



Jul 12, 2020

TAP media preparation

Joao Vitor Molino¹

¹University of Zürich



Works for me

This protocol is published without a DOI.



Joao Vitor Molino University of Zürich



ABSTRACT

This protocol describes the preparation of TAP media. Usually used for growing algae cells, as Chlamydomonas reinhardtii. The protocol is derived from the protocol descrived at

https://www.chlamycollection.org/methods/media-recipes/tap-and-tris-minimal/.

[Gorman, D.S., and R.P. Levine (1965) Proc. Natl. Acad. Sci. USA 54, 1665-1669]

EXTERNAL LINK

https://www.chlamycollection.org/methods/media-recipes/hutners-trace-elements/

THIS PROTOCOL ACCOMPANIES THE FOLLOWING PUBLICATION

Molino JVD, Carvalho JCMd, Mayfield SP (2018) Comparison of secretory signal peptides for heterologous protein expression in microalgae: Expanding the secretion portfolio for Chlamydomonas reinhardtii. PLoS ONE 13(2): e0192433. doi: 10.1371/journal.pone.0192433

PROTOCOL CITATION

Joao Vitor Molino 2020. TAP media preparation. protocols.io https://protocols.io/view/tap-media-preparation-big6kbze

MANUSCRIPT CITATION please remember to cite the following publication along with this protocol

Molino JVD, Carvalho JCMd, Mayfield SP (2018) Comparison of secretory signal peptides for heterologous protein expression in microalgae: Expanding the secretion portfolio for Chlamydomonas reinhardtii. PLoS ONE 13(2): e0192433. doi: 10.1371/journal.pone.0192433

FXTFRNALLINK

https://www.chlamycollection.org/methods/media-recipes/hutners-trace-elements/

Microalgae, Recombinant, electroporation, plasmid, Media, Algae, Chlamydomonas reinhardtii

LICENSE

This is an open access protocol distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

CREATED

Jul 12, 2020

LAST MODIFIED

Jul 12, 2020

PROTOCOL INTEGER ID

39166

GUIDELINES

All steps described in this protocol are intended to be conducted in a research laboratory.

SAFETY WARNINGS

Use EPIs at all times.

Concentrated acetic acid solution used during preparation.

DISCLAIMER:

DISCLAIMER - FOR INFORMATIONAL PURPOSES ONLY; USE AT YOUR OWN RISK

The protocol content here is for informational purposes only and does not constitute legal, medical, clinical, or safety advice, or otherwise; content added to protocols.io is not peer reviewed and may not have undergone a formal approval of any kind. Information presented in this protocol should not substitute for independent professional judgment, advice, diagnosis, or treatment. Any action you take or refrain from taking using or relying upon the information presented here is strictly at your own risk. You agree that neither the Company nor any of the authors, contributors, administrators, or anyone else associated with protocols.io, can be held responsible for your use of the information contained in or linked to this protocol or any of our Sites/Apps and Services.

BEFORE STARTING

- Prepare stock solutions
- Separate flasks to distribute the prepared media for autoclavation
- Separate magnetic stirrer for mixing
- Large enough becker for media preparation
- Pippetes/volume measuring apparatus for components

Components mixing

- 1 1. Add approximatelly [M]90 % volume of ddH₂0 to a large enough becker.
 - 2. Add a magnetic bar and start and keep mixing with a magnetic stirrer during the preparation.
 - 3. Place pH probe electrode in the solution for pH monitoring, and for 11 L final media volume
 - 4. Add 110 mL 2M Tris base (e.g. Trizma)
 - 5. Add 110 mL Solution A
 - 6. Add 11 mL Phosphate solution
 - 7. Add 11 mL Hutner's trace solution
 - 8. Add 11 mL Glacial acetic acid, then add drops until pH7 is reached
 - 9. Stop mixing, and add dd H₂0 until **1 L final volume** is reached
 - 10. Start mixing again until complete mixing is achieved. (A few minutes should suffice).

Componets concentration and informations.

Stock solution	Component	Amount g mL for 1L	Molecular weight	Final mM
Solution A	NH4Cl	15	53,491	7,01
	MgS04.7H20	4	120	0,83
	CaCl2 . 2H2O	2	147,015	0,34
Phosphate solution	K2HP04	28,8	174,2	0,0620
	KH2PO4	14,4	136	0,0397
Tris Solution	Tris	2,42	121,14	19,97
Acetic acid	Acetic Acid Glacial	~1,05	60,05	~17,5

Hutner's trace composition and protocol for preparation can be found here.

Liquid transfer

2 1. Transfer the newlly prepared media to flasks for autoclavation

Typically:

1L flasks with blue caps

Erlenmeyers with a max volume capacity of 100 mL are filled with 50 mL, capped with alluminium foil (2 layers) Erlenmeyers with a max volume capacity of 500 mL are filled with 250 mL, capped with alluminium foil (2 layers)

Autoclavation

- 3 1. Place flasks inside the autoclave (For flasks with lid, make sure it is loosen enough to allow vapor passage)
 - 2. Set autoclavation to § 121 °C for at least © 00:15:00 , 15 psi.
 - 3. After autoclavation, wait media to cool down and it is ready for use.