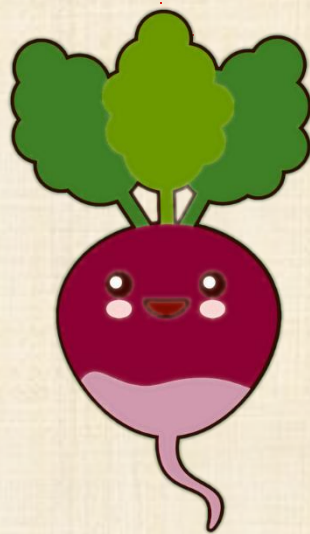


Gamtamoksliai tyrimai mokykloje

Laboratorinis darbas

Augalų ląstelių
membranų pralaidumo tyrimas

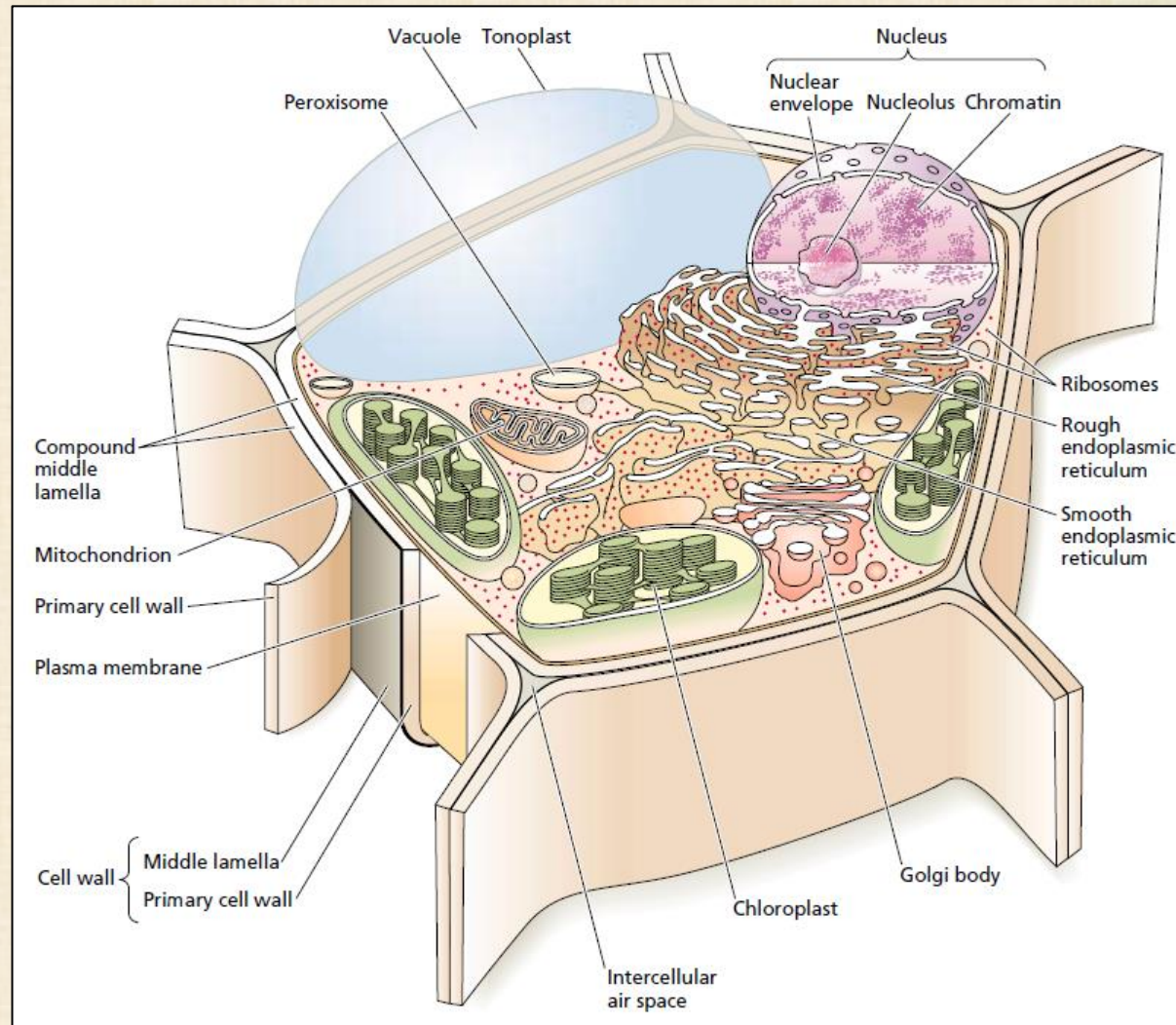


2023



Gyvybės mokslų
centras

Augalo ląstelė



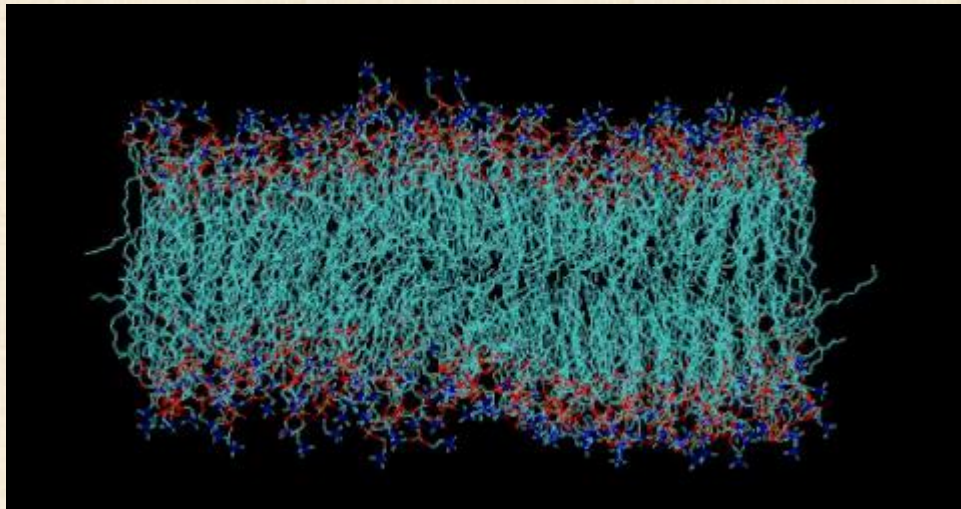
Ląstelės membrana

- Kam reikalingos membranos?
- Kaip membranų struktūra siejasi su jų funkcija?
- Kokias medžiagas membranos praleidžia, o kokių – ne?
- Kokios medžiagos gali paveikti membranų pralaidumą?
- Jei pasikeičia membranos pralaidumas, kokie struktūriniai pokyčiai tai nulemia?
- Kaip skiriasi skirtingas funkcijas atliekančios membranos?



<https://i.pinimg.com/originals/bc/22/7f/bc227f656550425132cd1a1eb372252f38g>

Membranų tyrimų istorija



<http://203.250.122.194/lecture/histology/histofigs/chapter11/00cell%20membrane.jpg>

- Membrana neatskirta nuo sienelės
- Membrana – citoplazmos „sukietėjimas“?
- Membrana sudaryta iš lipidų!

<https://images.hive.blog/p/54TLbcUcnRm3sWQK3HKkuAMedF1JSX7yKgEqYjnyTKPwrcxa37dyWUy4WgdEDNpPe8FKEejPRz7ucvfSgP5wXnyeyevzp3eh4PgtU12vF4BrQKrcNXzuf5LGXrcJPmtg4iaXHseuT?format=match&mode=fit>

Membranų tyrimų istorija

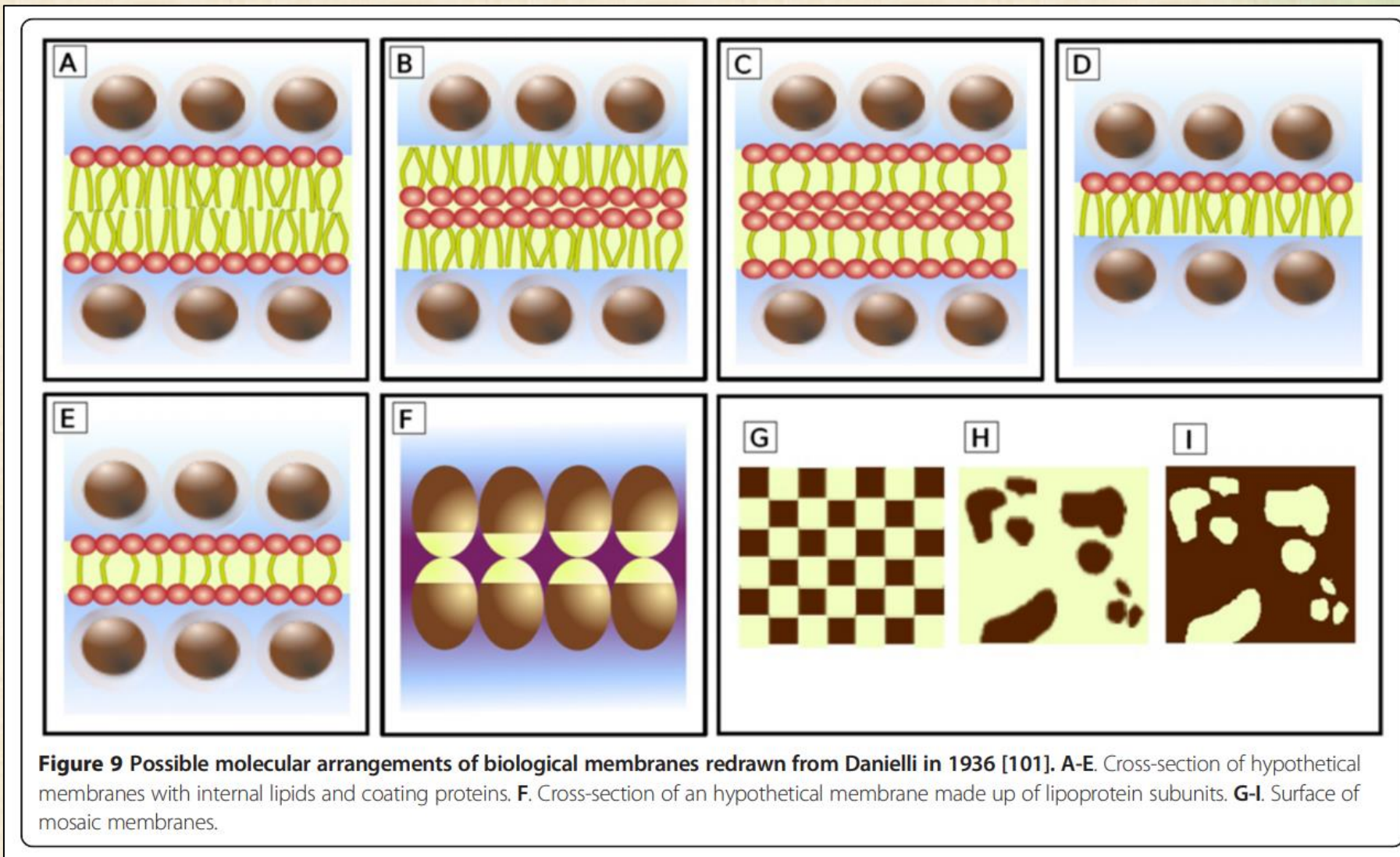
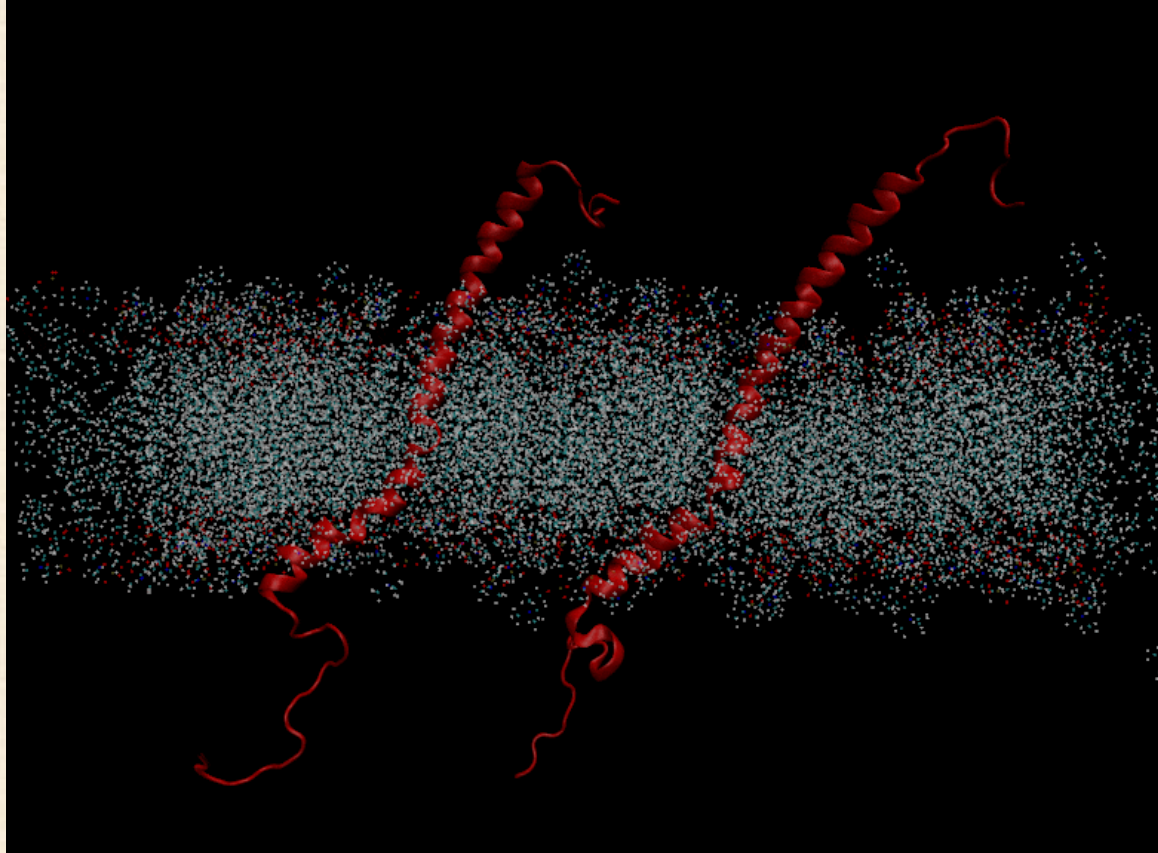
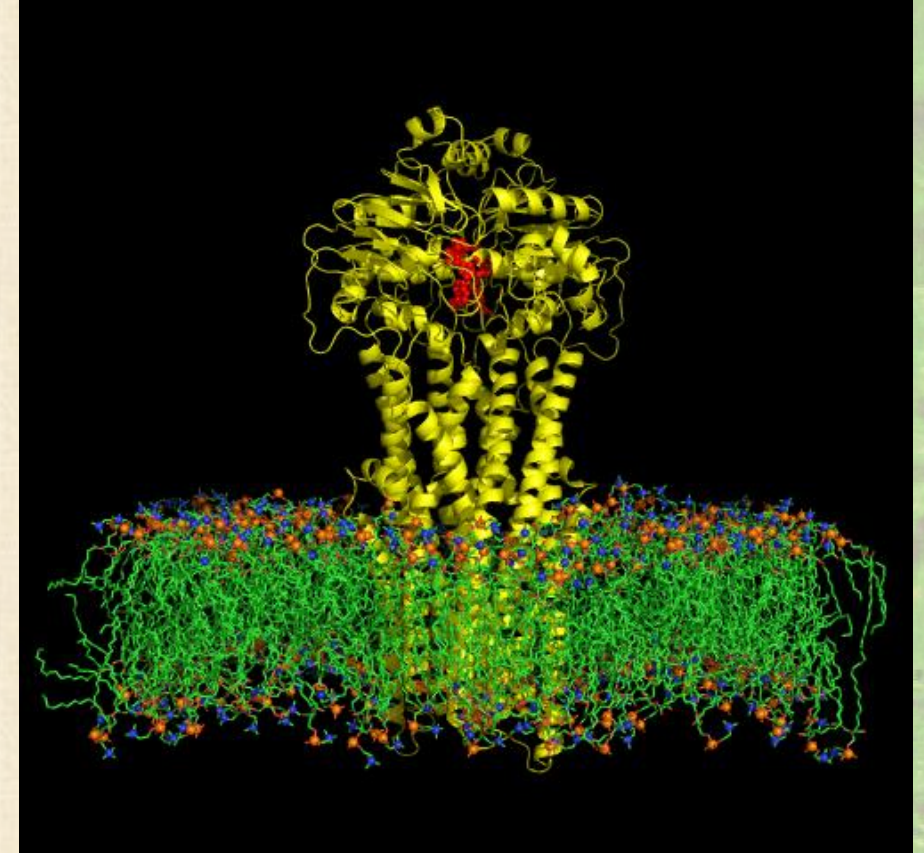


Figure 9 Possible molecular arrangements of biological membranes redrawn from Danielli in 1936 [101]. A-E. Cross-section of hypothetical membranes with internal lipids and coating proteins. **F.** Cross-section of an hypothetical membrane made up of lipoprotein subunits. **G-I.** Surface of mosaic membranes.

Membranos



<https://cs.lbl.gov/assets/Images/News/2013/Integrin-March-21/Beta-Integrin-Cytoplasmic-Interaction.gif>



<https://scifivewiki.files.wordpress.com/2016/05/md.gif>

Skystos mozaikos modelis - 1972



Membranos

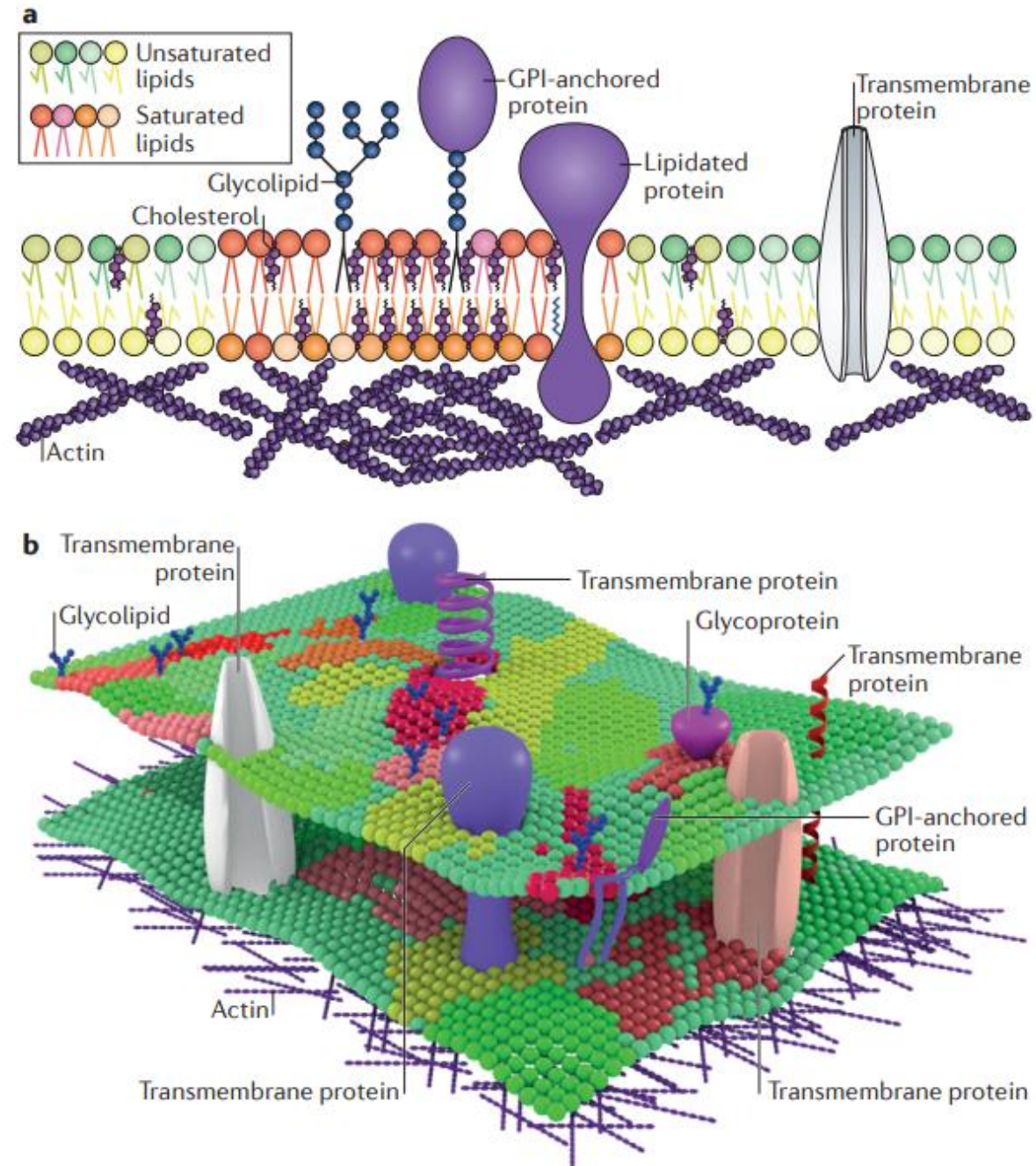
TABLE 11–1 Major Components of Plasma Membranes in Various Organisms

	Components (% by weight)			Sterol type	Other lipids
	Protein	Phospholipid	Sterol		
Human myelin sheath	30	30	19	Cholesterol	Galactolipids, plasmalogens
Mouse liver	45	27	25	Cholesterol	—
Maize leaf	47	26	7	Sitosterol	Galactolipids
Yeast	52	7	4	Ergosterol	Triacylglycerols, steryl esters
Paramecium (ciliated protist)	56	40	4	Stigmasterol	—
<i>E. coli</i>	75	25	0	—	—

Note: Values do not add up to 100% in every case because there are components other than protein, phospholipids, and sterol; plants, for example, have high levels of glycolipids.

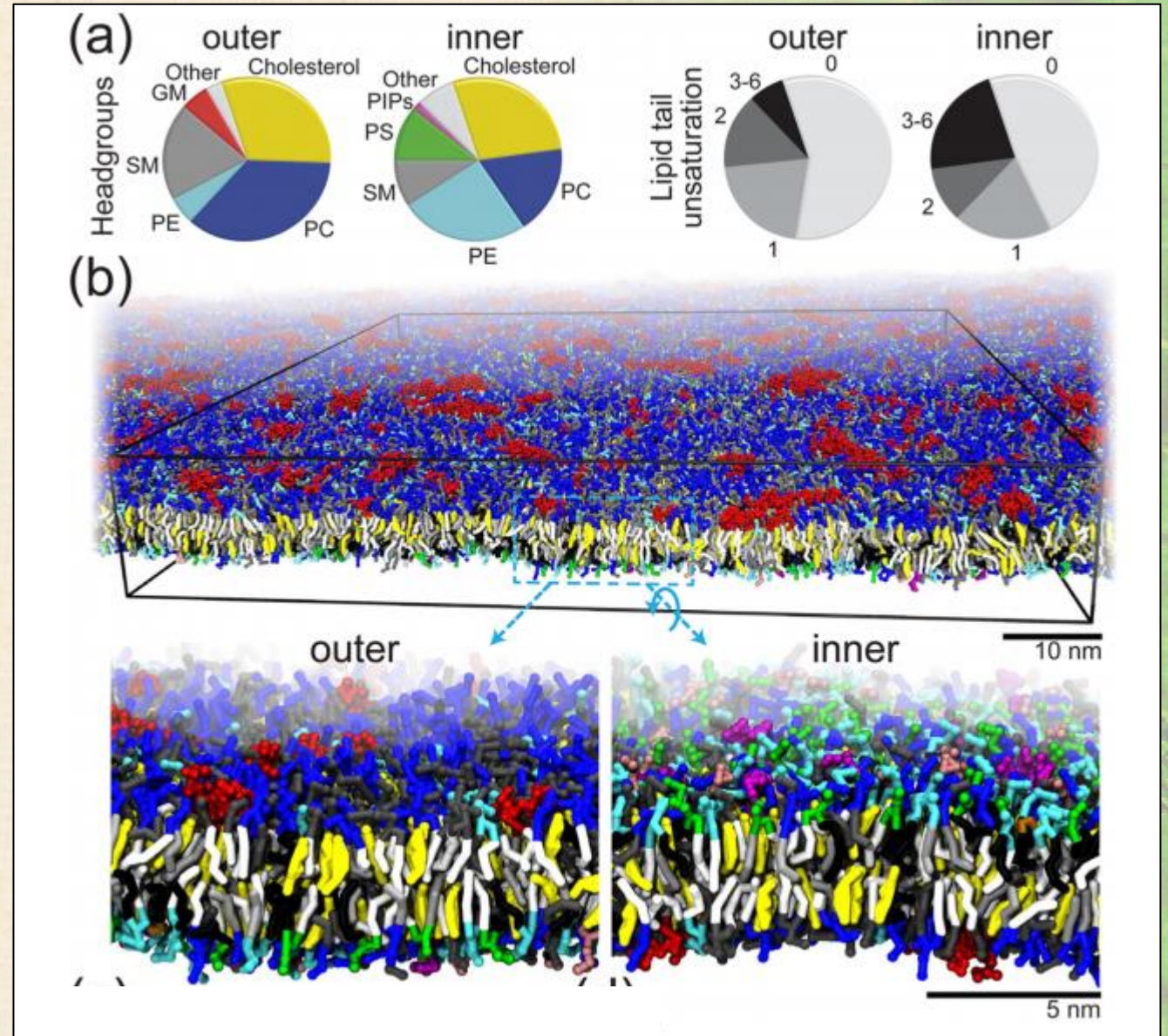
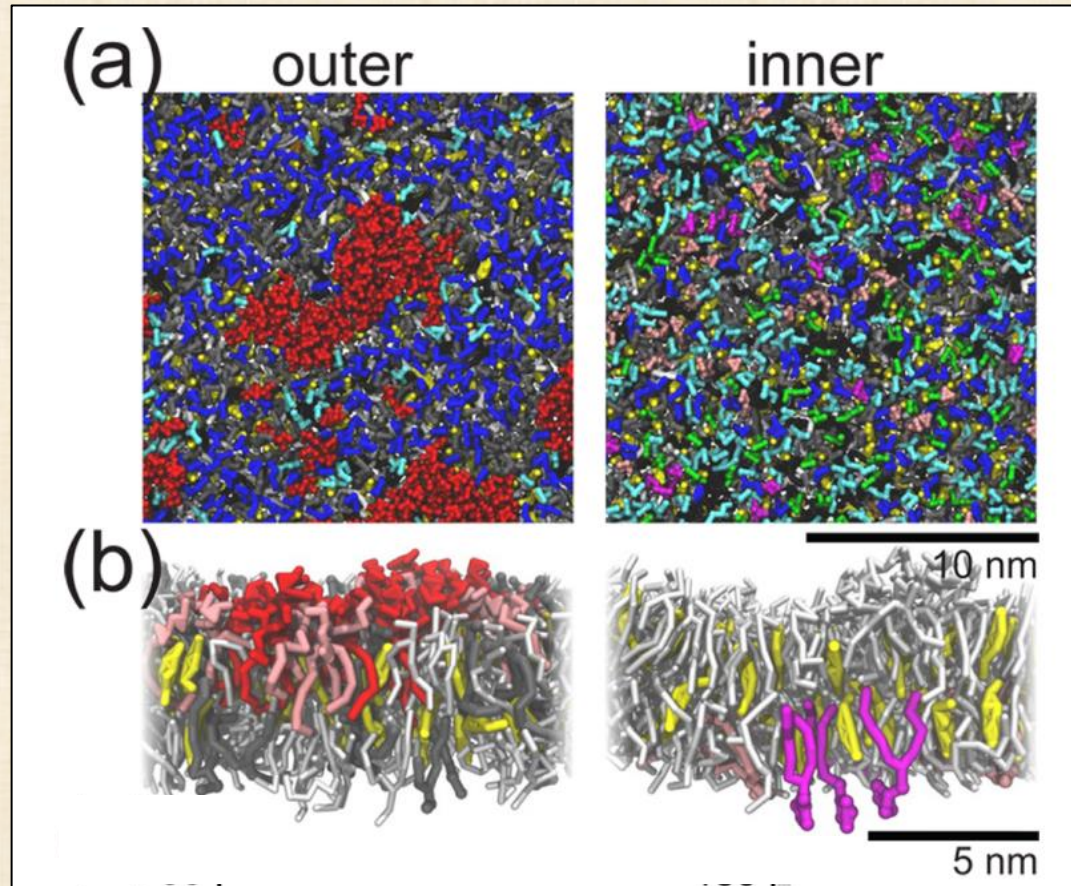


Membranos



Sezgin et al., 2017

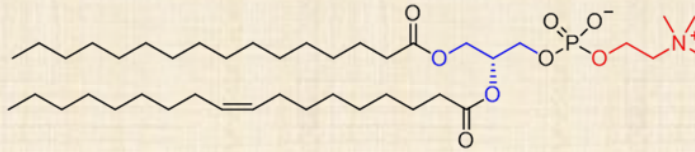
Membranos



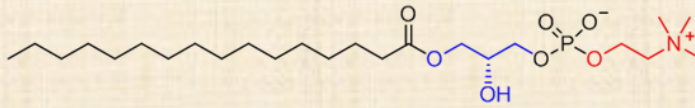
Ingólfsson et al., 2014

Fosfolipidų įvairovė

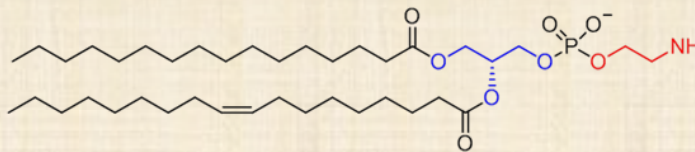
Phosphatidylcholine (PC)



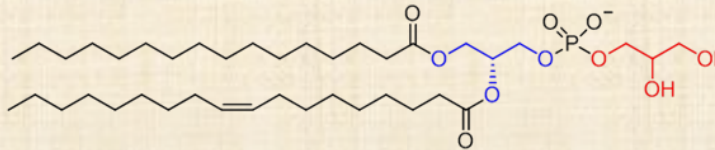
Monoacyl PC (LPC)



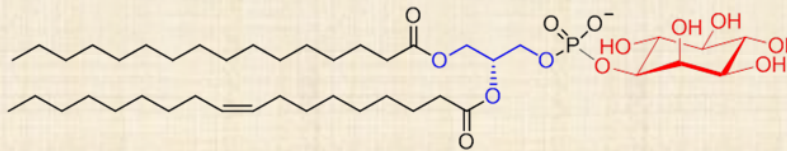
Phosphatidylethanolamine (PE)



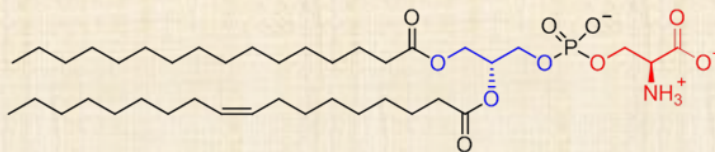
Phosphatidylglycerol (PG)



Phosphatidylinositol (PI)



Phosphatidylserine (PS)



Zwitterionic
at physiological pH

Zwitterionic
at physiological pH

Negatively charged
at basic pH

Negatively charged
at physiological pH

https://www.phospholipid-research-center.com/wp-content/uploads/2021/11/Phospholipid_chemical-structure_pH.png

Membranos

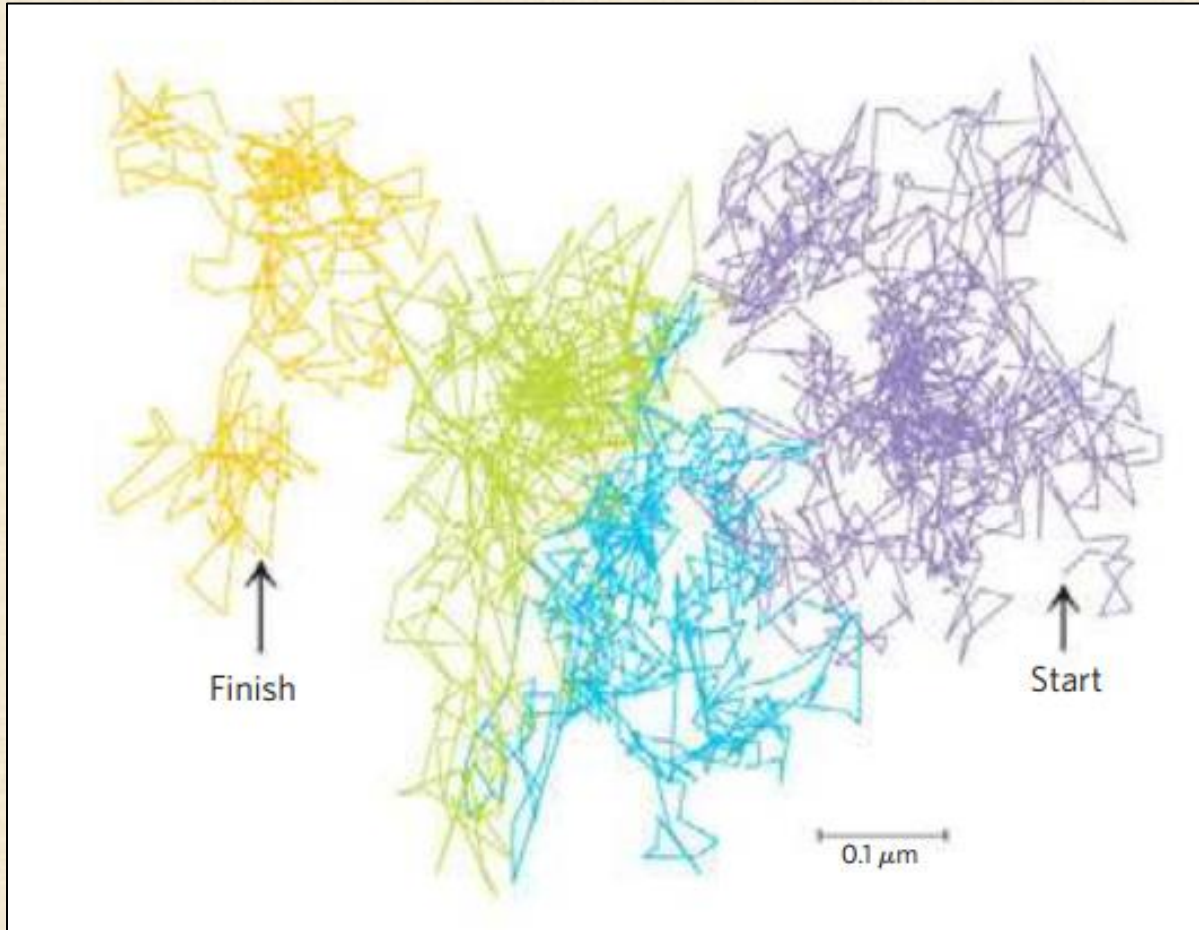


FIGURE 11-19 Hop diffusion of individual lipid molecules. The motion of a single fluorescently labeled lipid molecule in a cell surface is recorded on video by fluorescence microscopy, with a time resolution of $25\ \mu\text{s}$ (equivalent to 40,000 frames/s). The track shown here represents a molecule followed for 56 ms (2,250 frames); the trace begins in the purple area and continues through blue, green, and orange. The pattern of movement indicates rapid diffusion within a confined region (about 250 nm in diameter, shown by a single color), with occasional hops into an adjoining region. This finding suggests that the lipids are corralled by molecular fences that they occasionally jump.

Lehninger Principles of Biochemistry



Augalų membranos

Table 1 Lipid content of plant plasma membrane, expressed as percent of total lipids

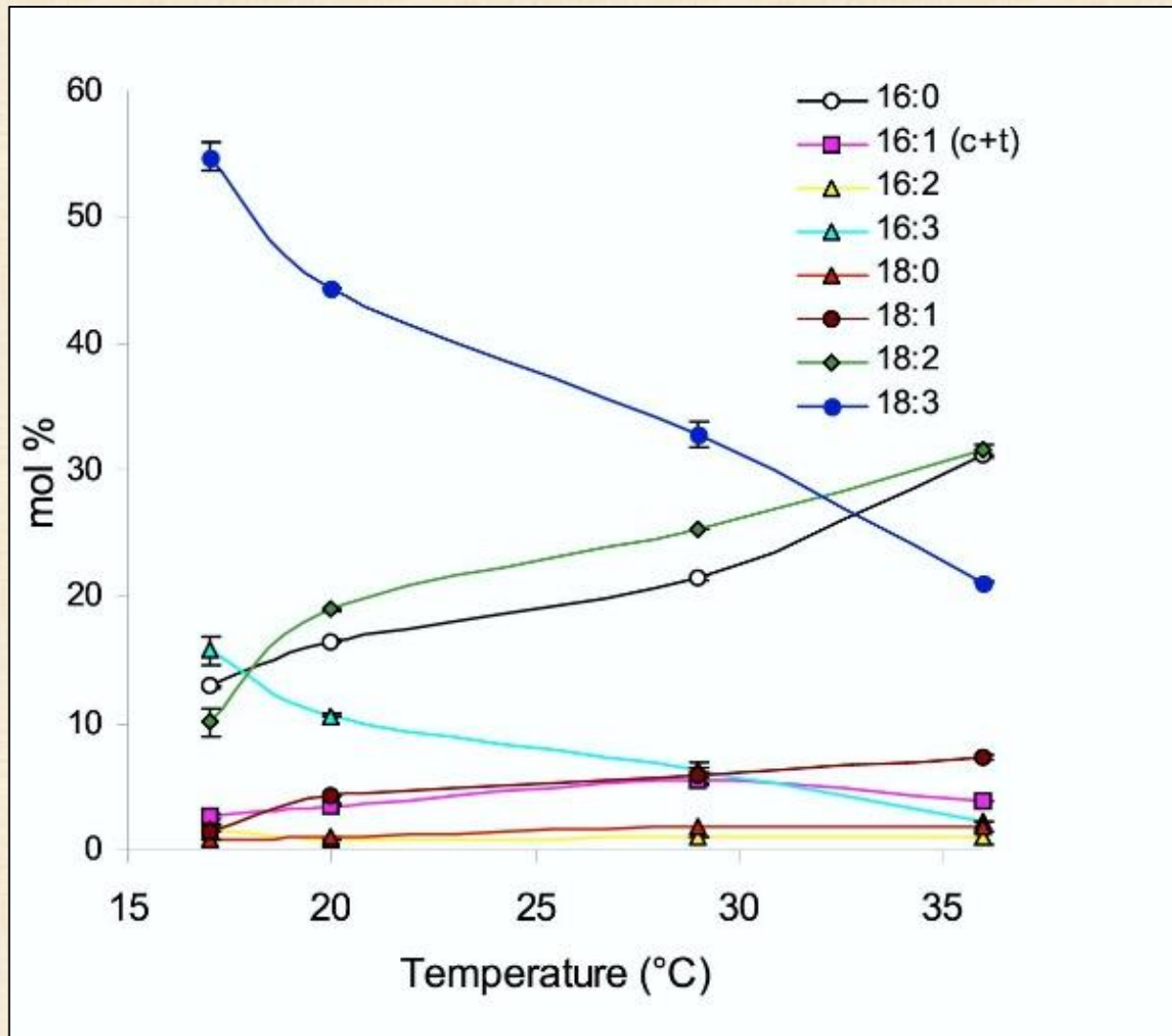
	<i>Vigna radiata</i> hypocotyl	<i>Secale cereale</i> leaves	<i>Secale cereale</i> leaves	<i>Avena sativa</i> coleoptyle	<i>Avena sativa</i> roots	Spring cut leaves	Winter cut leaves	<i>Hordeum vulgare</i> roots	<i>Zea mays</i> roots	<i>Arabidopsis thaliana</i> leaves	<i>Solanum tuberosum</i> leaves	<i>Solanum commersonii</i> leaves	<i>Nicotiana tabacum</i> BY-2 cells	<i>Nicotiana tabacum</i> leaves
PL	48.9	31.7	36.6	41.7	50.1	28.2	28.9	45	43.9	46.8	46.4	48.3	40.3	38.5
SL	6.8	16.2	16.4	26.1	10.1	27.2	30.4	8	6.8	7.3	6.5	6.1	11.3	20.6
St	43.6	52.1	46.6	32.2	39.7	41.3	39.1	43	49.3	46	45	41.6	26.7	22.6
Other	0.7	—	0.4	—	—	2.7	1.7	4	—	—	2.1	4	6.6	5.8
St/PL	0.9	1.6	1.3	0.8	0.8	1.5	1.3	0.9	1.1	1	1	0.9	0.7	0.6
References	Yoshida and Uemura (1986)	Lynch and Steponkus (1987)	Uemura and Steponkus (1994)	Sandstrom and Cleland (1989)	Sandstrom and Cleland (1989)	Uemura and Steponkus (1994)	Uemura and Steponkus (1994)	Brown and Dupont (1989)	Bohn et al. (2007)	Uemura et al. (1995)	Palta et al. (1993)	Palta et al. (1993)	Mongrand et al. (2004)	Mongrand et al. (2004)

PL phospholipid, SL sphingolipids, St sterols

Furt et al., 2011



Augalų membranos



RR kiekis augaluose,
augintuose skirtingose
temperatūrose

Arabidopsis thaliana

Falcone et al., 2004

Burokai

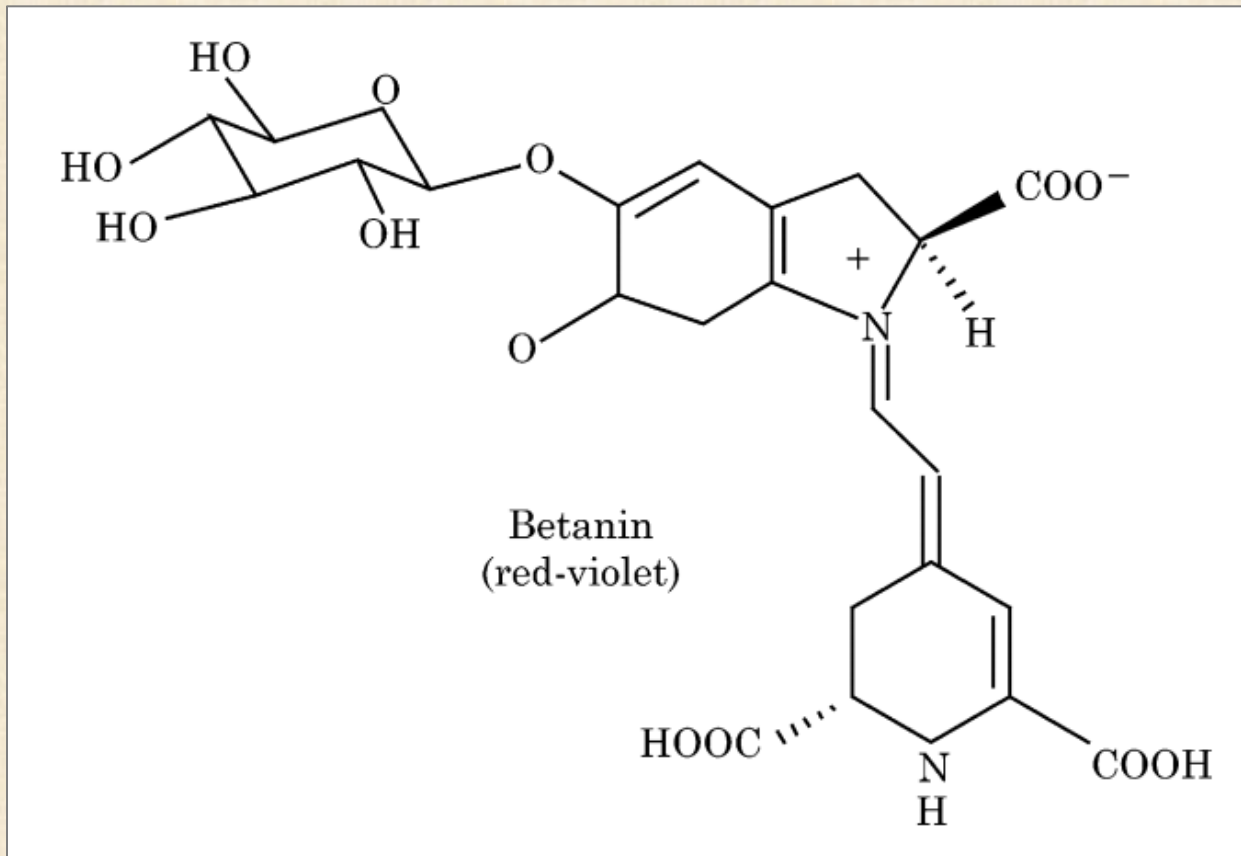


<https://www.thegreenlifestyletv.com/otoxyral/2019/06/beetroot-benefits.jpg>

Vilmantas Pupkis, Gamtamoksliai tyrimai mokykloje, 2023



Betacianinai



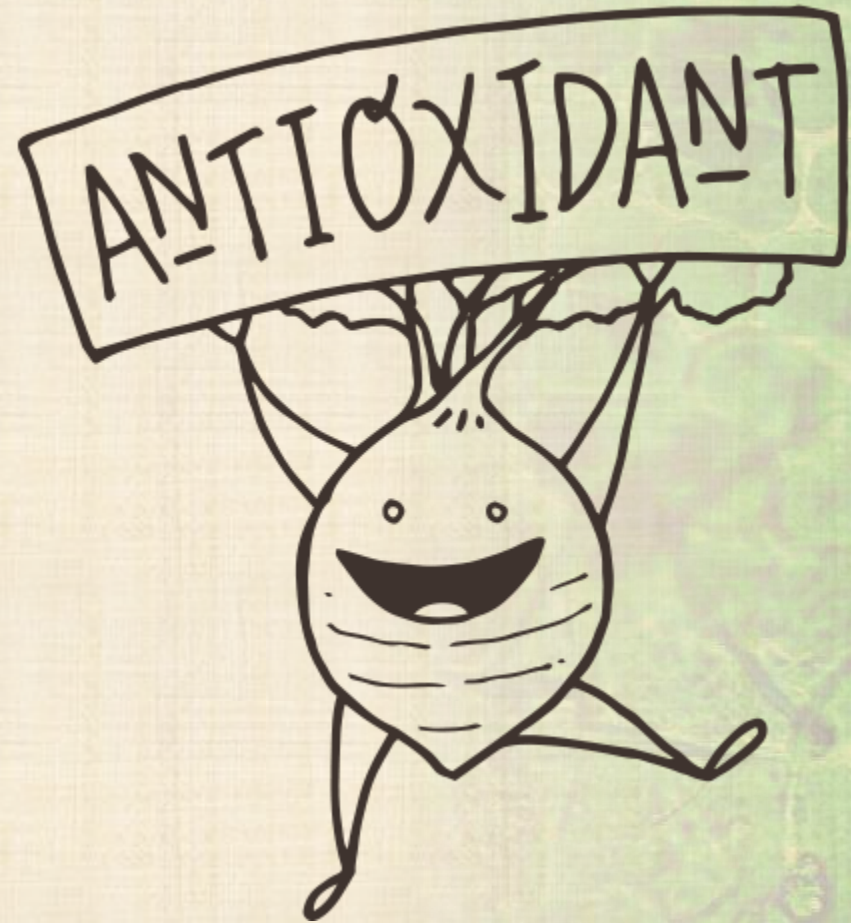
Antioksidantai,
susiję su atsaku į stresą

Sepúlveda-Jiménez et al., 2004

Betacianinai



https://cdn.shopify.com/s/files/1/2236/0863/v10/assets/beetroot_3.png



https://cdn.shopify.com/s/files/1/2236/0863/v10/assets/beetroot_2.png



Membranų pralaidumo tyrimas

Tikslas:

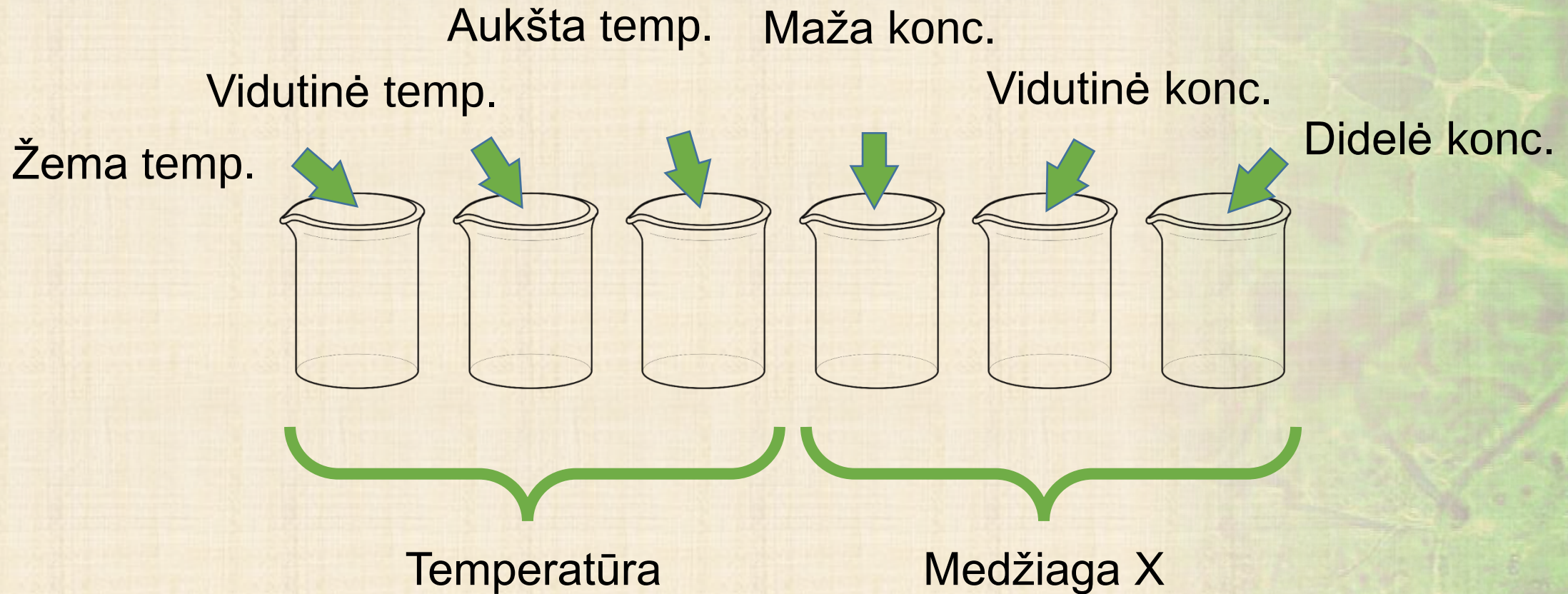
Įvertinti, kokios aplinkos sąlygos gali pažeisti augalų membranas.

Užduotys:

- 1) Nustatyti, kaip temperatūra veikia raudonojo burokėlio (*Beta vulgaris*) membranų pralaidumą.
- 2) Nustatyti, kaip įvairios cheminės medžiagos veikia raudonojo burokėlio (*Beta vulgaris*) membranų pralaidumą.



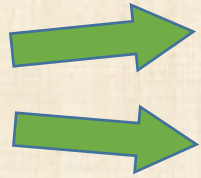
Membranų pralaidumo tyrimas



Skirtingos sąlygos

Membranų pralaidumo tyrimas

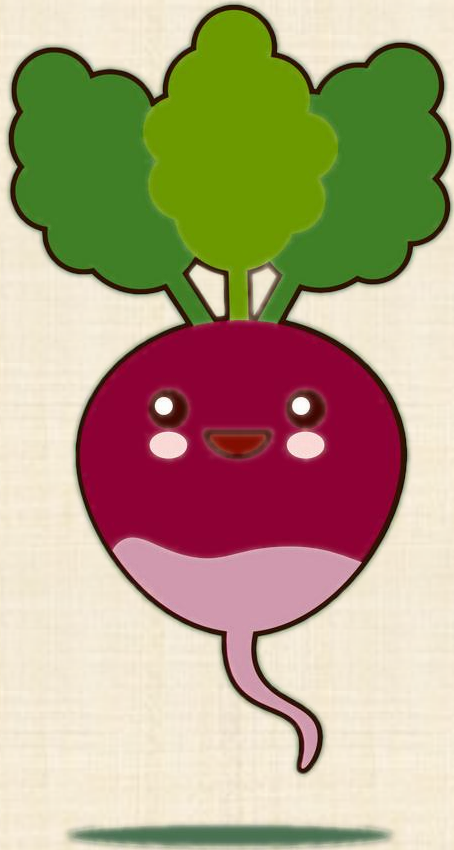
Skirtingos sąlygos



- Skirtingos temperatūros vanduo;
 - Skirtingos medžiagos / jų koncentracijos
-
- 3 % vandenilio peroksido tirpalas;
 - 9 % maistinės acto rūgšties tirpalas;
 - 33 % etanolio tirpalas;
 - dezinfekcinis skystis;
 - gėrimas „Sprite“;
 - 100 mM citrinos rūgšties tirpalas;
 - indų ploviklio tirpalas;
 - ...

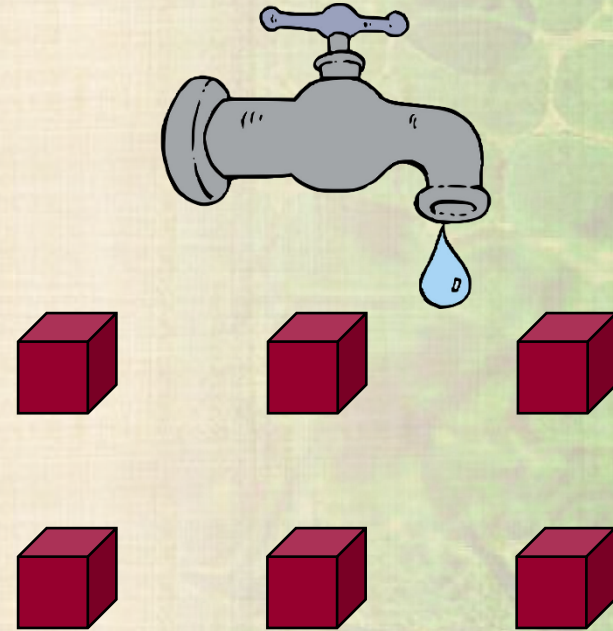
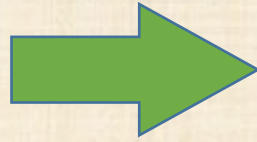
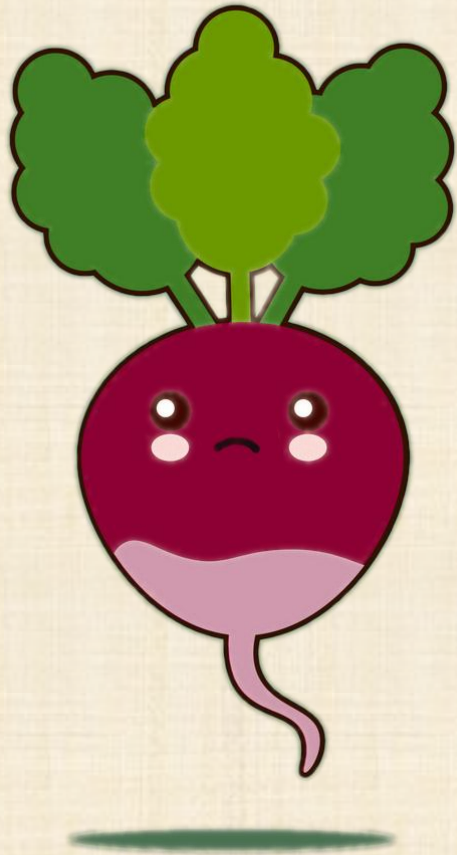


Membranų pralaidumo tyrimas



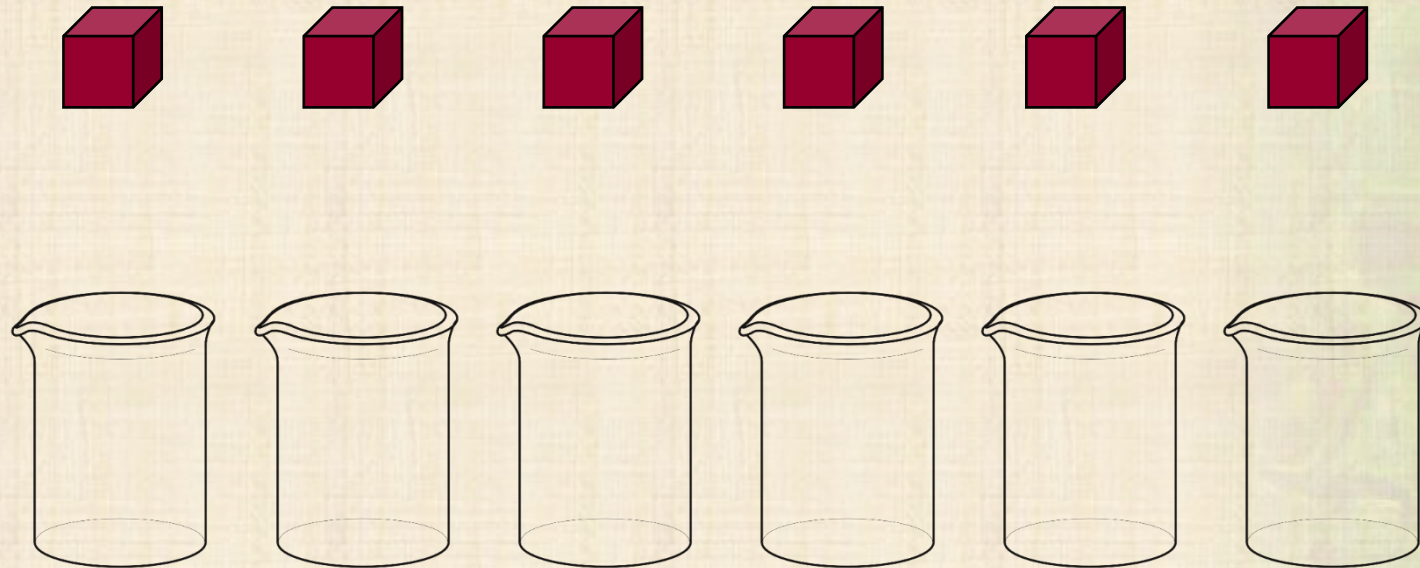
<https://cdn2.vectorstock.com/i/1000x1000/63/71/cute-vegetable-cartoon-character-beet-with-leaves-vector-17186371.jpg>

Membranų pralaidumo tyrimas



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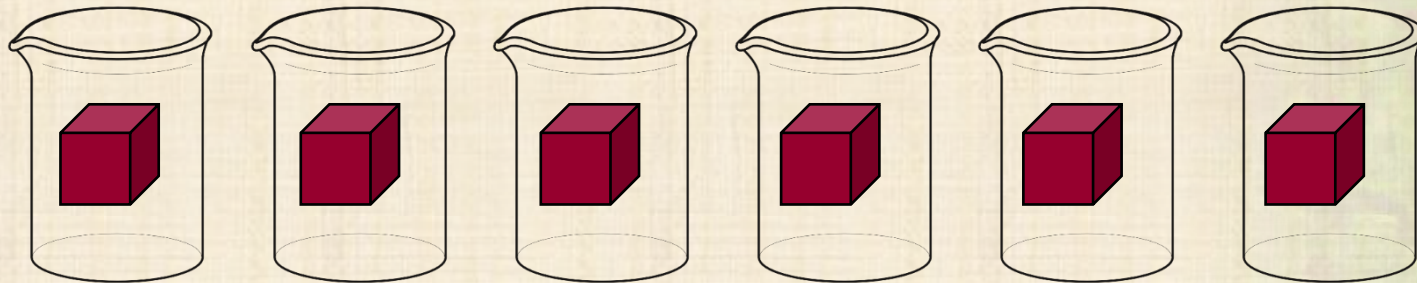
Membranų pralaidumo tyrimas



Skirtingos sąlygos



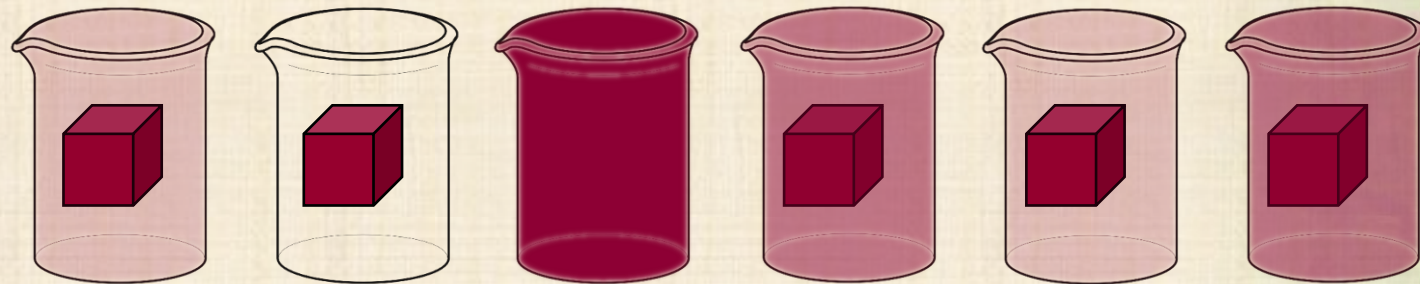
Membranų pralaidumo tyrimas



Skirtingos sąlygos



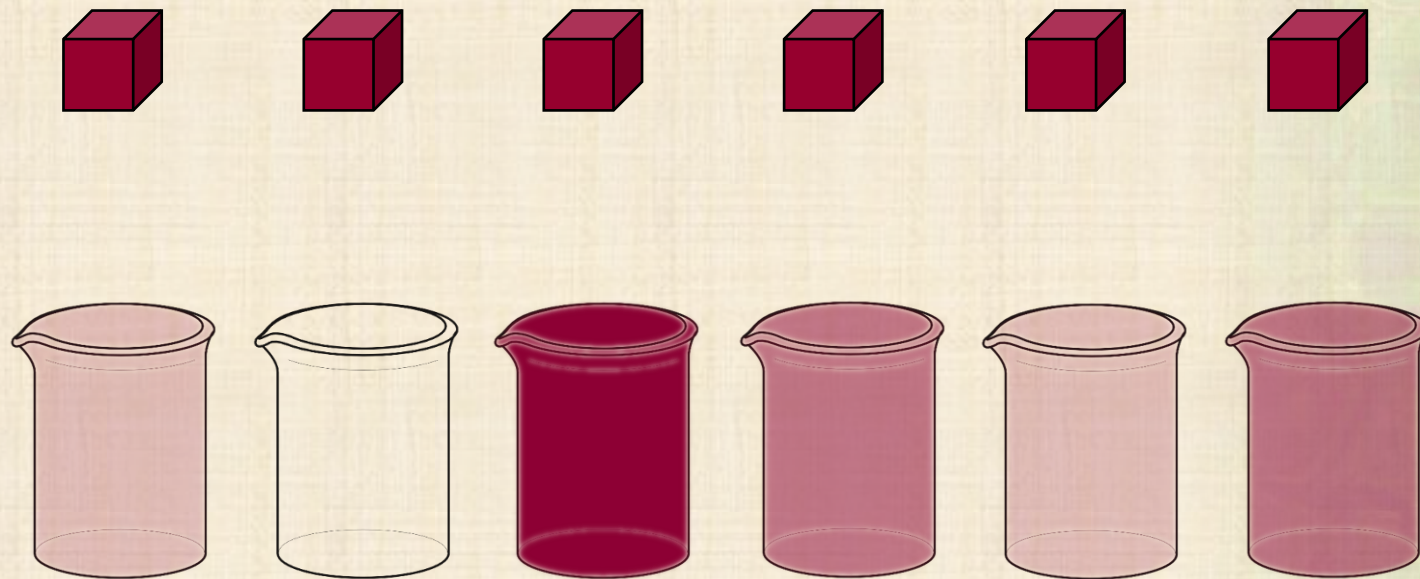
Membranų pralaidumo tyrimas



Skirtingos sąlygos



Membranų pralaidumo tyrimas

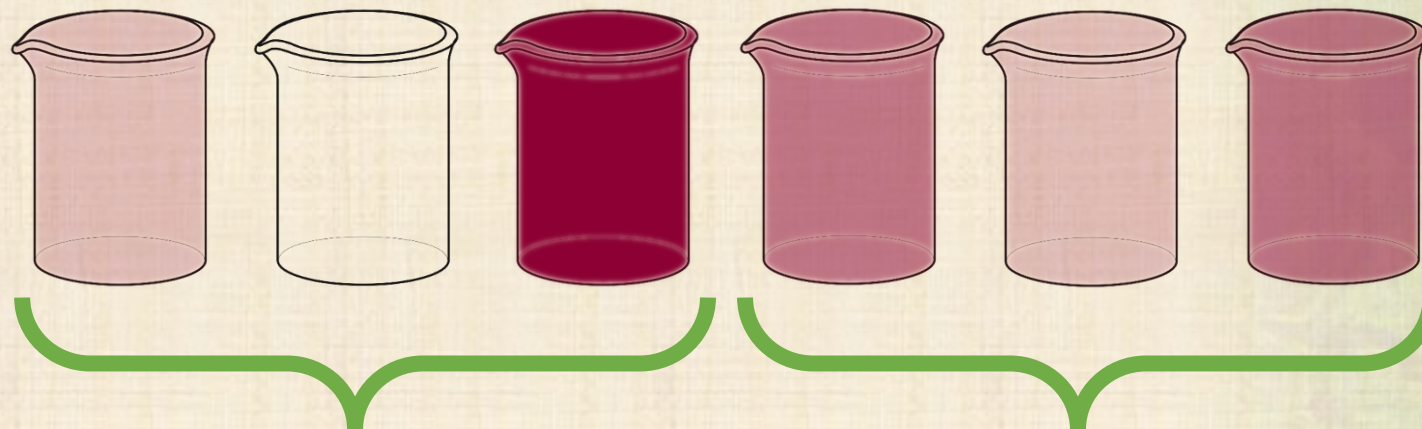


Skirtingos sąlygos



Membranų pralaidumo tyrimas

Tirpalų spalvos (betacianinų koncentracijos) įvertinimas



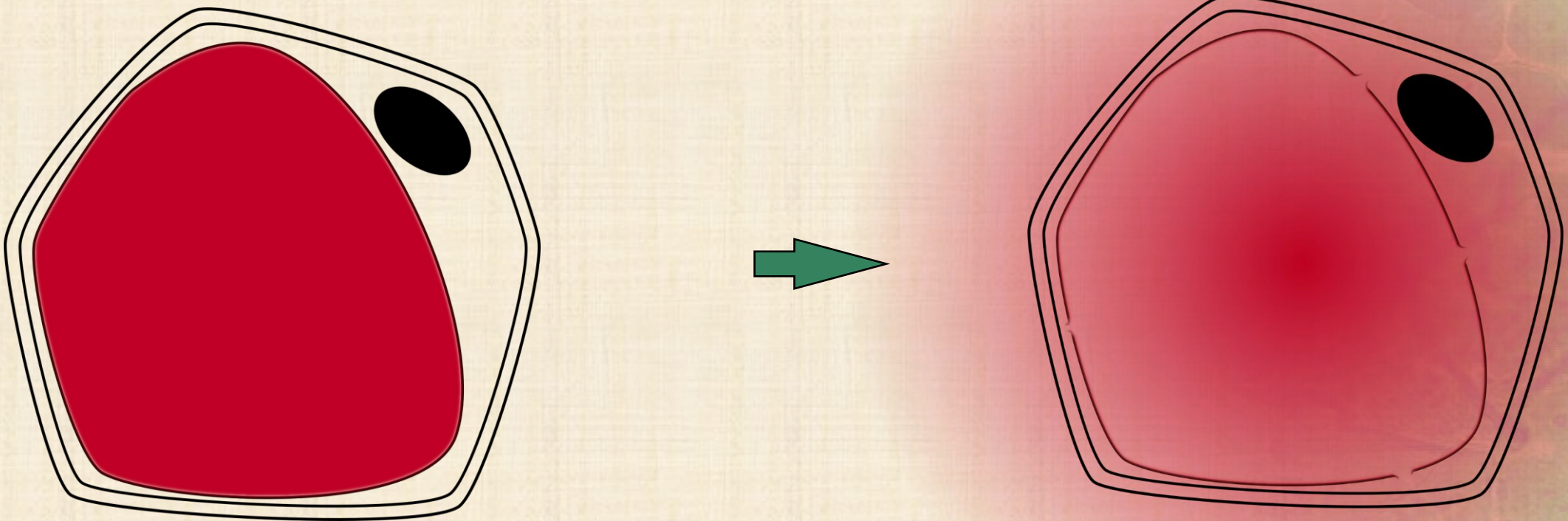
Temperatūra

Medžiaga X

Skirtingos sąlygos



Membranų pralaidumo tyrimas



Pažeidus membranas, betacianinai išteka į terpę



Membranų pralaidumo tyrimas

Lamberto-Bugero-Bero dėsnis

Ekstinkcijos koeficientas
(medžiagos savybė)

Sugertis



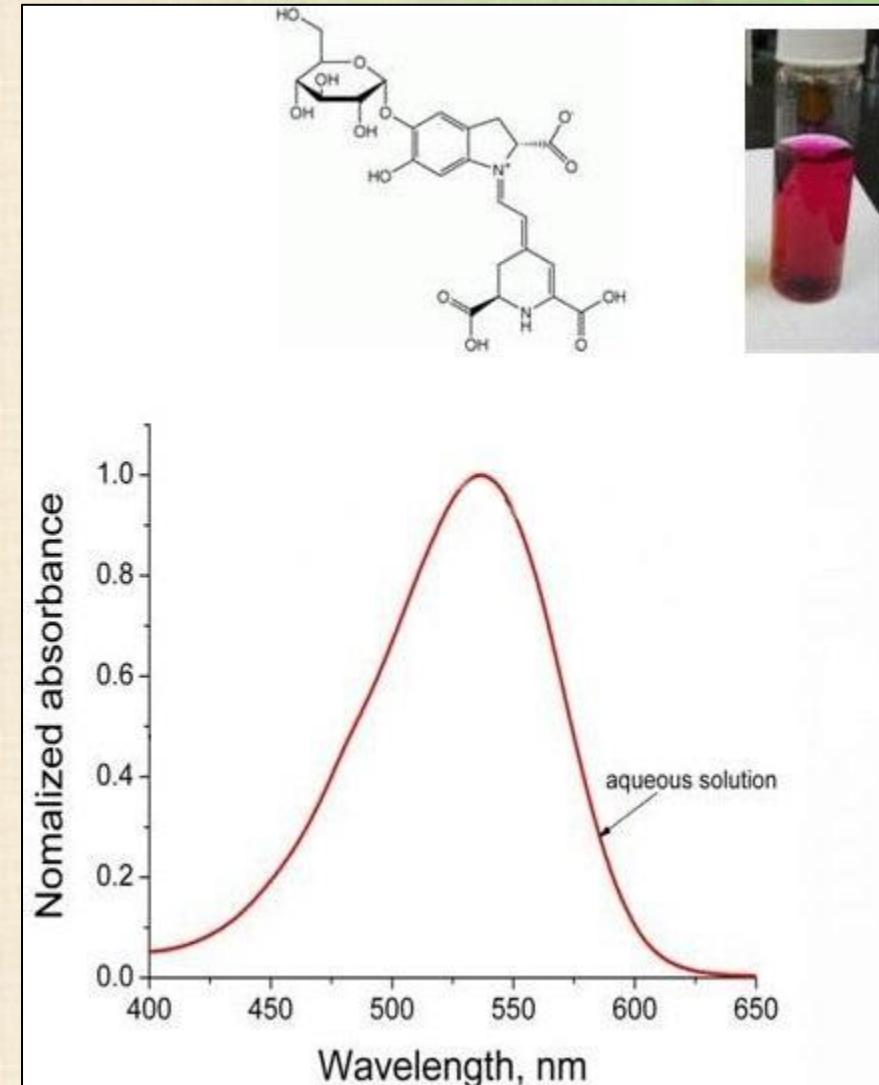
$$A = \varepsilon \cdot l \cdot c$$



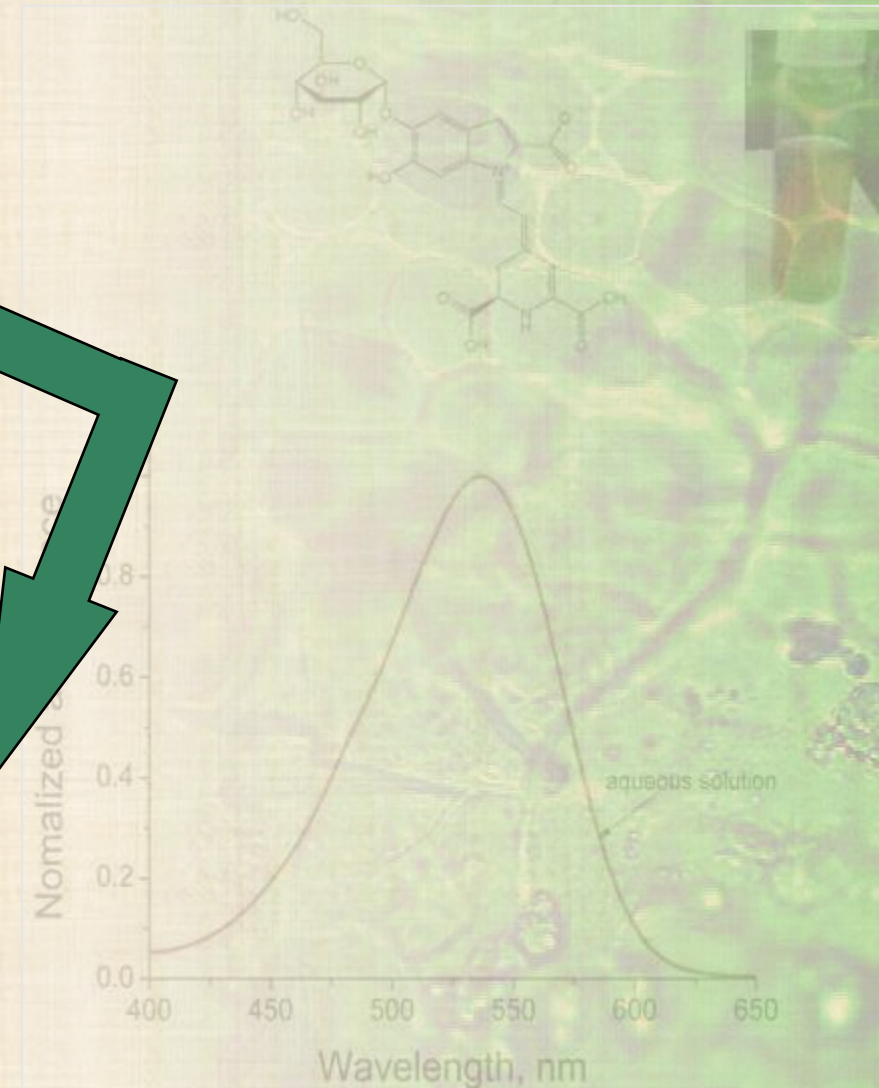
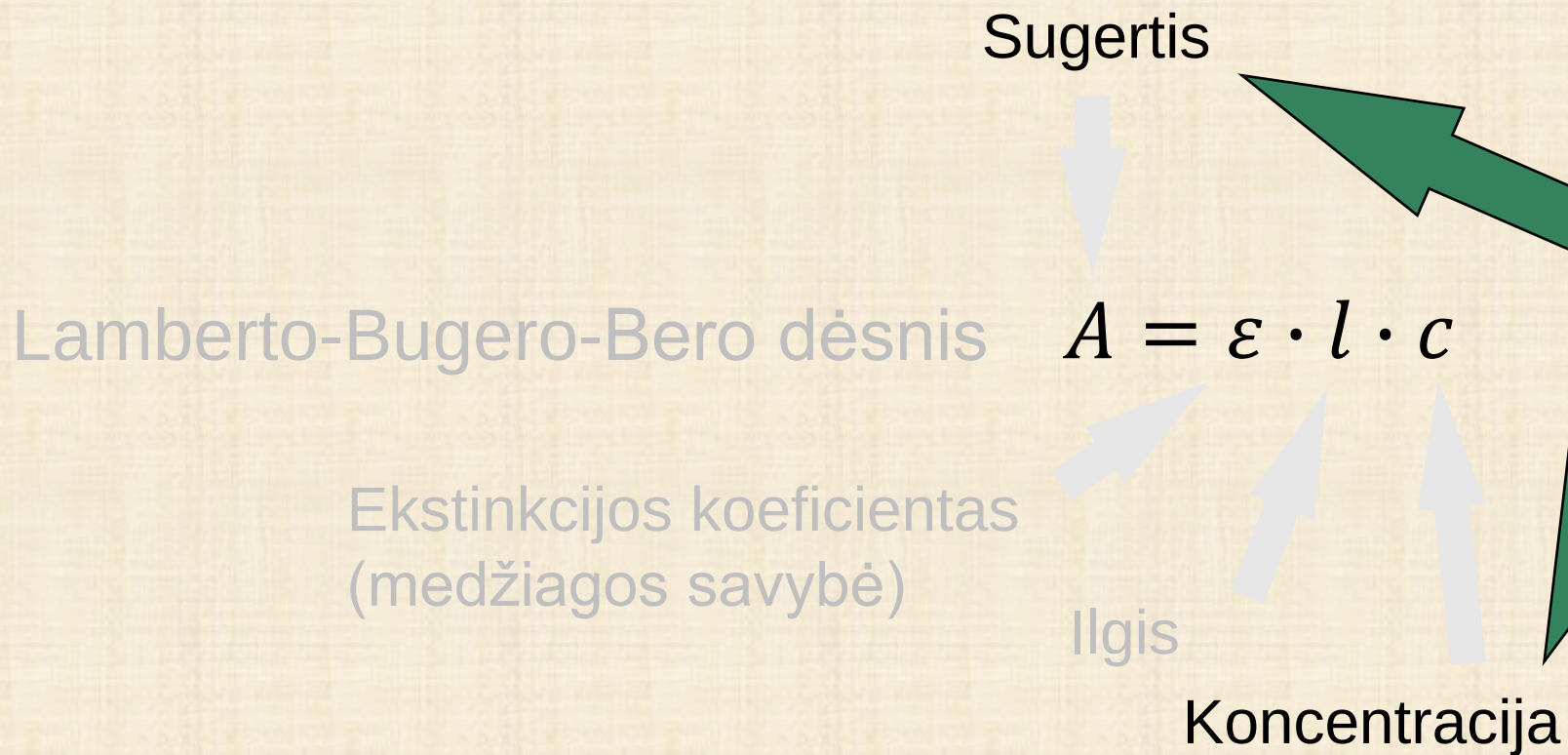
Ilgis



Koncentracija

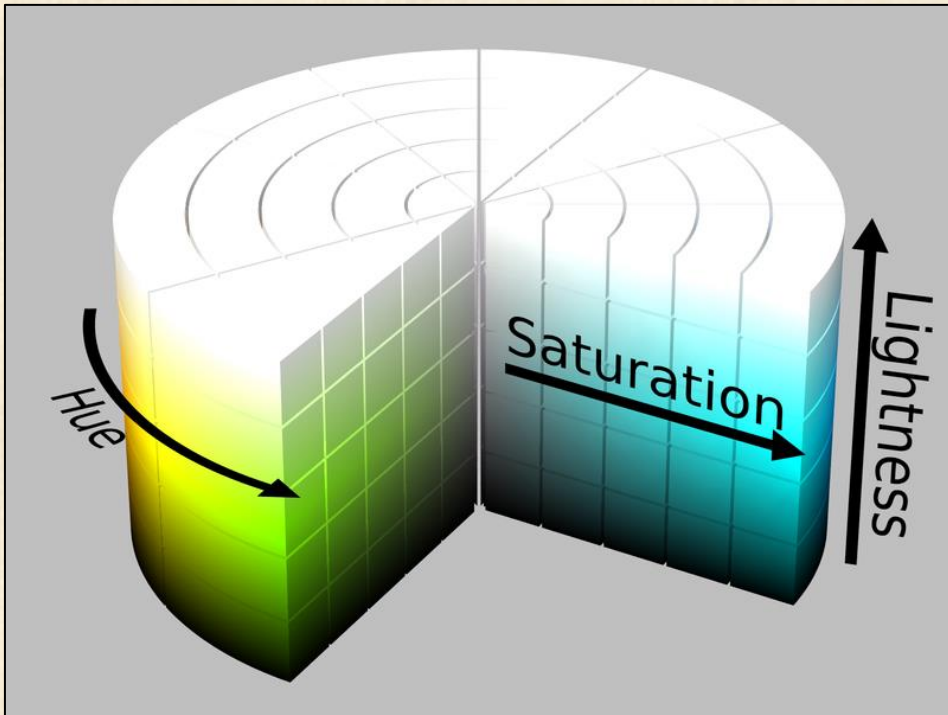


Membranų pralaidumo tyrimas



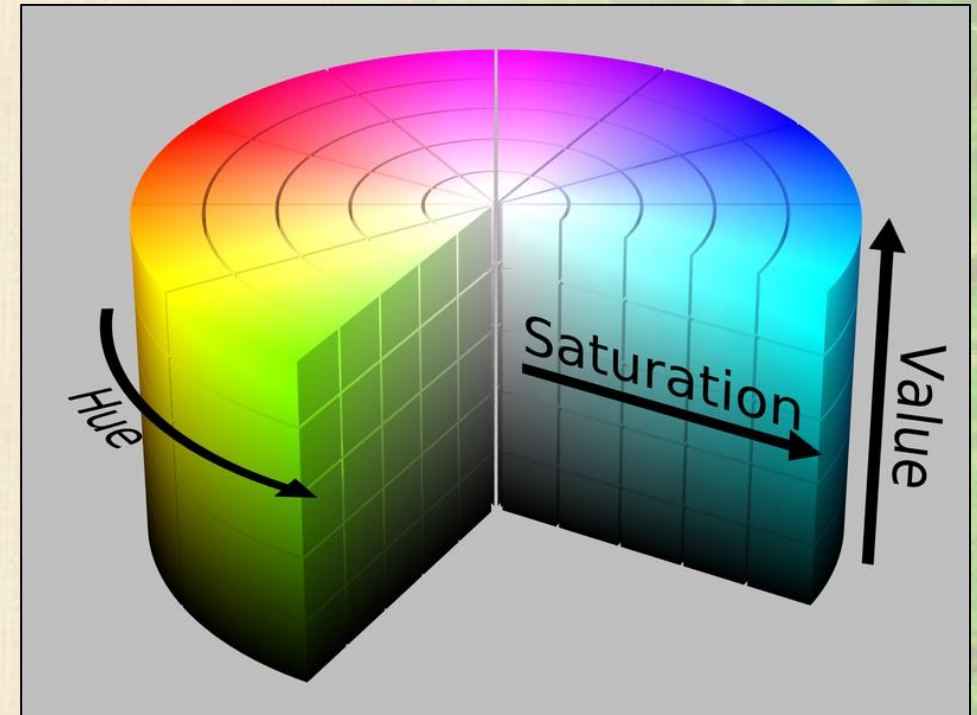
Membranų pralaidumo tyrimas – spalvų formatai

HSL



https://upload.wikimedia.org/wikipedia/commons/thumb/6/6b/HSL_color_solid_cylinder_saturation_gray.png/800px-HSL_color_solid_cylinder_saturation_gray.png

HSV



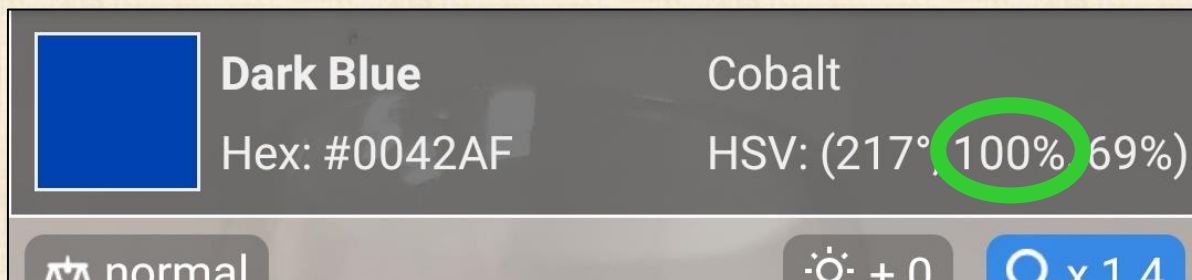
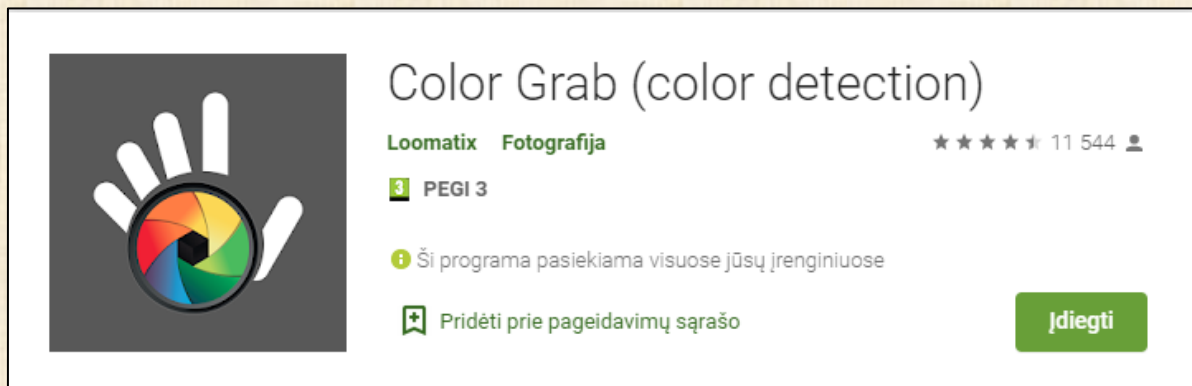
https://upload.wikimedia.org/wikipedia/commons/thumb/3/33/HSV_color_solid_cylinder_saturation_gray.png/800px-HSV_color_solid_cylinder_saturation_gray.png



Membranų pralaidumo tyrimas

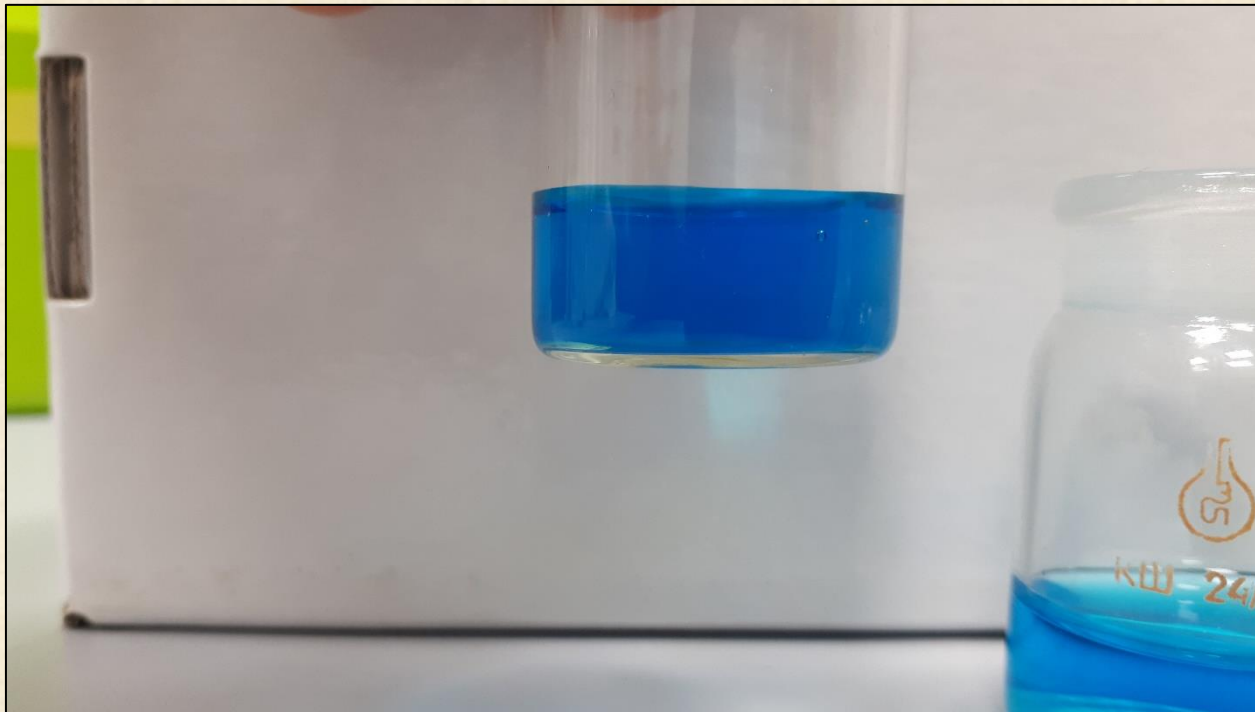
Spalvos nustatymas mobiliąja programėle:

- *Color Grab* (Android OS)
- *Color Detector* (iOS)



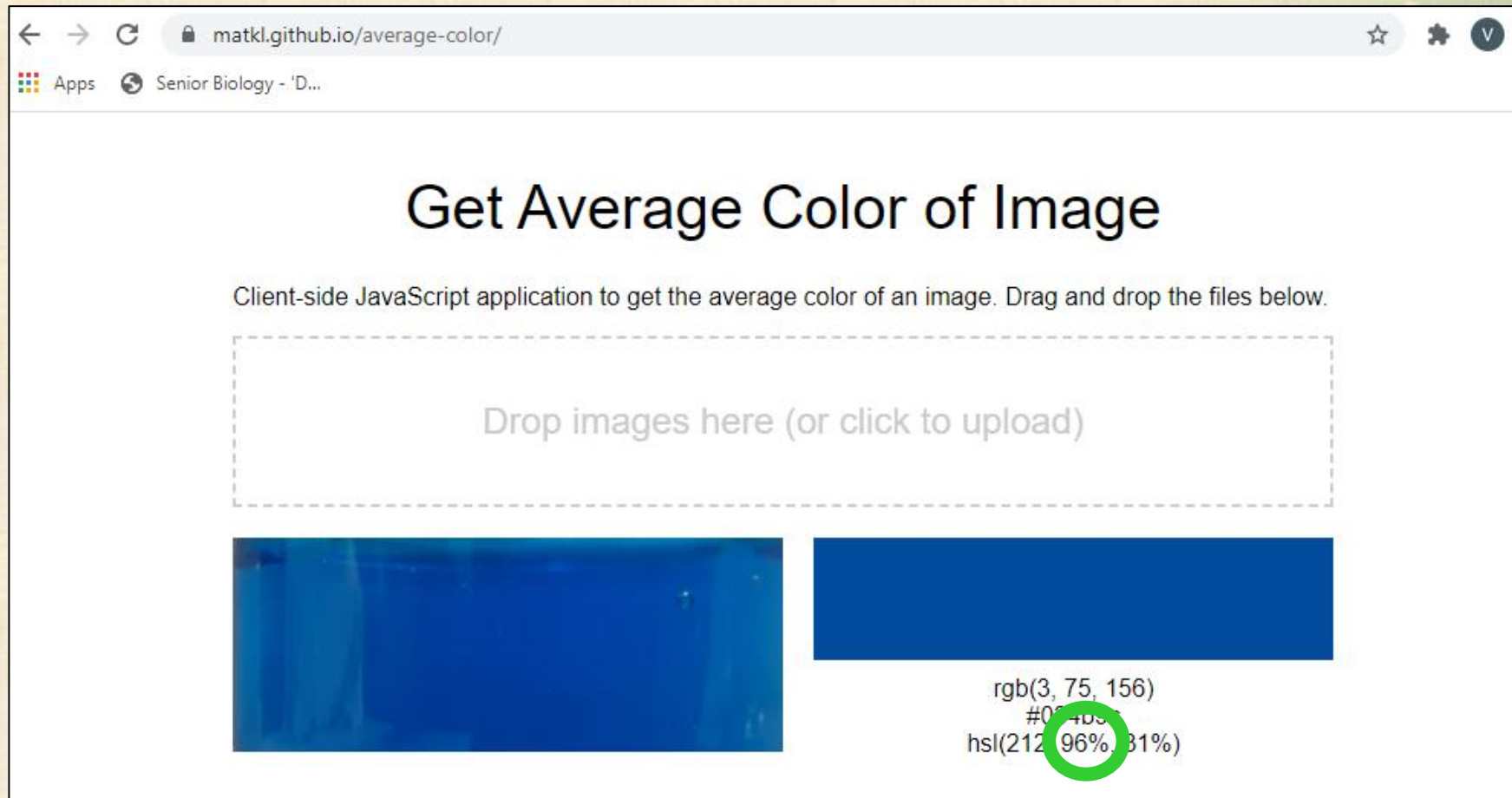
Membranų pralaidumo tyrimas

Arba spalvos nustatymas, naudojantis internetu



Membranų pralaidumo tyrimas

Arba spalvos nustatymas, naudojantis internetu: <https://matkl.github.io/average-color/>



Membranų pralaidumo tyrimas

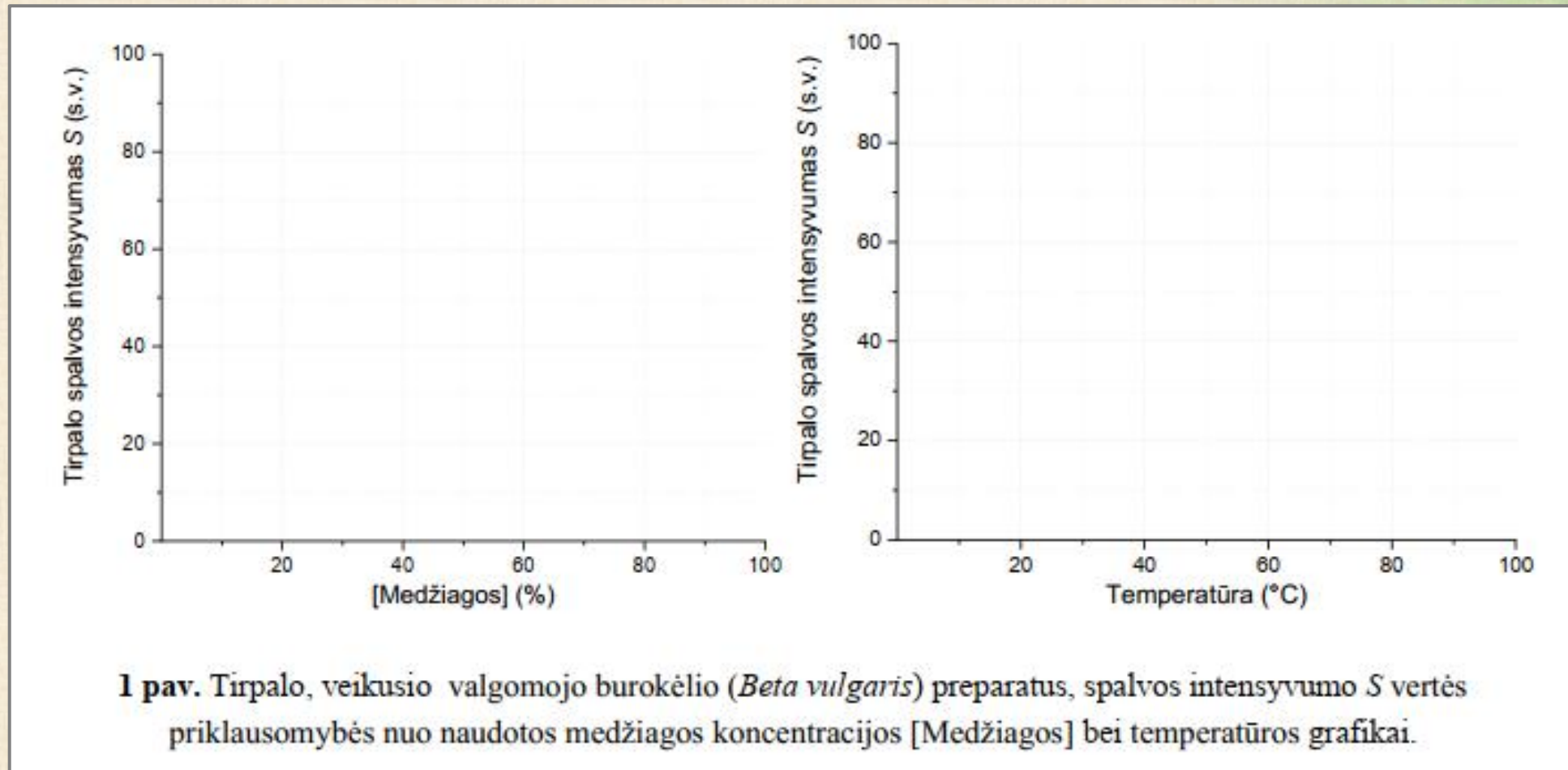
1 lentelė. Tirpalų spalvos.

Tirpalas	Tirpalo spalvos „S“ vertė
...	...

Kodėl stebėti tokie rezultatai?



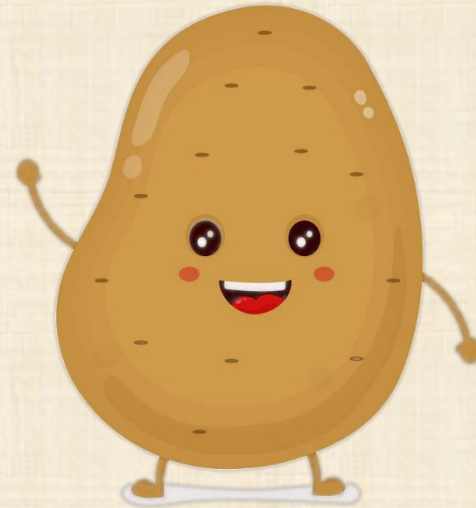
Membranų pralaidumo tyrimas



O ką prisimenam?

QUIZIZZ

[join my quiz.com](https://joinmyquiz.com)



Kas dar gali veikti membranų pralaidumą?

- Organiniai tirpikliai
 - Rūgštys
 - Dezinfekantai
 - Valikliai
 - ...
-
- Kai kurios medžiagos (balikliai, šarmai) gali ne tik pažeisti membranas, bet ir pigmentus, tuomet nematysime efekto.



Smalsiems

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- Sezgin, E., Levental, I., Mayor, S., & Eggeling, C. (2017). The mystery of membrane organization: composition, regulation and roles of lipid rafts. *Nature reviews Molecular cell biology*, 18(6), 361.

