound class or subclass and instrument type or other metadata and inspect individual clusters of nodes (Supplementary Fig. 9). For example, we observed a cluster that belonged to dart frogs from the Dendrobatidae superfamily, whereas the long-chain ketones are found in cheese and beer (Fig. 2e and Supplementary Fig. 10a).

The output from GNPS can be exported for use in statistical analysis environments and for data visualization (for example, Supplementary Figs. 7–10), including molecular cartography. GNPS/MassIVE lowers the expertise threshold required for analysis and encourages Findable, Accessible, Interoperable and Reusable (FAIR) practices by promoting re-use of GC–MS data. To highlight the broader utility of GNPS GC–MS-based analysis, videos were created (Supplemental Videos 1–6). This work aims to democratize scientific analyses. GC–MS is often the only mass spectrometry method in non-metabolomics laboratories or laboratories with fewer resources, including those in developing countries. GNPS-based GC–MS allows free access to data and reference data and to powerful computing infrastructures.

**Online content**

Any methods, additional references, Nature Research reporting summaries, source data, extended data, supplementary information, acknowledgements, peer review information; details of author contributions and competing interests; and statements of data and code availability are available at https://doi.org/10.1038/s41587-020-0700-3.

Received: 13 January 2020; Accepted: 9 September 2020; Published online: 9 November 2020

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Methods

Tutorials and general note. The tools are accessible through http://gnps.ucsd.edu. The documentation to use the GC–MS interface can be found at https://ccms-ucsd.github.io/GNPSDocumentation/gcanalysis/. The tutorials for the deconvolution can be accessed at https://ccms-ucsd.github.io/GNPSDocumentation/gc-ms-deconvolution/, and the library search and molecular networking instructions can be found at https://ccms-ucsd.github.io/GNPSDocumentation/gc-ms-library-molecular-network/.

The tutorial for spectral libraries upload can be found at https://ccms-ucsd.github.io/GNPSDocumentation/spectralupload/.

The GNPS workflows can be launched with recommended default settings or adjusted according to user needs. The ranges and effect of settings are described in the tutorial.

The results can be inspected and quality filters applied according to user criteria. The tutorial also describes how users can use various other aspects of GNPS functionality that include:

- Data upload and storage
- Data sharing
- Sharing analysis by sharing workflows
- Reproducing analyses
- Saving and sharing reference spectra
- Using GNPS analysis links for publishing
- Using GNPS/MassIVE repository for providing access to data along with the publication when required by the journal

The video tutorials for GNPS use for GC–MS data and examples of networking application videos can be accessed as follows: Tutorial for the use of GNPS for analysis of GC–MS data: https://www.youtube.com/watch?v=KIOMZ2hG6t0

GNPS for GC–MS in biology: using molecular networking to find incorrect annotations: https://www.youtube.com/watch?v=tD5s0L3HbK

GNPS for GC–MS in biochemistry: using networking to combine different data sets: https://www.youtube.com/watch?v=bDvZLi2Gw

Use of the GNPS GC–MS workflows. GNPS GC–MS environment. The GNPS leverages the repository infrastructure and now has expanded to include GC–MS–specific deconvolution, reference spectra matching and molecular networking tools. The new analysis workflows not only solved the scaling of analysis, but also configured to promote data analysis reproducibility, as an analysis performed in GNPS is retained in the account-specific job tab and can be shared as a hyperlink. The user’s own or someone else’s shared analysis can be precisely reproduced by clicking the ‘clone’ button. In addition, we have enabled the community to upload and share reference spectra that then continuously accumulate, leading to continuous improvements of annotations. GNPS also gives the ability to explore all public data sets together with studies in one’s private space for a particular research problem (for example, drug discovery). There are no other publicly available GC–MS–deconvolution and annotation infrastructures that also work with the data in a repository. The scalability, reproducibility, capture of knowledge and ability to efficiently reuse data in the public domain make the GC–MS infrastructure in GNPS unique compared to other existing open or commercial resources. GNPS promotes FAIR use practices for mass spectrometry data.

Deconvolution. Currently, 1D EI GC–MS data are amenable. We recommend using a maximum of ten files in the data set for deconvolution with MSHub. If the user only has fewer than ten files, spectral deconvolution and alignment should be performed using alternative methods (for example, ZMmine, Open Chrom, AMDIS, MZmine/ADAP MS–MDIAL, BinBase, XCMS/XCMS Online, MetaAlign, SpecAlign, SpecConnect, PARAFAC2, MeDDB or eRah). After using one of those tools, molecular networking can be performed in the same fashion as for MSHub (a detailed description is given in the Supplementary Notes), as the library search GNPS workflow accepts input from other tools into the GNPS/MassIVE environment. GNPS directly supports deconvolution output from ZMmine/ADAP and MS–MDIAL. The quantitative table of the deconvolution output can be used for statistical analysis with external tools.

Library search. Once the .mgf file is generated by GNPS-MSHub or imported from another deconvolution tool, the spectral features can be searched against public libraries (currently GNPS has Fiehn, HMDB, MoNA and VocBinBase) or the user’s own private or commercial libraries (such as NIST and Wiley) and the freely available reference data of 19,908 spectra for 19,708 standards released with this manuscript. Users can also upload their own libraries to GNPS as well as share them with the community. Although the possible candidate annotations can be further narrowed by retention index (RI), they should still be considered level 3, a molecular family, annotation according to the 2007 Metabolomics Standards Initiative. Calculation of RIs is included and encouraged but not enforced. When multiple annotations can be assigned, GNPS provides all candidate matches within the user’s filtering criteria.

Filtering the results. The balance score is a new metric that will be available when MSHub deconvolution is used. A fragmentation pattern of a compound found too many times in different measurements would result in a high balance score. Missing or chimeric peaks would change randomly across files and would result in a low balance score. Even when a compound is present in few samples, as long as the spectral patterns (irrespective of compound abundances) are conserved across samples, it would result in a high balance score.

Cosine and balance score should be jointly used as spectral matching filters for processing of the final results. The effect of filtering can be seen in Fig. 1m–n and Supplementary Fig. 3d,e. For the test data set shown in Fig. 1m,n, the lowest false discovery rate (FDR) of the top match was achieved with the combined threshold values of cosine >0.9 and balance score >60% (Fig. 1m). A more conservative balance score value of >80% essentially ensures the lowest observed FDR, even for poor cosine scores (these refer to match scores). Conversely, even a high match score by itself might still result in unacceptably high FDR if the balance score is poor (Fig. 1m,n). The high match score reflects that a library spectrum exists that is similar to the query spectrum, whereas a high balance score is reflective of the high confidence in deconvolution of the spectral pattern. A wise deconvolution pattern, as defined by the balance score, is likely to give better matches against the spectral library. Selecting higher values of both metrics ensures that the best spectra are used and are matched to most likely annotations. The ‘optimal’ thresholds—that is, the values that minimize mis-annotations without being excessively restrictive—are data specific, but we recommend using the above values as a good starting point.

Molecular networks. No matter how the spectral library is searched in GC–MS, owing to the absence of a parent mass, a list of spectral matches is more likely to contain mis-annotations, both related (isomers and isobars) or, less frequently, entirely unrelated compounds. However, to spot mis-assignments at the molecular family level, we propose exploring deconvoluted GC–MS data via molecular networking, a strategy that has been effective for liquid chromatography with tandem mass spectrometry (LC–MS/MS) data. In the case of EI, unlike in LC–MS/MS where the precursor ion mass is known, the molecular ion is often absent. For this reason, the molecular networks are created through spectral similarity of the deconvoluted fragmentation spectrum without considering the molecular ion. We explored molecular networking patterns for the EI data (Supplementary Fig. 7) and observed that the EI-based cosine similarity networks are predominantly driven by structural similarity based on chemical class annotations (Supplementary Fig. 7a). These EI networks can be used to visualize chemical distributions and guide annotations (Supplementary Fig. 8). Some examples of molecular networking applications are discussed in the Supplemental Videos.

Three-dimensional mapping of volatilome. The sample collection and GC–MS analysis are described in the “Skin volatilome analysis” section of the Supplementary Notes. Feature tables from the deconvolution jobs for head space volatiles and injection were downloaded from GNPS and combined into a single table. The coordinates for the three-dimensional model were picked for all of the sampled spots and added into the feature table as described in the tutorial (https://ccms-ucsd.github.io/GNPSDocumentation/gcanalysis/). The chemical distributors were then visualized using iGV. The chemical annotations of features have been cross-referenced from the library search jobs as described in the tutorial. Using balance filters at 50% and >0.9 cosine, we arrived at annotations that, once visualized, revealed the distributions of skin volatiles (Fig. 2f–i). For example, squalene was found on all locations but less on the feet. Hexanoic acid was most abundant on the chest and armpits. Globulol, a perfume ingredient that this individual used on the chest, was most intense on the chest, whereas phenylene dibenzoate, a skincare ingredient, was found on the face and hands. The three-dimensional model, the feature table used for mapping and the snapshots shown in Fig. 2f–i are available at https://github.com/aaksenov1/Human-volatilome-3D-mapping.

Generation of molecular networks. The data were collected across multiple studies as described in the Supplementary Notes. All of the data sets (Supplementary Table 1) were processed on the GNPS MSHub deconvolution workflow as described in the tutorial. The figures were generated as described in the Supplementary Notes.

Testing and validation. All modules were tested and validated individually to determine possible fail points and the results validated by manually reviewing the annotations that are obtained. The full pipeline was also tested for a variety of data sets, including those collected for this study (the GC–MS analysis for validation studies’ section of the Supplementary Notes) and data from several previously published studies and unpublished public data. A variety of GC–MS
data are represented, including different types of mass analyzers (both high- and low-resolution instruments), different modes of sample introduction and analysis of both derivatized and non-derivatized samples. The goal was to ensure that both feature finding and library matching workflows are operational for all of these scenarios and that the results are consistent with those expected. We manually verified that the molecules that are known to be present in the data set are indeed identified and reported by the workflow. The testing information is summarized in Supplementary Table 1.

Comparison of deconvolution tools. We compared the deconvolution performance of MSHub alongside MZmine2/ADAP and MS-DIAL. These tools were chosen because they satisfy the following criteria: they are open, specifically designed for GC-MS data, can perform multi-file processing, and are routinely used by the metabolomics community and are actively being developed and maintained. Detailed descriptions of the procedure and parameters are given in the Supplementary Notes.

Generating input files with the alternative workflows. The MZmine2/ADAP and MS-DIAL workflows are the alternative options to perform spectral deconvolution on GC-MS data explicitly supported to be compatible with the GNPS library search workflow. For better integration, we added a new module to MZmine (version 2.52 and later) to export the quantification table (.csv) and the spectra summary file (.mgf) for the GNPS GC-MS workflow. Furthermore, a new MZmine module was also developed to enable the conversion of the Kovats RI marker file compatible with the GNPS workflow. Detailed directions are given in the GNPS documentation: https://ccms-ucsd.github.io/GNPSDocumentation/gc-ms-deconvolution/.

Generation of plots. All plots were generated in Python 3.7.3, using NumPy 1.16.4, Pandas 0.25.0, RDKit 2019.03.4 and lxml 4.3.4 for data analysis purposes and Matplotlib 3.1.0 and Seaborn 0.9.0 for visualization purposes. The detailed description is given in the Supplementary Notes.

Reporting Summary. Further information on research design is available in the Nature Research Reporting Summary linked to this article.

Data availability
All of the data used in the preparation of this manuscript are publicly available at the MassIVE repository at the University of California, San Diego Center for Computational Mass Spectrometry website (https://massive.ucsd.edu). The data set accession numbers are: #1 (MSV000080430), #2 (MSV000085136), #3 (MSV000080434), #4 (MSV000084036), #5 (MSV000084032), #6 (MSV000080438), #7 (MSV000084042), #8 (MSV000084039), #9 (MSV000084040), #10 (MSV000084041), #11 (MSV000083598), #12 (MSV000080892), #14 (MSV000080892), #15 (MSV000080892), #16 (MSV000083437), #17 (MSV000083658), #18 (MSV000083743), #19 (MSV000084226), #20 (MSV000083859), #21 (MSV000083824), #22 (MSV000084349), #23 (MSV000081340), #24 (MSV000084340), #25 (MSV000084376), #26 (MSV000084383), #27 (MSV000084339), #28 (MSV000081161), #29 (MSV000084350), #30 (MSV000084377), #31 (MSV000084145), #32 (MSV000084144), #33 (MSV000084379), #34 (MSV000084380), #35 (MSV000084246), #36 (MSV000084276), #37 (MSV000084277) and #38 (MSV000084212).

All of the GNPS analysis jobs for all of the studies are summarized in Supplementary Table 1.

Code availability
The source code of the MShub software, including low- and high-resolution data processing versions, is available online at GitHub (version used in GNPS) (https://github.com/CCMS-UCSD/GNPS_Workflows/tree/master/mshub-gc-tools/mshub-gc-proc) and at BitBucket (standalone version in MShub developers’ repository, both high and low resolution: https://bitbucket.org/aAnalytica/mshub_process/src/master/). Scripts used to parse, filter, organize data and generate the plots in the manuscript are available online at GitHub (https://github.com/bittremenix/GNPS_GC_fig). Script for merging individual .mgf files into a single file for creating global network is available at GitHub (https://github.com/bittremenix/GNPS_GC_Molnet/mshub-process/src/merge_mgf.py).

The three-dimensional model, the feature table with coordinates used for the mapping and the snapshots shown in Fig. 4a–d are available at https://github.com/aaksonov1/Human-voalite3D-3d-mapping/. The GC-MS-adopted MolNetEnhancer code with an example Jupyter notebook can be found at https://github.com/madeleineernst/ MolNetEnhancer. Source data are provided with this paper.

Acknowledgements
The conversion of the data from different repositories was supported by grant R03 CA211211 on reuse of metabolomics data, to build enabling chemical analysis tools for the ocean symbiosis program, and the development of a user-friendly interface for GC-MS analysis was supported by the Gordon and Betty Moore Foundation through grant GBMF7622. The University of California, San Diego Center for Microbiome Innovation supported the campus-wide seed grant awards for data collection that enabled the development of some of this infrastructure. P.C.D. was supported by the National Science Foundation (grant no. IOS-1656475) and the National Institutes of Health (grant nos. R01 GM140392 and R01 GM107550). K.V. and L.L. are very grateful for the support of the Vodafone Foundation as part of the DRUGS/DreamLab project. The MSHub platform development was supported by NIH/NIAAA grant (R21 AA028432) on integrated machine learning for mass spectrometry data in liver disease, Intelligently Limited and Vodafone Foundation’s DRUGS/CORENA-AI projects on network machine learning for drug repositioning and discovery of hyperfoods with antiviral/anticancer molecules. M.E. was supported by the University of Cordoba. L.F.N. was supported by the NIH (R01 GM107550) and the European Union’s Horizon 2020 Research and Innovation Programme (MSCA-Ge.740768). A.B. was supported by the National Institute of Justice Award (2015–DN–B9–K047). Additional support for data acquisition and data storage was provided by the Center for Computational Mass Spectrometry (PA41 GM130484).

The collection of data from the HomeChem Project was supported by the Sloan Foundation. G.B.H., S.L.F.D., I.L., K.V. and I.B. are grateful for the support of the O.G cancer breath analysis study by the National Institute for Health Research London Invitro Diagnostic Co-operative and the NIH Imperial Biomedical Research Centre, the Rosetrees and Stonegate Trusts and the Imperial College Charity. D.V. acknowledges support from ERC-Consolidator grant 724228 (LEMAN). I.B. acknowledges the contribution of Q. Wen and M. Colavita in the production of the training video. C. Callewaert was supported by the Research Foundation Flanders, with support from the industrial research fund of Ghent University. W.B. was supported by the Research Foundation Flanders. A.A.K. acknowledges the support of the Programa de Investigación de Innovación Tecnológica Nacional (CONICET-Argentina). The work of P.L. and P.L.B. on the data set 30 was supported by Metaboxpe, part of the ‘Platform 3a’ funded by the European Regional Development Fund, the French Ministry of Research, Higher Education and Innovation, the Provence-Alpes-Côte d’Azur region, the Departmental Council of Vaucluse and the Urban Community of Avignon. S.A. and A.J.L. acknowledge the PlantSysX project by the European Union’s Horizon 2020 Research and Innovation Programme (SGA-CSA nos. 664621 and 739582 under FPA no. 664620). V.V. acknowledges support from the National Institute on Alcohol Abuse and Alcoholism award R24AA022057. M. Gupta and R.C. acknowledge the support of the Krupp Endowed Fund grant. A portion of mass spectra in the public reference library was produced within the framework of the State Task for the Topchiev Institute of Petrochemical Synthesis RAS and with the support of the RUDN University Program 5-100. R.S.B. acknowledges support of the State Task for the Topchiev Institute of Petrochemical Synthesis RAS. L.N.K. acknowledges support of the RUDN University Program 5-100. I.M. acknowledges support of the Israel Science Foundation (project no. 1947/119) and European Research Council under the European Union’s Horizon 2020 Research and Innovation Programme (project no. 640384). J.S. has been supported by NIH/NIAAAR03AR072182, the Colton Center for Autoimmunity, the Rheumatology Research Foundation, the Riley Family Foundation and the Snyder Family Foundation. J. Manasson acknowledges support from the 2017 Group for Research and Assessment of Psoriasis and Pсорiatric Arthritis Pilot Research Grant and NIH/NIAAMS T32AR065915. R.C. is grateful to the Azrili Foundation for the award of an Azrili Fellowship. J.J.v.d.H. acknowledges support from an ASDi eScience grant (ASDI2017:030) from the Netherlands eScience Center–NLeSC. B.A. was supported by the National Science Foundation through the Graduate Research Fellowship Program. GC-MS analyses for collection of the MSV000083743 data set were supported by the Pacific Northwest National Laboratory, Laboratory Directed Research and Development Program, and were contributed by the Microbiomes in Transition Initiative; data were collected in the Environmental Molecular Sciences Laboratory, a national scientific user facility sponsored by the Department of Energy (DOE) Office of Biological and Environmental Research and located at the Pacific Northwest National Laboratory (PNNL). PNNL is operated by the Battelle Memorial Institute for the DOE under contract DEAC05-76RL01830. M. Gupta and B.C. acknowledge the support of the Krupp Endowed Fund grant. R.C. was also funded by T32AR064194-07. The authors are grateful to R. da Silva for his contribution to developing the first prototype of the EI data network and his continuous assistance with further development and testing of the infrastructure. The authors are also grateful to M. Vance and D. Farmer, who organized the sampling for Human-Volatilome 3D-mapping-. The GC–MS-adapted MolNetEnhancer code with the snapshots shown in Fig. 4a–d are available at https://github.com/aaksonov1/Human-voalite3D-3d-mapping/. The GC–MS-adopted MolNetEnhancer code with an example Jupyter notebook can be found at https://github.com/madeleineernst/ MolNetEnhancer. Source data are provided with this paper.

Author contributions
P.C.D., A.A.A., M.W. and L.F.N. developed the concept of GNPS for GC-MS data. K.V. designed and supervised MSHub platform development. I.L., D.V. and V.V.
K.V. developed the MSHub platform. M.W., Z.Z. and A.A.A. developed the workflows. A.A.A., Z.Z., M.W., B.B.M. and R.S.B. performed infrastructure testing and benchmarking. A.A.A., Z.Z. assessed EI-based molecular networking. W.B. generated plots for MSHub algorithm performance testing and benchmarking against existing deconvolution tools. Z.Z., A.A. and M.E. generated molecular network plots. M.E. and JJJ.v.d.H. adapted the MolNetEnhancer workflow for GC–MS molecular networks. A.S., X.D., A.A.A. and B.B.M. conducted comparative testing of MSHub with existing deconvolution tools. A.A.A., A.V.M., M.P., K.L.J. and K.D. conducted three-dimensional skin volatolome mapping studies. S.L.F.D., I.B. and G.B.H. conducted the esophageal and gastric breath analysis cancers detection study. A.A.A., Z.Z., M.P. and M.W. converted and added public libraries to GNPS. A.A.A., A.V.M., S.L.F.D., C. Callewaert, B.B.M., M. Gonzalez, C. Carazzone, A.A., J.T.M., R.A.Q., A.B., A.A.O., D.P., A.M.S., S.P.C., T.O.M., M.C.B., C.D.N., E.Z., E.H.-F., R.G., M.M.M., I.M., S.E., P.L.B., B.A., R.D., R.I., Y.G., S.P., A.F., G.D., B.I.B., A.F., N.S.P., K.G., C.S., R.C., M. Giana, J. Manasson, J.U.S., D.K.B., S.A. and A.R.F. generated GC–MS data. M.N.-E., A.A.A., M. Gonzalez, B.B.M., A.S. and L.N.K. produced training videos. M.N.-E., A.A.A., M. Gonzalez, K.N.M. and R.S.B. produced training videos. M.N.-E., A.A.A., M. Gonzalez, K.N.M. and R.S.B. produced training videos. P.C.D., A.A.A., W.B., K.V., R.M. and R.K. wrote the paper.

Competing interests
P.C.D. is a scientific advisor for Sirenas, Galileo and Cybele. P.C.D. is scientific adviser and cofounder of Enveda and Omecta; this has been approved by UC San Diego. M.W. is a consultant for Sirenas and the founder of Omecta Labs. A.A.A. is a consultant for Omecta Labs.

Additional information
Supplementary information is available for this paper at https://doi.org/10.1038/s41587-020-0700-3.
Correspondence and requests for materials should be addressed to P.C.D. or K.V.
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  - A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
  - For null hypothesis testing, the test statistic (e.g. F, t, r) with confidence intervals, effect sizes, degrees of freedom and P value noted
    
    Give P values as exact values wherever suitable.
  - For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
  - For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
  - Estimates of effect sizes (e.g. Cohen's d, Pearson's r), indicating how they were calculated

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection

Collection of GC-MS data was carried out using software supplied by the manufacturer for each individual instrument.

Data analysis

The source code of the MShub software used for deconvolution of all data is available online at Github (version used in GNPS) [https://github.com/CCMS-UCSD/GNPS_Workflows/tree/master/mshub-gc/tools/mshub-gc/proc] and at BitBucket (standalone version in MShub developers' repository: https://BitBucket.org/Analytica/mshub_process/src/master/). Scripts used to parse, filter, organize data and generate the plots in the manuscript are available online at Github [https://github.com/bitremieux/GNPS_GC_fig]. Script for merging individual .mgf files into a single file for creating global network is available at Github: [https://github.com/bitremieux/GNPS_GC/blob/master/src/merge_mgf.py].

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Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A list of figures that have associated raw data
- A description of any restrictions on data availability

All of the data used in preparation of this manuscript are publicly available at the MassIVE repository at the UCSD Center for Computational Mass Spectrometry website [https://massive.ucsd.edu]. The dataset accession numbers are: #1 (MSV000084033), #2 (MSV000084033), #3 (MSV000084034), #4 (MSV000084036), #5 (MSV000084032), #6 (MSV000084038), #7 (MSV000084042), #8 (MSV000084039), #9 (MSV000084040), #10 (MSV000084037), #11 (MSV000084211), #12 (MSV000085398), #13 (MSV000080892), #14 (MSV000080892), #15 (MSV000080892), #16 (MSV000083537), #17 (MSV000083658), #18 (MSV000083743), #19 (MSV000084226), #20 (MSV000083859), #21 (MSV000083294), #22 (MSV000084349), #23 (MSV000081340), #24 (MSV000084348), #25 (MSV000084378), #26
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Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

| Sample size | Multiple datasets including previously collected and/or published studies were co-analyzed; the sample size in each dataset was contingent on the original study goals. The evaluation of MSHub algorithm performance was carried out for datasets of varying sizes ranging from minimum amenable of five samples to several thousands |
|-------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Data exclusions | No data points were excluded from the analysis. |
| Replication | All analyses can be directly replicated by cloning the workflow jobs within the GNPS environment. Links to all of the jobs with source files are included in the Table S1 submitted along with the manuscript. |
| Randomization | N/A |
| Blinding | N/A |

Reporting for specific materials, systems and methods

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| Materials & experimental systems | Methods |
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| ☑ Palaeontology | ☑ MRI-based neuroimaging |
| ☑ Animals and other organisms | |
| ☐ Human research participants | |
| ☑ Clinical data | |

Human research participants

Policy information about studies involving human research participants

| Population characteristics | A single volunteer has been enrolled into the skin volatilome mapping study. The volunteer has had no diagnosed medical conditions, including skin disease and has been carrying out normal daily routine prior to sampling. |
|---------------------------|------------------------------------------------------------------------------------------------------------------|
| Recruitment | The recruitment was carried out under the UC San Diego IRB #171662. |
| Ethics oversight | All of the sampling for the skin volatilome mapping has been conducted under UC San Diego IRB #171662. |

Note that full information on the approval of the study protocol must also be provided in the manuscript.