

Project 03: Proteome-wide screen for RNA-dependent proteins

PD Dr. Maiwen Caudron-Herger / Tutorin: Michela Pozzi

Research Group "RNA-Protein Complexes & Cell Proliferation"

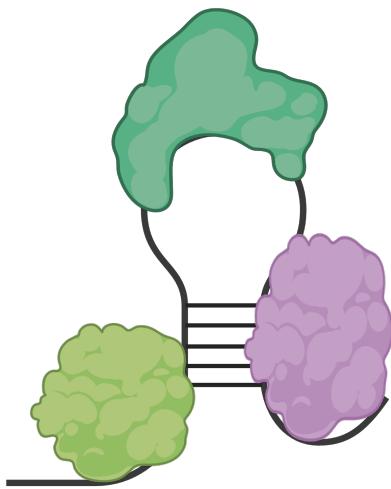
German Cancer Research Center (DKFZ), Heidelberg



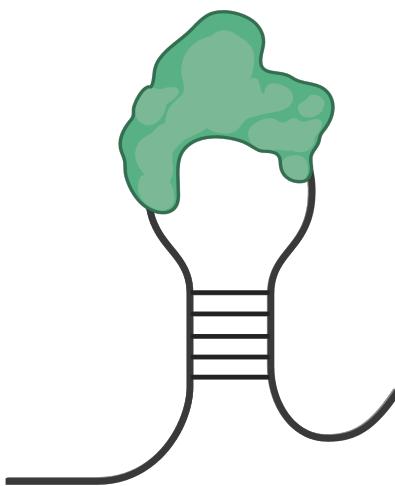
GERMAN
CANCER RESEARCH CENTER
IN THE HELMHOLTZ ASSOCIATION

RNA-binding proteins bind to RNA

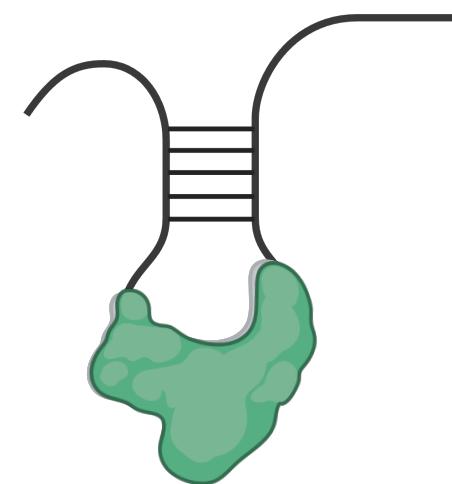
Ribonucleoprotein complex



RBPs regulate the fate of RNA



RNA regulates the fate of RBPs



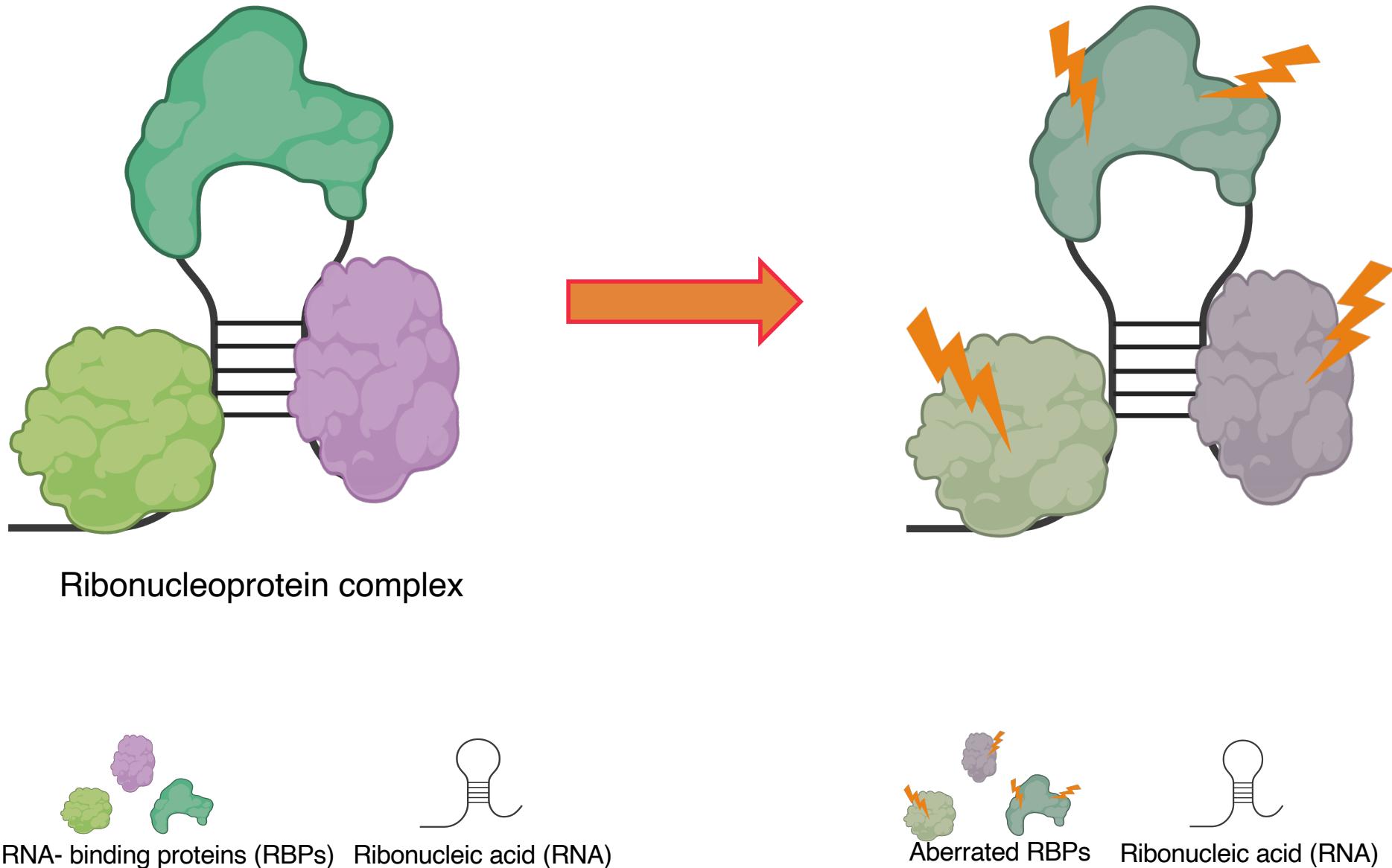
- Transcription
- Splicing
- Translation
- Degradation
- Function
- Interaction
- Localization
- Conformation



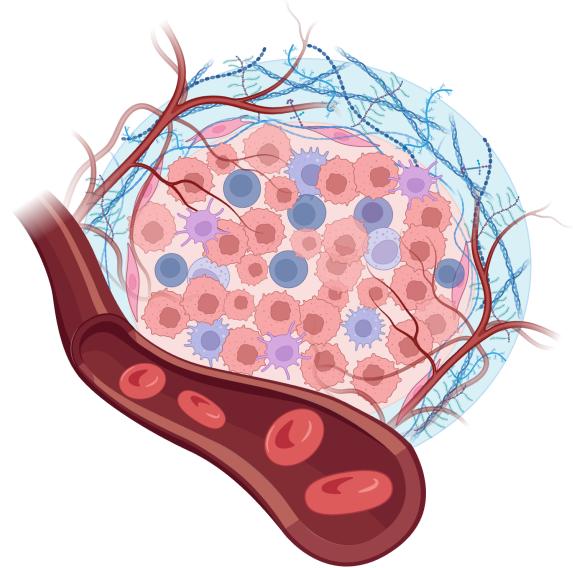
RNA- binding proteins (RBPs) Ribonucleic acid (RNA)

The images were created using Biorender

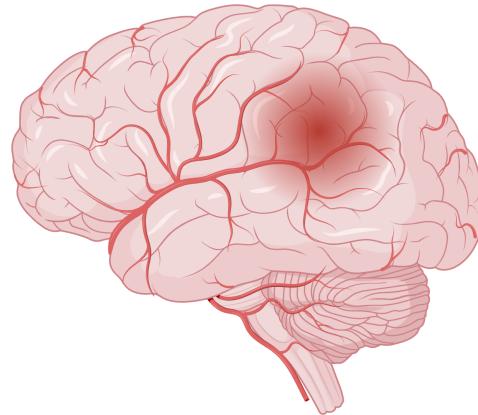
RNA-binding proteins can be aberrated



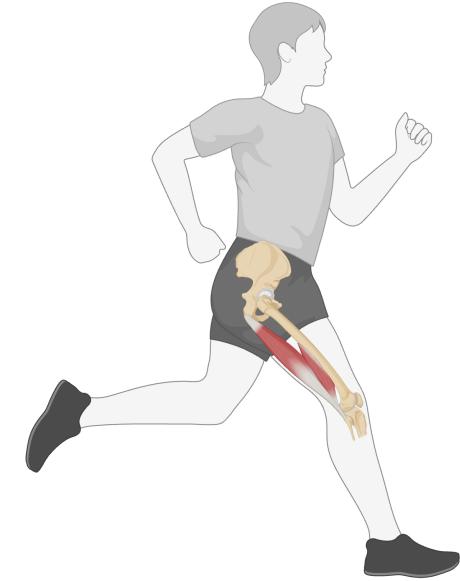
Dysregulated RBPs cause pathological human diseases



Cancer



Neurological disorders



Muscular atrophies

RNA-binding proteins (RBPs) have faced a growing interest



Multiple strategies



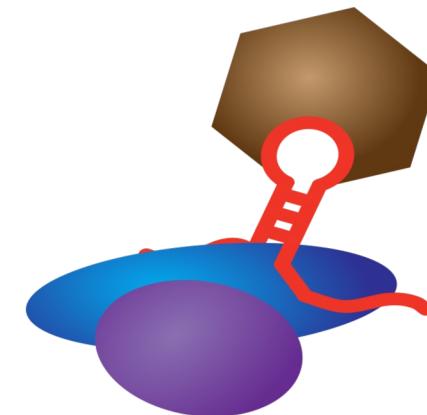
Catalogues of RBPs
in various species

RNA-Dependent Proteins (R-DeeP)

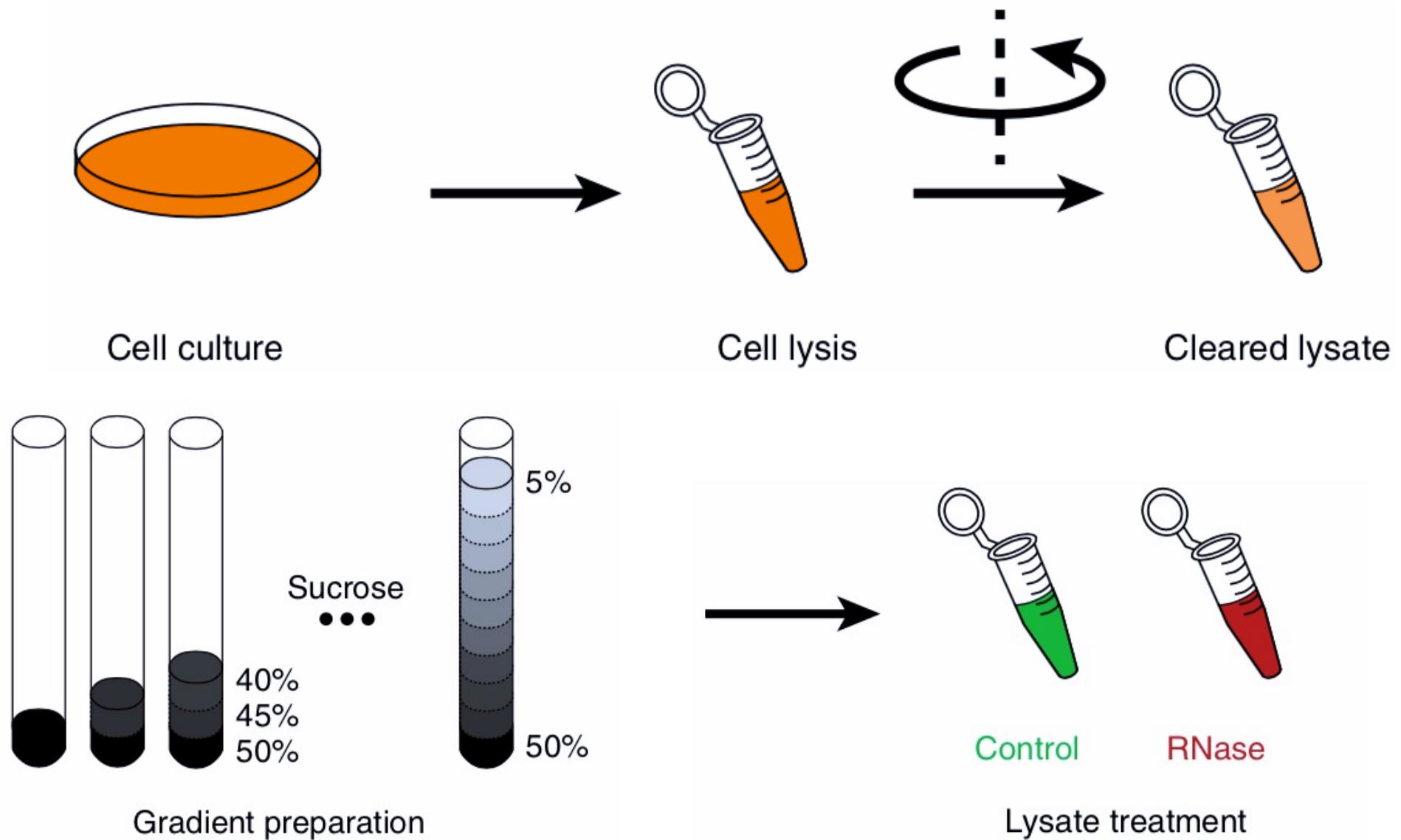
RNA dependence:
Proteins and protein complexes,
whose molecular interactions depend on RNA

**RNA-dependent
Proteins**

{ **RNA-binding protein
(RBP)**
**RBP-binding protein
(if RBP bound to RNA)**

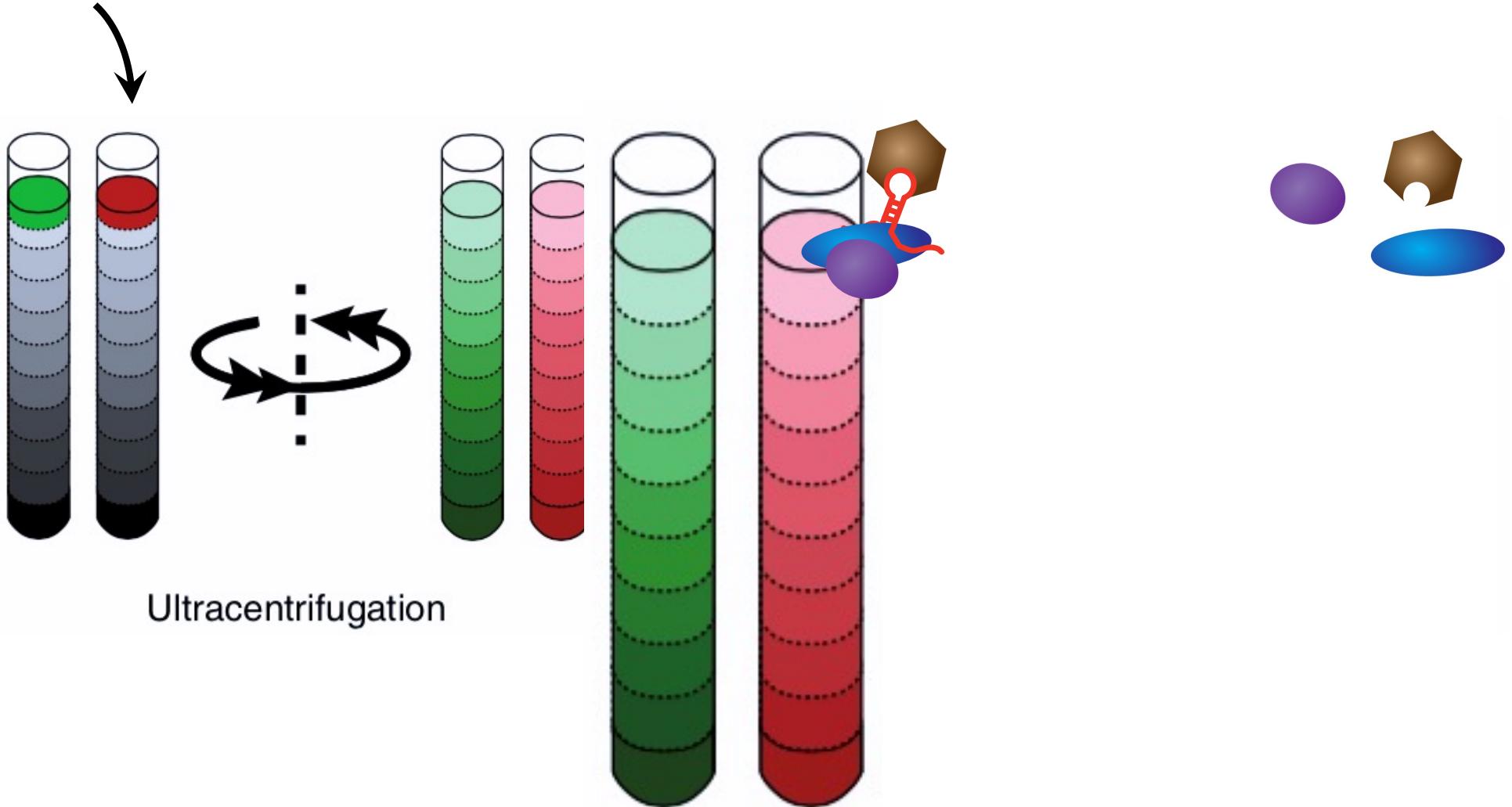


R-DeeP Screen: Density Gradient Fractionation



Caudron-Herger et al. Nat Protoc 2020

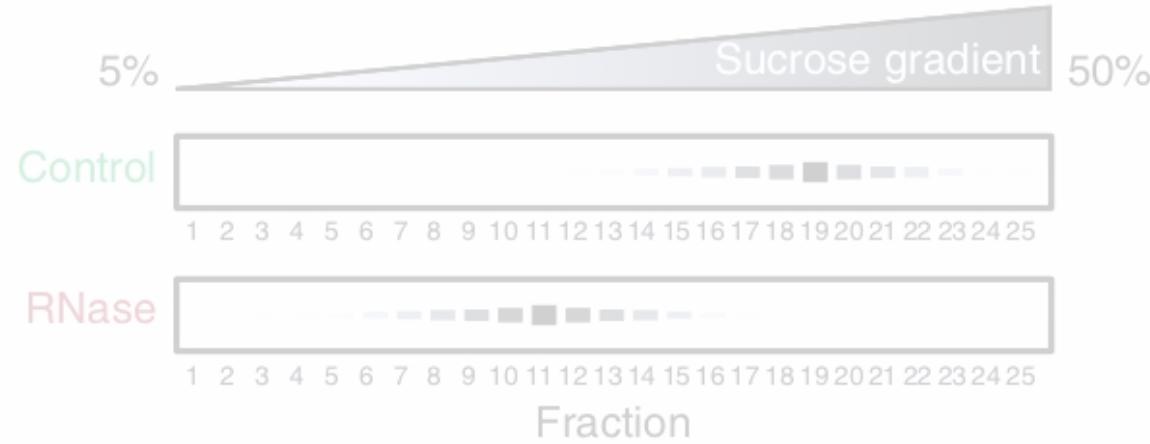
R-DeeP Screen: Density Gradient Fractionation



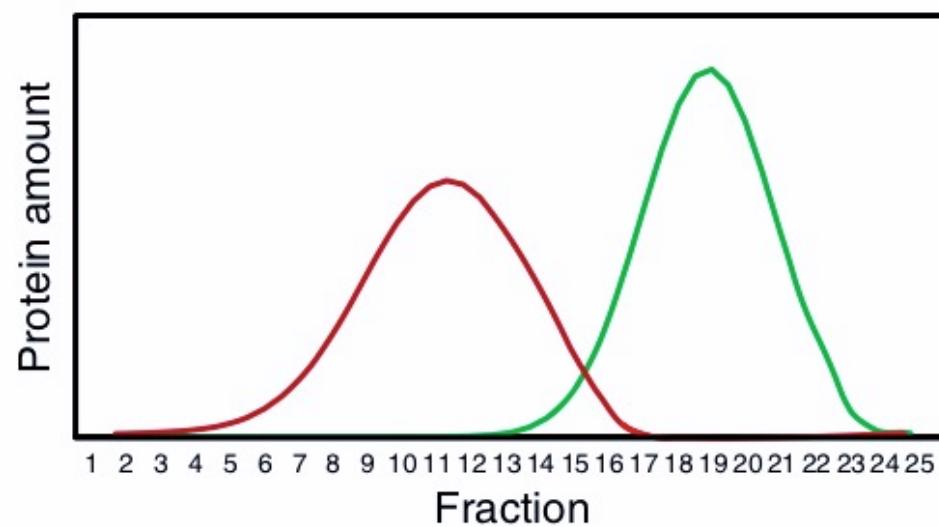
Caudron-Herger et al. Nat Protoc 2020

R-DeeP Screen: Western blot analysis

Western Blot Analysis



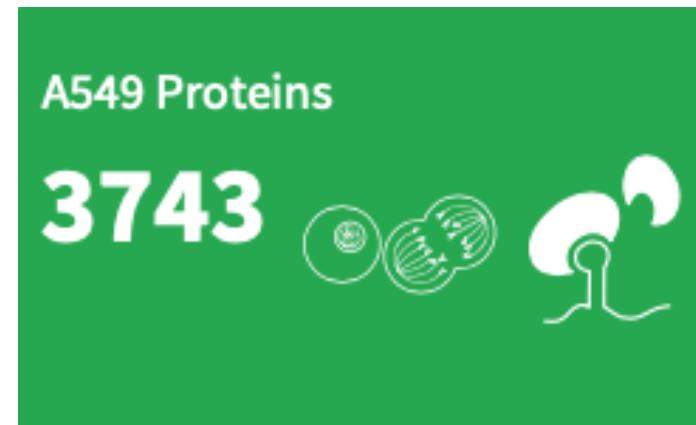
Mass Spectrometry Analysis



Caudron-Herger et al. Nat Protoc 2020

R-DeeP Datasets

Unsynchronized = mixed of different cell cycle phases



Synchronized = specific cell cycle phase



Analysis of the Mass Spectrometry Data

Dataset

Proteins ↓

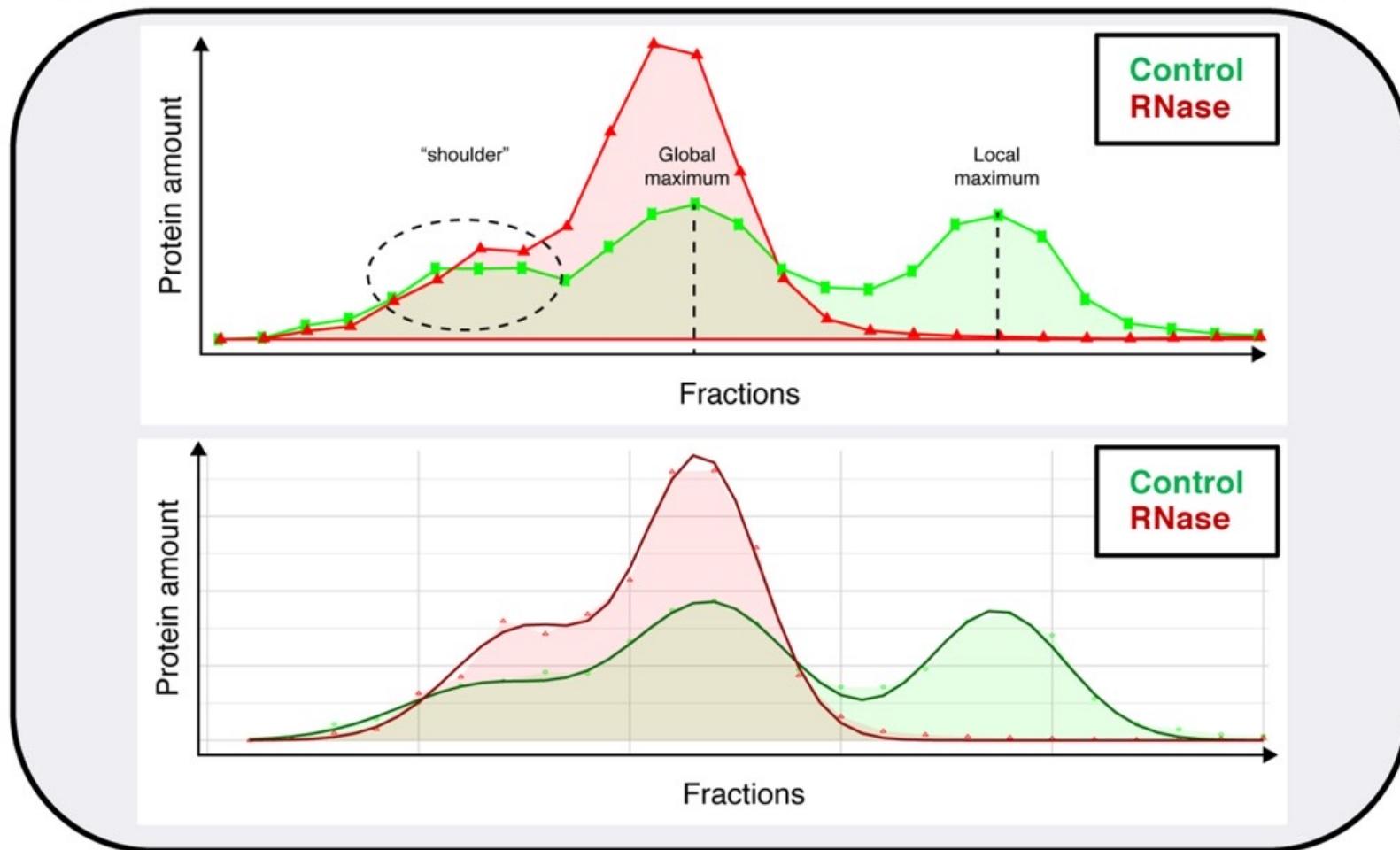
Fractions →

	ctrl1.01	rnase1.01	ctrl2.01	rnase2.01	ctrl3.01	rnase3.01	ctrl1.02	rnase1.02	ctrl2.02	rnase2.02
AHNK_HUMAN	1776.21	6255.75	7425.759	7407.75	8496.14	119191.498	30490.92	26878.59	19694.137	792765.57
NUCL_HUMAN	245790.45	131631.78	228390.701	143404.11	234347.133	375159.18	77925.52	87250.307	46215.575	650379.04
DDX21_HUMAN	4160.08	838.901	4565.32	1065.21	3444.76	5682.59	0	0	0	0
HNRPU_HUMAN	123166.896	8726.286	121454.669	13419.524	166212.35	776750.31	25644.232	12747.712	38348.274	3024203.4
NOLC1_HUMAN	26928.23	6739.17	26026.81	6646.33	29827.73	191081.3	20934.977	28179.84	17917.394	1062686
FLNB_HUMAN	0	0	0	0	0	0	0	0	0	0
DDX5_HUMAN	0	0	0	0	0	0	0	0	0	9494.41
SPTN1_HUMAN	0	0	0	0	0	0	0	0	0	0
ECHA_HUMAN	0	0	0	0	0	0	0	0	0	0
TFR1_HUMAN	0	0	0	0	0	0	0	0	0	0
TBB2A_HUMAN	3761.29	3529.95	4407.34	1927.89	5859.63	83841.4	8245.419	2545.65	7718.404	218850.1
LMNA_HUMAN	866.512	1743.31	2393.261	4908.105	8372.076	1049019.69	7399.795	4731.085	20638.237	3273084.03
EIF3A_HUMAN	0	0	0	0	0	0	0	0	0	0
HNRPQ_HUMAN	1300.142	1239.218	3609.4	0	5447.885	210971.49	106951.274	119616.866	118844.273	1199009.1
HS90A_HUMAN	0	0	0	0	490.622	24909.3	16227.356	8584.171	27297.006	926919.28
G3P_HUMAN	42420.682	73567.54	27846.01	76105.85	62379.33	241602.9	102092.28	60758.99	118202.88	1316499

Part 1:

- **Description of the dataset**
- **Cleaning / Reproducibility check**
- **Normalization**

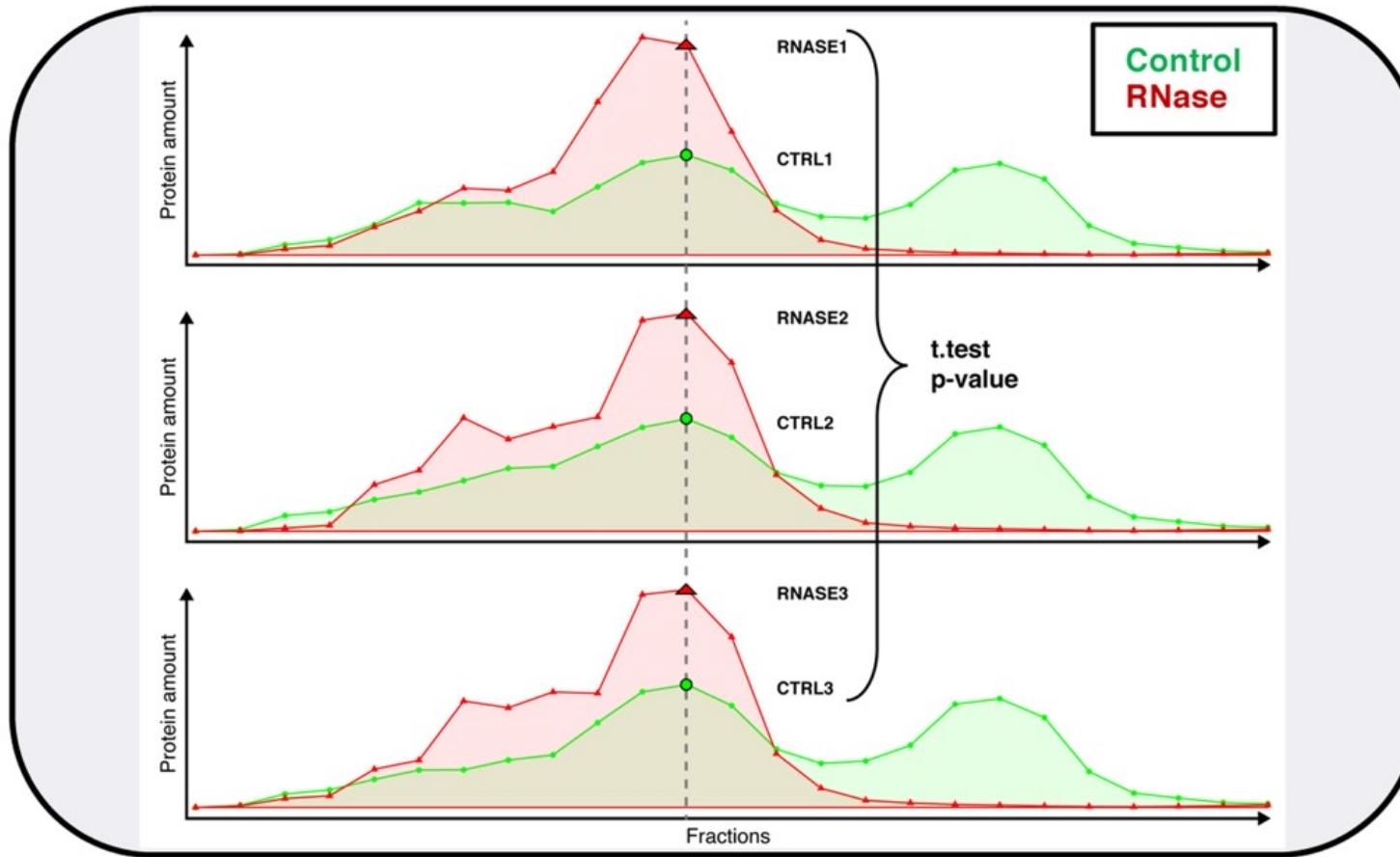
Analysis of the Mass Spectrometry Data



Part 2:

- Find the maxima for each protein in control and RNase-treated samples

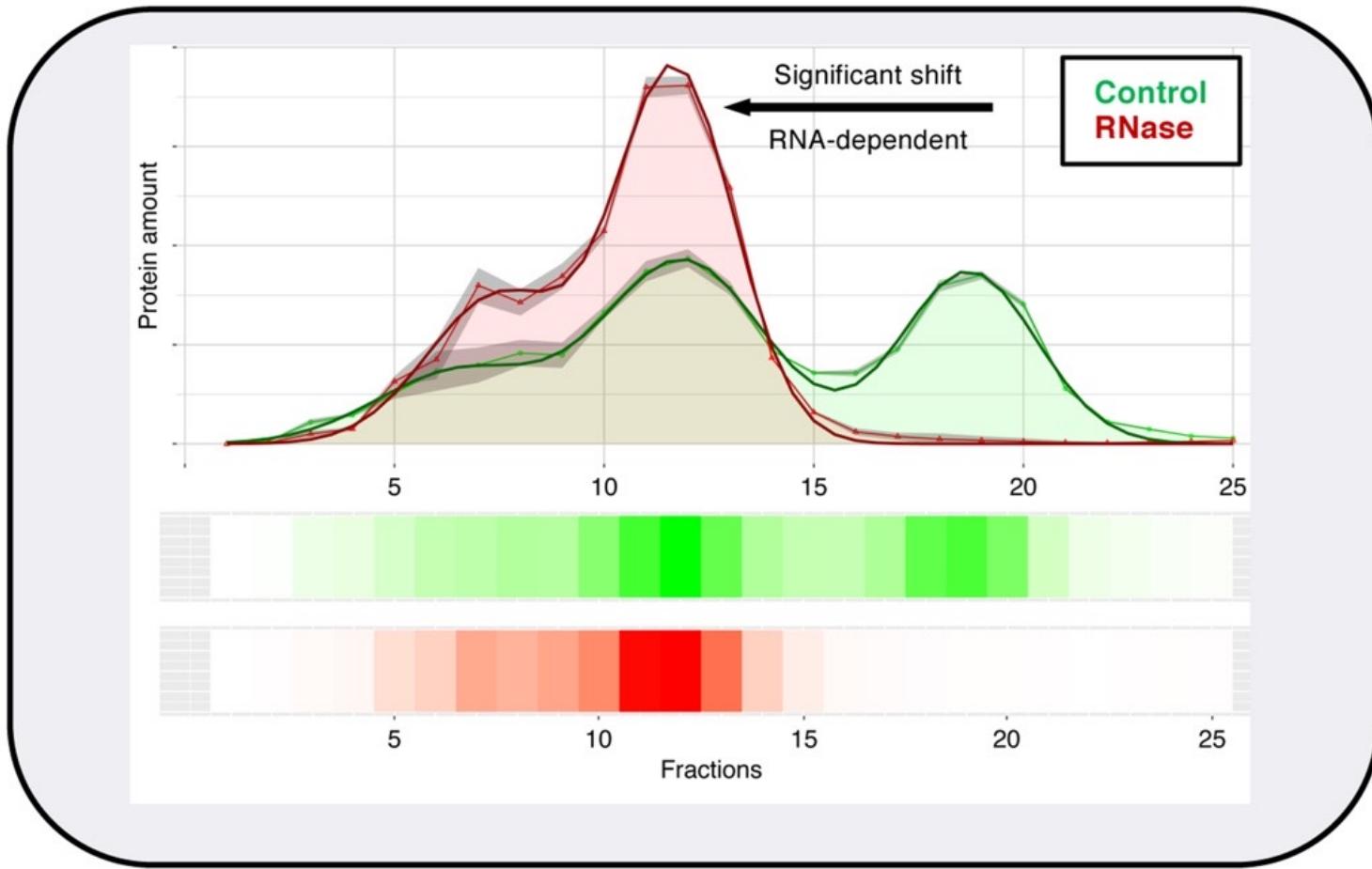
Analysis of the Mass Spectrometry Data



Part 3:

- Evaluate the differences at the maxima
- Define selection criteria for RNA-dependent proteins

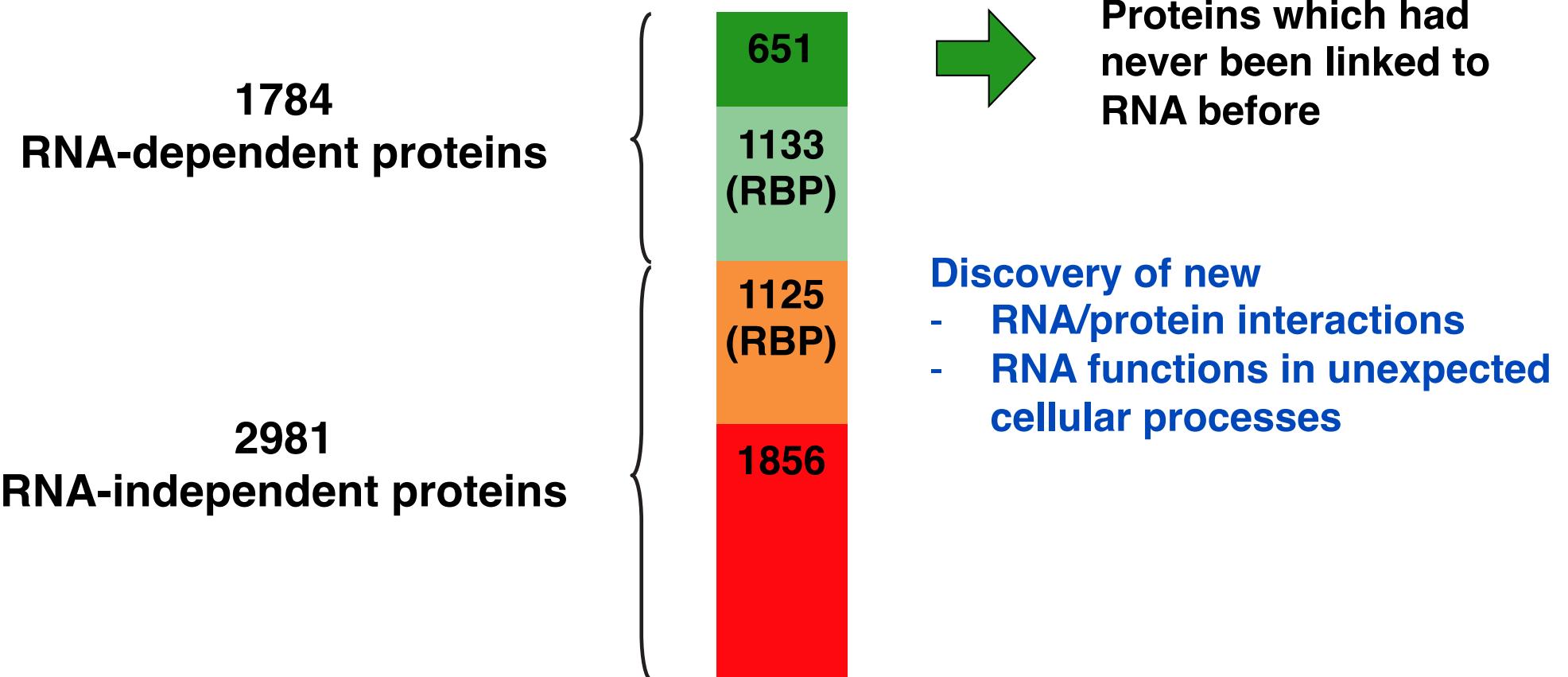
Analysis of the Mass Spectrometry Data



Part 4:

- **Apply selection criteria**
- **Produce graphics for specific proteins or groups**
- **Perform a linear regression**

Part 5: further analysis of the results



Caudron-Herger et al., Mol Cell, 2019

R-DeeP: Database for RNA-dependent Proteins

<http://R-DeeP3.dkfz.de>

 = A Database for Screens on RNA-Dependent Proteins i

[Home](#)

[Advanced search](#)

[Single search](#) <

Unsynchronized cells

- HeLa S3 cells
- A549 cells

Synchronized cells

- HeLa S3 cells - Interphase
- HeLa S3 cells - Mitosis

[Documentation](#)

[References](#)

[About](#)

Welcome to R-DeeP 3.0

R-DeeP 3.0 is a database on RNA-dependent proteins in unsynchronized HeLa S3 and A549 cells, and during either mitosis or interphase synchronized HeLa S3 cells resulting from the concept of RNA dependence and its experimental conversion

Advanced Search

Search for RNA-dependent proteins in all R-DeeP 3.0 cell lines and enter lists of proteins to your search.

R-DeeP 3.0 Proteins
8708 

Single Search

Search for RNA-dependent proteins in a specific cell line

Proteins of Unsynchronized Cells

HeLa S3 Proteins
4765 

A549 Proteins
3743 

Proteins of Synchronized Cells

HeLa S3 - Interphase Proteins
7069 

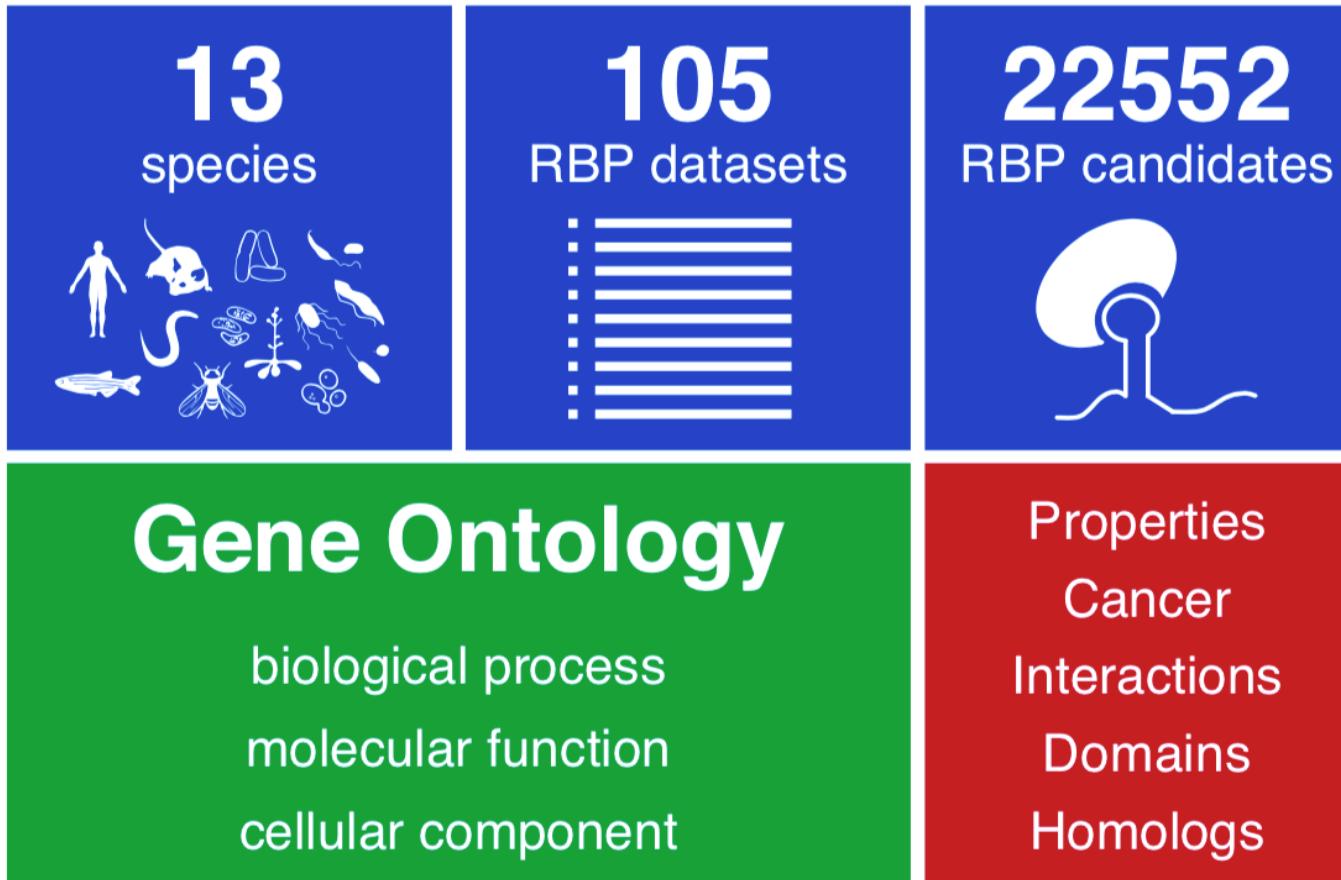
HeLa S3 - Mitosis Proteins
7152 

Rajagopal V, Seiler J et al., Nature Communications, 2025

RBP2GO: a new pan-species RBP database



RBP2GO.dkfz.de



or **RBP2GO-2-beta.dkfz.de** (detailed protein domain information)

Caudron-Herger et al., NAR 2021

Wassmer et al., NAR 2024

UniProt BLAST Align Peptide search ID mapping SPARQL

Release 2025_01 | Statistics 📈 💾 📧 Help

Find your protein

UniProtKB ▾ | Examples: Insulin, APP, Human, P05067, organism_id:9606 Advanced | List Search

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Proteins
UniProt Knowledgebase

Reviewed (Swiss-Prot) 572,970
Unreviewed (TrEMBL) 252,633,200

Species
Proteomes

Protein sets for species with sequenced genomes from across the tree of life

Protein Clusters
UniRef

Clusters of protein sequences at 100%, 90% & 50% identity

Sequence archive
UniParc

Non-redundant archive of publicly available protein sequences seen across different databases

Help

Questions?

- Information in the GitHub repository
 - Project description
 - References
 - Link to databases
 - Folien
- Per email: m.caudron@dkfz.de

Viel Erfolg und viel Spaß!

