Scientific and Regulatory Perspectives on Mass Spectrometry-based Strategies During Early Stages of Therapeutic Development for COVID-19

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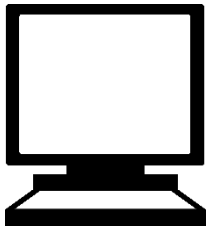
FDA/CDER

CASSS Mass Spec 2021

Pharmaceutical Quality

A quality product of any kind consistently meets the expectations of the user.



Pharmaceutical Quality

A quality product of any kind consistently meets the expectations of the user.

Drugs are no different.


FDA
Patients expect safe and effective medicine with every dose they take.

Pharmaceutical quality is

assuring *every* dose is safe and effective, free of contamination and defects.


It is what gives patients confidence in their *next* dose of medicine.

Disclaimer

The views and opinions expressed should not be used in place of regulations, published FDA guidances, or discussions with the Agency.

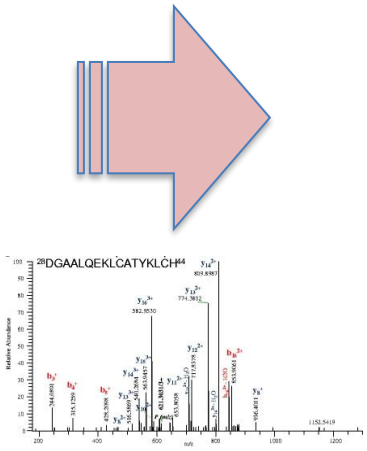
Outline of presentation

- How MS analyses fit into biotech control strategies
- Applications of MS during the characterization and control of biologic drugs
- Current opportunities and challenges with MS in the context of COVID-19

How MS analyses fit into biotech control strategies

Control strategies for biotech products

- Product Characterization
- Mechanism of Action Studies
- Pre-formulation Studies
- Small-scale Manufacturing
- Process development
- Method Qualification



- Control of Raw Materials
- Control of Cell Banks
- Drug Substance
- Drug Product
- Release testing
- Justified Specifications
- Quality Management System
- Process Validation
- Method Validation
- cGMP
- Risk Analysis Trending
- Product & Process Lifecycle Management
- Drug Intermediate
- In-Process Testing
- Reference Standards
- Formulation
- Stability testing

Applications of MS during the characterization and control of biologic drugs

Applications of MS for biotechnology product analysis

- Identification
- Characterization
- Comparability (process change by same manufacturer)
- Comparative analytical assessment (biosimilar vs reference product)
- Surveillance for contamination/adulteration
- Process improvement
- New product knowledge (new critical post-translational modification or other structural attributes)
- PK/PD measurement

(Rathore D et al, Exp Rev Proteomics, 2018; Namuswe F, CASSS MS, 2019)

J. Am. Soc. Mass Spectrom. (2017) 28:786–794
DOI: 10.1007/s13361-016-1531-9

FOCUS: 28th SANIBEL CONFERENCE, CHARACTERIZATION OF PROTEIN THERAPEUTICS BY MS: RESEARCH ARTICLE

A Retrospective Evaluation of the Use of Mass Spectrometry in FDA Biologics License Applications

Sarah Rogstad,[1] Anneliese Faustino,[1] Ashley Ruth,[2] David Keire,[1] Michael Boyne,[2] Jun Park[3]

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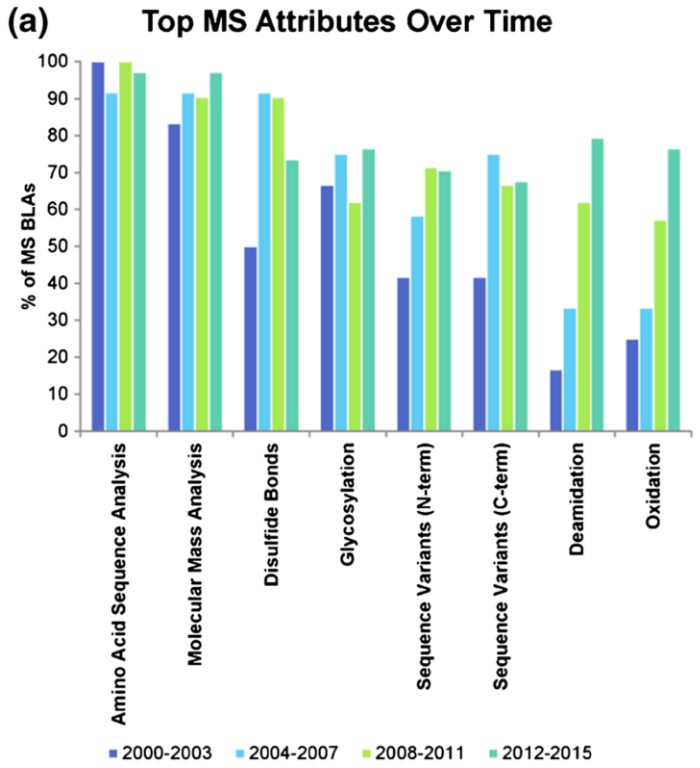
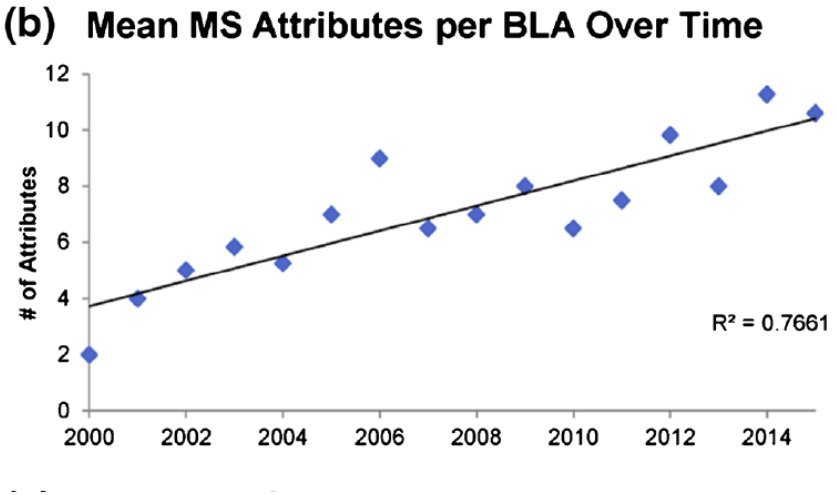
[2]Biotechlogic, Inc., Glenview, IL 60025, USA
[3]Office of Biotechnology Products, Center for Drug Evaluation and Research, U.S. Food and Drug Administration, Silver Spring, MD 20993, USA





BLAs approved between 2000-2015 using MS

EXPERT REVIEW OF PROTEOMICS
2018, VOL. 15, NO. 5, 431–449
https://doi.org/10.1080/14789450.2018.1469982

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FDA

REVIEW

The role of mass spectrometry in the characterization of biologic protein products

Deepali Rathore[a,b], Anneliese Faustino*[a], John Schiel[c], Eric Pang[d], Michael Boyne[a,e] and Sarah Rogstad[a]

[a]Division of Pharmaceutical Analysis, Office of Testing and Research, Center for Drug Evaluation and Research, U.S. Food and Drug Administration, Silver Spring, MD, USA; [b]Center for Biomedical Mass Spectrometry Research, Medical College of Wisconsin, Milwaukee, WI, USA; [c]Biomolecular Measurement Division, National Institute of Standards and Technology, Institute for Bioscience and Biotechnology Research, Rockville, MD, USA; [d]Office of Lifecycle Drug Products, Office of Pharmaceutical Quality, Center for Drug Evaluation and Research, U.S. Food and Drug Administration, Silver Spring, MD, USA; [e]COUR Pharmaceuticals Development Company, Northbrook, IL, USA

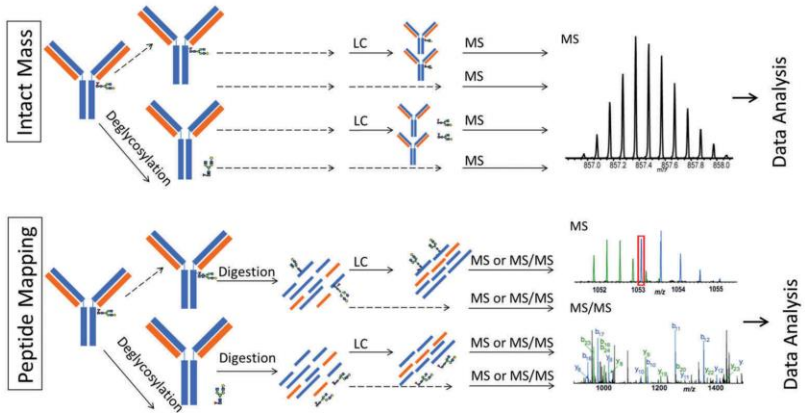


Figure 2. Routine MS-based mAb analysis. Common options for MS analysis of a glycosylated mAb are shown. Analysis can be conducted via either peptide mapping or intact mass analysis. Either of these methods can be completed with or without deglycosylation and LC separation. Deglycosylation can be done using PNGaseF, which cleaves glycans from the protein, while enzymatic digestion breaks the protein down into peptides. Peptides or protein isoforms can then be separated using LC. Intact mass analysis is conducted using full MS scans only; however, additional top-down techniques utilize MS/MS for further sequence information [73]. Adapted with permission from Boyne, Michael T. et al. 'Tandem mass spectrometry with ultrahigh mass accuracy clarifies peptide identification by database retrieval.' *Journal of proteome research* 8.1 (2009): 374–379. *PMC*. Web. 26 July 2016. Copyright 2016 American Chemical Society.

Table 4. Major MS attributes for analysis. Thirty-two specific MS attributes were found to be analyzed at varying levels across BLAs. Adapted with permission from [48].

MS attribute	% of MS BLAs	MS attribute	% of MS BLAs
Amino acid sequence analysis	97.5	Sequence variants (Amino Acid Substitutions)	8.9
Molecular mass	92.4	Covalent dimers	7.6
Disulfide bonds	77.2	Methionine/Cysteine formylation	7.6
Glycosylation	70.9	Phosphorylation	5.1
Sequence variants (C-term)	64.6	Truncation	5.1
Sequence variants (N-term)	64.6	Acetylation	3.8
Deamidation	58.2	Aggregation	3.8
Oxidation	57.0	Folding/HOS	3.8
Size variants	27.8	Host cell proteins (HCPs)	3.8
Free thiols	25.3	Partial reduction	3.8
Glycation	22.8	PEGylation	3.8
Charge variants	19.0	Translucent particles	3.8
Other impurities	17.7	Zinc	3.8
Proteolysis/ Fragmentation	13.9	Glutathionylation	1.3
Succinimidation	12.7	Methylation	1.3
Isomerization	10.1	Norleucine incorporation	1.3
Other	10.1	Phosphogluconylation	1.3

- Sequence fidelity
- Structure, higher order structure
- Post-translational modifications
- Product and process-related impurities

Biosimilars and Comparative Analytical Assessments

- 100% usage of MS in approved 351(k) BLAs reviewed as of July 2019
- Used for peptide mapping, intact mass, subunit analysis, glycan profiling
- LC-MS, LC-MS/MS, MALDI-TOF, HDX-MS
- Ionization by ESI, LC-ESI, MALDI, nanoESI
- Amino acid sequence, molecular weight, disulfide bonds, PTMs, product- and process-related impurities, higher order structure

- https://www.fda.gov/drugs/biosimilars/biosimilar-product-information
- Namuswe F, CASSS MS 2019.

Early product development

- The core regulatory expectation is to demonstrate that the method is fit for the intended purpose
  - 21 CFR 211.165(e) and 211.194(a)(2)
- IND-enabling studies to characterize the primary sequence, HOS
- Extended characterization studies to identify variants, impurities related to product and process, other components of drug intermediate, substance and product
- Optimization of purification and production steps where advanced analytics can inform on the identity, criticality and robustness of key product and process attributes

As product development proceeds

- General method development and validation expectations are similar for MS-based methods
  - ICHQ2R(1) and FDA Guidance for Industry on analytical procedures and method validation for drugs and biologics
- Amount of information on method and suitability typically varies with the phase of development and intended purpose
- Setting meaningful specifications that reflect current process knowledge and clinically relevant product lots
  - ICHQ6B
  - FDA MAPP 5017.2 on Establishing Impurity Acceptance Criteria As Part of Specifications for NDAs, ANDAs, and BLAs Based on Clinical Relevance

As product development proceeds

- Lifecycle management
  - Method performance
  - Method changes and revalidation
  - Bridging studies during method replacement
  - Comparison to orthogonal methods
  - Method transfer
  - Reference standard
  - Comparability studies

Case study #1

MS for control of a process-related impurity

- A specific host cell protein, critical for the stability of the therapeutic protein.
- CQA is monitored at DS release and stability testing.
- MS provides relative quantitation, compared to reference.
- Specifications are wide but reflect current manufacturing and product knowledge

Sponsor is successfully monitoring this impurity while gaining additional knowledge and preparing for a manufacturing scale-up and comparability study.

Case study #2

MS for control of multiple attributes

- MAM proposed for DS/DP release and stability for multiple attributes/product-related impurities.
- Impurities and attributes are generated by different chemical/degradation/PTM pathways.
- However, insufficient information as provided to support replacing conventional methods with MAM.
- Additional studies were provided to compare with conventional methods with multiple lots and different samples. Extended characterization data was supportive.
- New Peak Detection – LOQ/LOD were determined, and function of new peak determined
- The stability indicating nature of the NPD was verified.
- Agency agreed with sponsors strategy to phase out conventional methods over time.

MS in the context of COVID-19 drug development

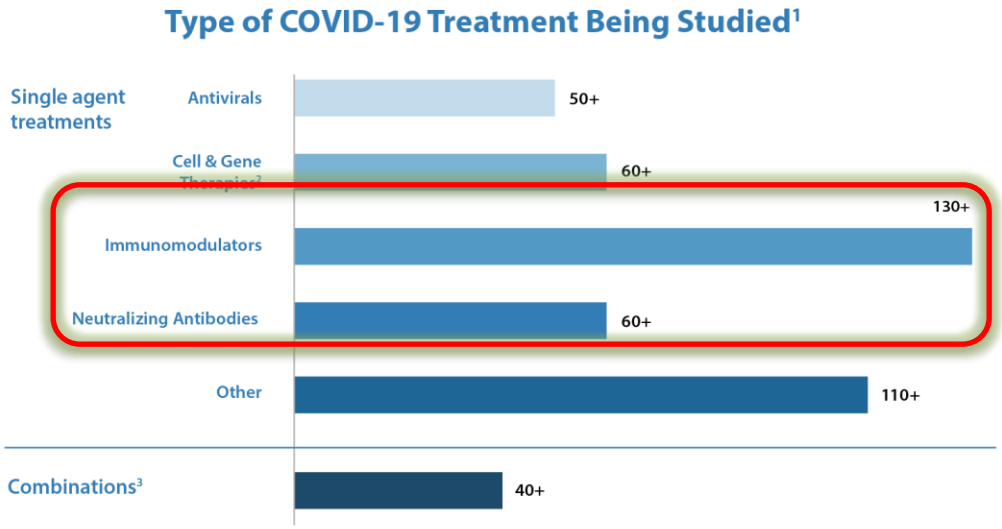
Coronavirus Treatment Acceleration Program (CTAP)

640+
Drug development programs in planning stages[1]

470+
Trials reviewed by FDA[2]

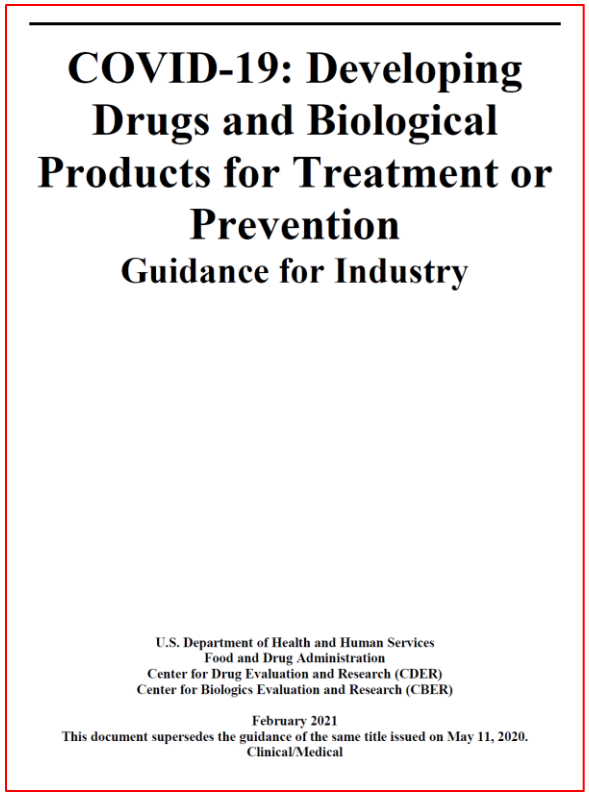
11
COVID-19 treatments currently authorized for Emergency Use[3]

1
Treatments currently approved by FDA for use in COVID-19



https://www.fda.gov/drugs/coronavirus-covid-19-drugs/coronavirus-treatment-acceleration-program-ctap

FDA Guidance on developing COVID-19 biological products

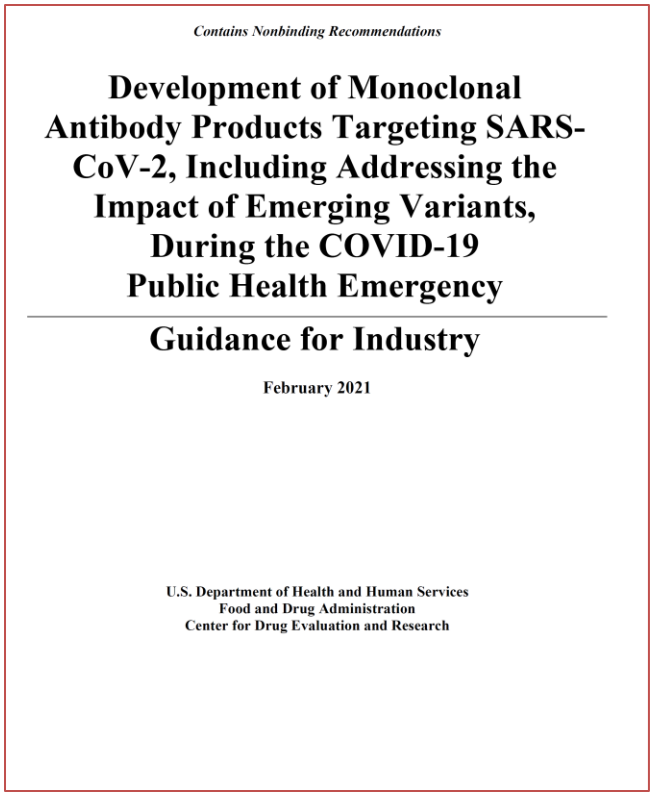


COVID-19: Developing Drugs and Biological Products for Treatment or Prevention

Guidance for Industry

U.S. Department of Health and Human Services
Food and Drug Administration
Center for Drug Evaluation and Research (CDER)
Center for Biologics Evaluation and Research (CBER)

February 2021
This document supersedes the guidance of the same title issued on May 11, 2020.
Clinical/Medical



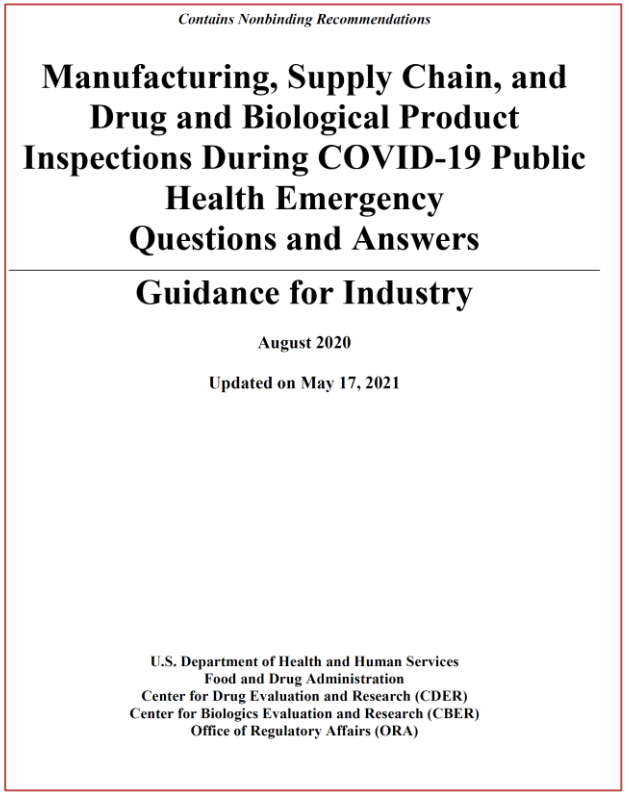
Contains Nonbinding Recommendations

Development of Monoclonal Antibody Products Targeting SARS-CoV-2, Including Addressing the Impact of Emerging Variants, During the COVID-19 Public Health Emergency

Guidance for Industry

February 2021

U.S. Department of Health and Human Services
Food and Drug Administration
Center for Drug Evaluation and Research



Contains Nonbinding Recommendations

Manufacturing, Supply Chain, and Drug and Biological Product Inspections During COVID-19 Public Health Emergency Questions and Answers

Guidance for Industry

August 2020

Updated on May 17, 2021

U.S. Department of Health and Human Services
Food and Drug Administration
Center for Drug Evaluation and Research (CDER)
Center for Biologics Evaluation and Research (CBER)
Office of Regulatory Affairs (ORA)

https://www.fda.gov/emergency-preparedness-and-response/coronavirus-disease-2019-covid-19/covid-19-related-guidance-documents-industry-fda-staff-and-other-stakeholders

Current opportunities and challenges with MS with respect to product quality

Challenges in the context of COVID-19

- Accelerated timelines
- Rapidly evolving molecular targets and drugs
- Characterization of impurities with unknown biological function
- Availability of key raw materials, reagents, parts, instruments and trained personnel at the right location.
- Rapid manufacturing scale-up to meet supply
  - General information - https://www.fda.gov/drugs/drug-supply-chain-integrity/fda-leads-effort-create-supply-chain-security-toolkit-medical-products
  - Guidance - https://www.fda.gov/regulatory-information/search-fda-guidance-documents/manufacturing-supply-chain-and-drug-and-biological-product-inspections-during-covid-19-public-health
- Site-specific, travel and geographical limitations

Opportunities for MS in the context of COVID-19

- The selection of analytical methods that have already been qualified or validated. This may include adapting potency assay[9] formats (including binding assays and/or neutralizing assays) with revised reagents that evaluate new variant protein(s) or pseudotyped virus/virus-like particles in the assay(s)

- The exploration of opportunities for less-experienced manufacturers to partner with those with more experience to leverage all available development tools

- The leveraging of related product quality data (e.g., formulation development) to support in-use stability and compatibility

- The use of prior development experience to anticipate the best dosage form, route of administration, and formulation (composition) selection

https://www.fda.gov/emergency-preparedness-and-response/coronavirus-disease-2019-covid-19/covid-19-related-guidance-documents-industry-fda-staff-and-other-stakeholders

Opportunities for MS in the context of COVID-19

- Selection of methods that have already been qualified or validated.
- The exploration of opportunities for less-experienced manufacturers to partner with those with more experience to leverage all available development tools
- The leveraging of related product quality data (*e.g.* formulation development, host cell protein profile) to support process development, in-use stability, compatibility
- The use of prior development experience to anticipate the best dosage form, route of administration and formulation selection

https://www.fda.gov/emergency-preparedness-and-response/coronavirus-disease-2019-covid-19/covid-19-related-guidance-documents-industry-fda-staff-and-other-stakeholders

Other applications of MS in COVID-19

Journal of proteome research
pubs.acs.org/jpr
Perspective

Open Science Resources for the Mass Spectrometry-Based Analysis of SARS-CoV-2

Wout Bittremieux,* Charlotte Adams, Kris Laukens, Pieter C. Dorrestein, and Nuno Bandeira

Cite This: *J. Proteome Res.* 2021, 20, 1464–1475 Read Online

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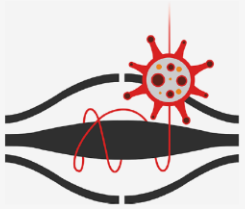
ABSTRACT: The SARS-CoV-2 virus is the causative agent of the 2020 pandemic leading to the COVID-19 respiratory disease. With many scientific and humanitarian efforts ongoing to develop diagnostic tests, vaccines, and treatments for COVID-19, and to prevent the spread of SARS-CoV-2, mass spectrometry research, including proteomics, is playing a role in determining the biology of this viral infection. Proteomics studies are starting to lead to an understanding of the roles of viral and host proteins during SARS-CoV-2 infection, their protein−protein interactions, and post-translational modifications. This is beginning to provide insights into potential therapeutic targets or diagnostic strategies that can be used to reduce the long-term burden of the pandemic. However, the extraordinary situation caused by the global pandemic is also highlighting the need to improve mass spectrometry data and workflow sharing. We therefore describe freely available data and computational resources that can facilitate and assist the mass spectrometry-based analysis of SARS-CoV-2. We exemplify this by reanalyzing a virus−host interactome data set to detect protein−protein



Journal of the American Society for Mass Spectrometry
pubs.acs.org/jasms
Account & Perspective

Mass Spectrometry Techniques in Emerging Pathogens Studies: COVID-19 Perspectives

Iqbal Mahmud and Timothy J. Garrett*

Cite This: *J. Am. Soc. Mass Spectrom.* 2020, 31, 2013–2024 Read Online

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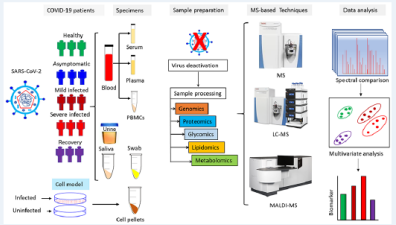
ABSTRACT: As corona virus disease 2019 (COVID-19) is a rapidly growing public health crisis across the world, our knowledge of meaningful diagnostic tests and treatment for severe acute respiratory syndrome coronavirus 2 (SARS-COV-2) is still evolving. This novel coronavirus disease COVID-19 can be diagnosed using RT-PCR, but inadequate access to reagents, equipment, and a nonspecific target has slowed disease detection and management. Precision medicine, individualized patient care, requires suitable diagnostics approaches to tackle the challenging aspects of viral outbreaks where many tests are needed in a rapid and deployable approach. Mass spectrometry (MS)-based technologies such as proteomics, glycomics, lipidomics, and metabolomics have been applied in disease outbreaks for identification of infectious disease



Rais et al. Clin Proteom (2021) 18:19
https://doi.org/10.1186/s12014-021-09325-x
Clinical Proteomics

REVIEW Open Access

Mass spectrometry-based proteomics in basic and translational research of SARS-CoV-2 coronavirus and its emerging mutants

Yasmine Rais, Zhiqiang Fu and Andrei P. Drabovich*

Trends in Analytical Chemistry 142 (2021) 116328
Contents lists available at ScienceDirect
Trends in Analytical Chemistry
journal homepage: www.elsevier.com/locate/trac

Mass spectrometry analytical responses to the SARS-CoV2 coronavirus in review

Justin H. Griffin, Kevin M. Downard*
Infectious Disease Responses Laboratory, Prince of Wales Clinical Research Sciences, Sydney, Australia

Other applications of MS in COVID-19

REGEN-COV: **LC-MRM-MS** (A liquid chromatography-multiple reaction monitoring-mass spectrometry) assay was used to measure the concentrations of REGENCOV in two-dose groups of ambulatory patients.

analytical chemistry

pubs.acs.org/ac Article

Liquid Chromatography-Multiple Reaction Monitoring-Mass Spectrometry Assay for Quantitative Measurement of Therapeutic Antibody Cocktail REGEN-COV Concentrations in COVID-19 Patient Serum

Xuefei Zhong,[‖] Shruti Nayak,[‖] Lili Guo, Shivkumar Raidas, Yunlong Zhao, Rachel Weiss, Matthew Andisik, Chinnasamy Elango, Giane Sumner, Susan C. Irvin, Michael A. Partridge, Hong Yan, Sook Yen E, Haibo Qiu, Yuan Mao,* Albert Torri, and Ning Li

Cite This: https://doi.org/10.1021/acs.analchem.1c01613 Read Online

Other applications of MS in COVID-19

In research and development of small molecule therapies for COVID, an affinity purification–mass spectrometry (MS) approach has been widely used to identify potential inhibitors of SARSCoV-2.

Tiwari, et al: (2020) *Drug Discovery Today*, 25(8):1535-44

Reviews • POST SCREEN

Discovering small-molecule therapeutics against SARS-CoV-2

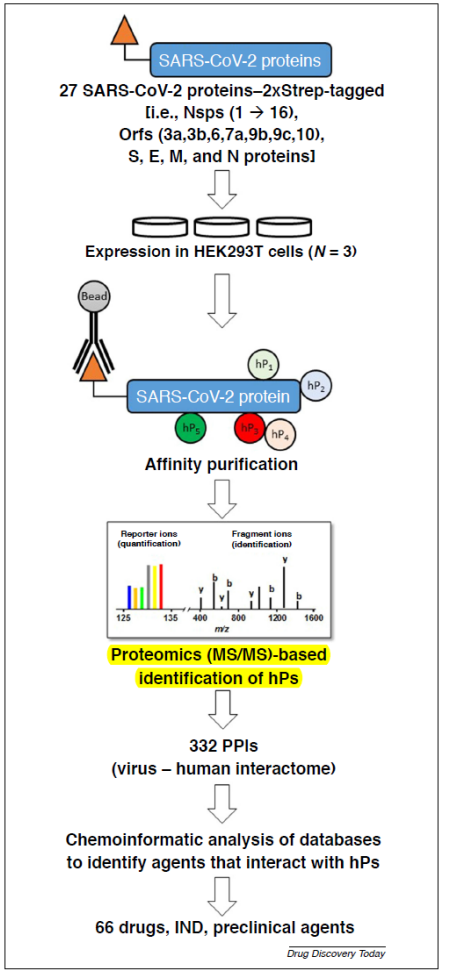
Vaibhav Tiwari[1,2], Jacob C. Beer[2], Nehru Viji Sankaranarayanan[3,4], Michelle Swanson-Mungerson[1,2] and Umesh R. Desai[3,4]

[1] Department of Microbiology and Immunology, College of Graduate Studies, Midwestern University Downers Grove, IL 6051, USA
[2] Chicago College of Osteopathic Medicine, Midwestern University, Downers Grove, IL 60515, USA
[3] Department of Medicinal Chemistry, School of Pharmacy, Virginia Commonwealth University, Richmond, VA 23298, USA
[4] Institute for Structural Biology, Drug Discovery and Development, Virginia Commonwealth University, Richmond, VA 2321, USA

Article *Gordon, et al:* (2020) *Nature*, 583:459–468

A SARS-CoV-2 protein interaction map reveals targets for drug repurposing

https://doi.org/10.1038/s41586-020-2286-9 A list of authors and affiliations appears at the end of the paper



Opportunities for MS in the context of COVID-19

Friday, April 17, 2020

NIH to launch public-private partnership to speed COVID-19 vaccine and treatment options

Health agencies, leading pharmaceutical companies to join forces to accelerate pandemic response.

The National Institutes of Health and the Foundation for the NIH (FNIH) are bringing together more than a dozen leading biopharmaceutical companies, the Health and Human Services Office of the Assistant Secretary for Preparedness and Response, the Centers for Disease Control and Prevention, the U.S. Food and Drug Administration and the European Medicines Agency to develop an international strategy for a coordinated research response to the COVID-19 pandemic. The planned Accelerating COVID-19 Therapeutic Interventions and Vaccines (ACTIV) partnership will develop a collaborative framework for prioritizing vaccine and drug candidates, streamlining clinical trials, coordinating regulatory processes and/or leveraging assets among all partners to rapidly respond to the COVID-19 and future pandemics. This is part of the whole-of-government, whole-of-America response the Administration has led to beat COVID-19.

"We need to bring the full power of the biomedical research enterprise to bear on this crisis," said NIH Director Francis S. Collins, M.D., Ph.D. "Now is the time to come together with unassailable objectivity to swiftly advance the development of the most promising vaccine and therapeutic candidates that can help end the COVID-19 global pandemic."

Coordinated by the FNIH, ACTIV government and industry partners will

Participating Organizations

Government

- National Institutes of Health
- HHS Office of the Assistant Secretary for Preparedness and Response
- U.S. Food and Drug Administration
- Centers for Disease Control and Prevention
- European Medicines Agency

Non-Profit

- Foundation for the National Institutes of Health

Industry

- AbbVie
- Amgen

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Multimedia

NIAID's Novel Coronavirus 19 Flickr album

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https://www.fda.gov/drugs/coronavirus-covid-19-drugs/coronavirus-treatment-acceleration-program-ctap#dashboard

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