

Review-Form

General Information about This Paper:

Paper ID: ABS1560

Paper Title: Detailed optimization of ribosome isolation method with rate zonal centrifugation in *Arabidopsis thaliana* leaves

Review Period: Three Weeks SINCE 27.01.2016

Paper Quality: Mark your evaluation in the suitable column. 5 indicates the best and 1 implies the worst.

ASPECT	BEST	5	4	3	2	1	WORST
Scope	Relevant			X			Irrelevant
Organization:	Excellent	X					Poor
Clarity:	High	X					Low
Length:	Too Short	X					Too Long
References:	Adequate		X				Incomplete
Correctness:	Correct		X				Incorrect
Significance:	High			X			Low
Originality:	High			X			Low
Attachments:	Helpful		X				Unnecessary
If Survey Coverage:	Broad				X		Shallow
Contribution:	Significant				X		No new
Expression	Clearly		X				Vague
Grammar	Good			X			Poor

Please make very detailed technical and editorial comments and suggestions directly on the manuscript. Your comments are an invaluable aid to the author(s) to help in improving the overall technical quality, utility, and readability of the material. Particular attention should be given to details that guide possible revisions, or that clearly explain reasons for rejection. Please summarize comments that appear on the manuscript to help the author(s) focus on the major issues you have raised in your review.

What are the contributions of this paper?

Proposes shortened protocol for *Arabidopsis thaliana* ribosomes subunits purification procedure.

Recommendation ()

~~A) Accept~~

~~B) Revise and Accept (Minor Revision)~~

C) Major Revision and Resubmit

~~D) Reject~~

Comments

Confidential Comments to the Editor-in-Chief

Comments to the Author(s)

A) If you agree to accept this paper, please illustrate your reasons why this paper is qualified to be published in the journal in detail, or provide revision suggestions if you have any.

- Why this paper is qualified:

- Revision suggestions:

B) If you think this paper needs major modification and resubmission, please provide summary and detail revision suggestions (on research base, research technique, paper presentation, grammar, jargon use, typesetting, reference, etc.). Please point out the section(s) where you think an error/ flaw occurs.

- Detail revision suggestions:

The manuscript should be carefully edited, in the field of spelling and grammar, lots of misspellings. All species names in latin should be given in italics- check the whole text. Please check my corrections on the manuscript (in the graph section also).

Minor revisions:

Please clarify why the chloramphenicol has been used in the purification buffer, as it is used in the case of Procaryotes, while in case of Eucaryotes the cycloheximide in the final concentration of 100 µg/ml is used or 1 mM puromycin.

Major revisions:

The basic novelty of the manuscript refers to the purification protocol conditions. Authors write: “Suc-density gradients and fractionated as described in the previous study (Kawaguchi et al., 2004). But the experiment parameters and condition were made some change.” Authors admit that their contribution is not impressive. If the change is only slightly visible – it is usually irrelevant...The essential change makes a contribution. More detailed comment about research are given as follows:

- 1) Authors write: “1.5h centrifugal time is perfect for separation of ribosomes”. This statement is made on the basis of only two time points checked. The timecourse for Suc-gradient centrifugation should be widened. Necessary to give more experimental points of time, because the conventional protocols give usually one time point and the authors upgraded it with only one more timecourse checking. Afterwards: rather surprising that spin velocity enhanced about 2 times results 10x shortening the spin time, referring to the other popular protocols (see Stupina et all. 2008, 2013 and others.).
- 2) Authors write :“Suitable concentration range of sucrose gradient promotes the quality of isolation”. This is no new. It is a basic principle of the method. Once more only two values of gradients-spans are checked. If Authors want to support the novelty of this work, more detailed study should be added.
- 3) Overloading the gradient with the sample volume is the obvious phenomenon. Can be omitted using standard sample concentrating techniques (e.g. Amicon Ultra 100 K column). Resolution capacity dramatically falls with the overloaded sample. Only 2 volumes checked, varying of 100%- too big span.

C) If you think this paper should be rejected, please expound the reasons why it is not of sufficient quality/novelty or seriously flawed to be published in the journal.

Would you be willing to review a revision of this manuscript? Yes (X) No ()

Thank you very much for your contribution !