HaDeX: an R package and web-server for analysis of data from HDX-MS experiments

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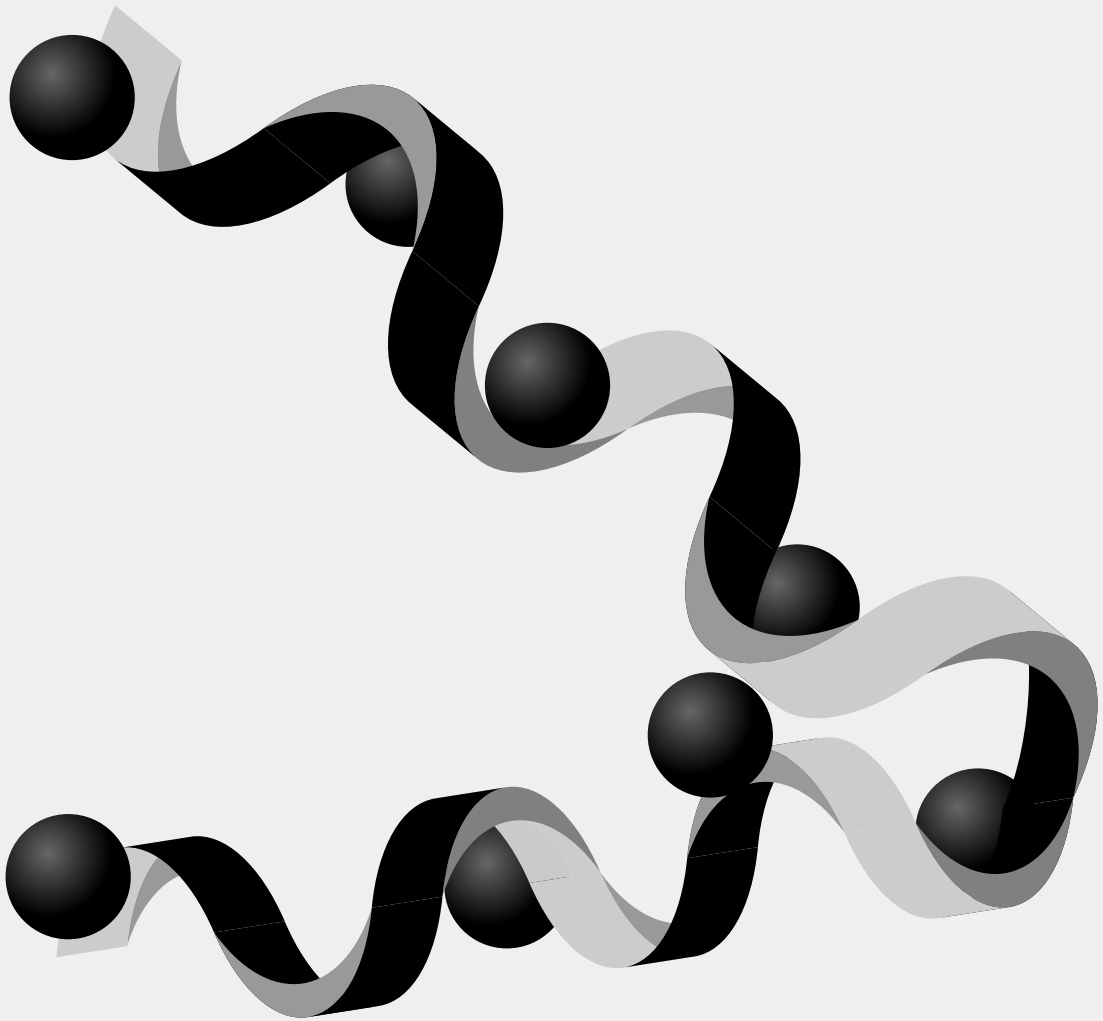
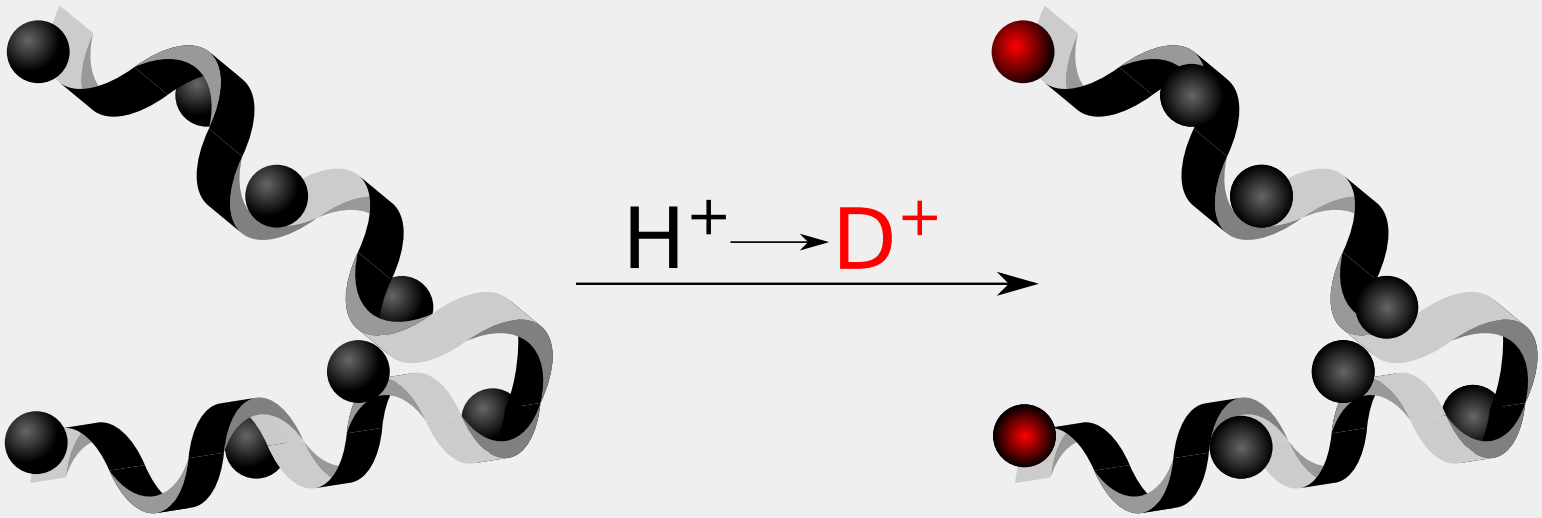
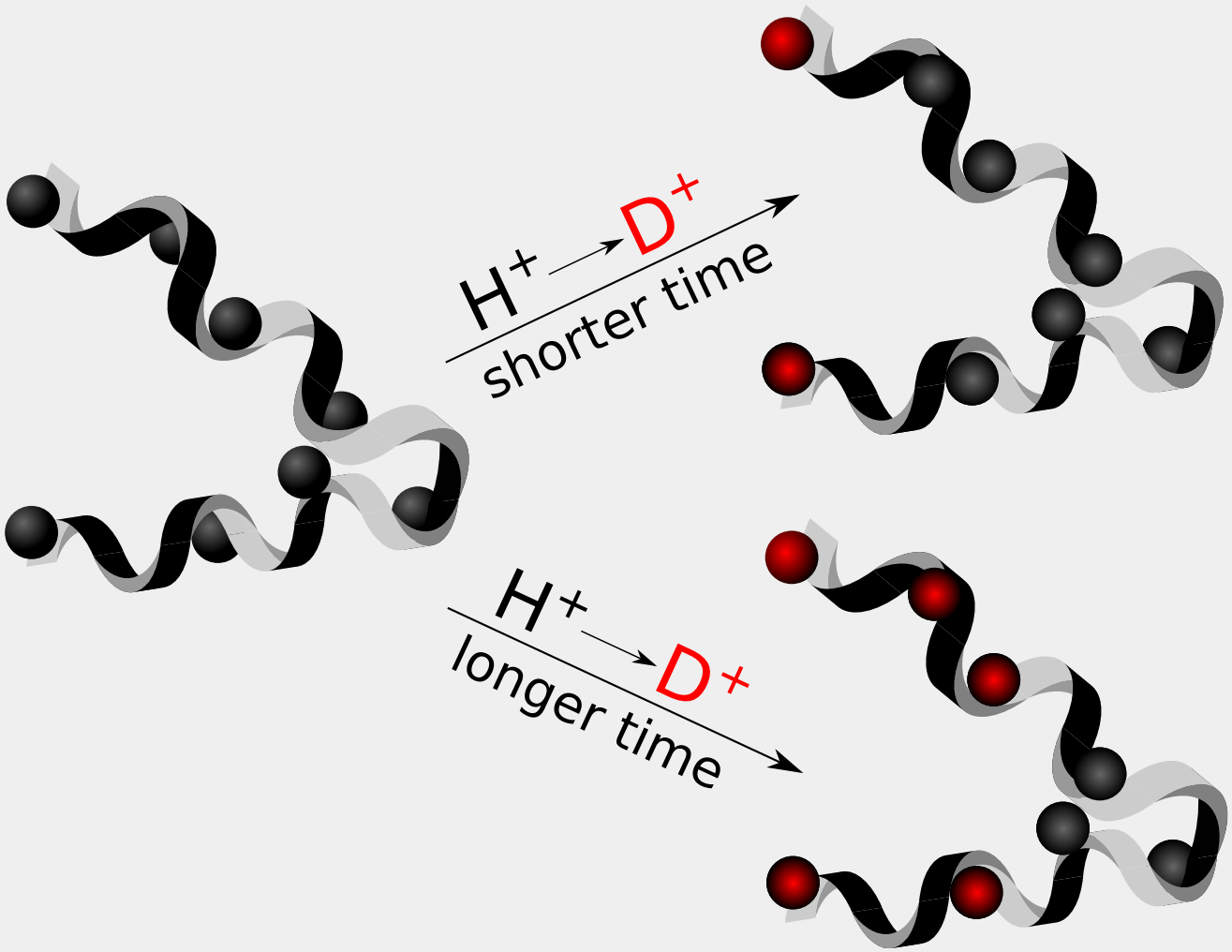
Outline

HDX-MS



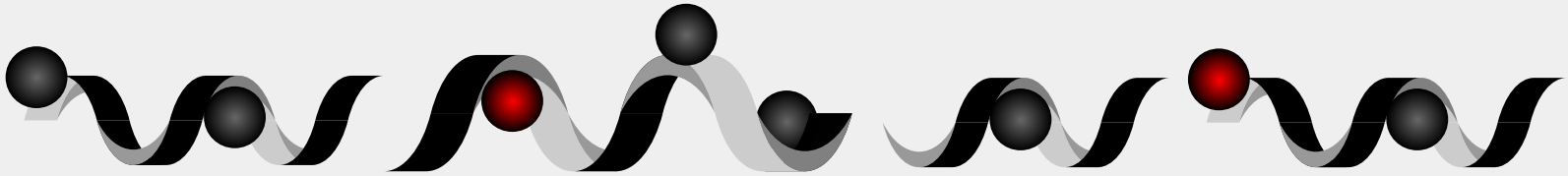


After the incubation in heavy water (D_2O), the most exposed amide hydrogens of the protein backbone are being replaced by deuters.

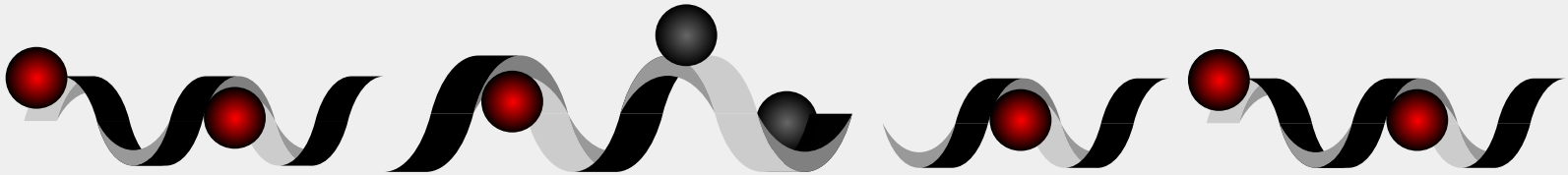


The longer incubation time, the more protected hydrogens are being replaced by deuters.

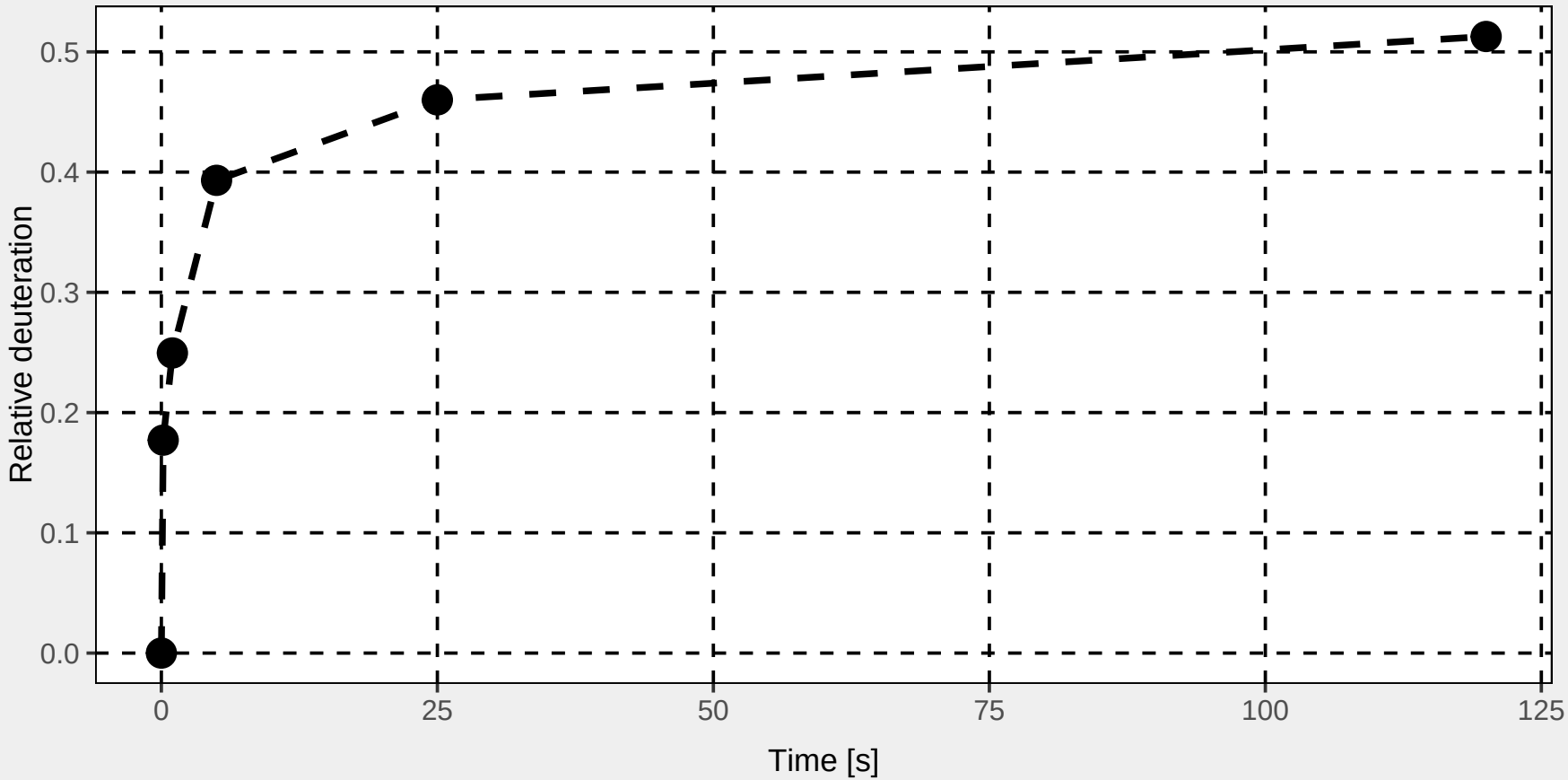
shorter incubation time



longer incubation time

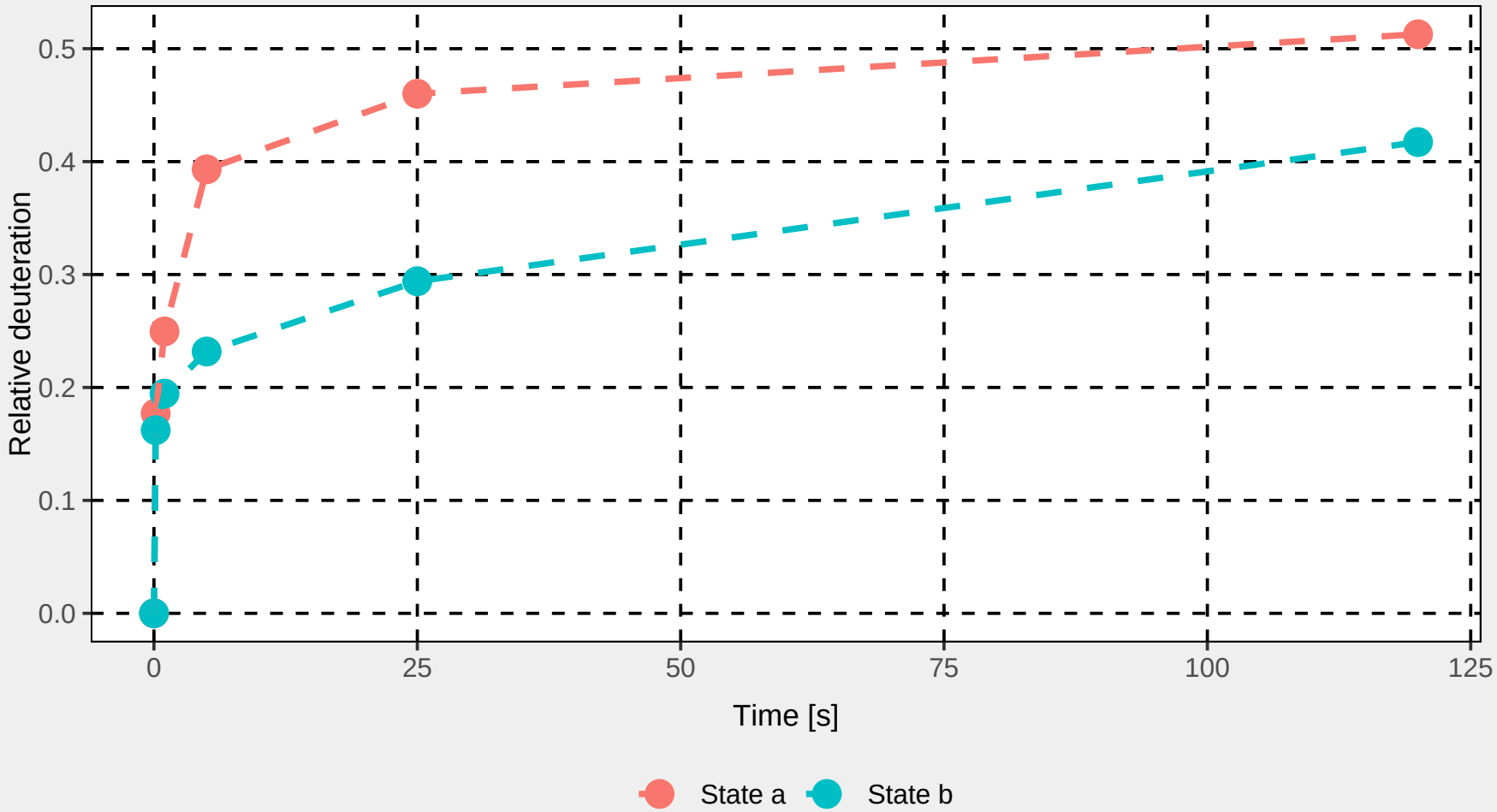


After the incubation, proteins are digested by a combination of proteases.

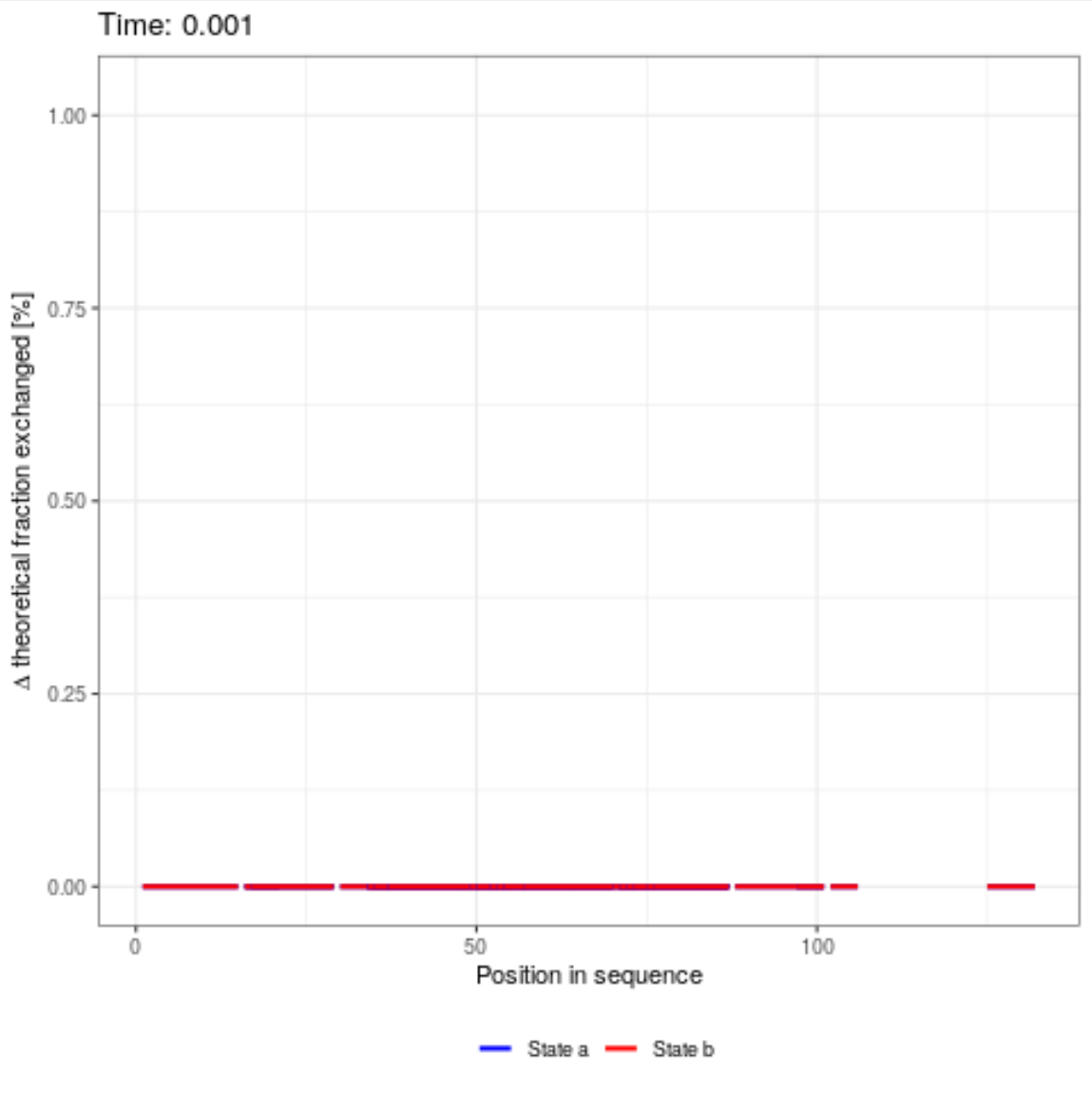


Thanks to the mass spectrometry, we are able to compute the m/z of each peptide depending on the duration of the incubation, thus we can compute a relative deuteration of a peptide.

Multi-state analysis

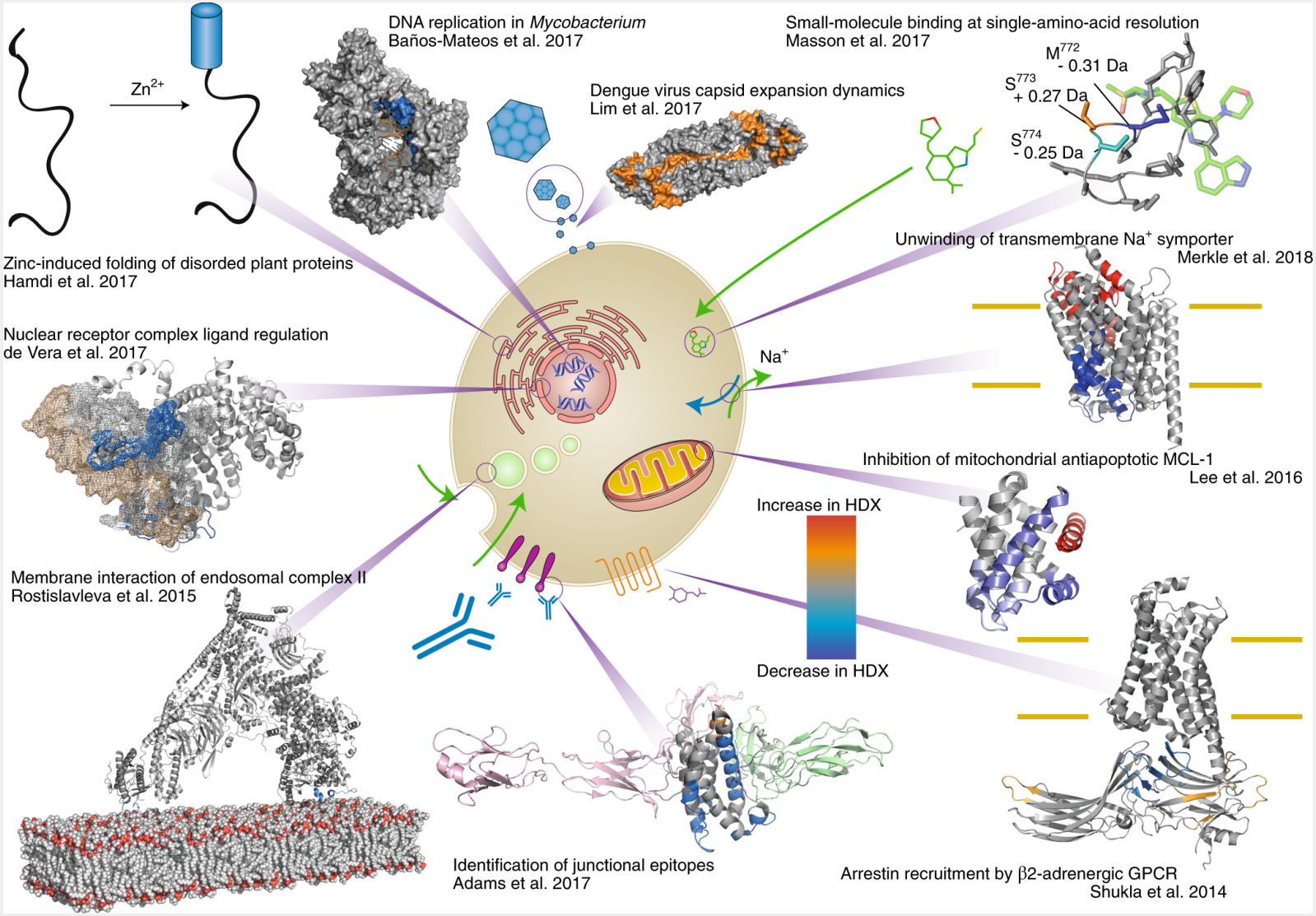


Peptides may come from proteins in differents states, i.e. bounded by different cofactors.



During the analysis of the HDX-MS data we have to consider both time of the incubation and position of a peptide in a sequence.

Applications



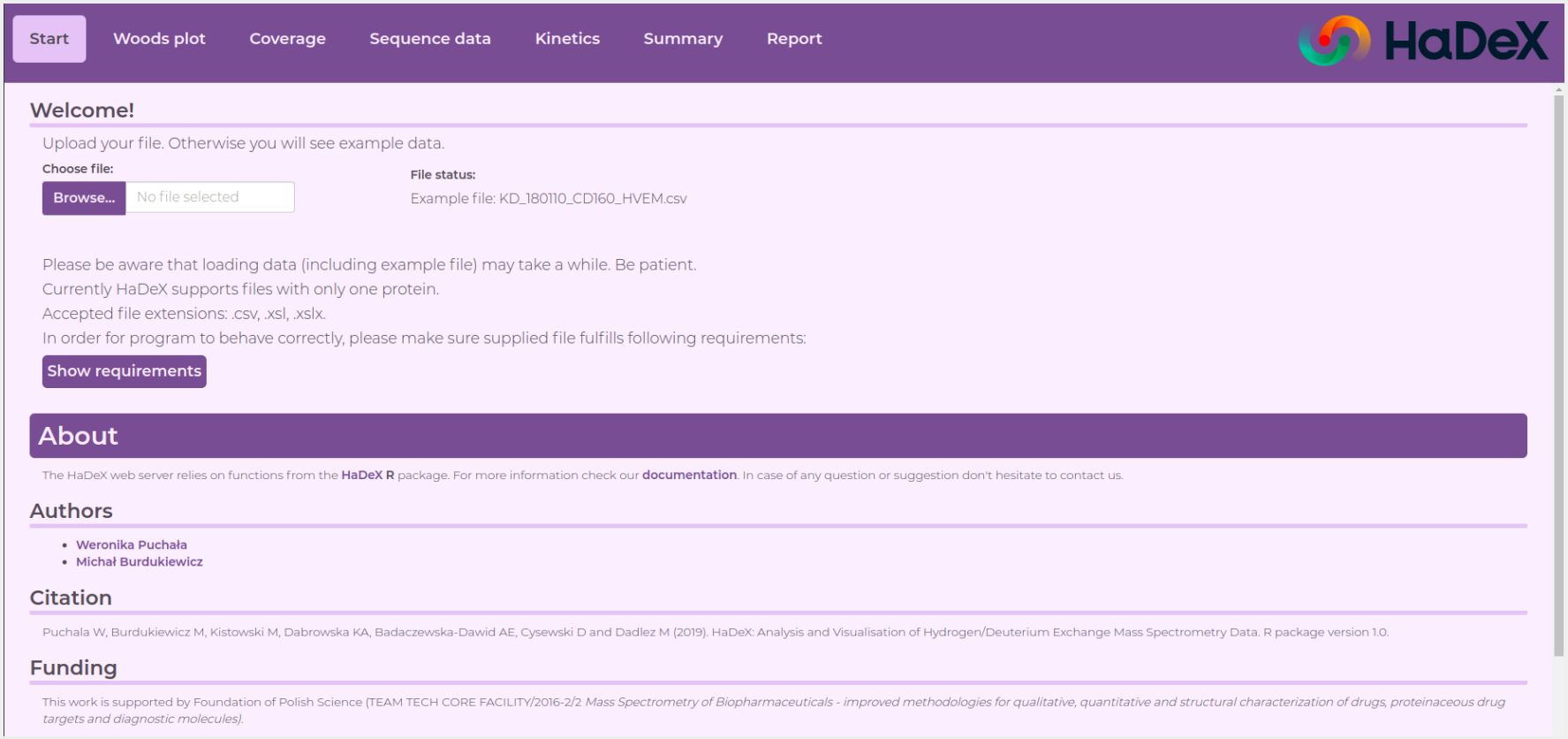
Source: Masson et al. (2019).

HaDeX workflow

Aims

HaDeX:

- easy,
- comprehensive,
- reproducible.


Start
Woods plot
Coverage
Sequence data
Kinetics
Summary
Report
HaDeX
Welcome!
Upload your file. Otherwise you will see example data.
Choose file:
Browse...
No file selected
File status:
Example file: KD_180110_CD160_HVEM.csv
Please be aware that loading data (including example file) may take a while. Be patient.
Currently HaDeX supports files with only one protein.
Accepted file extensions: .csv, .xsl, .xslx.
In order for program to behave correctly, please make sure supplied file fulfills following requirements:
Show requirements
About
The HaDeX web server relies on functions from the HaDeX R package. For more information check our documentation. In case of any question or suggestion don't hesitate to contact us.
Authors
Weronika Puchała
Michał Burdukiewicz
Citation
Puchala W, Burdukiewicz M, Kistowski M, Dabrowska KA, Badaczewska-Dawid AE, Cysewski D and Dadlez M (2019). HaDeX: Analysis and Visualisation of Hydrogen/Deuterium Exchange Mass Spectrometry Data. R package version 1.0.
Funding
This work is supported by Foundation of Polish Science (TEAM TECH CORE FACILITY/2016-2/2 Mass Spectrometry of Biopharmaceuticals - improved methodologies for qualitative, quantitative and structural characterization of drugs, proteinaceous drug targets and diagnostic molecules).

Start | Woods plot | Coverage | Sequence data | Kinetics | Summary | Report

HaDeX

Select parameters for the plot.

Theoretical calculations

Choose values type:

relative

absolute

Comparison plot parameters:

Choose time parameters:

IN	CHOSEN	OUT
0.001	1	1440

Choose states for comparison:

CD160

CD160_HVEM

Adjust colors

Woods plot parameters:

State 1	State 2
CD160	CD160_HVEM

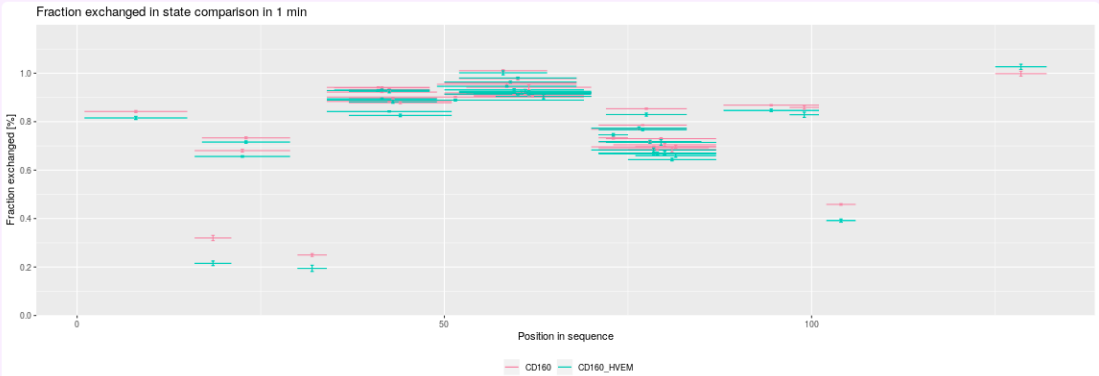
Confidence limit 1:	Confidence limit 2:
98%	99%

Adjust plot:

Choose y range for comparison plot:

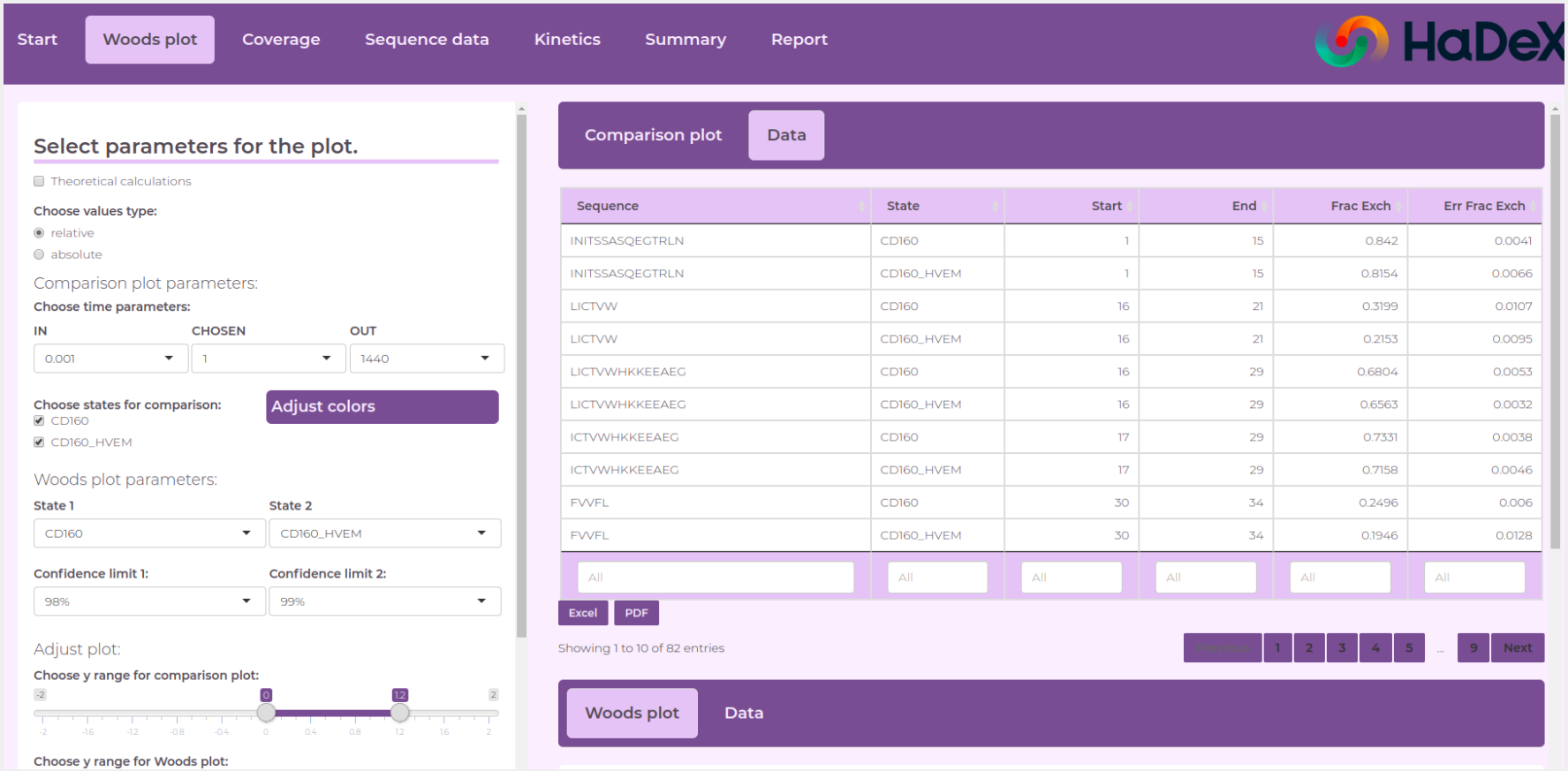
Choose y range for Woods plot:

Comparison plot | Data

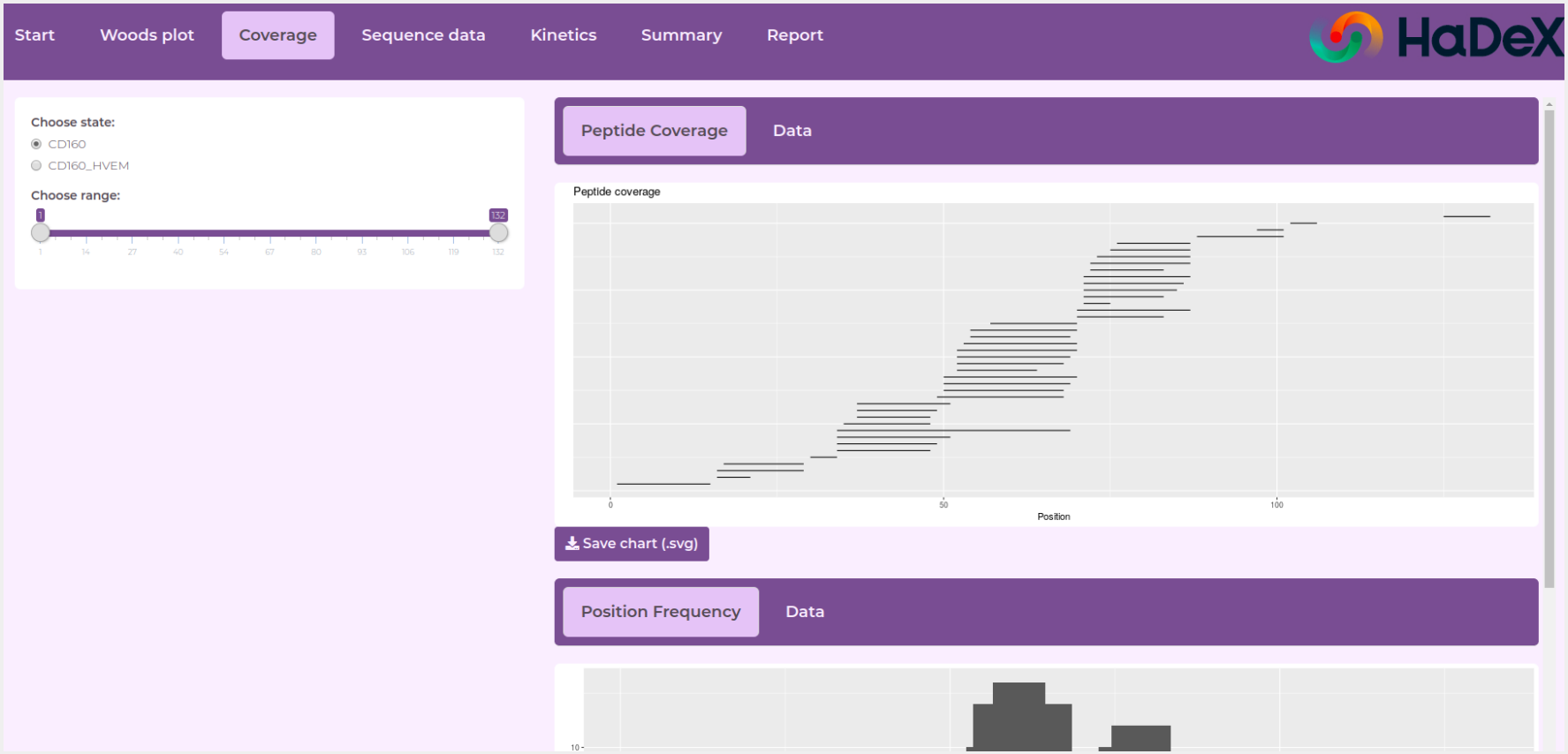


Save chart (.svg)

Woods plot | Data



Uncertainties derived by error propagation (Joint Committee for Guides in Metrology, 2008).


Start
Woods plot
Coverage
Sequence data
Kinetics
Summary
Report
HaDeX
Choose state:
CD160
CD160_HVEM
Choose range:
1
132
1
14
27
40
54
67
80
93
106
119
132
Peptide Coverage
Data
Peptide coverage
0
50
100
Position
Save chart (.svg)
Position Frequency
Data
10

Start | Woods plot | Coverage | Sequence data | Kinetics | Summary | Report | HaDeX

Protein name

db_CD160

Reconstructed sequence

INITSSASQEGTRLNLICTVWHKKEEAEGFVVFLCKDRSGDCSPETSLKQLRLKRDPGIDGVGEISSQLMFTISQVTPLHSGTYQCCARSQKSGIRLQGHFFSILFxxxxxxxxxxxxxxxxxFSHNEGTL

Name	Value
Length	132
Coverage	86.36%
Cys	5

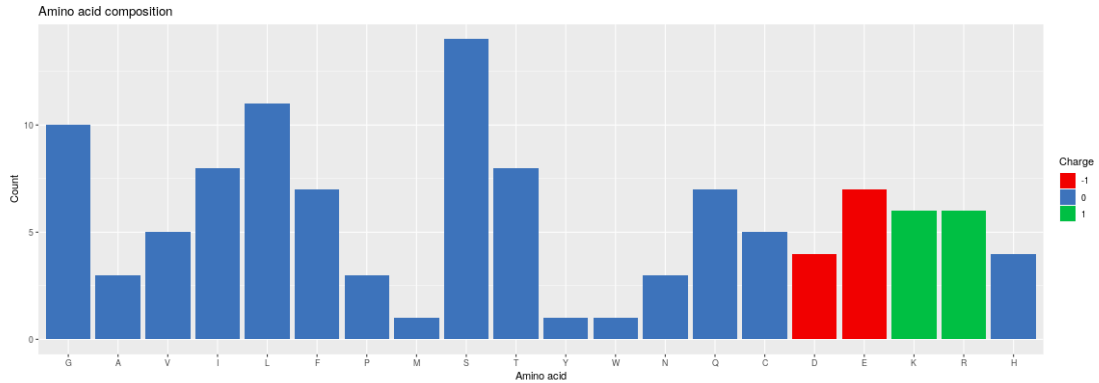
Hydro-
- [x] Hydrophilic
- [x] Hydrophobic

Correct sequence length:

132

If C-terminal of is not covered by peptides, enter its correct position.

Sequence length from the file is 132 .



Save chart (.svg)

Start | Woods plot | Coverage | Sequence data | Kinetics | Summary | Report

HaDeX

Select parameters for the plot.

Theoretical calculations

Choose values type:

relative

absolute

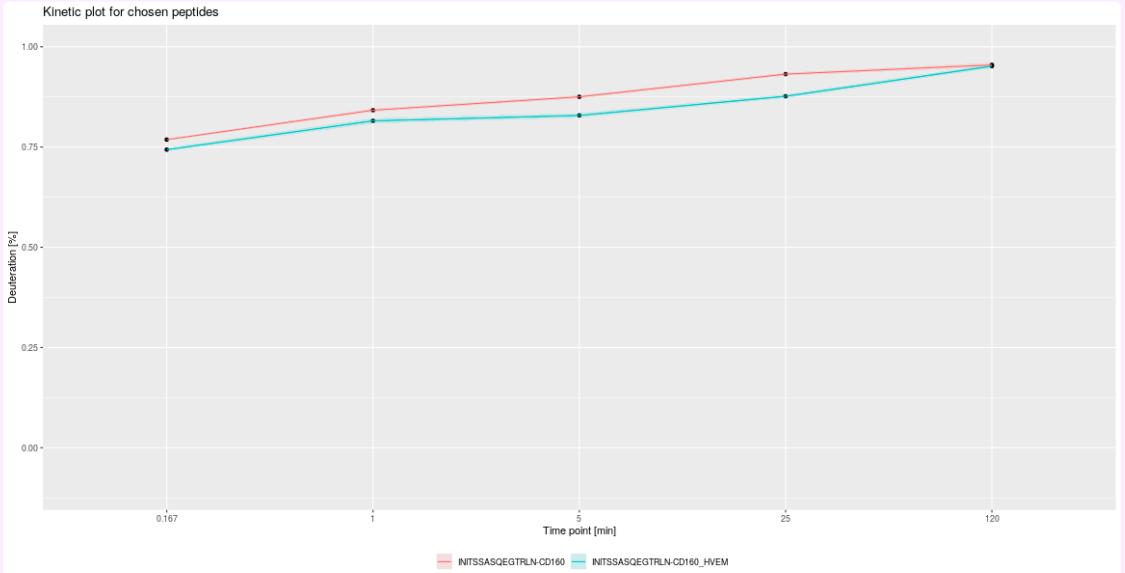
Choose time parameters:

IN: 0.001 | OUT: 1440

Choose peptide:

Sequence	State	Start	End
INITSSASQEGTRLN	CD160	1	15
INITSSASQEGTRLN	CD160_HVEM	1	15
LICTVW	CD160	16	21
LICTVW	CD160_HVEM	16	21
LICTVWHKKEEAEG	CD160	16	29
LICTVWHKKEEAEG	CD160_HVEM	16	29
ICTVWHKKEEAEG	CD160	17	29
ICTVWHKKEEAEG	CD160_HVEM	17	29
FVVFL	CD160	30	34
FVVFL	CD160_HVEM	30	34

Kinetic plot | Data



Save chart (.svg)

Start | Woods plot | Coverage | Sequence data | Kinetics | **Summary** | Report

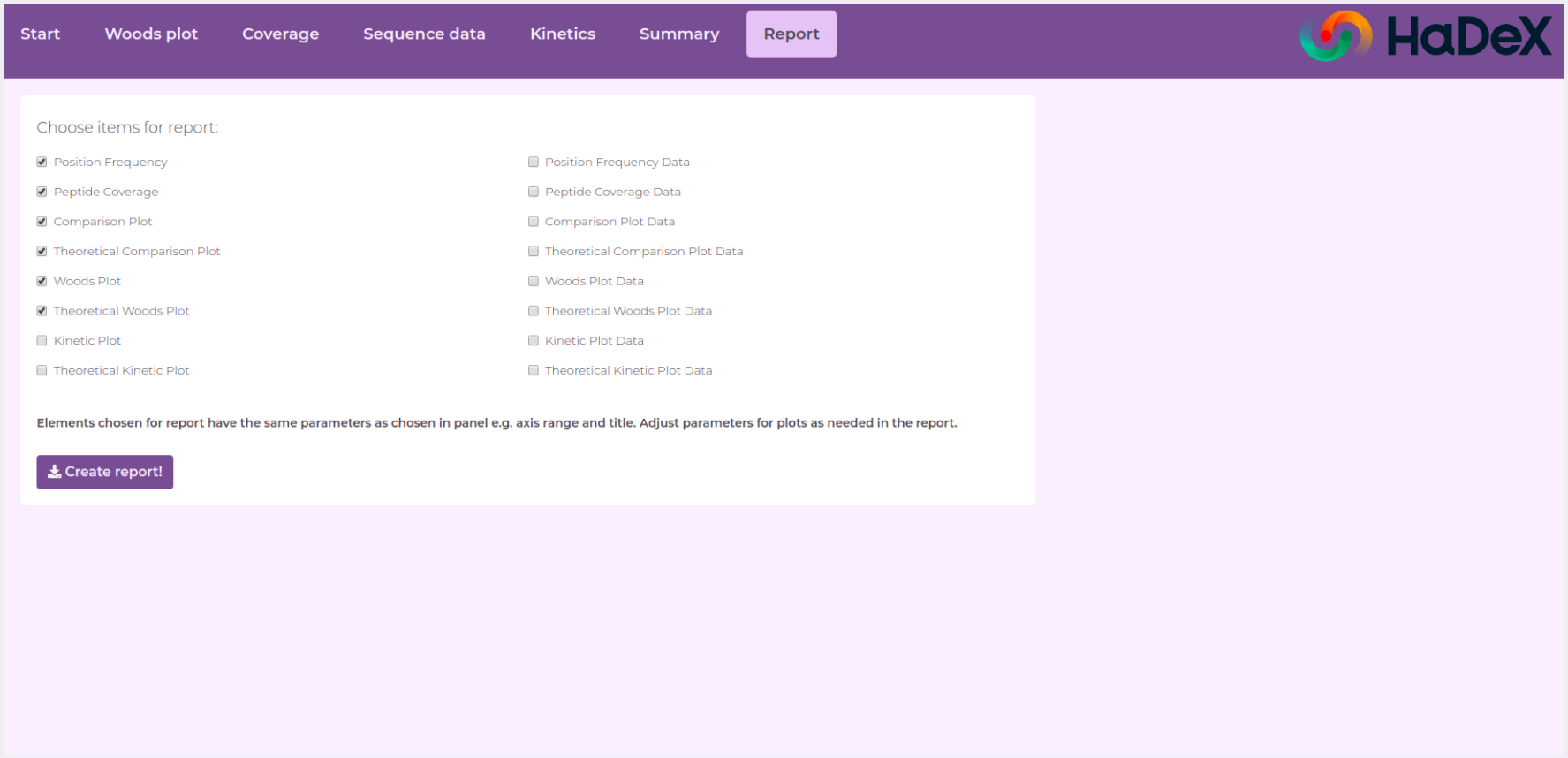
HaDeX

Name	Value
HDX time course	8
Number of peptides	41
Sequence coverage	0.8636
Average peptide length	14.6829
Redundancy	4.5606
Replicates	4
Significant differences in HDX	0.98 | 0.99
All	All

Excel PDF

Showing 1 to 7 of 7 entries

This table was adapted from the HDX Data Summary (Masson, G.R., Burke, J.E., Ahn, N.G., Anand, G.S., Borchers, C., Brier, S., Bou-Assaf, G.M., Engen, J.R., Englander, S.W., Faber, J., et al. (2019). Recommendations for performing, interpreting and reporting hydrogen deuterium exchange mass spectrometry (HDX-MS) experiments. Nat Methods 16, 595–602).


Start
Woods plot
Coverage
Sequence data
Kinetics
Summary
Report
HaDeX
Choose items for report:
Position Frequency
Peptide Coverage
Comparison Plot
Theoretical Comparison Plot
Woods Plot
Theoretical Woods Plot
Kinetic Plot
Theoretical Kinetic Plot
Position Frequency Data
Peptide Coverage Data
Comparison Plot Data
Theoretical Comparison Plot Data
Woods Plot Data
Theoretical Woods Plot Data
Kinetic Plot Data
Theoretical Kinetic Plot Data
Elements chosen for report have the same parameters as chosen in panel e.g. axis range and title. Adjust parameters for plots as needed in the report.
Create report!

Availability

- a web-server (`http://mslab-ibb.pl/shiny/HaDeX/`),
- the R package (`https://CRAN.R-project.org/package=HaDeX`),
- a standalone software (`https://sourceforge.net/projects/HaDeX/`).

Summary

- rapidly developing technologies require a flexible framework,
- the methods of data analysis should follow the development of both technology and expectations of its users.

Acknowledgements

HaDeX developers:

- Weronika Puchala (main developer),
- Dominik Rafacz (frontend developer).

Funding

- Foundation of Polish Science TEAM TECH CORE FACILITY/2016-2/2 Mass Spectrometry of Biopharmaceuticals - improved methodologies for qualitative, quantitative and structural characterization of drugs, proteinaceous drug targets and diagnostic molecules.

MI² Data Lab

MI² Data Lab (https://mi2.mini.pw.edu.pl/), Faculty of Mathematics and Information Science, Warsaw University of Technology.

Contact: michalburdukiewicz@gmail.com.
Presentation:
https://github.com/HaDeXversum/Talks-and-posters.

References I

Joint Committee for Guides in Metrology (2008). JCGM 100: Evaluation of Measurement Data - Guide to the Expression of Uncertainty in Measurement. Technical report, JCGM.

Masson, G. R., Burke, J. E., Ahn, N. G., Anand, G. S., Borchers, C., Brier, S., Bou-Assaf, G. M., Engen, J. R., Englander, S. W., Faber, J., Garlish, R., Griffin, P. R., Gross, M. L., Guttman, M., Hamuro, Y., Heck, A. J. R., Houde, D., Iacob, R. E., Jørgensen, T. J. D., Kaltashov, I. A., Klinman, J. P., Konermann, L., Man, P., Mayne, L., Pascal, B. D., Reichmann, D., Skehel, M., Snijder, J., Strutzenberg, T. S., Underbakke, E. S., Wagner, C., Wales, T. E., Walters, B. T., Weis, D. D., Wilson, D. J., Wintrode, P. L., Zhang, Z., Zheng, J., Schriemer, D. C., and Rand, K. D. (2019). Recommendations for performing, interpreting and reporting hydrogen deuterium exchange mass spectrometry (HDX-MS) experiments. Nature Methods, 16(7):595–602.