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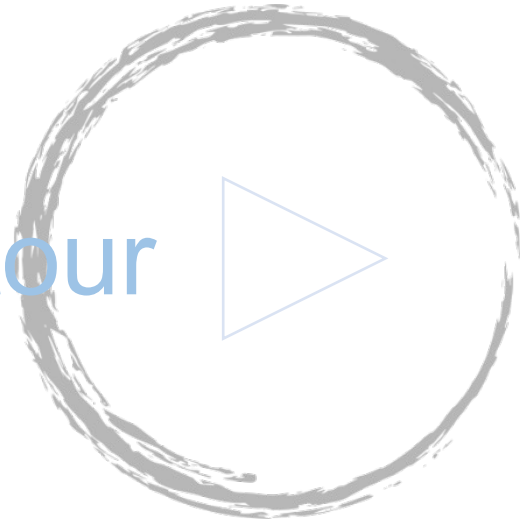

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In silico methods

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- quantitative analysis of proteins
- analysis of biological activity of proteins
- analysis of physical and chemical properties of proteins

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- Biuret
- Warburg-Christian
- Smith
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Bradford assay

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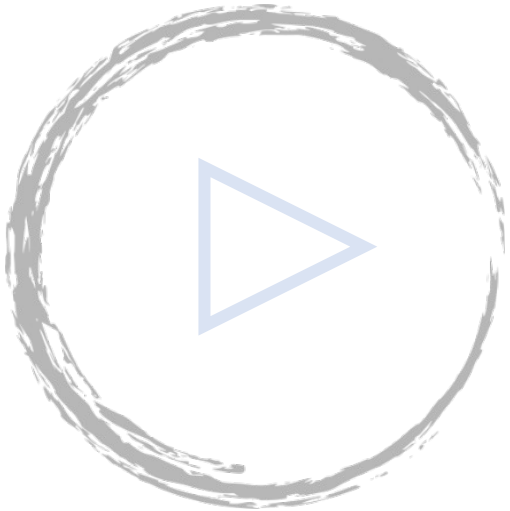

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Bradford assay

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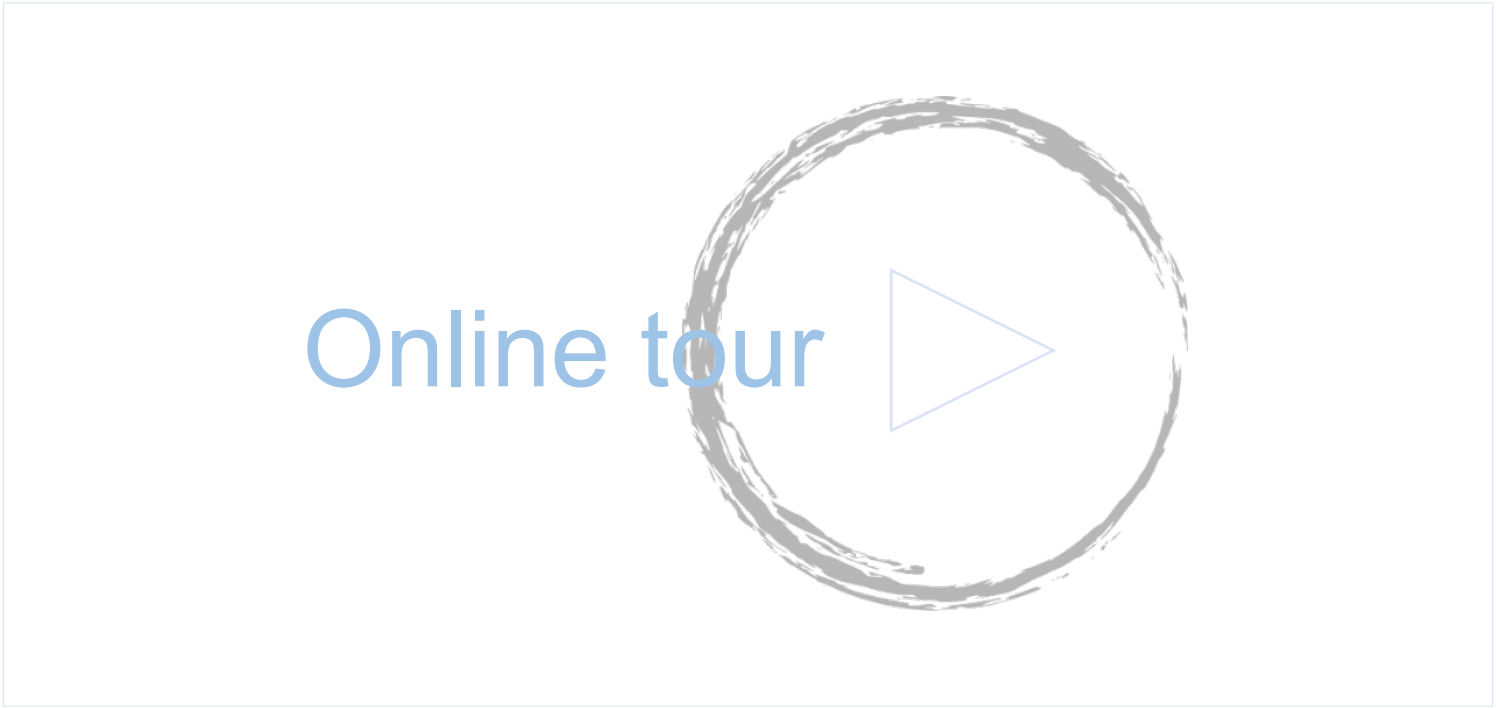

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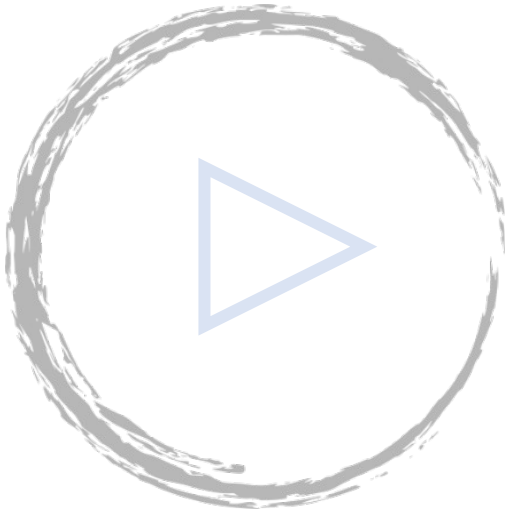

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Quantitative analysis of proteins

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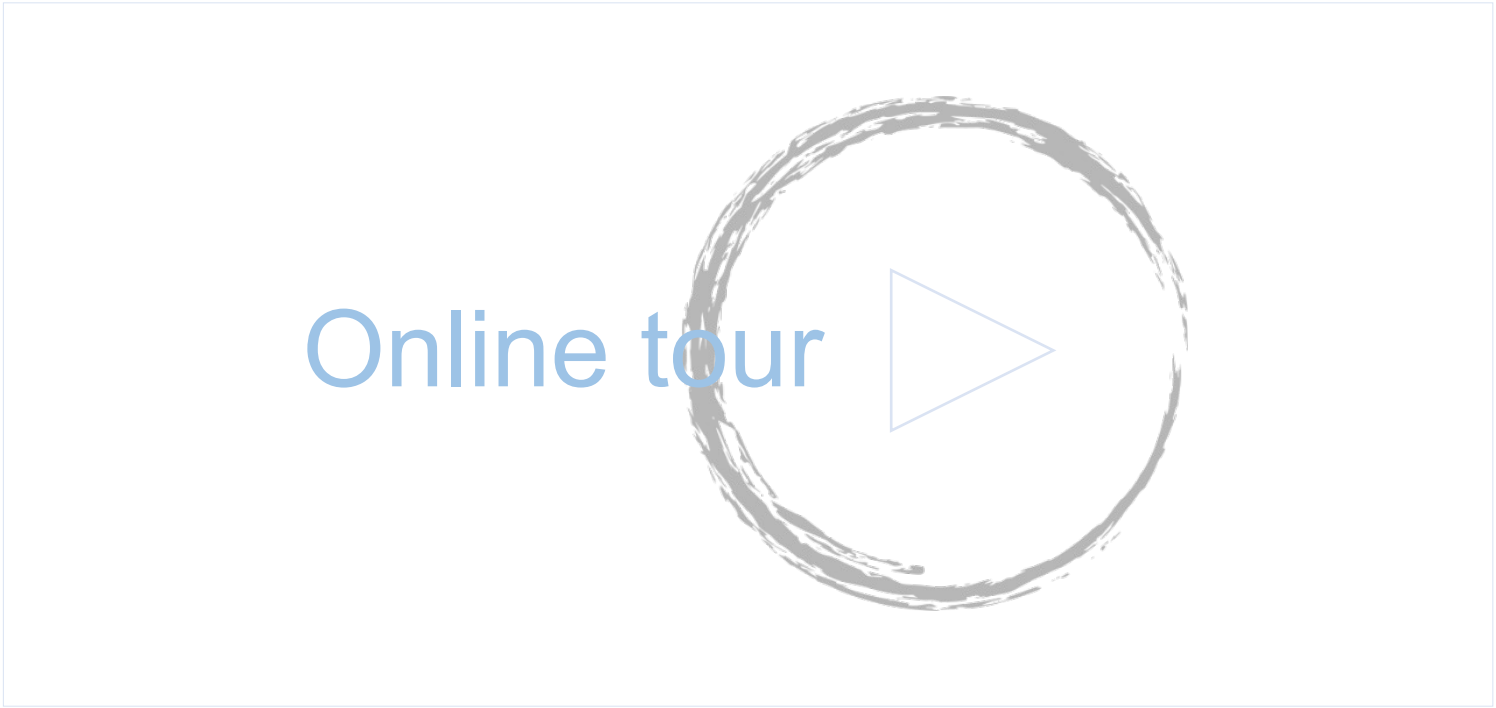

Relevant research & **E**asy learning

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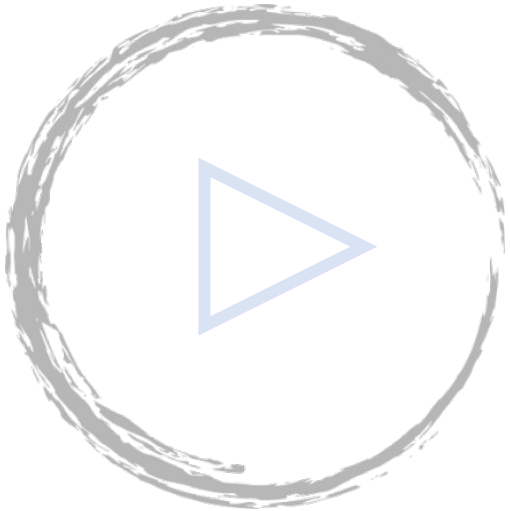

Method principle

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Bradford Assay/ general information /

Method for quantify the protein content in sample. This method has multiple applications in experimental sciences. Chemical basis of the Bradford method (1976) is based on the absorbance shift observed in an acidic solution of dye Coomassie® Brilliant Blue G-250. When added to a solution of protein, the dye binds to the protein resulting in a colour change from a reddish brown to blue.

+add information

References:

1. Bradford MM A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. // Analytical Biochemistry. 1976. № 72. С. 248-254.
2. Pedrol, Nuria & Tamayo, Pilar. (2001). Protein Content Quantification by Bradford Method. 10.1007/0-306-48057-3_19.

Bradford Assay/ general information / chemical basis /

The dye has been assumed to bind to protein via electrostatic attraction of the dye's sulfonic groups to the protein. The bound points are primarily arginine residues, but the dye also binds to a lesser degree to histidine, lysine, tyrosine, tryptophan and phenylalanine

+add information

References:

1. Compton and Jones, 1985

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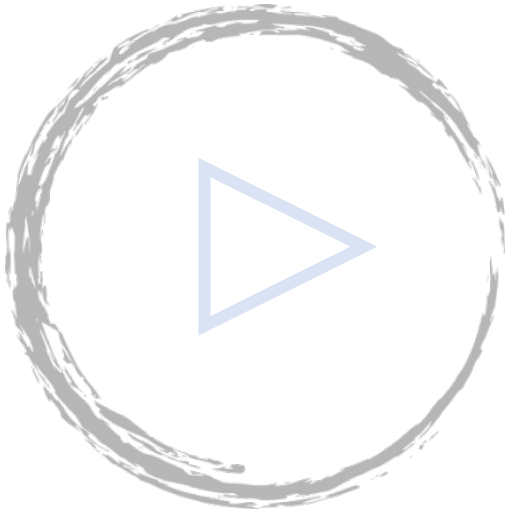

Method principle

Procedure

…

Bradford Assay/ protocol/

Safety info

1. Select a Protein Standard and sample volume

2. Remove the 1x Bradford dye reagent from 4°C storage and let it warm to ambient temperature. Invert the 1x dye reagent a few times before use.

3. Prepare the protein standard if needed.

4. Put your unknown sample to the tube (or cuvette)

5. Add the 1x dye reagent to each tube (or cuvette) and vortex (or invert).

6. Incubate at room temperature for at least 5 min. Samples should not be incubated longer than 1 hr at room temperature.

7. Set the spectrophotometer to 595 nm. Zero the instrument with the blank sample (not required for microplate readers). Measure the absorbance of the standards and unknown samples.

8. Data analysis

References:

Bradford Assay/ protocol/

Set the parameters	Download	Visualization	Safety info	+add information

1. Select a Protein Standard and sample volume

2. Remove the 1x Bradford dye reagent from 4°C storage and let it warm to ambient temperature. Invert the 1x dye reagent a few times before use.

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7. Set the spectrophotometer to 595 nm. Zero the instrument with the blank sample (not required for microplate readers). Measure the absorbance of the standards and unknown samples.

8. Data analysis

References:

Bradford Assay/ protocol/ parameters /

Sample volume	...
Standard protein	...

apply parameters

Bradford Assay/ protocol/ parameters /

Sample volume	· 1 ml cuvette · 250 µl microplate · 5 ml cuvette
Standard protein	

apply parameters

Bradford Assay/ protocol/ parameters /

Sample volume	1 ml
Standard protein	BSA

apply parameters

Bradford Assay/ protocol/

Set the parameters	Download	Visualization	Safety info	+add information	Ready to collaborate

1. Prepare BSA Standard and your samples.
2. Remove the 1x Bradford dye reagent from 4°C storage and let it warm to ambient temperature. Invert the 1x dye reagent a few times before use.

3. Put 100 µl of each BSA standard to the own tube (or cuvette)

4. Put 100 µl of the sample to the own tube (or cuvette)

5. Add 500 µl of 1x Bradford dye reagent to each tube (or cuvette) and vortex (or invert).

6. Incubate at room temperature for at least 5 min. Samples should not be incubated longer than 1 hr at room temperature.

7. Set the spectrophotometer to 595 nm. Zero the instrument with the blank sample (not required for microplate readers). Measure the absorbance of the standards and unknown samples.

8. Data analysis

References

1. Bradford MM A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. // Analytical Biochemistry. 1976. № 72. C. 248-254.

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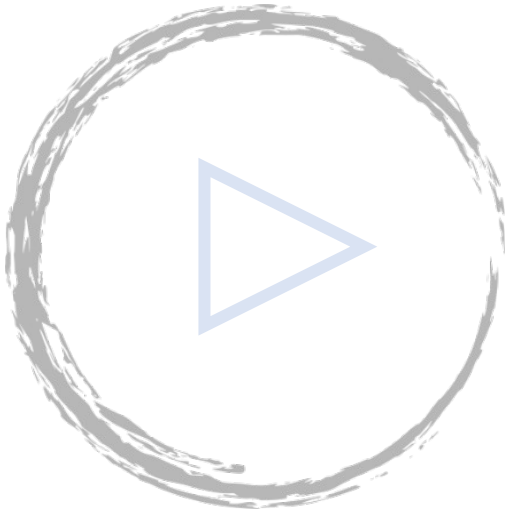

Method principle

Procedure

…

Bradford Assay/ equipment and reagents required /

Materials:

- Diluted protein sample

Equipment:

- Spectrophotometer
- Micropipette
- Plastic tubes or cuvette

Reagents:

- Bradford dye reagent
- Standard protein

Create a list of required equipment and reagents

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Find method …

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Bradford assay

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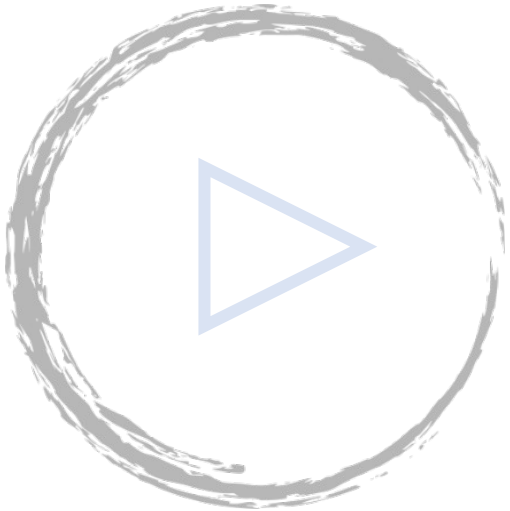

Method principle

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Bradford Assay/ application /

quantitative analysis of proteins

Find method …

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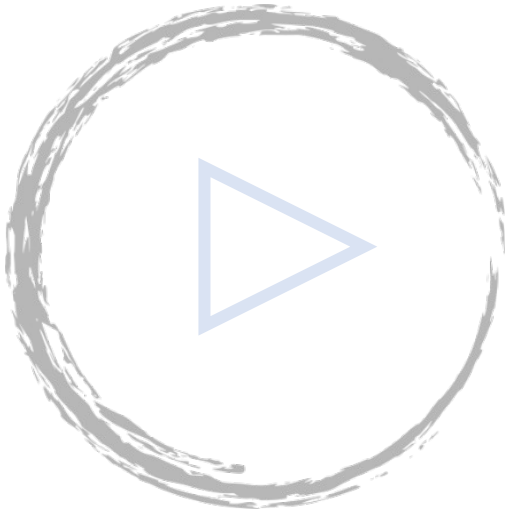

Method principle

Procedure

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Bradford Assay/ Method advantages and disadvantages

time	complexity	sensitivity	reproducibility	accuracy	cost
15 min	+	+	+++	+++	+

Compare with other methods

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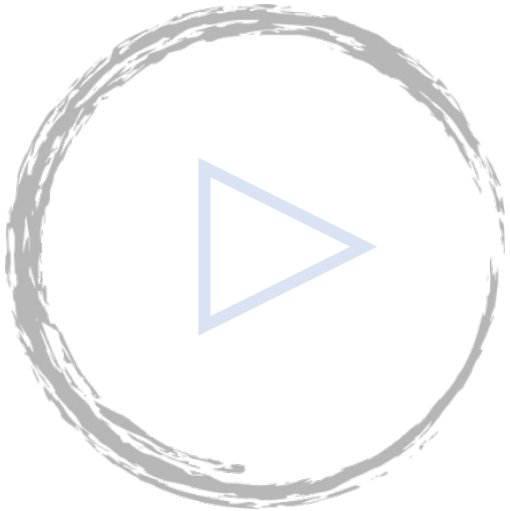

Method principle

Procedure

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Bradford Assay/ Troubleshooting

1. Reagents compatible with the Quick Start Bradford protein assay when using the standard procedure

Bradford Assay/ Troubleshooting / Compatible reagents

Acetone, 10%
Acetonitrile, 10%
Ammonium sulfate, 1 M
Ampholytes, 3–10, 0.5%
ASB-14, 0.025%
Ascorbic acid, 50 mM
Bis-Tris, pH 6.5, 0.2 M
β-mercaptoethanol, 1 M
Calcium chloride, 40 mM
CHAPS, 10%
CHAPSO, 10%
Deoxycholic acid, 0.2%
DMSO, 5%
Dithioerythritol (DTE), 10 mM
Dithiothreitol (DTT), 10 mM
Eagle's MEM
Earle's salt solution
EDTA, 0.2 M
EGTA, 0.2 M
Ethanol, 10%
Glucose, 20%
Glycerol, 5%
Glycine, 0.1 M
Guanidine-HCl, 2 M
Hank's salt solution
HCl, 0.1 M
HEPES, 0.1 M
Imidazole, 0.2 M
Magnesium chloride, 1 M
MES, 0.1 M
Methanol, 10%
Modified Dulbecco's PBS
MOPS, 0.1 M
NAD, 2 mM
Nonidet P-40, 0.25%
Octyl β-glucoside, 0.5%
Octyl β-thioglucopyranoside, 1%
PBS
Phenol Red, 0.5 mg/ml
PIPES, 0.2 M
PMSF, 2 mM
Potassium chloride, 2 M
Potassium phosphate, 0.5 M
SB 3–10, 0.1%
SDS, 0.025%
Sodium acetate, pH 4.8, 0.2 M
Sodium azide, 0.5%
Sodium bicarbonate, 0.2 M
Sodium carbonate, 0.1 M
Sodium chloride, 2.5 M
Sodium citrate, pH 4.8 or 6.4, 0.2 M
Sodium hydroxide, 0.1 M
Sodium phosphate, 0.5 M
Sucrose, 10%
TBP, 5 mM
TBS (25 mM Tris, 0.15 M NaCl, pH 7.6), 0.5x
TCEP, 20 mM
Thio-urea, 1 M
Tricine, pH 8, 50 mM
Triethanolamine, pH 7.8, 50 mM
Tris, 1 M
Tris-glycine (25 mM Tris, 192 mM glycine)
Tris-glycine-SDS, (25 mM Tris, 192 mM glycine, 0.1% SDS), 0.5x
Triton X-100, 0.05%
Tween 20, 0.01%
Urea, 4 M

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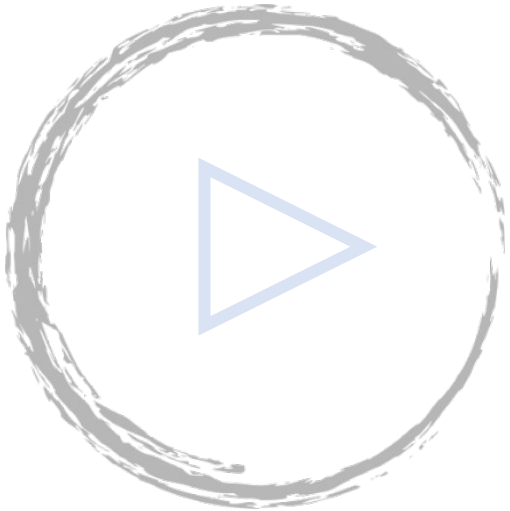

Method principle

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Bradford Assay/ Find collaboration

Country

City

Organization

Search

Bradford Assay/ Find collaboration

Saint-Petersburg, Russia

Sate University (2)	contact
ITMO(1)	contact
SPSPU(1)	contact

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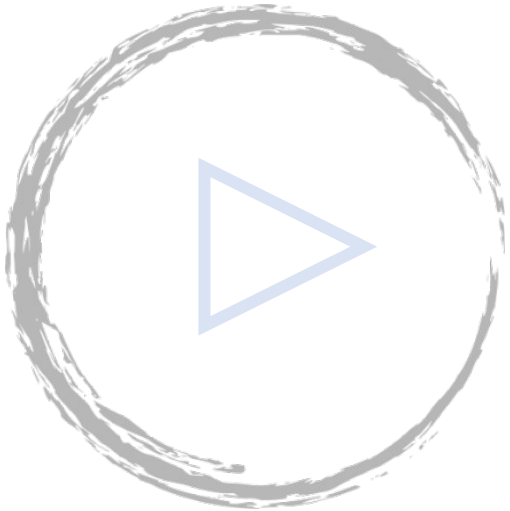

Method principle

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Bradford Assay/ Education

Country

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Form

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Bradford Assay/ Education

Sate University (2)	Course program
ITMO(1)	Course program
SPSPU(1)	Course program

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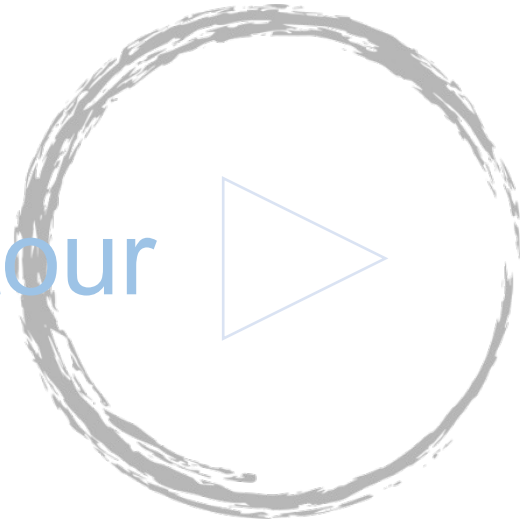

Relevant research & **E**asy learning

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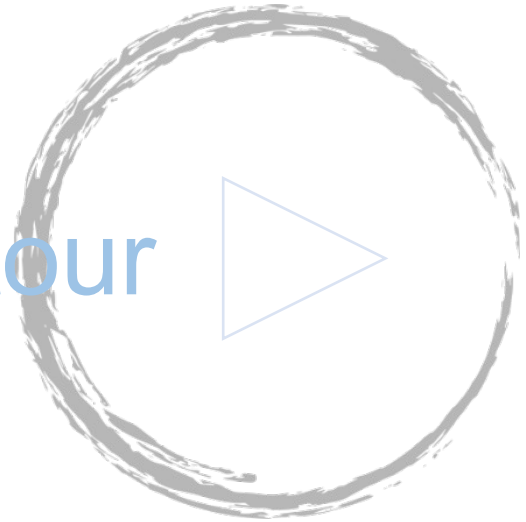

Relevant research & **E**asy learning

Name: Yankelevich Irina

Education: Saint-Petersburg State Chemical-Pharmaceutical University

Degree: PhD

Position: researcher

Sphere of interests: immunology, biotechnology, recombinant technology

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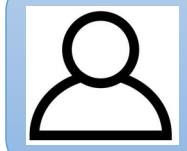
(5) | Professional Area (3) | My projects | Lab mates | Saved protocols



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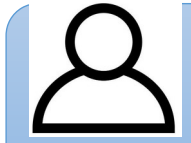

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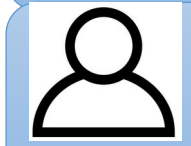

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Molecular biology  (233)

Immunology  (280)

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- Requests in my professional competence**(1)**
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1. TLR-4 expression in brain
2. Corticostatins

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- **Protocols**
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Masha

John

Peter

1. *Bradford assay*
2. *SDS PADE electrophoresis*
3. *E. coli transformation*