



bjmhkk36 ▾

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TAP media preparation V.(bjmhkk36)

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Works for me

This protocol is published without a DOI.

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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 described 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-bjmhkk36>

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

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KEYWORDS

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

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40329

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

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BEFORE STARTING

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

Components mixing

1. Add approximately **190 % volume** of ddH₂O to a large enough vessel.
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

For **1 L final media volume**

1. Add **10 mL 2M Tris base (e.g. Trizma)**
2. Add **10 mL Solution A**
3. Add **1 mL Phosphate solution**
4. Add **1 mL Hutner's trace solution**
5. Add **1 mL Glacial acetic acid**, then add drops until **pH7** is reached
6. Stop mixing, and add dd H₂O until **1 L final volume** is reached
7. Start mixing again until complete mixing is achieved. (5 minutes should suffice).

Components concentration and informations.

Stock solution	Component	Amount for Stock (g) *Check final volume for each in column A	Molecular weight (g/mol)	Final media concentration (mM)
Solution A (1000ml)	NH ₄ Cl	40	53.49	7.48
	MgSO ₄ · 7H ₂ O	10	246.47	0.406
	CaCl ₂ · 2H ₂ O	5	147.01	0.34
Phosphate solution (250mL)	K ₂ HPO ₄	27	174.2	0.620
	KH ₂ PO ₄	14	136.086	0.41
Tris Solution (1000mL)	Tris	242.28	121.14	20
Acetic acid	Acetic Acid Glacial		60.05	~17.5

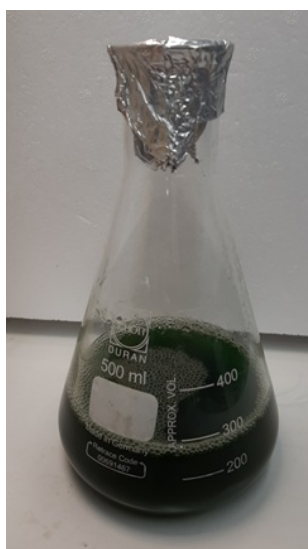
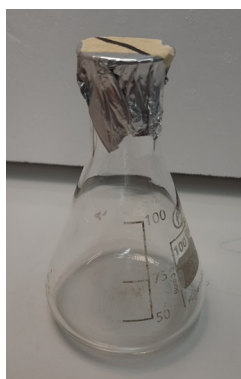
Hutner's trace composition and protocol for preparation can be found [here](#).

Liquid transfer

- 2 1. Transfer the newly 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 aluminium foil (2 layers)
- Erlenmeyers with a max volume capacity of 500 mL are filled with 250 mL, capped with aluminium 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.