Based on the reviewers’ and editorial comments the manuscript has be revised. Additions to the manuscript are in red type and deletions in blue type.

Editorial comments:

Changes to be made by the Author(s) regarding the written manuscript:

1. Please take this opportunity to thoroughly proofread the manuscript to ensure that there are no spelling or grammar issues. The JoVE editor will not copy-edit your manuscript and any errors in the submitted revision may be present in the published version.

The manuscript was proofed to correct any typographical errors.

2. Figure 1: Please include a scale bar for all images taken with a microscope to provide context to the magnification used. Define the scale in the appropriate figure Legend.

The scale has been added to the figure and the figure legend.

3. Please shorten the figure and table legends. The Discussion of the Figures should be placed in the Representative Results. Details of the methodology should not be in the Figure Legends, but rather the Protocol.

All figure and table legends were shorten.

4. Please revise the title to be more concise if possible.

5. Please provide an email address for each author.

The email addresses for each author has been added.

6. Please rephrase the Long Abstract to more clearly state the goal of the protocol.

The long abstract was revised with the goal of the protocol included as the first sentence.

7. Please rephrase the Introduction to include a clear statement of the overall goal of this method.

A paragraph was added to the introduction stating the goal of the protocol.

8. Please use SI abbreviations for all units: L, mL, µL, h, min, s, etc.

The manuscript was edited to include SI abbreviations.

9. Please revise the protocol text to avoid the use of any personal pronouns (e.g., "we", "you", "our" etc.).

Personal pronouns were not used in the manuscript.

10. Please revise the protocol to contain only action items that direct the reader to do something. The actions should be described in the imperative tense in complete sentences wherever possible. Avoid usage of phrases such as “could be,” “should be,” and “would be” throughout the Protocol. Any text that cannot be written in the imperative tense may be added as a “Note.”

The protocol section was extensively revised to include only action items.

11. Please add more details to your protocol steps. There should be enough detail in each step to supplement the actions seen in the video so that viewers can easily replicate the protocol. Please ensure you answer the “how” question, i.e., how is the step performed? Alternatively, add references to published material specifying how to perform the protocol action.

Additional details were added to the protocol steps.

12. In the JoVE Protocol format, “Notes” should be concise and used sparingly. They should only be used to provide extraneous details, optional steps, or recommendations that are not critical to a step. Any text that provides details about how to perform a particular step should either be included in the step itself or added as a sub-step. Please consider moving some of the notes about the protocol to the discussion section. For example:

2.2: Please describe how to sterilize the soil mixture in the step itself, not in “Note”.

2.3: Please specify the soil mixture needed and the size of the pots in the step itself, not in “Note”.

Most of the notes were removed from the protocols and critical information was included in the step.

13. Please revise the Protocol steps so that individual steps contain only 2-3 actions per step and a maximum of 4 sentences per step. Use sub-steps as necessary.

The protocol steps were revised to be less than 4 sentences and sub-steps were added as needed.

14. 4.1: Please provide the composition of potting media.

The composition of the potting media was added.

15. Line 273: Should step 5.1 be 4.1 instead?

This changed was made.

16. 4.5: Please describe how to process boll samples to obtain seeds.

This information was added as sub-steps.

17. After you have made all the recommended changes to your protocol (listed above), please highlight 2.75 pages or less of the Protocol (including headings and spacing) that identifies the essential steps of the protocol for the video, i.e., the steps that should be visualized to tell the most cohesive story of the Protocol.

Approximately 2.25 pages of the protocol were highlight after editing.

18. Discussion: Please also discuss critical steps within the protocol and any limitations of the technique.

The discussion was modified to include this information.

19. References: Please do not abbreviate journal titles. Please include volume and issue numbers for all references.

Journal titles were edited and the issue numbers were added.

Reviewers' comments:

Reviewer #1:

Manuscript Summary:

The manuscript is well written and will be useful for researches who are searching for sources of needed resistance to the reniform nematode.

Minor Concerns:

The method does not provide a measure of nematode reproduction which can be important if the source of resistance is reducing eggs production and not nematode numbers. However, the authors point out that the protocol can be modified to collect this data if desired. In Table 1 need to spell out FGR (Females per gram of root) or use a footnote. It is spelled out in Table 2.

Table 1 was modified.

Reviewer #2:

Manuscript Summary:

This study provides a simple and safe protocol to evaluate reniform nematode resistance. In addition, it is more important to describe a vegetative propagation method which is a non-destructive screening protocol to obtain next generation seeds. From this point, it is a very valuable method for nematode resistance breeding program. Moreover, the method is reliable and repeatable. The paper is well written. The only concern is about the sections of introduction and discussion which should focus on the method itself. Therefore, it will be better to shorten the two sections. In conclusion, the manuscript can be accepted after the issue is dealt with.

Major Concerns:

Since the major purpose of this study is to provide the detail steps of evaluating nematode resistance, the introduction and discussion sections should be shortened a bit.

The introduction and discussion were shortened and revised to focus on the protocol.

Minor Concerns:

Line 57: Change "300 plants species" to "300 plant species".

This change was made.