

Dear Author(s),

This document is divided into a number of sections in which you can add your comments to the video, voiceover, and online text/PDF. Please be aware that our policy is to do a single complimentary revision, so it is critical that all participants in this project offer their comments collectively. In addition, please make sure that your comments are easily interpreted and transparent.

Have fun!

Protocol Name: Transforming, Genome Editing and Phenotyping the Nitrogen-Fixing Tropical Cannabaceae Tree *Parasponia andersonii*

Date: 9 July 2019

Authors and Affiliations

Please fill in any missing author information not included in the video.

Order	Author	Affiliation	Email

Video Comments

Please fill in any comments you wish to make using the table below using the example as a guide. If you need more space to write, please do so below the table. **DO NOT ADD CORRECTIONS TO THE NARRATION HERE. PLEASE DO THIS IN THE AUDIO COMMENTS SECTION.**

	Time code	Comment	Requested Change	Adjustment appropriate
<i>Example</i>	2:52	Onscreen text says use 0.25 mM Fluo-4	Text should say use 0.50 mM Fluo-4	-
1. Original comment	00:02	Onscreen text says "TRANSFORMING, GENOME EDITING AND PHENOTYPING THE NITROGEN-FIXING TROPICAL CANNABACEAE TREE PARASPONIA ANDERSONII"	Text should say "TRANSFORMING, GENOME EDITING AND PHENOTYPING THE NITROGEN-FIXING TROPICAL CANNABACEAE TREE <i>PARASPONIA ANDERSONII</i> "	NO, change is required
1b. Comment after reviewing changes	00:02	Onscreen text now says "TRANSFORMING, GENOME EDITING AND PHENOTYPING THE NITROGEN-FIXING TROPICAL CANNABACEAE TREE <i>PARASPONIA andersonii</i> " This is not the adjustment that we requested and is scientifically incorrect.	Text should say either "TRANSFORMING, GENOME EDITING AND PHENOTYPING THE NITROGEN-FIXING TROPICAL CANNABACEAE TREE <i>PARASPONIA ANDERSONII</i> " OR "TRANSFORMING, GENOME EDITING AND PHENOTYPING THE NITROGEN-FIXING TROPICAL CANNABACEAE TREE <i>Parasponia andersonii</i> "	
2.	00:02	In the author list, the "1" behind "Rene Geurts" is not superscripted.	Please change "Rene Geurts ¹ " to "Rene Geurts ¹ "	Yes
3.	00:02	The affiliations on screen are displayed until the bottom edge of the screen.	Please reduce the font size of the affiliations so that the use of space will be more	Yes

			aesthetically pleasing.	
4.	00:02	Onscreen the second line of the title is not aligned with the top and bottom lines.	Please remove the space before "AND PHENOTYPING THE NITROGEN-FIXING" to align the title to left side	Yes
5.	01:54	Onscreen text says "DO NOT USE HEATED LID"	Text should say "CYCLE BETWEEN 7 AND 28 °C – DO NOT USE HEATED LID"	Yes
6. Original comment	10:33	Onscreen table is not sharp	Please improve resolution of the table	NO, change is required
6b. Comment after reviewing changes	10:36	The onscreen table is still not sharp. This has to be improved.	For your convenience we have uploaded a sharp version of the table as .tiff file to the JoVE file upload page.	
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ADDITIONAL REMARKS:

- During the filming day, the introductory shots of talents (Titis Wardhani, Yuda Roswanjaya, and Marijke Hartog) were made. Why are these shots not used to introduce the talents in the video between 00:52 – 01:00?
OK, shots have been incorporated according to our requests.
- Text appears onscreen for 3 seconds which in some cases feels a bit short. If possible, please leave the text visible onscreen for 5 seconds at 01:54, 04:59, 08:31.
OK, display of onscreen information has been adjusted according to our requests.
- Please add 3 items to the credit title:
 - Researchers: Titis Wardhani
Yuda Roswanjaya
Marijke Hartog
Arjan van Zeijl
 - Filming location: Radix building, Wageningen University & Research, The Netherlands
 - This research is funded by The Netherlands Organization for Scientific Research (NWO-VICI grant 865.13.001; NWO-Open Competition grant 819.01.007) and The Ministry of Research, Technology and Higher Education of the Republic of Indonesia (RISET-PRO grant 8245-ID).
Not ok. Why it was decided not to add the three lines of information that we requested, without explaining this decision to us? We do understand that the 4 researchers (line a.) are already listed in the video and can accept if you do not want to list them again. Also, listing the filming location

is not a must (line b.). However, our research is funded by public money that was provided to us by the Dutch and Indonesian governments. We feel it is important to acknowledge this funding appropriately, as without this funding it would not have been possible to produce this JoVE movie. Therefore, we request that you add this line of information to the credit titles. We could accept if it is modified to read: "This research is funded by The Netherlands Organization for Scientific Research and The Ministry of Research, Technology and Higher Education of the Republic of Indonesia."

Audio Comments

This section is used to specify the changes that need to be made to the narration. Please follow the example below as a guide to list your changes. If there is a pronunciation change, please provide a phonetic pronunciation key.

	Time code	Comment	Step(s) in Shotlist	Rewritten Text or Corrected Pronunciation (highlight in bold)	Adjustment appropriate
<i>Example</i>	1:49	Original Script Text: "Then, show the participant their electromyography, or EMG patterns, which correspond to eight specific and unique polar plots."	2.2	Rewritten Script Text: "Then, show the participant the unique and specific polar plots, which correspond to their electromyography, or EMG (pronounced E-M-G) patterns. "	-
1.	01:09	Original script text: "To grow <i>Parasponia andersonii</i> (pronounce 'Pa-ra-spo-nee-ya an-der-SON-ee-eye') trees, begin by germinating the seeds [1-TXT]. Remove the seeds from <i>Parasponia</i> (pronounce "Pa-ra-spo-nee-ya") berries by squashing them on a piece of tissue paper [2] or rubbing against the inside of a tea sieve [3]."	2.1	Pronounce script text: "To grow <i>Parasponia</i> <i>andersonii</i> (pronounce 'Pa-ra-SPO-nee-ya an-der-SO-nee-eye') trees, begin by germinating the seeds [1-TXT]. Remove the seeds from <i>Parasponia</i> (pronounce 'Pa-ra-SPO-nee-ya') berries by squashing them on a piece of tissue paper [2] or rubbing against the inside of a tea sieve [3]." Please listen to the correct pronunciation of " <i>Parasponia andersonii</i> " at 00:20.	NO, the pronunciation of <i>Parasponia</i> has not improved. However, based on the overall sound this new audio fragment can be clearly discriminated from the other audio fragments. Therefore, we request that you return to the audio fragment that was used in the first assembly of the movie.
2.	08:47	Original script text:	5.4	Pronounce script text:	NO, same as point 1.

		"Plant 1 to 3 <i>Parasponia andersonii</i> plantlets to each pot and prepare several pots with plantlets transformed with a control construct [1-TXT]. Weigh several of the pots [2] and cover the bottom of each pot to shield the roots from light exposure [3]."		"Plant 1 to 3 <i>Parasponia andersonii</i> plantlets to each pot and prepare several pots with plantlets transformed with a control construct [1-TXT]. Weigh several of the pots [2] and cover the bottom of each pot to shield the roots from light exposure [3]."	Please return to the audio fragment that was used in the first assembly of the movie.
3.	09:52	Original script text: "This technique has been used to successfully transform petioles and segments of <i>Parasponia andersonii</i> stems with <i>Agrobacterium</i> strain AGL1[1]. First, the tissue explants are co-cultivated with <i>Agrobacterium</i> for 2 days [2]. Prolonged co-cultivation results in bacterial over-colonization and should be prevented [3]."	6.1	Pronounce script text: "This technique has been used to successfully transform petioles and segments of <i>Parasponia andersonii</i> stems with <i>Agrobacterium</i> strain AGL1[1]. First, the tissue explants are co-cultivated with <i>Agrobacterium</i> for 2 days [2]. Prolonged co-cultivation results in bacterial over-colonization and should be prevented [3]."	NO, same as point 1. Please return to the audio fragment that was used in the first assembly of the movie.
4.					
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Online Text/PDF Protocol

Please use this table to address changes that need to be made to the online text/PDF document. Both the online text and PDF are generated from the HTML template of your article. Since the PDF is generated from the HTML by our conversion software, it may contain formatting errors. For major structural changes or more than 10 spelling or grammatical mistakes, we will require re-upload of the entire document.

	Protocol Step	Comment	Requested Change (highlight in bold)
<i>Example</i>	1.1	Step says "Centrifuge lysate at 2,000 x g."	Please correct to "Centrifuge lysate at 4,000 x g."
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