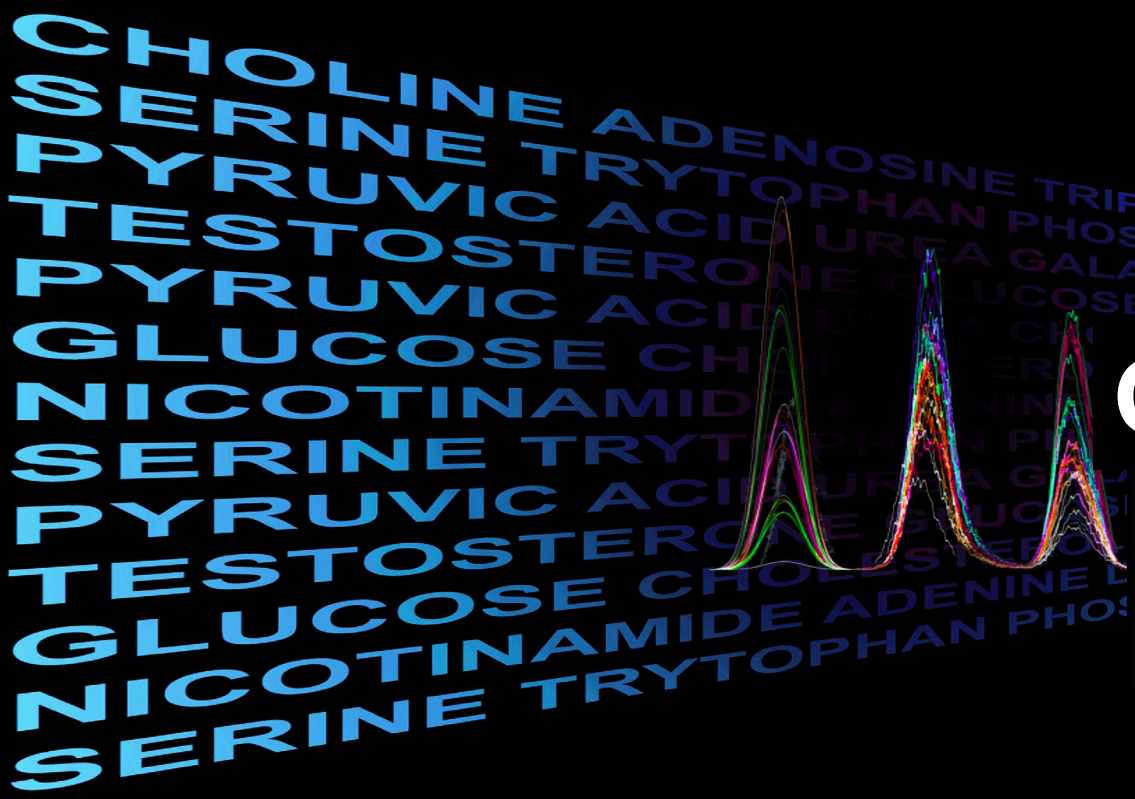
METLIN Ion Mobility (downloadable)

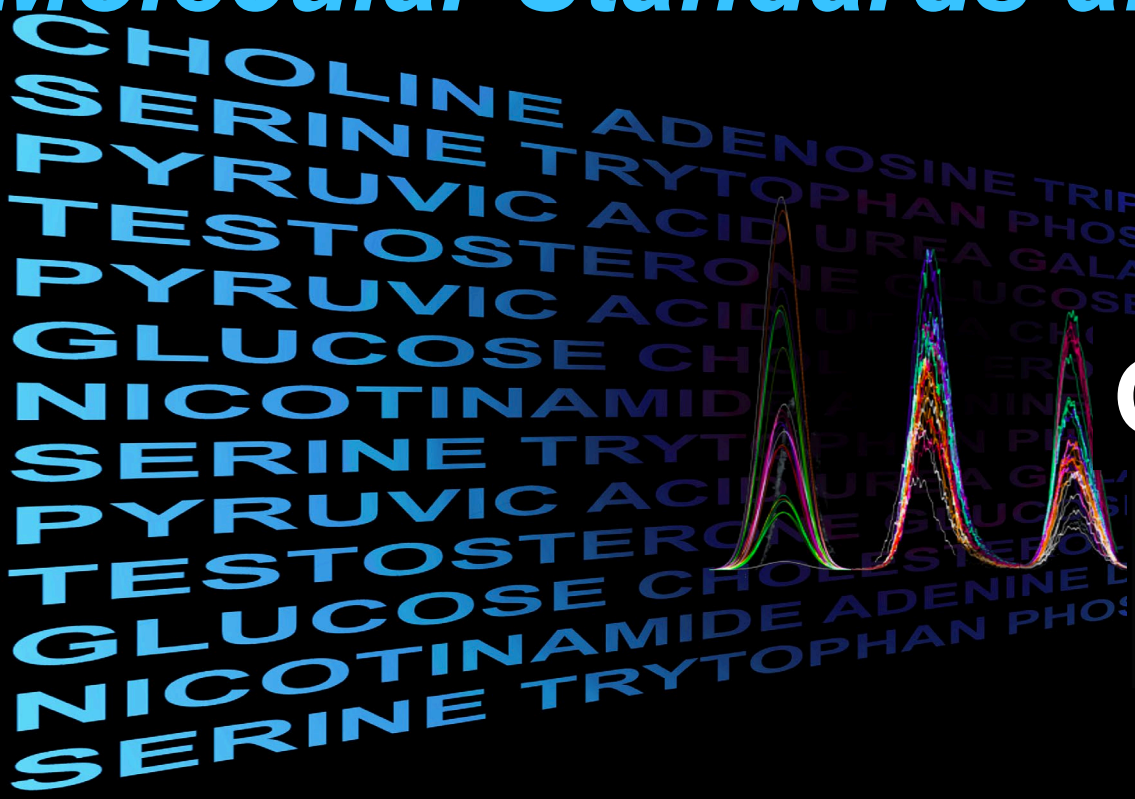


Gary Siuzdak

Scripps Research

METLIN Ion Mobility (downloadable)

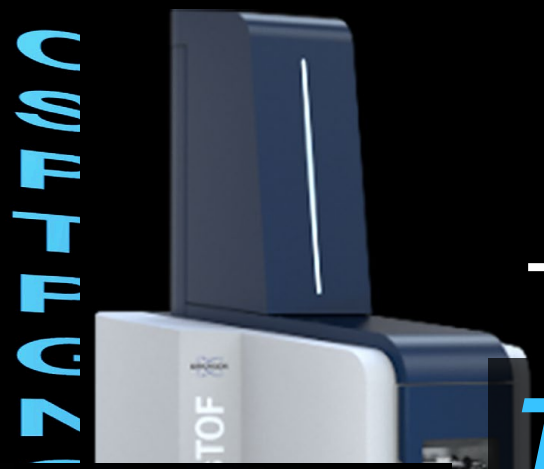
How to Analyze a Million Molecular Standards and Stay Sane

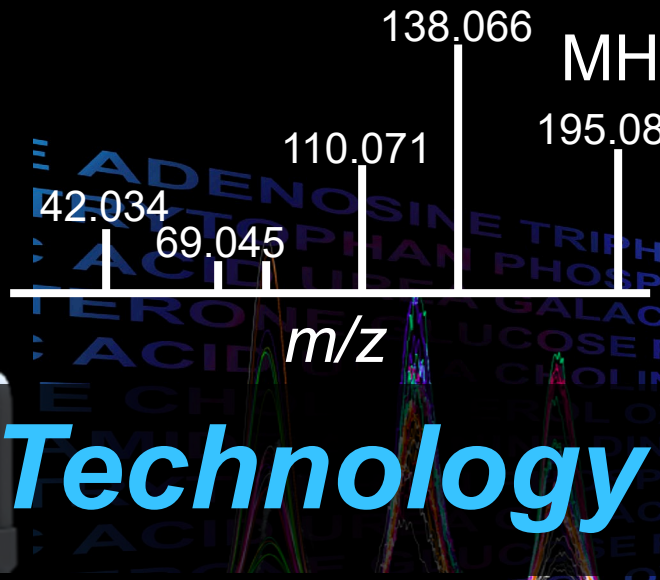


Gary Siuzdak

Scripps Research

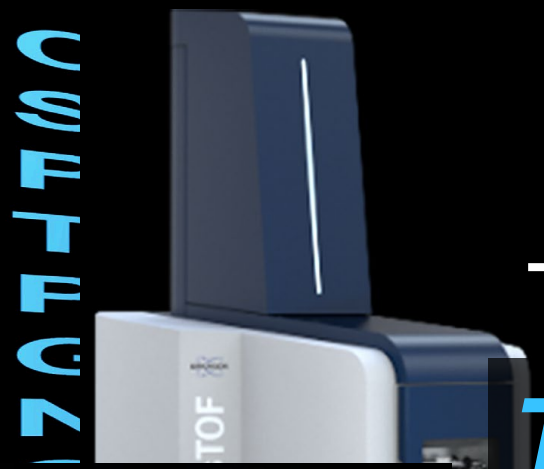
CHOLINE ADENOSINE TRIPHOSPHATE CHOLESTEROL TESTOSTERONE GLUCOSE
SERINE TRYTOPHAN PHOSPHOCHOLINE ACYLCARNITINE THREONINE GLYCEROL
PYRUVIC ACID UREA GALACTOSE CHOLINE ADENOSINE CHOLINE MALIC ACID
TESTOSTERONE GLUCOSE PHOSPHATE CHOLESTEROL OXALOSUCCINIC ACID
PYRUVIC ACID CHOLINE ADENOSINE CHOLINE LACTIC ACID KETOGLUTARATE
GLUCOSE CHOLESTEROL OXALOSUCCINIC ACID GALACTOSE GLYCEROL FUMARATE
NICOTINAMIDE ADENINE DINUCLEOTIDE OXALOSUCCINIC ACID GALACTOSE GLYCEROL
SERINE TRYTOPHAN PHOSPHOCHOLINE ACYLCARNITINE THREONINE GLYCEROL
PYRUVIC ACID UREA GALACTOSE CHOLINE ADENOSINE CHOLINE MALIC ACID
TESTOSTERONE GLUCOSE PHOSPHATE CHOLESTEROL OXALOSUCCINIC ACID
GLUCOSE CHOLESTEROL OXALOSUCCINIC ACID GALACTOSE GLYCEROL FUMARATE
NICOTINAMIDE ADENINE DINUCLEOTIDE OXALOSUCCINIC ACID GALACTOSE GLYCEROL
SERINE TRYTOPHAN PHOSPHOCHOLINE ACYLCARNITINE THREONINE GLYCEROL

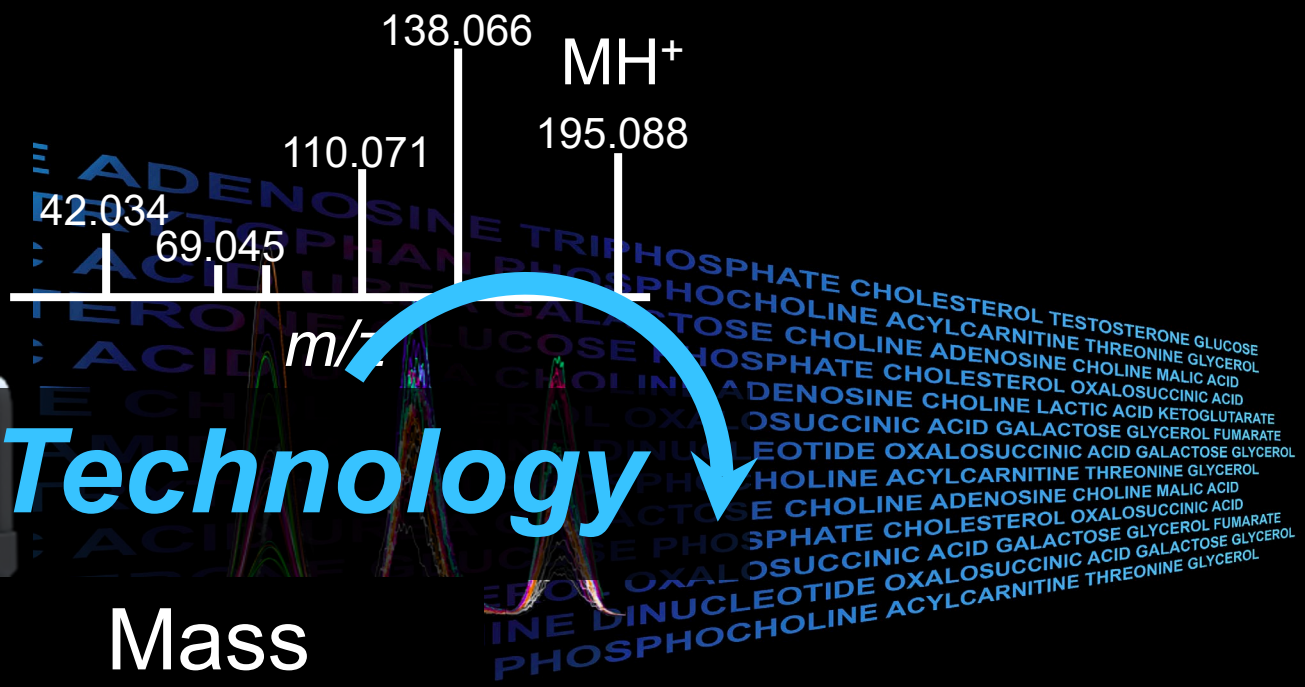




Technology

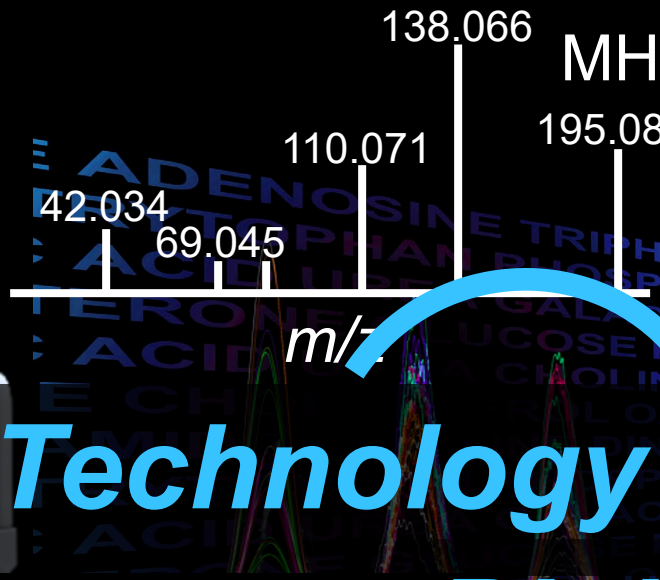
Mass
Spectrometry
&
Ion Mobility





Technology

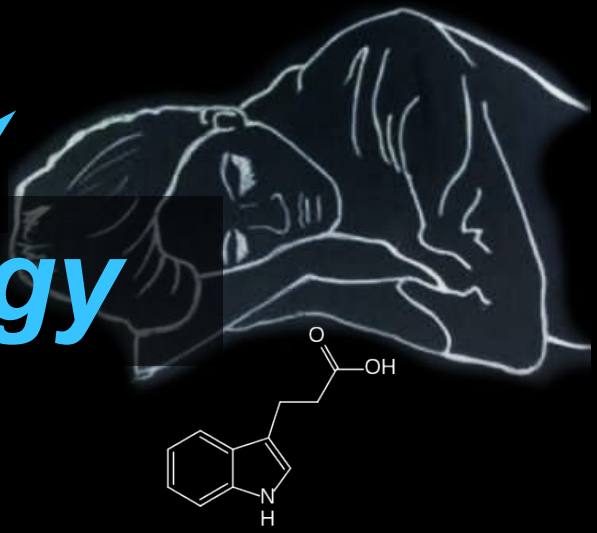
Mass
Spectrometry
&
Ion Mobility

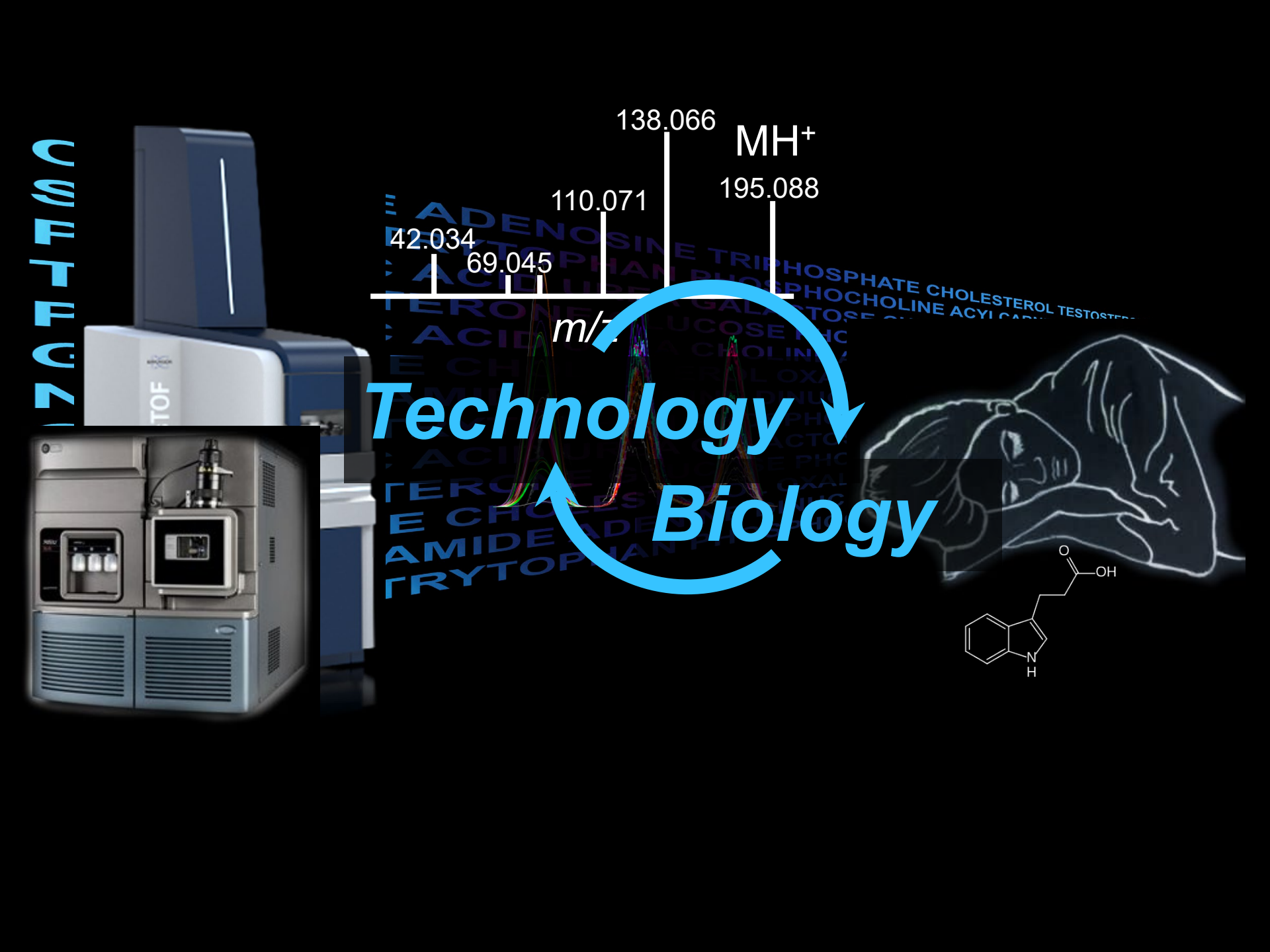


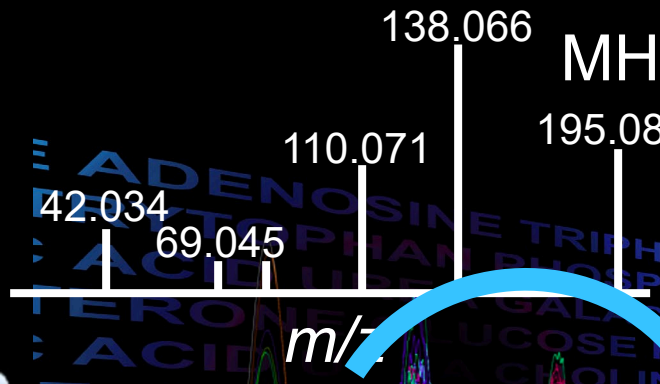
Technology

Biology

Mass Spectrometry & Ion Mobility

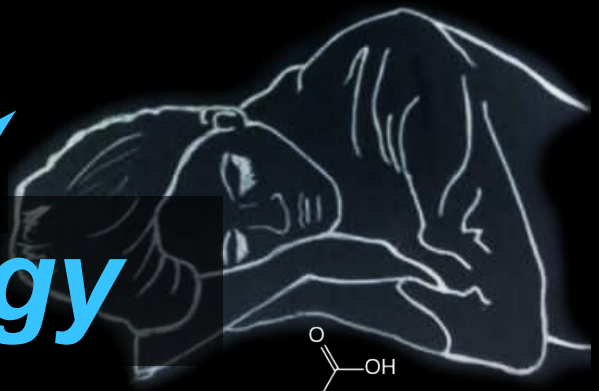


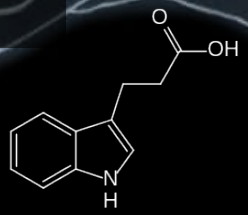

138.066
MH+
195.088
110.071
42.034
69.045
m/z
Technology
Biology
OH
O
N
H



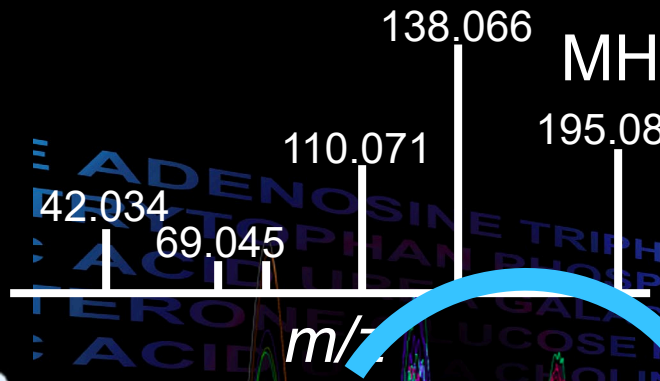
Technology

Biology



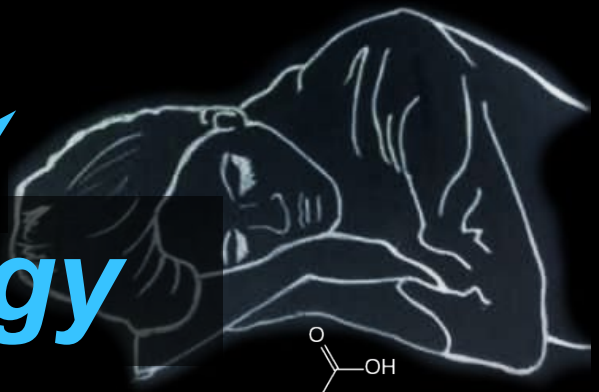


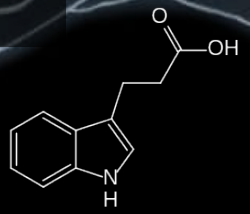
PNAS	*1994*
Science	*1995*
Ther. Drug Dis.	*2005*
Nature Biotech.	*2012*
Nature Methods	*2020*
Cell Metabolism	*2022*
J. Amer. Soc. MS	*2022*



Technology

Biology





PNAS	*1994*
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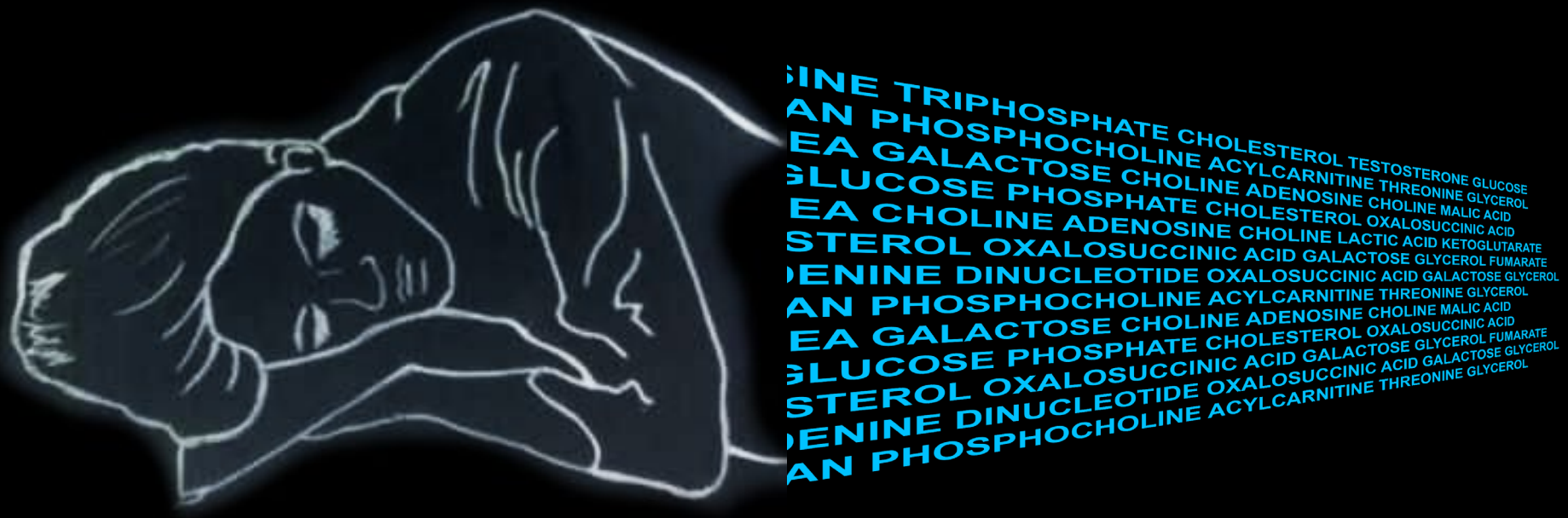
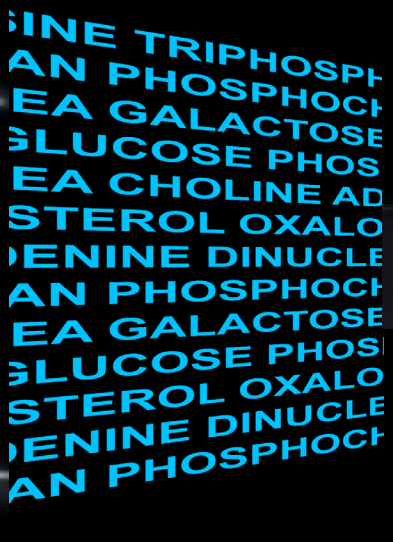
Nature Biotech.	*2012*
Nature Methods	*2020*
Cell Metabolism	*2022*
J. Amer. Soc. MS	*2022*

One Simple Motivation



Sleep

PNAS 1994, Science 1995, & Anal. Chem. 2006

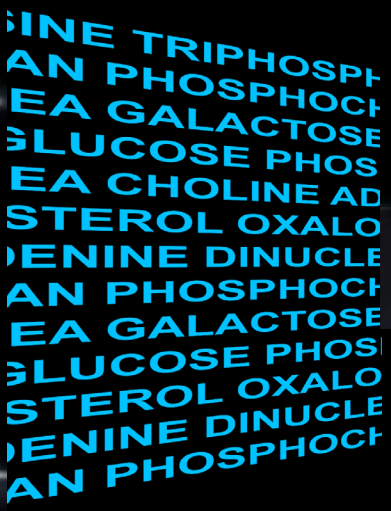


Richard Lerner

Sleep

PNAS 1994, Science 1995, & Anal. Chem. 2006

XCMSOnline.scripps.edu



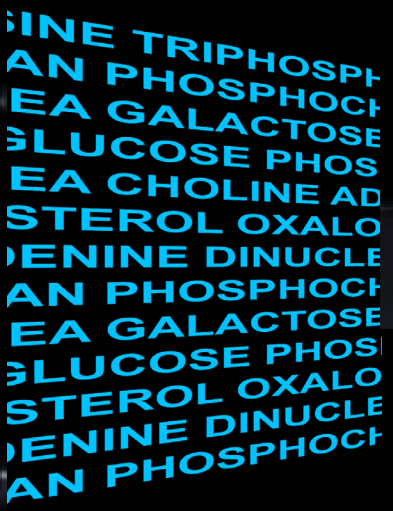
Richard Lerner

Sleep

PNAS 1994, Science 1995, & Anal. Chem. 2006

XCMSOnline.scripps.edu

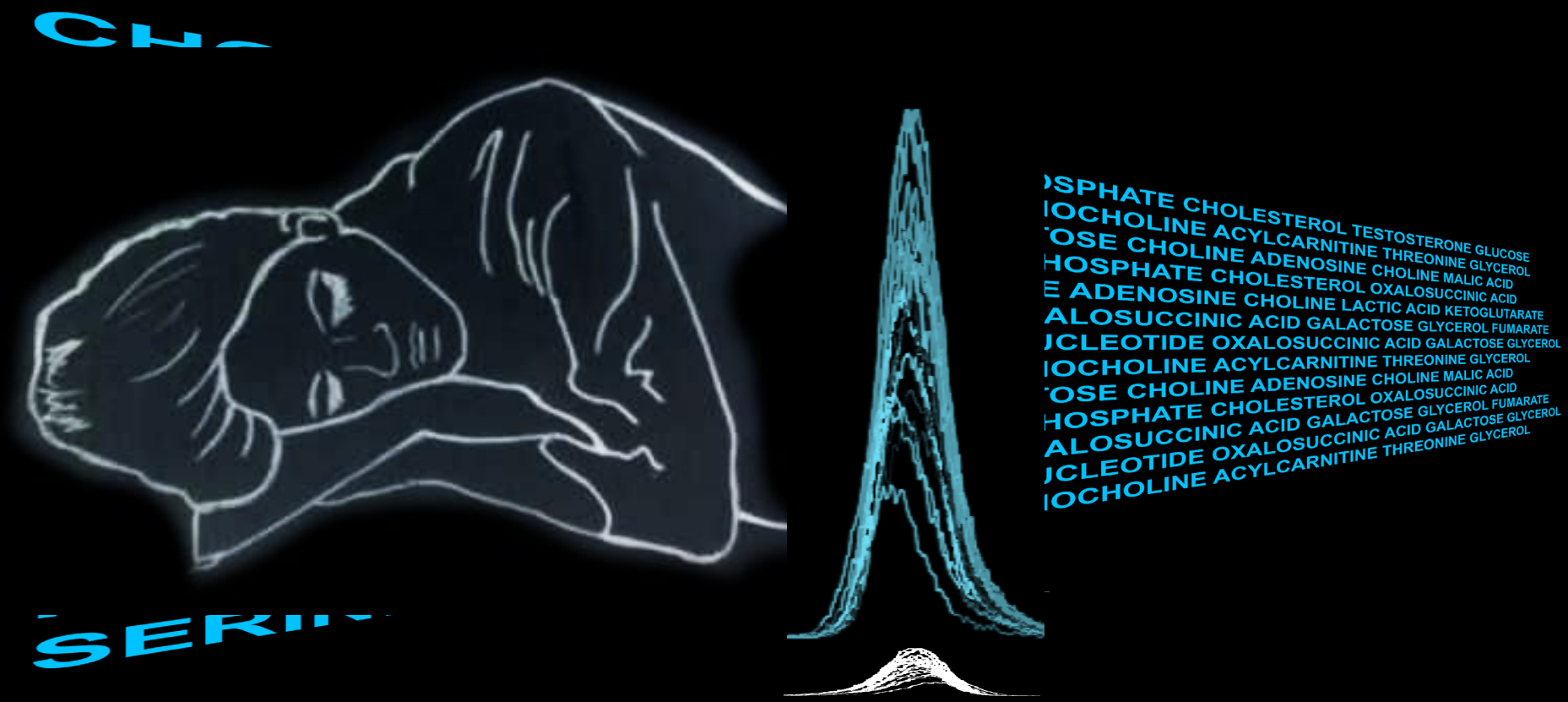
Original approach to process and statistically assess metabolomics data



Richard Lerner

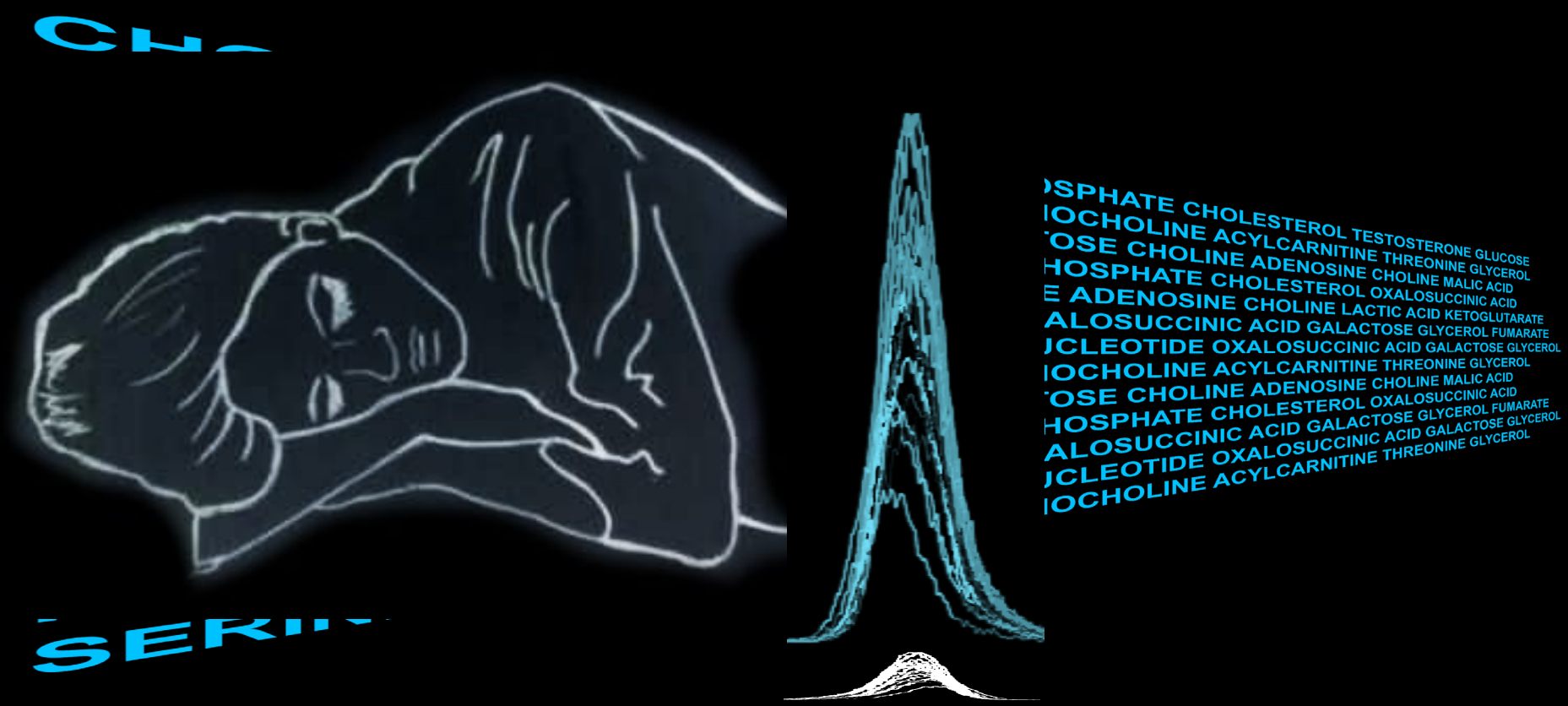
Sleep

PNAS 1994, Science 1995, & Anal. Chem. 2006



PNAS 1994, Science 1995

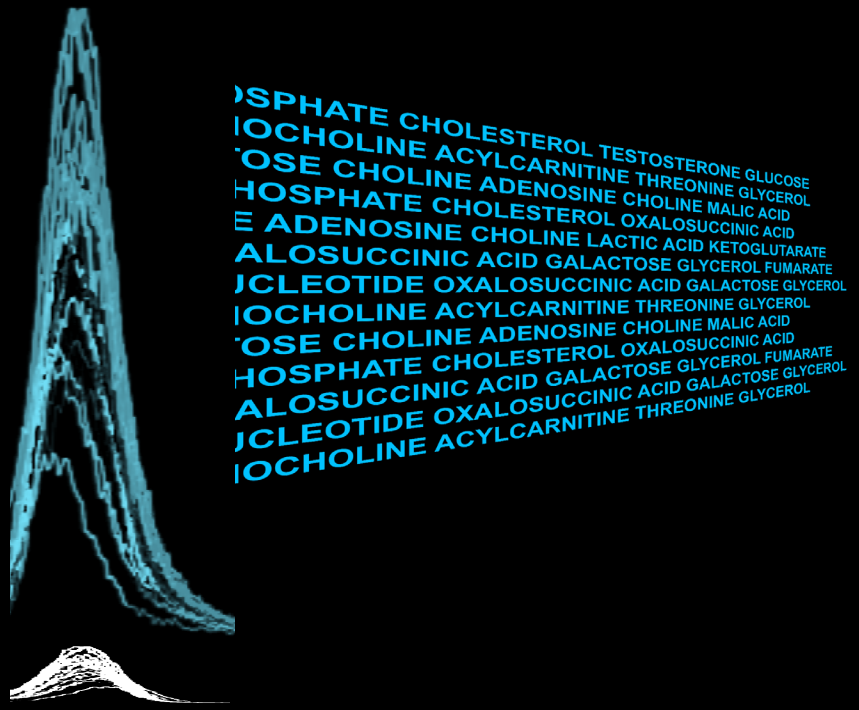
Metabolite Identification



PNAS 1994, Science 1995

Metabolite Identification

- Accurate *m/z*
- Tandem MS/MS
- NL Interpretation
- Isotope Pattern
- Preparative LC
- Synthesis



PNAS 1994, Science 1995

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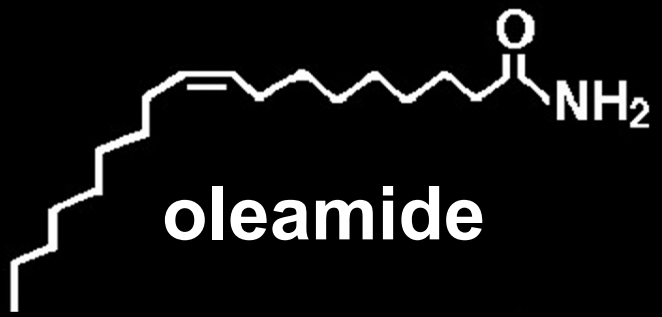
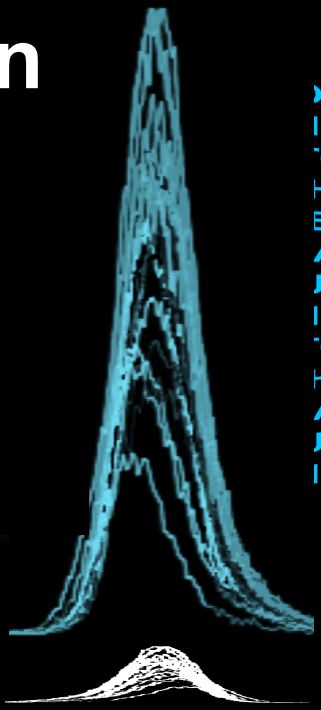
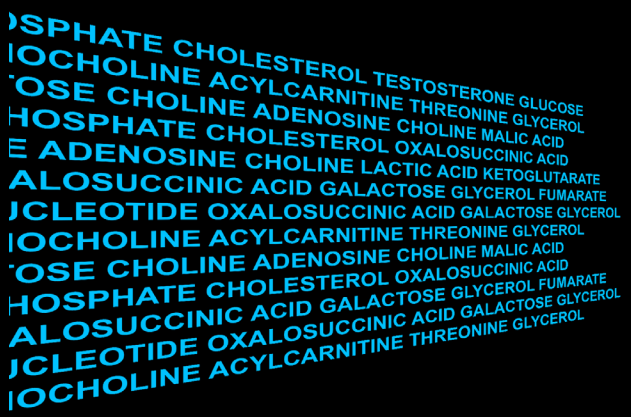


oleamide





PNAS 1994, Science 1995

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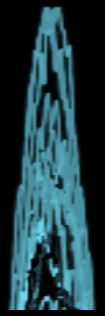

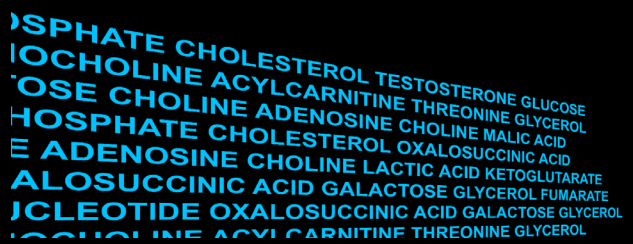

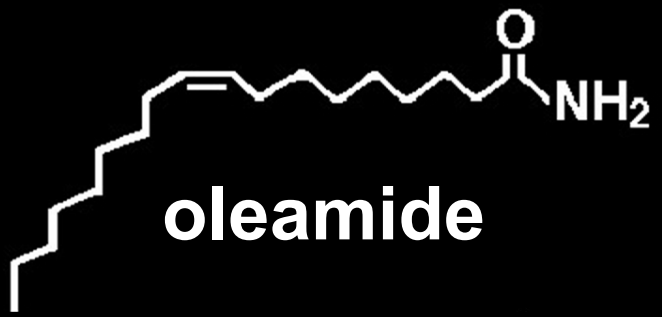

oleamide

Induces a sleep-like state

PNAS 1994, Science 1995

Metabolite Identification

- Accurate *m/z*
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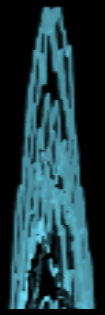
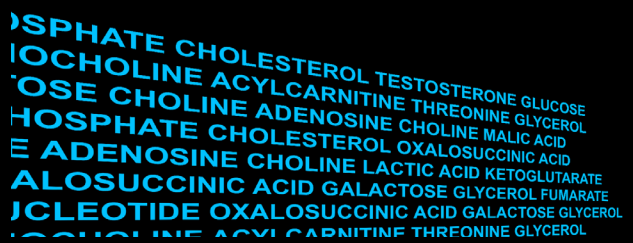
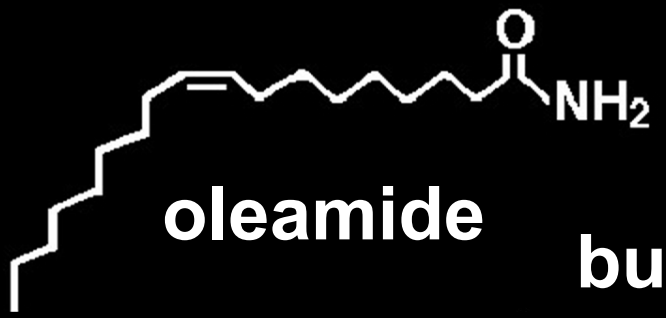
- Isotope Pattern
- Preparative LC
- Synthesis







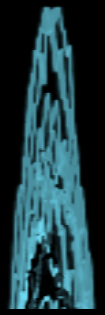
oleamide

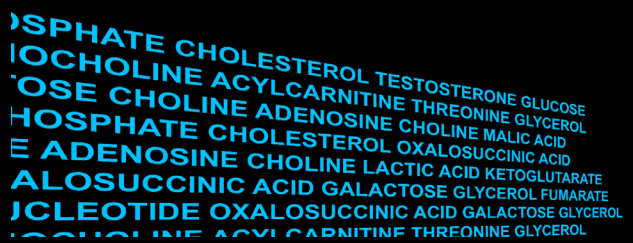
Induces a sleep-like state

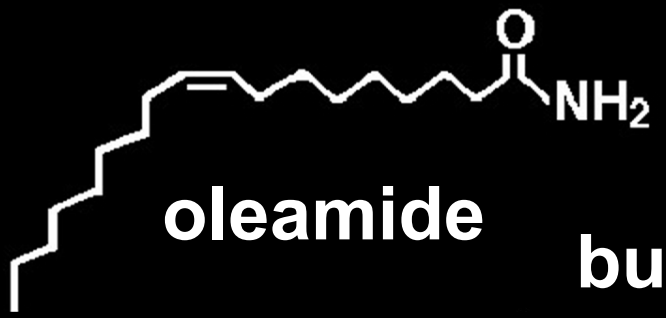
but it took 8 months to identify

Metabolite Identification

- Accurate *m/z*
- Tandem MS/MS
- NL Interpretation
- Isotope Pattern
- Preparative LC
- Synthesis







oleamide

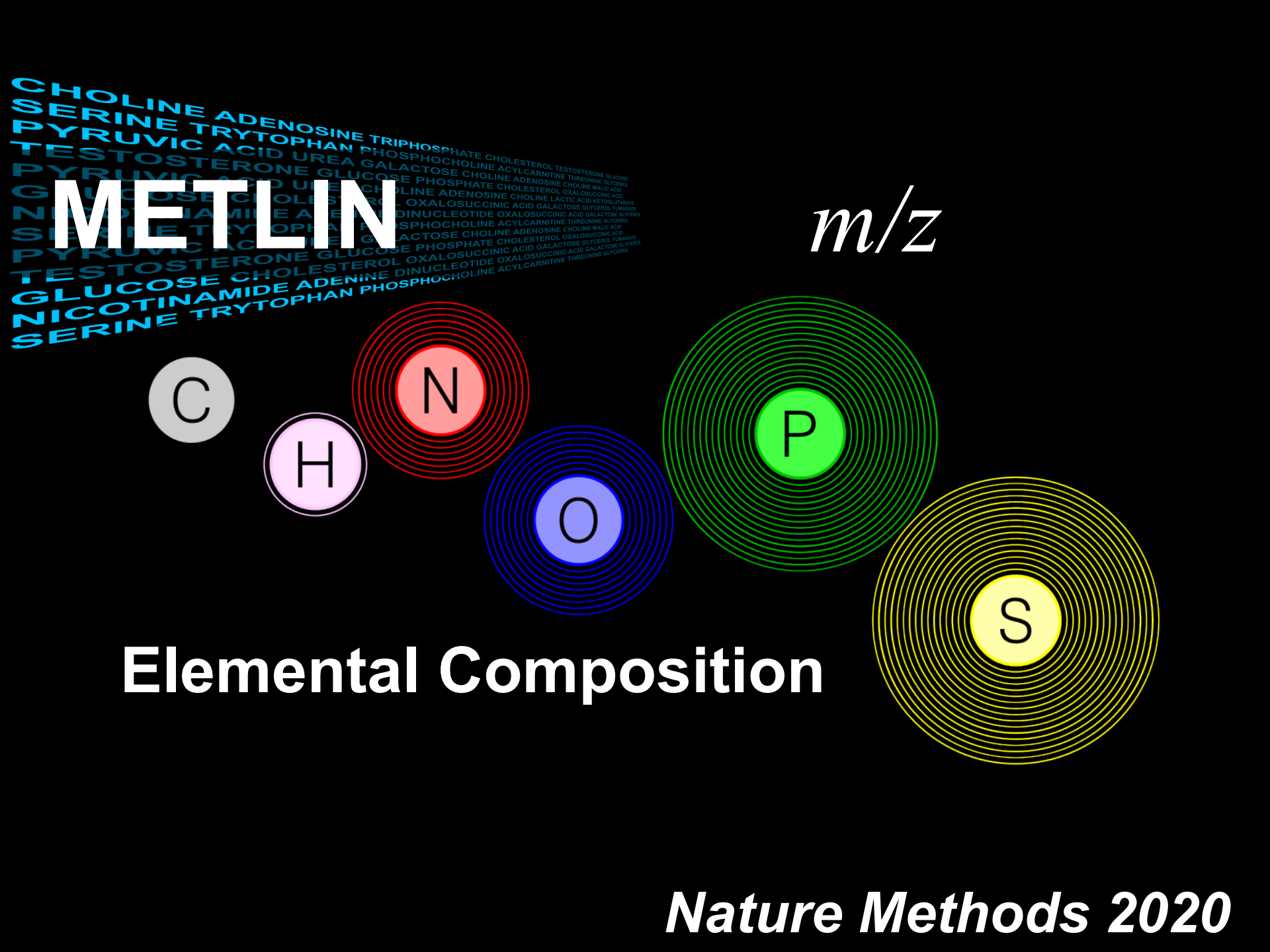
Induces a sleep-like state

but it took 8 months to identify

A better way to identify molecules is needed

Ther. Drug Dev. 2005

Nature Methods 2020


METLIN
m/z
C
H
N
O
P
S
Elemental Composition
Nature Methods 2020

m/z

$Frag_{int}$ vs *m/z*

Nature Methods 2020

METLIN

m/z

METLIN-MS^2

$Frag_{int}$ vs *m/z*

Oleamide

MS^2

m/z

$\mathrm{Frag}_{\mathrm{int}}$ vs m/z

$\mathrm{NL}_{\mathrm{int}}$ vs $\Delta m/z$

JASMS 2022

Nature Methods 2020

METLIN

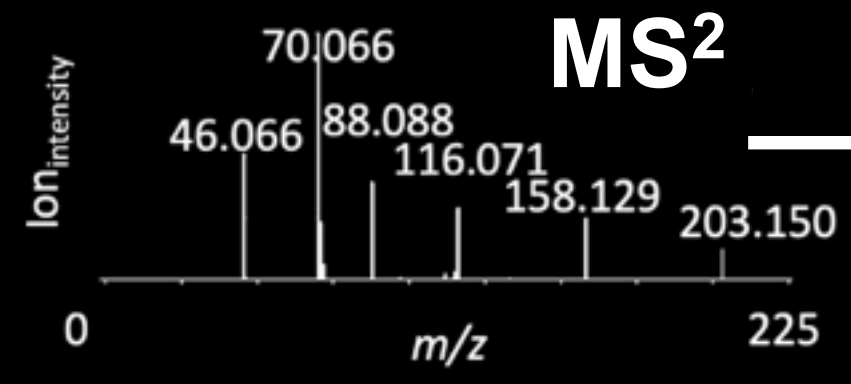
m/z

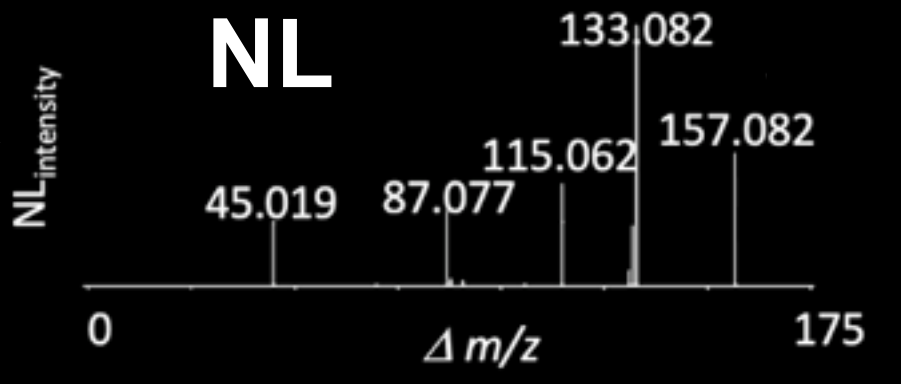
METLIN-MS^2

Frag_{int} vs m/z

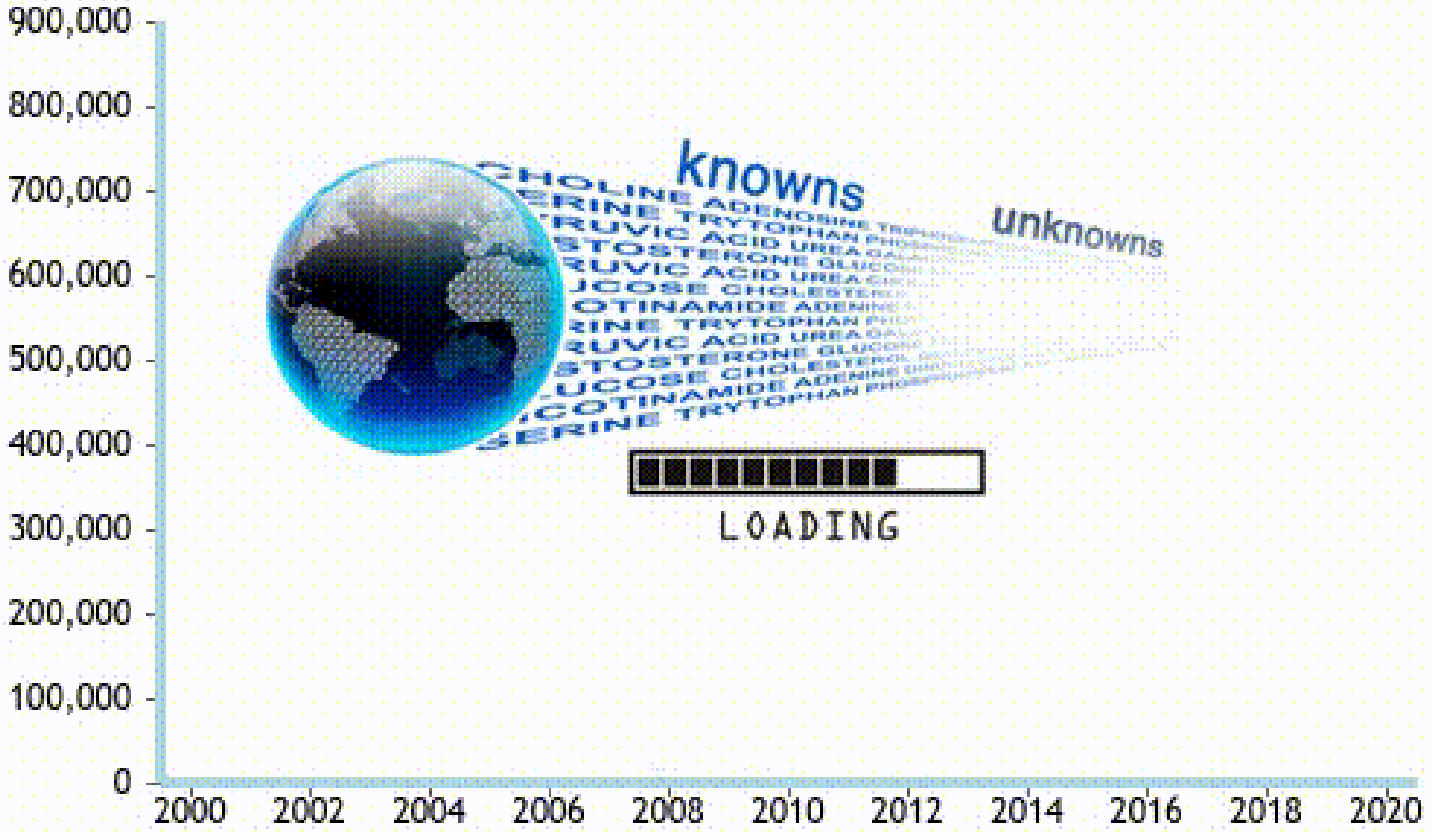
METLIN-NL

NL_{int} vs $\Delta m/z$

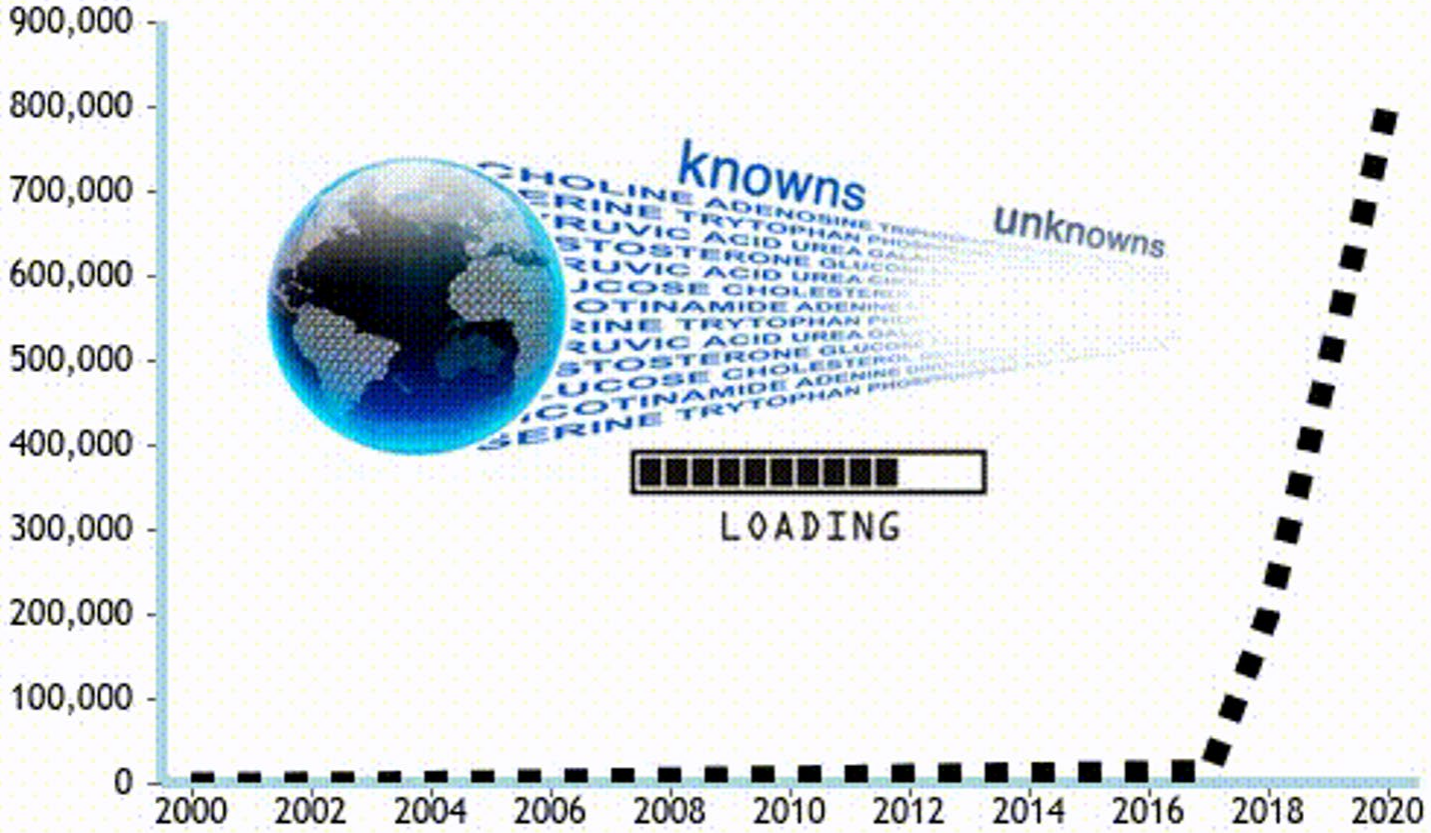




METLIN MS/MS Data

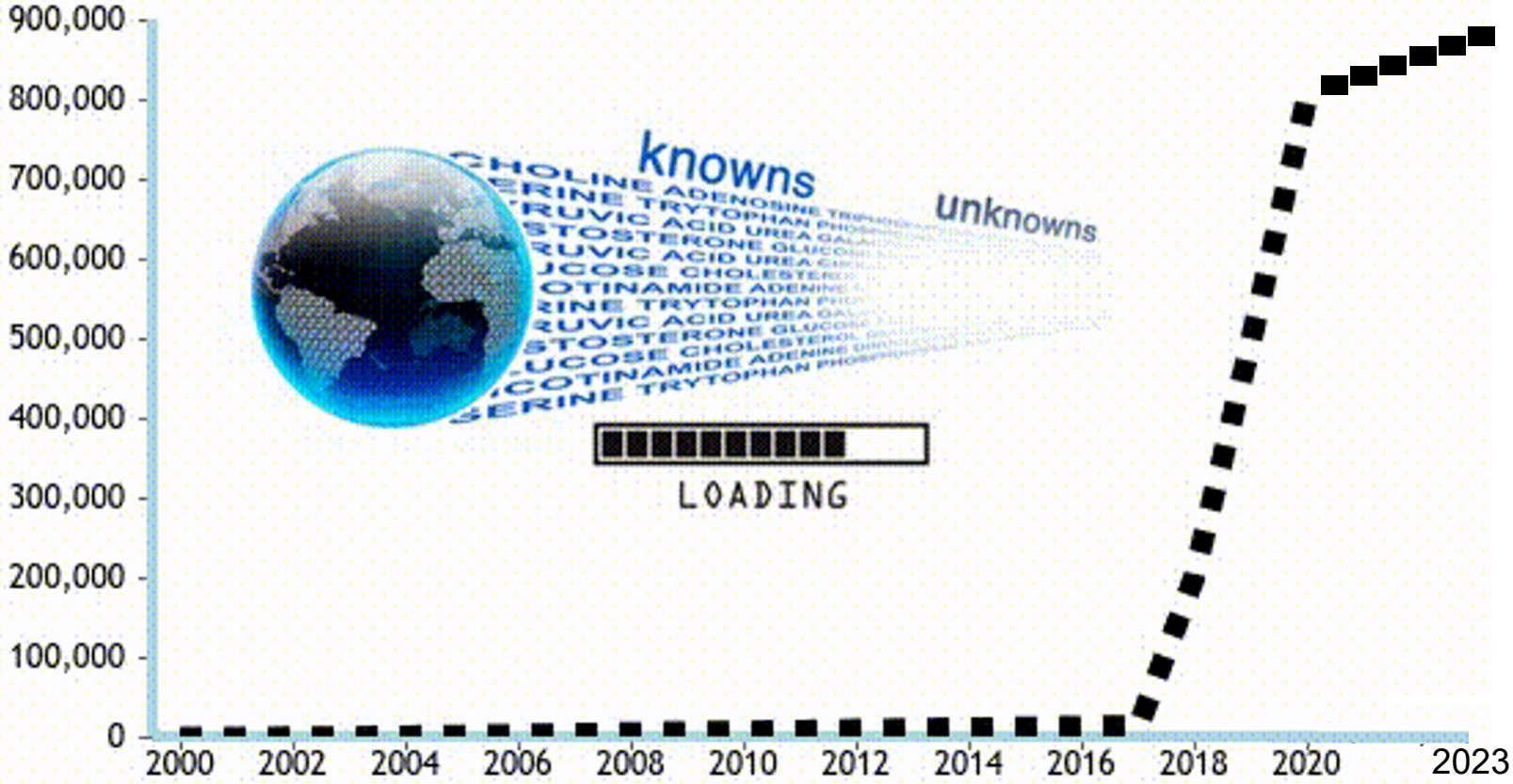


METLIN MS/MS Data



850,000

METLIN MS/MS Data



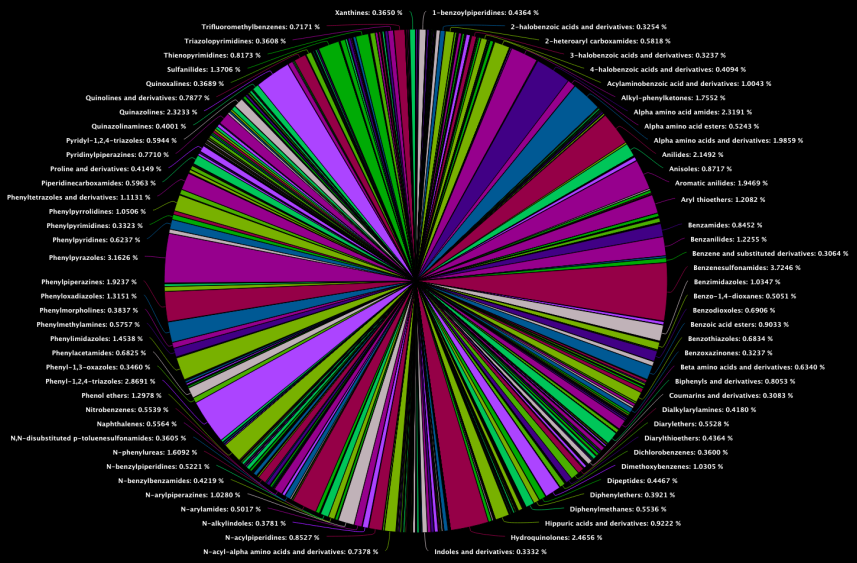
900,000

METLIN-MS^2 & NL Statistics

Nature Methods 2020
Cell Metabolism 2022

METLIN-MS^2 & NL Statistics

- 1.17 Million Standards Analyzed



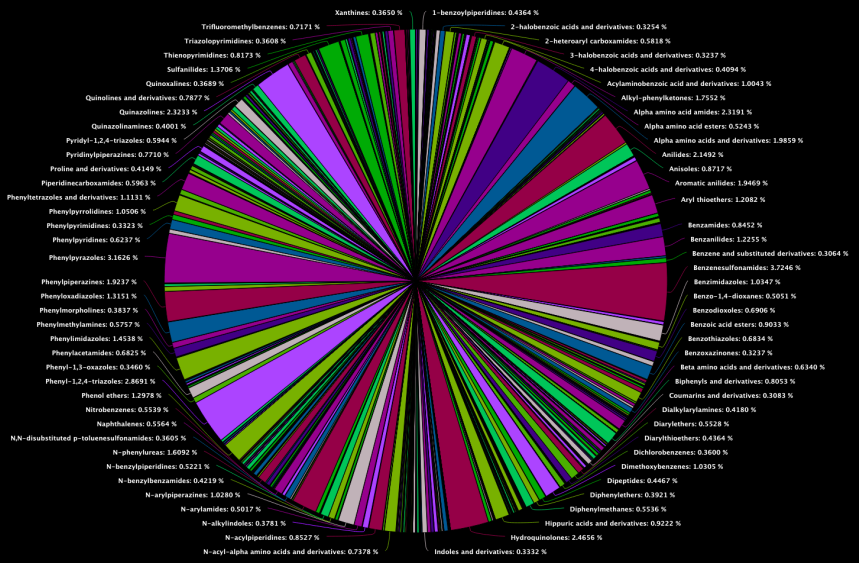
350 chemical classes

Nature Methods 2020

Cell Metabolism 2022

METLIN-MS^2 & NL Statistics

- 1.17 Million Standards Analyzed
- 77% Success: 23% failed filter
- 900K Standards each with MS/MS



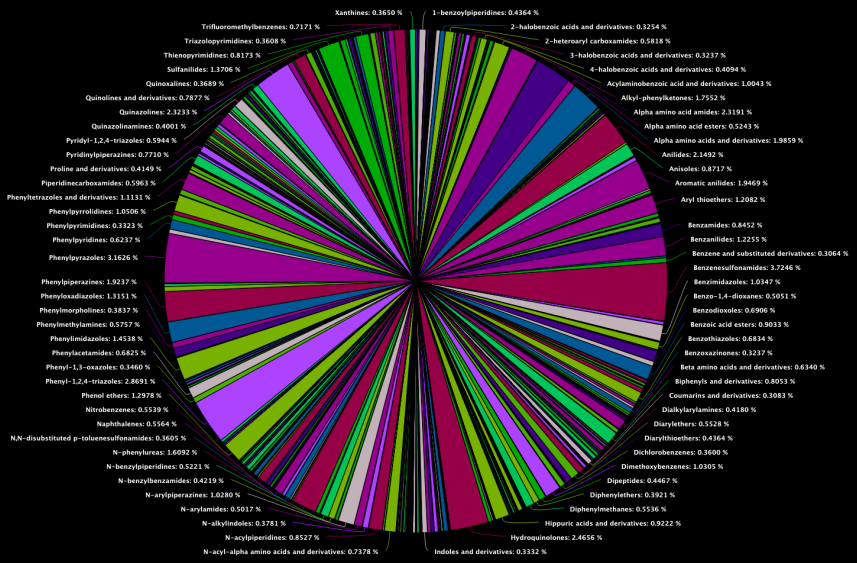
350 chemical classes

Nature Methods 2020

Cell Metabolism 2022

METLIN-MS^2 & NL Statistics

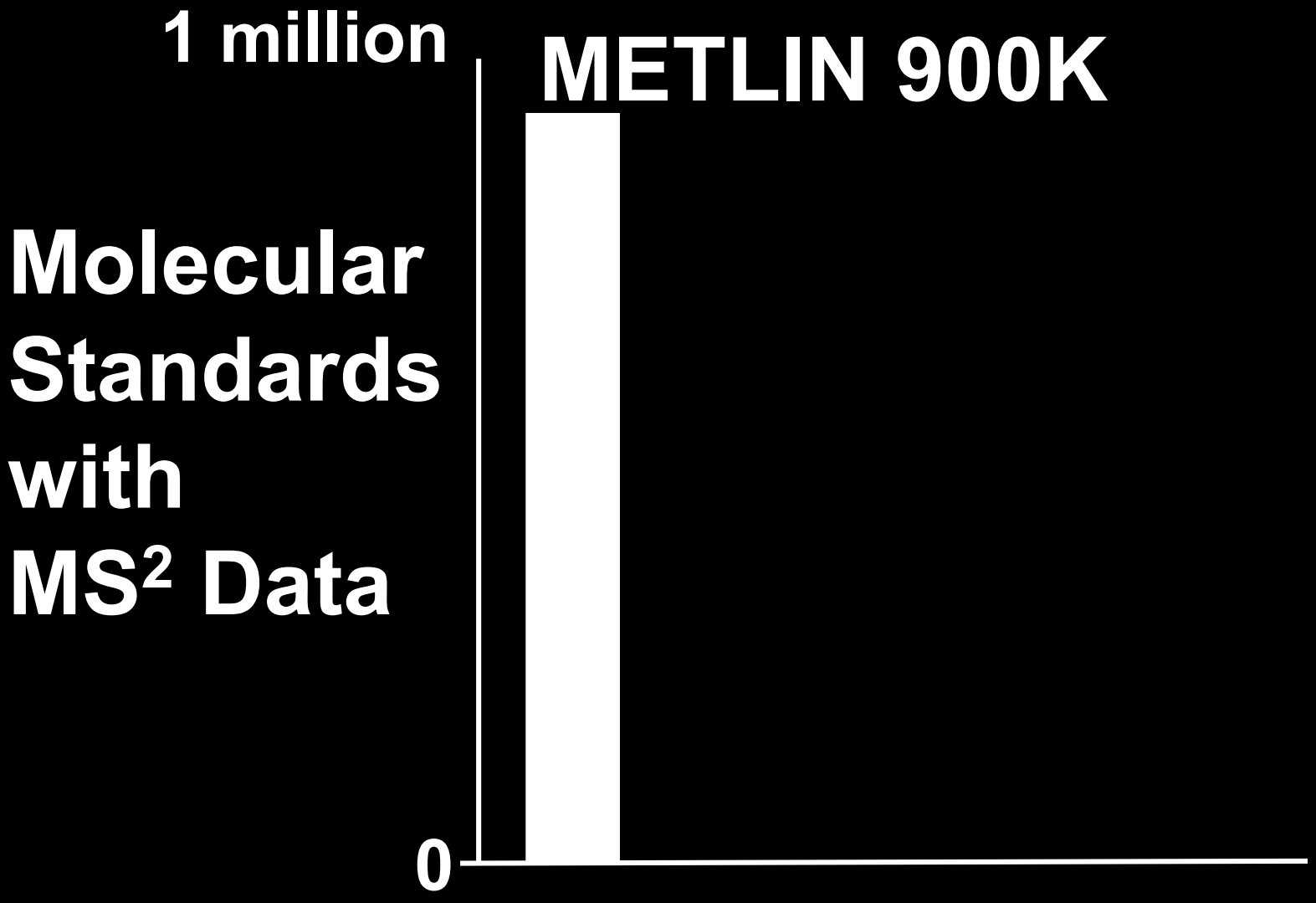
- 1.17 Million Standards Analyzed
- 77% Success: 23% failed filter
- 900K Standards each with MS/MS acquired at 4 collision energies in pos & neg ionization modes



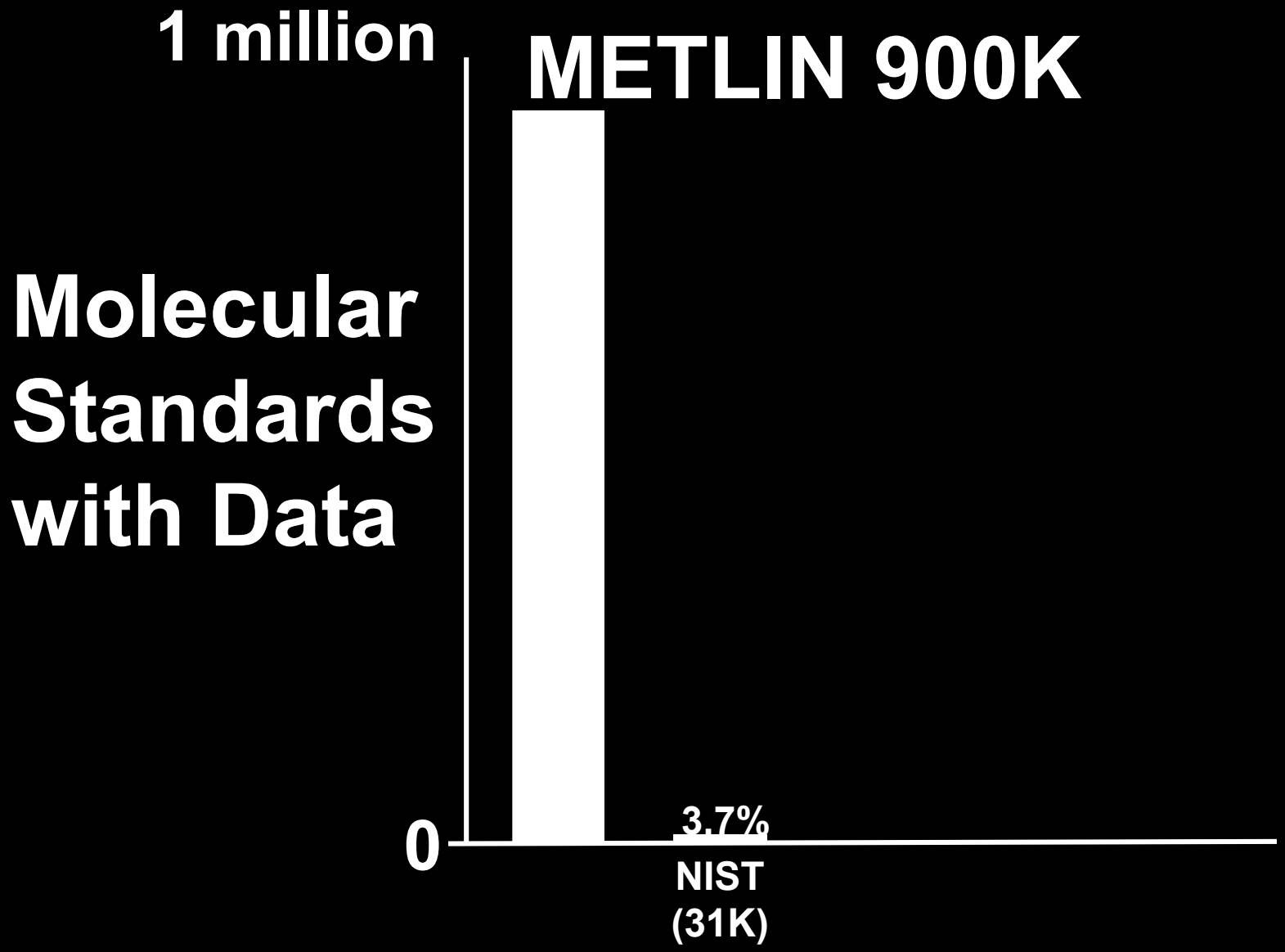
350 chemical classes

Nature Methods 2020
Cell Metabolism 2022

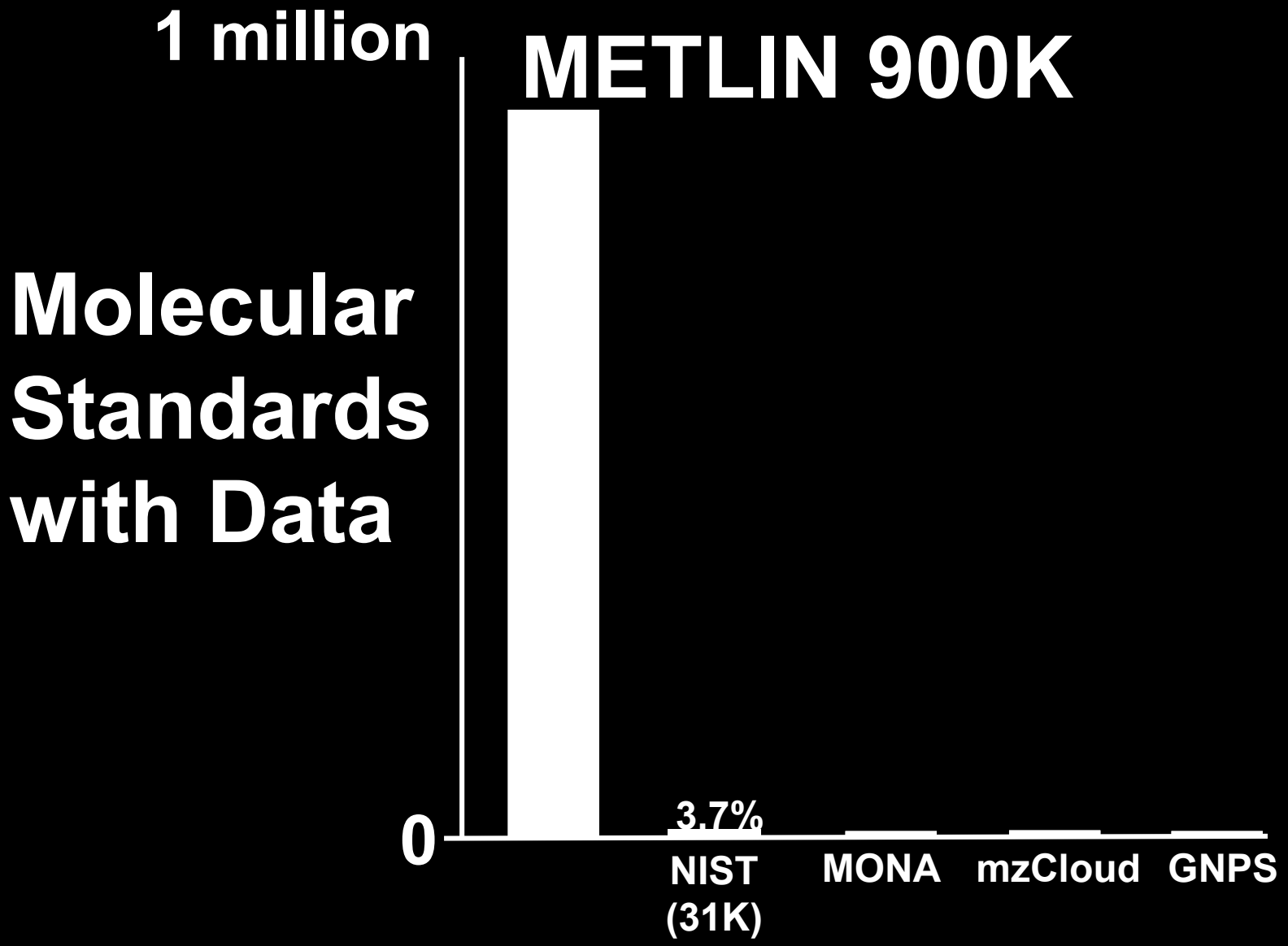
METLIN-MS^2 & NL Statistics



METLIN-MS^2 & NL Statistics



METLIN-MS^2 & NL Statistics

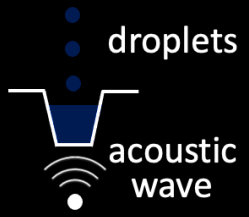



METLIN
m/z
METLIN-MS2
Frag int vs m/z
METLIN-NL
NL int vs Δm/z

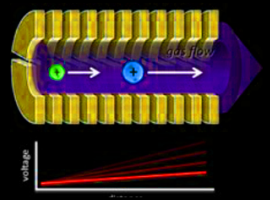

METLIN
m/z
METLIN-MS2
Frag int vs m/z
METLIN-NL
NL int vs Δm/z
METLIN-IMS
CCS values

METLIN - Ion Mobility Spectrometry

Step 1: Sample Plate Preparation

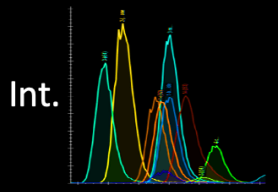


Step 2: Collect LC-IMS Data



TIMS and TWIMS

Step 3: Generate Raw IMS Data



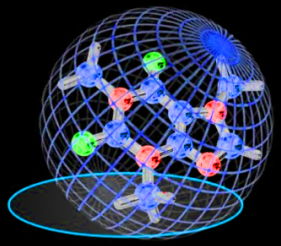
RT

Step 4: Skyline IMS Data Input to CCS Values

Skyline

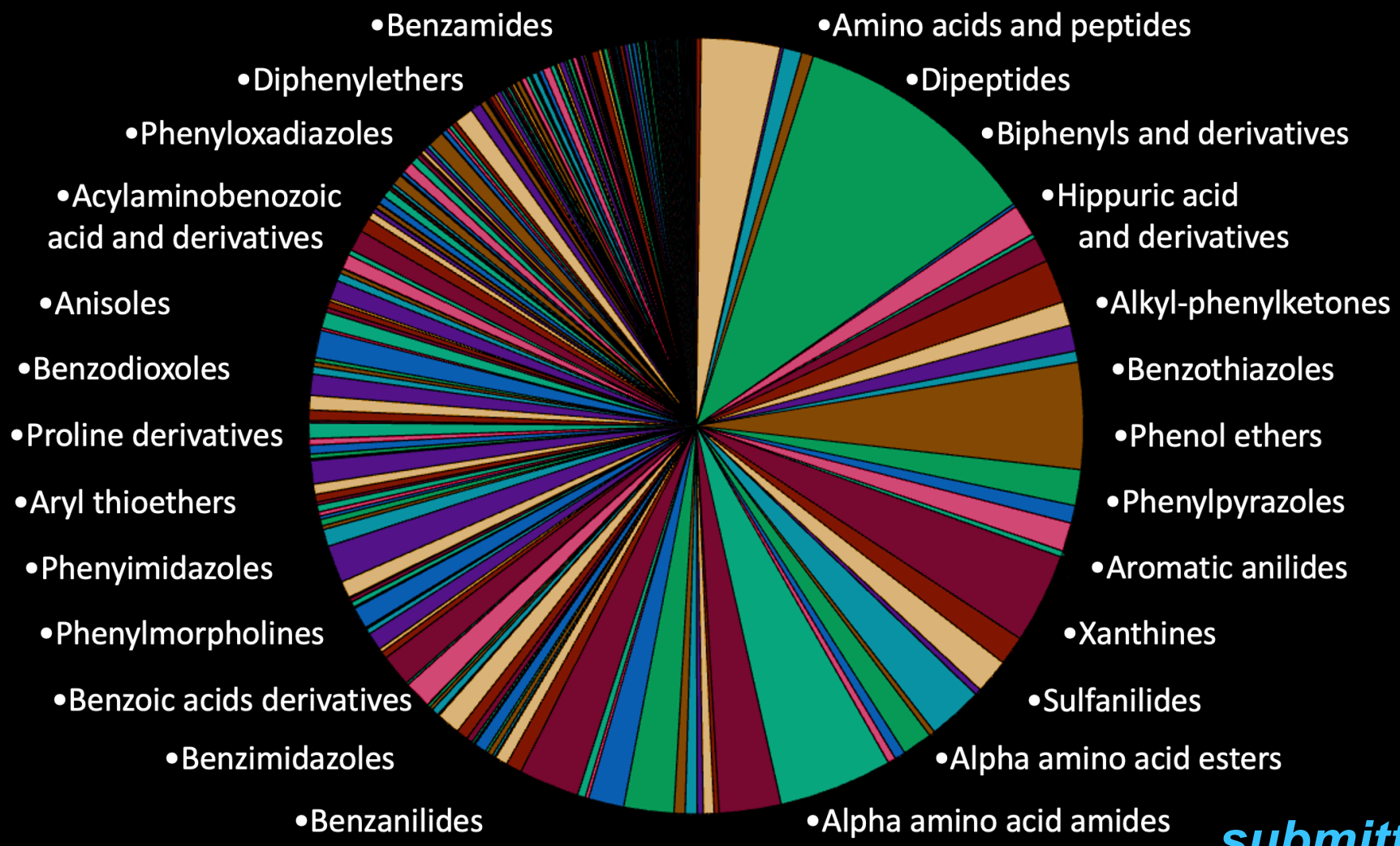
Automated 600,000 Data Sets Conversion to CCS

Step 5: Generate & Export CCS Values



submitted

METLIN - Ion Mobility Spectrometry



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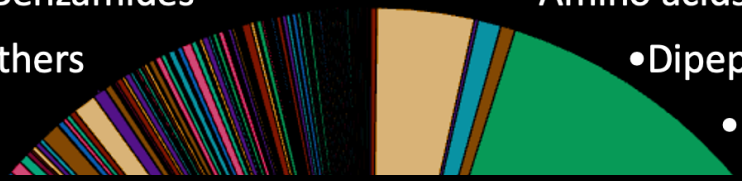

•Acylaminoben
acid and derivat
•Anisoles
•Benzodioxoles
•Proline derivative
•Aryl thioethers
•Phenyimidazoles
•Phenylmorphol
•Benzoic acids
•Benzir

ric acid
derivatives
kyl-phenylketones
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acid esters
es

160,000 CCS values on ~28,000 molecular standards

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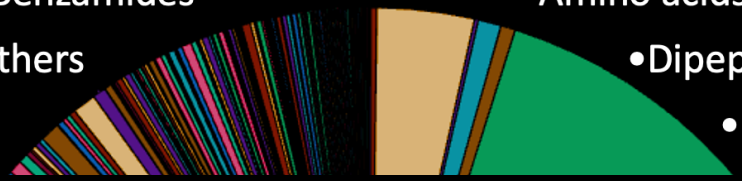

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MH+
MNa^+
MNH_4^+
$M\text{-}H^-$
$M+Cl^-$
$M+TFA^-$

submitted

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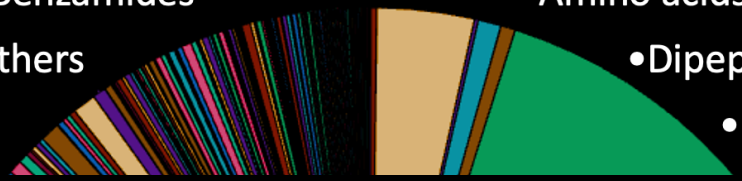

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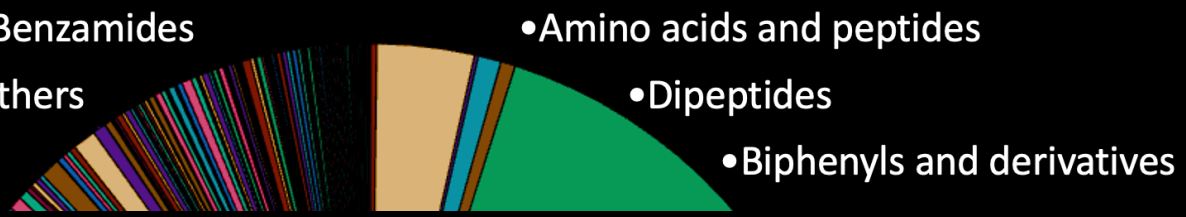

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In collaboration with Erin Baker & Skyline

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submitted

METLIN - Ion Mobility Spectrometry

Brian Pratt
Michael MacCoss
Brendan MacLean
(Skyline)

Heino Heyman
(Bruker)

The Two Hows?

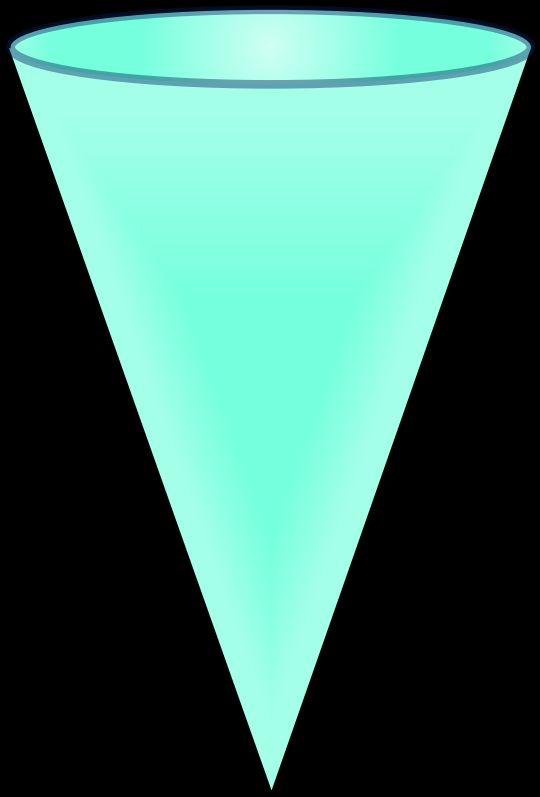

HPLC Column


HPLC Column


PHOTONIS
Waters
TITAN
X-GEN
Powder-Free Nitrile Examination Gloves
VWR
EMPTY
EMPTY

One Simple Motivation

30,000 features detected



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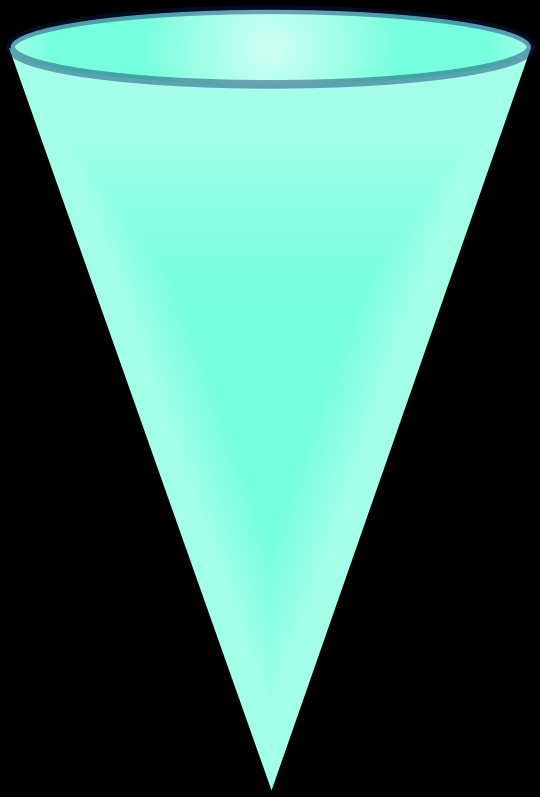

- Experimental Design
- Fold change
- Statistics (p-value)

One Simple Motivation

30,000 features detected

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- MS, IMS, & MS/MS data

One Simple Motivation

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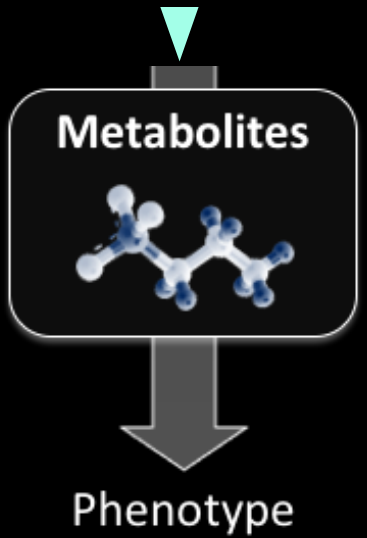

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- Fold change
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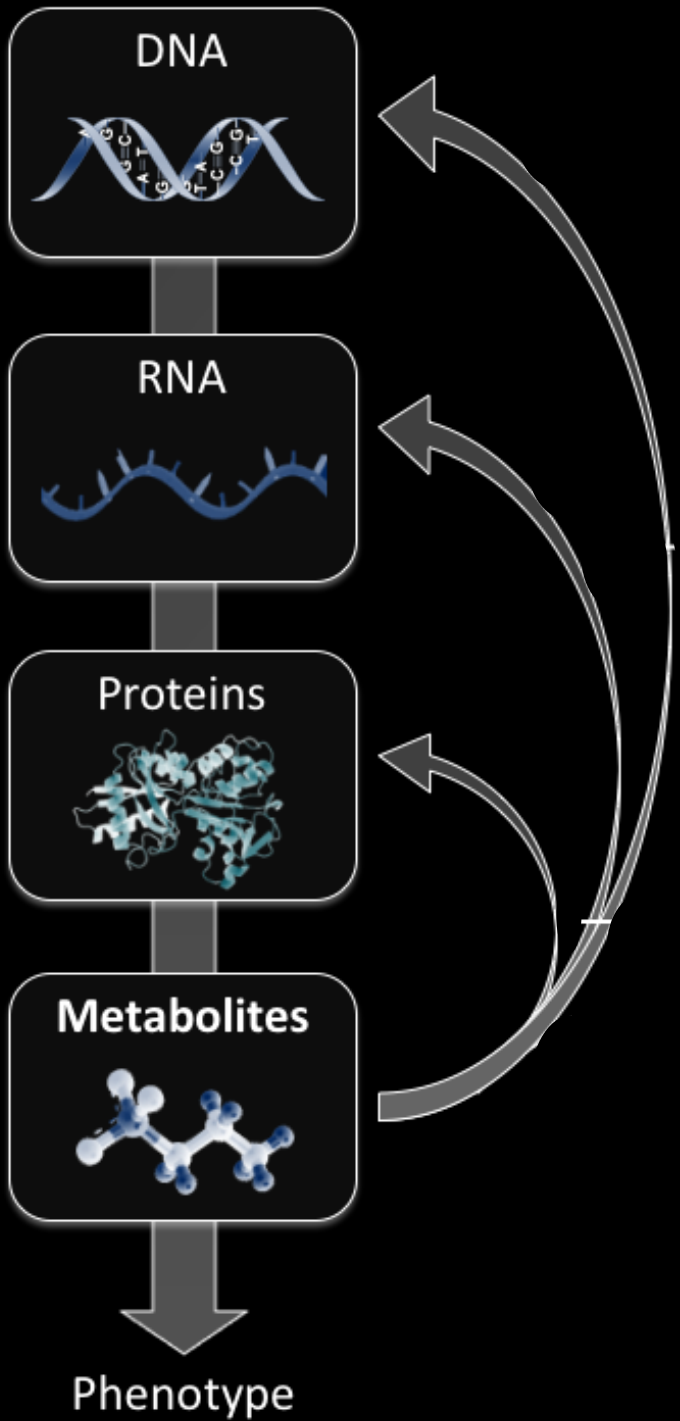
One Simple Motivation

Central Dogma of Biology

Nature Biotech. 2018
Nature Reviews 2019
Cell Metabolism 2022

Central Dogma of Biology

Nature Biotech. 2018
Nature Reviews 2019
Cell Metabolism 2022



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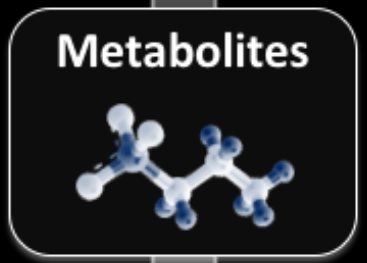
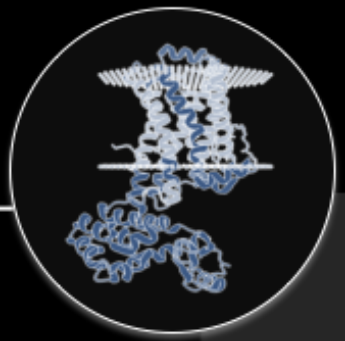


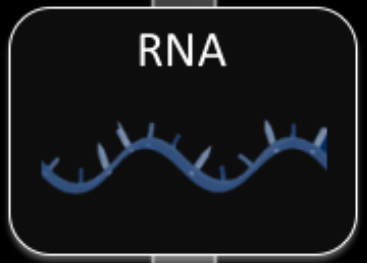


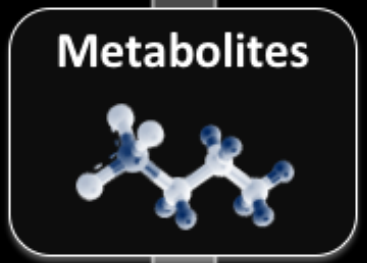
Gene expression

- Signal transduction control of transcription
- Epigenetic regulation by cofactors of chromatin enzymes

Phenotype

Nature Biotech. 2018
Nature Reviews 2019
Cell Metabolism 2022





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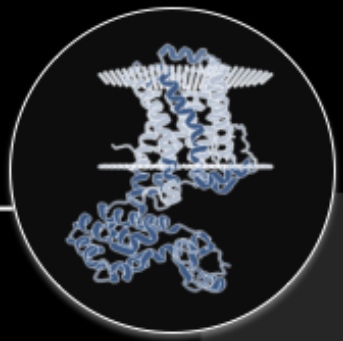

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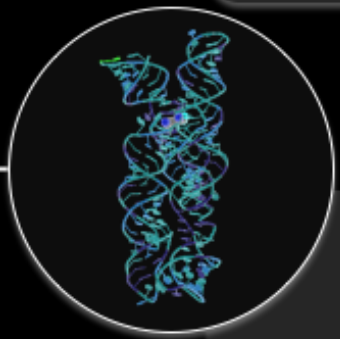

RNA metabolism

- Metabolite sensing by riboswitches
- Post-transcriptional modifications

Nature Biotech. 2018
Nature Reviews 2019
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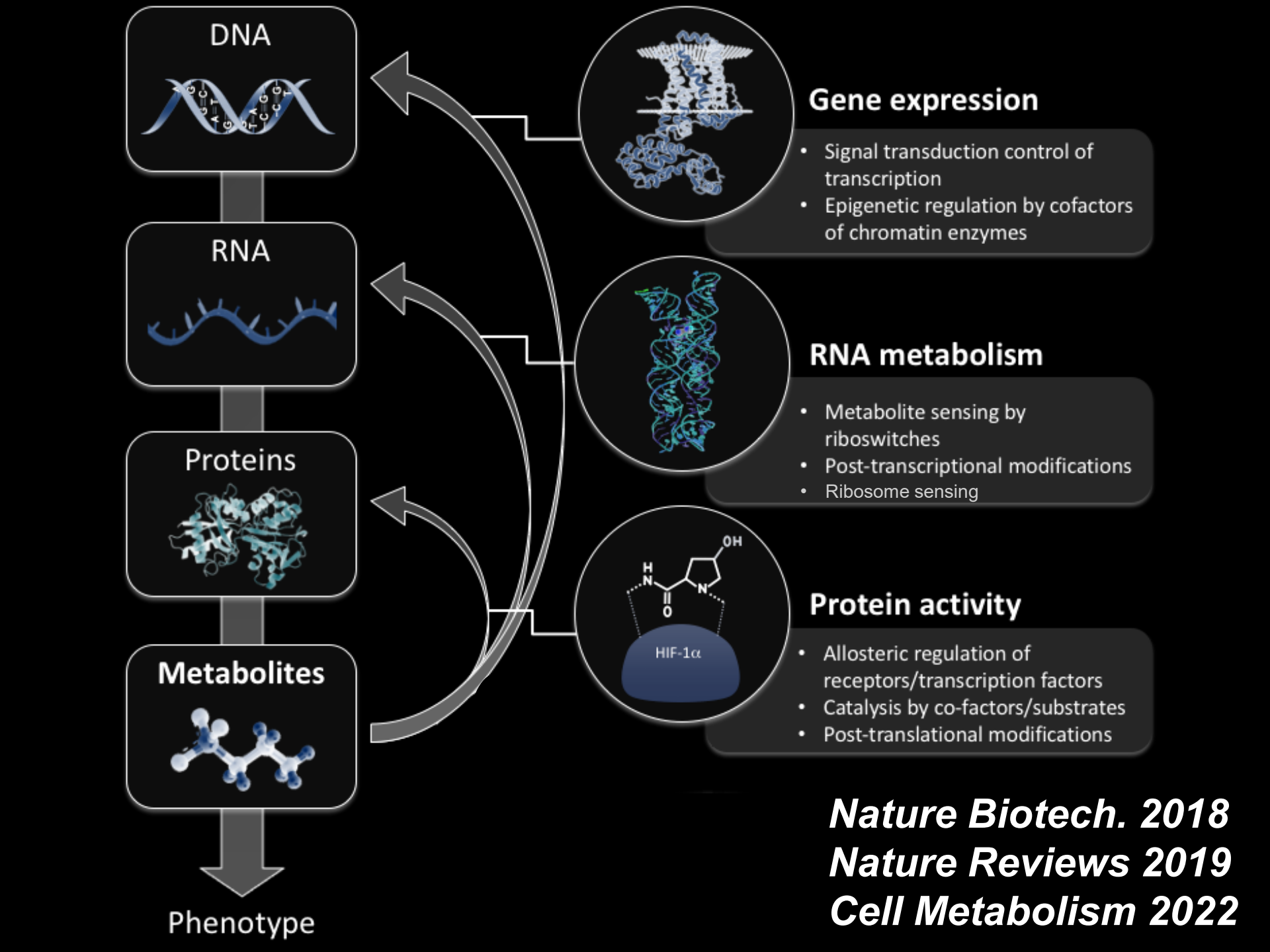
DNA
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Metabolites
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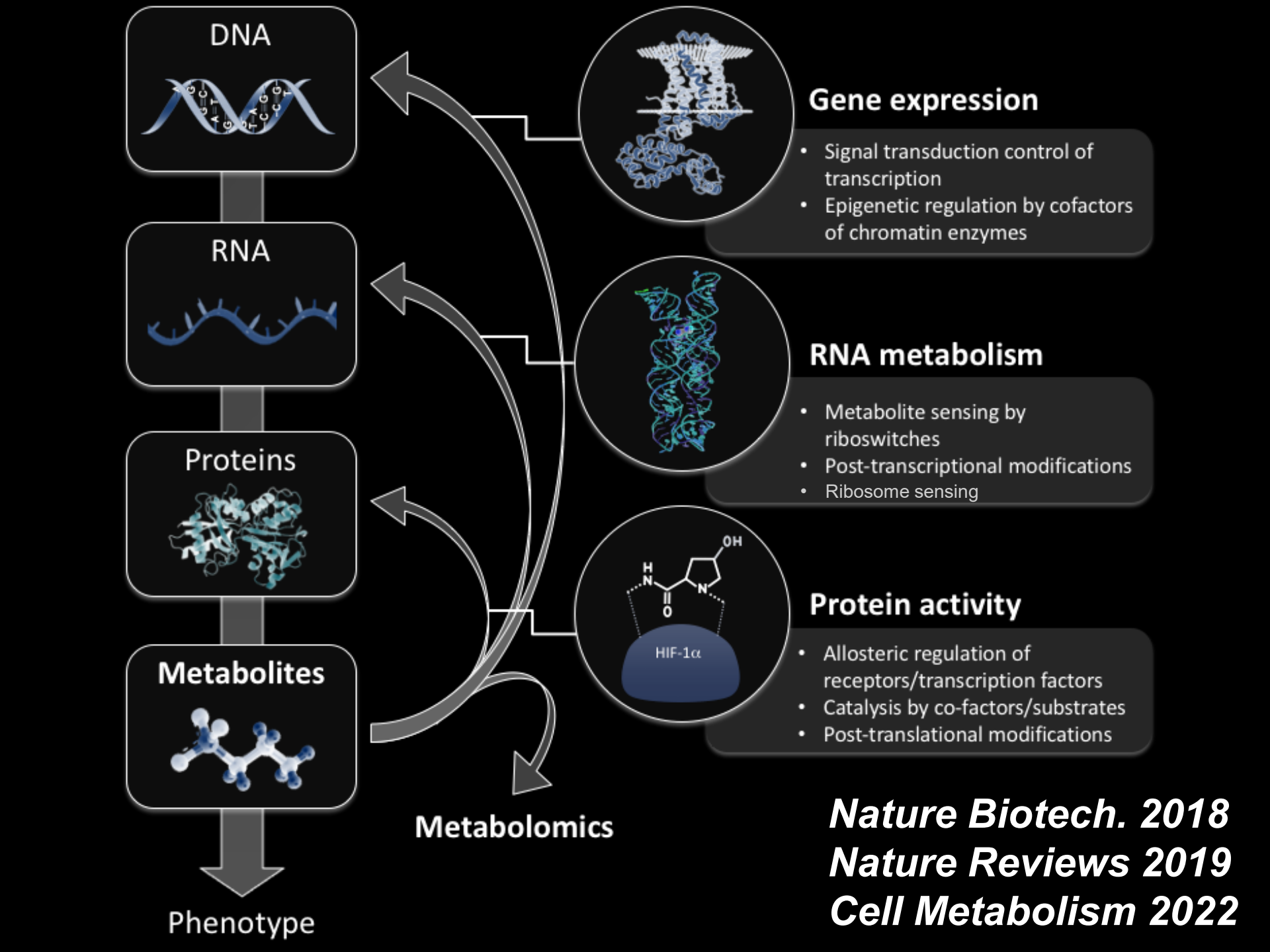
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DNA

RNA

Proteins

Metabolites

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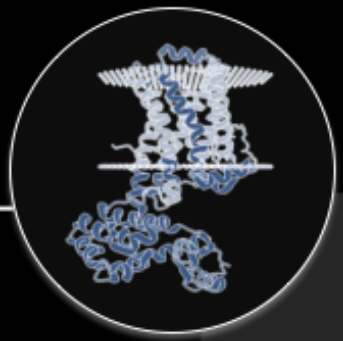

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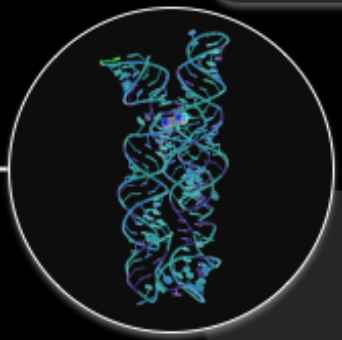

RNA metabolism

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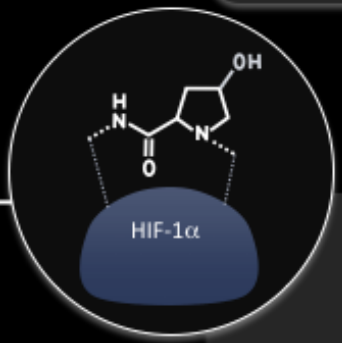

Protein activity

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Metabolomics

Nature Biotech. 2018
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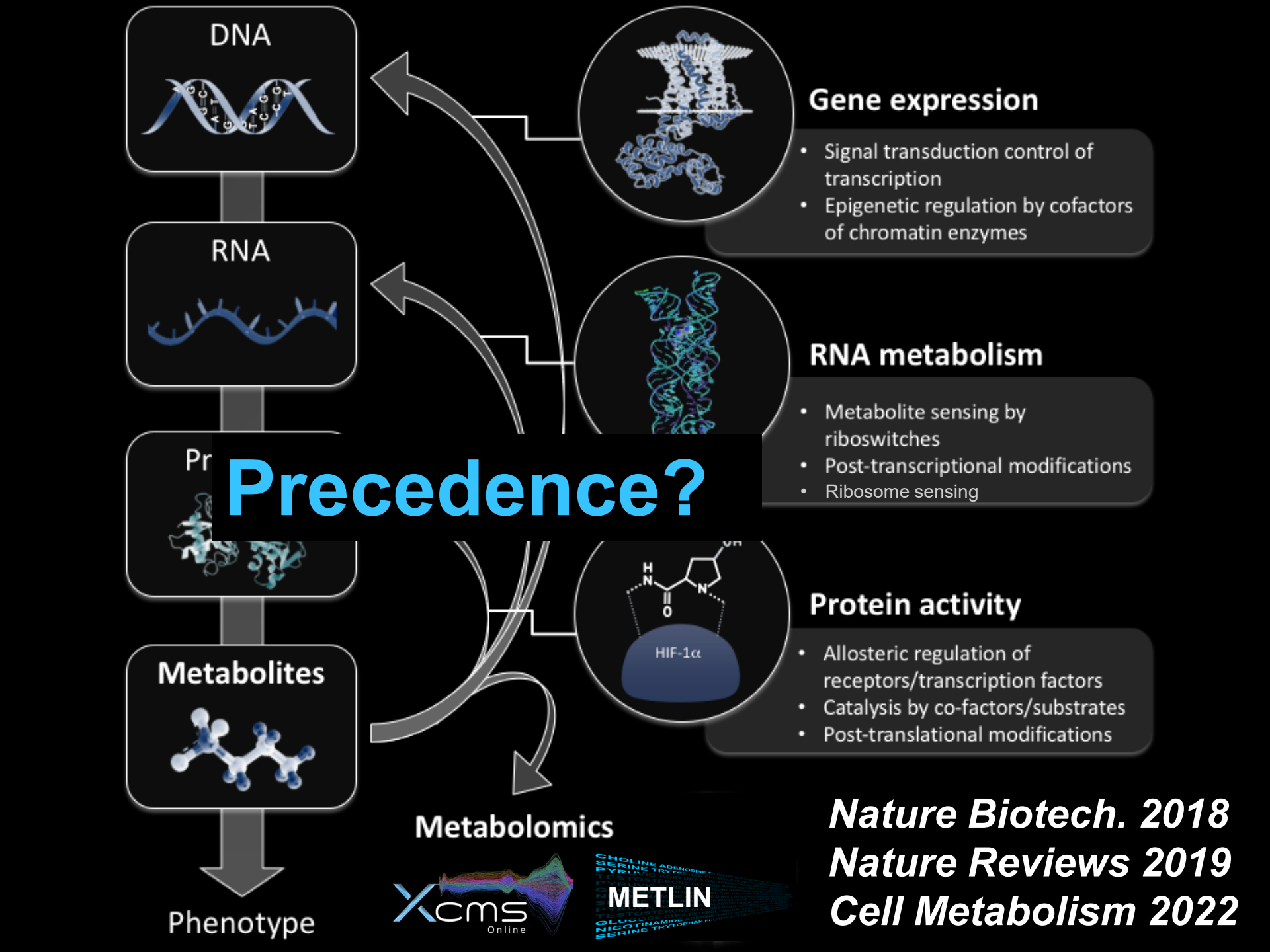
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Precedence?
Metabolomics
Xcms Online
METLIN
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Metabolite System Journal

Precedence?

Metabolite	System	Journal
Taurine	Multiple Sclerosis	Nature Chemical Biology
Microbial metabolites	Plant Assembly	Nature Microbiology
Itaconate	Anti-Inflammatory	Nature
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Ribonucleotide	Antiviral	Nature
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Kynurenine depletion	Cancer therapeutic	Nature Biotechnology
Histidine	Cancer therapeutic	Nature
Succinate	Thermogenesis	Nature
Glutamine	Sickle Cell Disease	NEJM
Apelin peptide	Sarcopenia	Nature Medicine
Sterols	Multiple Sclerosis	Nature
Succinate	Intestinal Remodeling	Cell
Inositol Phosphates	Viral Capsid Co-factors	Nature
5-Aminovaleric Acid	Reduces B-Oxidation	Scientific Reports
Nicotinamide	Acute Kidney Injury	Nature Medicine
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Albrioza just approved *2022 for ALS (Lou Gehrig's disease)*

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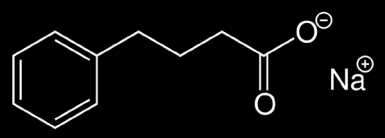
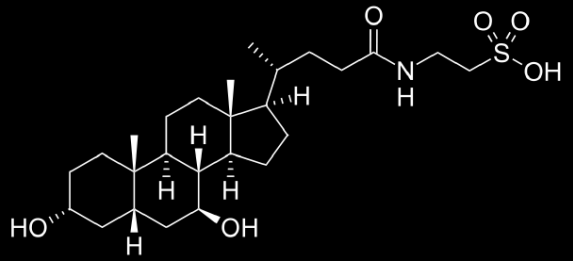
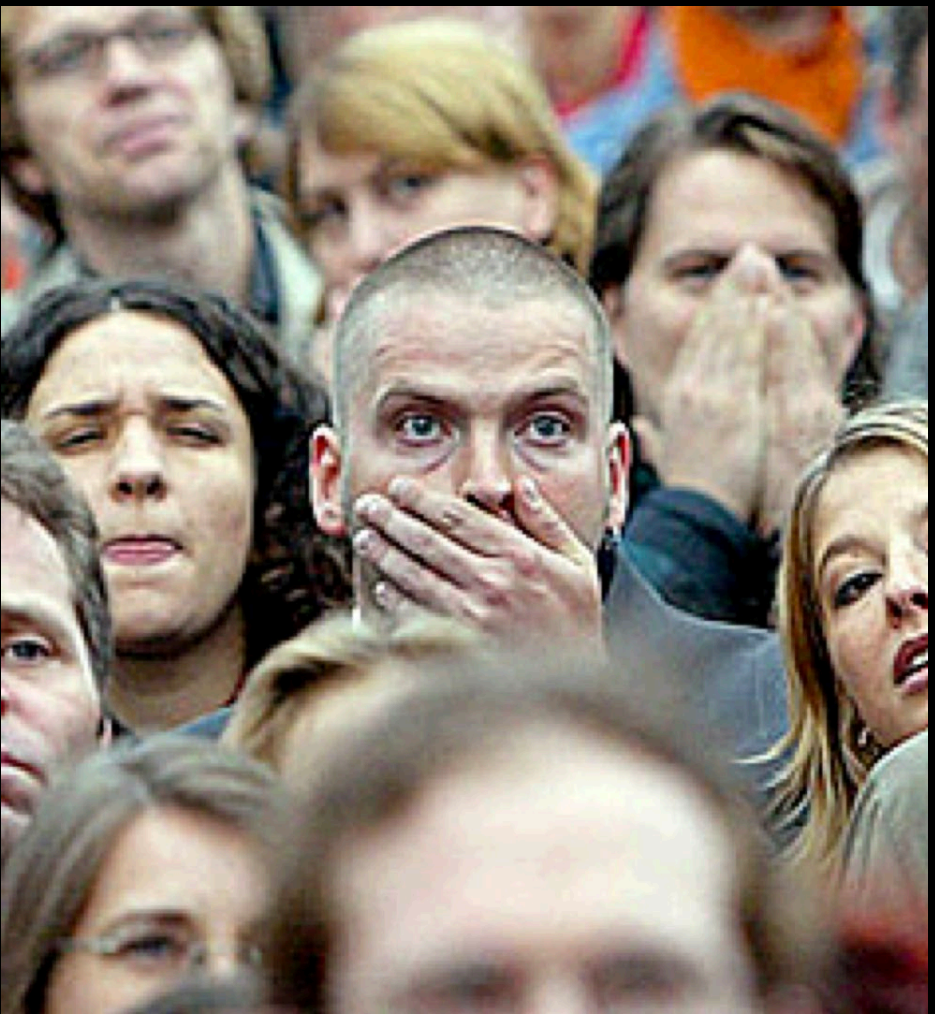
phenylbutyrate/ursodoxicoltaurine



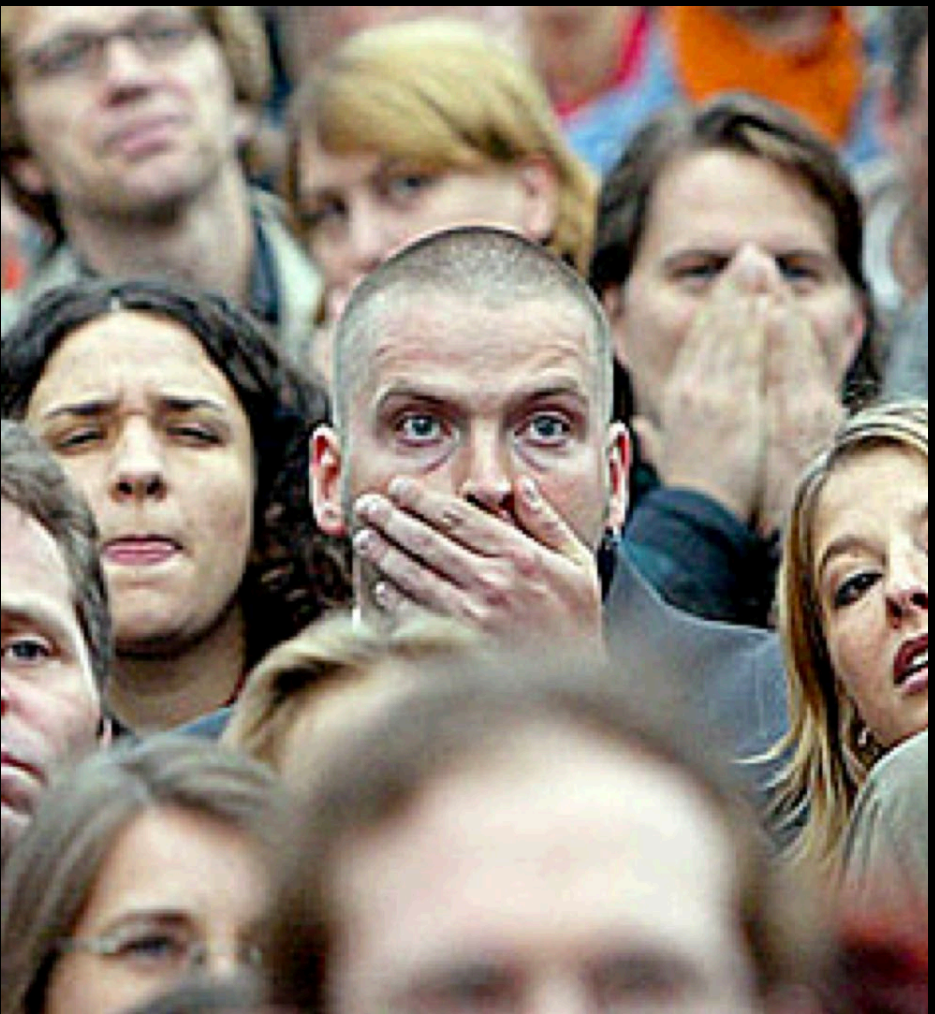


One Simple Motivation



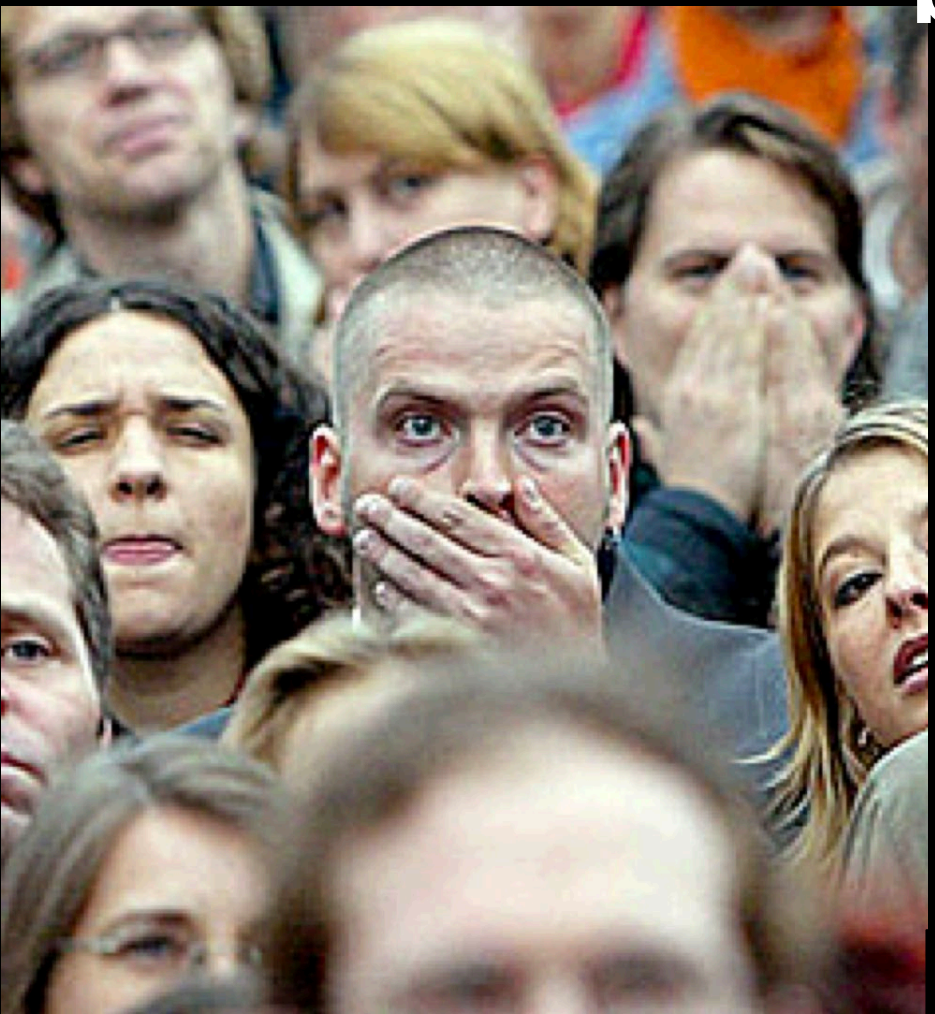
One Simple Motivation

Metabolomics



One Simple Motivation

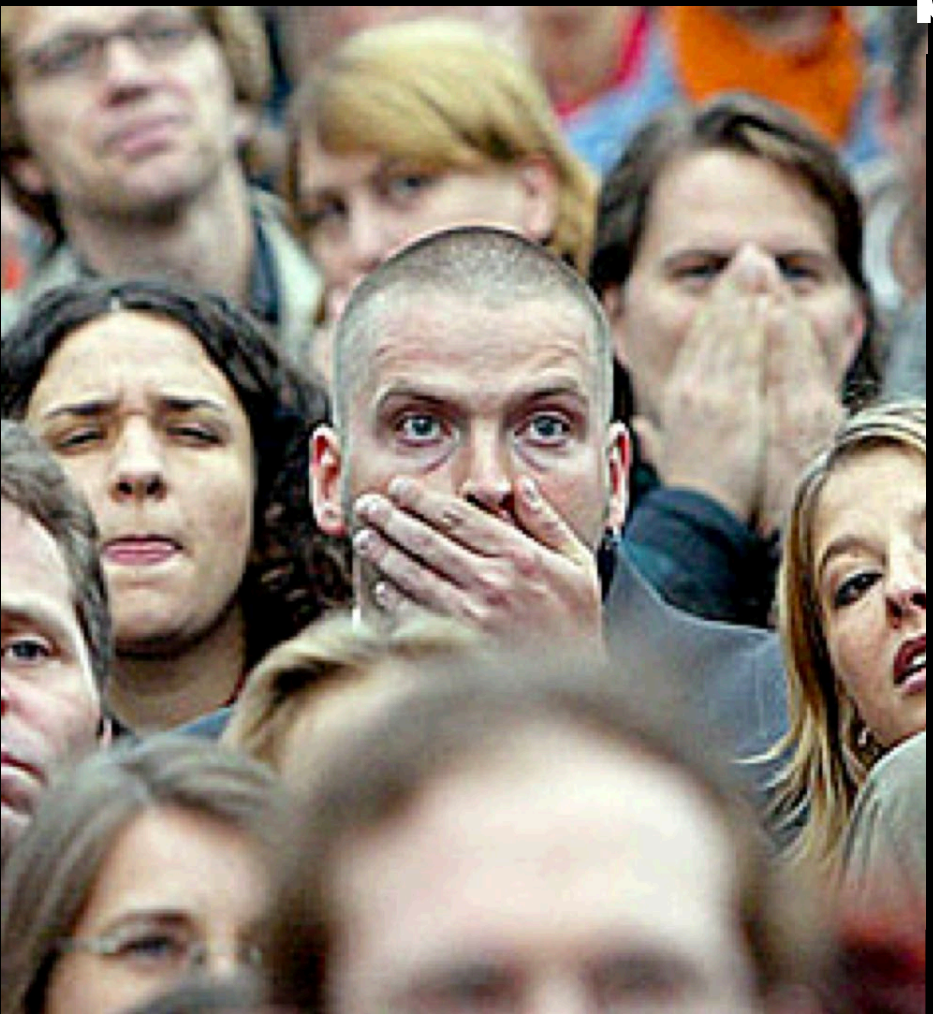
Metabolomics beyond biomarkers



One Simple Motivation

Metabolomics

beyond biomarkers



toward identifying endogenous metabolites that modulate physiology

Acknowledgements

Acknowledgements

NIH DOE DoD

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Winnie Heim

Aries Aisporna

Cayman CHEMICAL

Avanti® POLAR LIPIDS, INC.

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Winnie Heim

Aries Aisporna

Sleep

Richard Lerner

Ben Cravatt

Acknowledgements