**Nictation Scoring GUI User Guide**

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Text

Description automatically generated with low confidence

**TOGGLE**: change the score being recorded: quiescent, crawling, waving, standing, censored

**<** : step backward one frame

**||** : pause playback

**>** : step forward one frame

**>>** : playback in real time

**>>>** : fast forward

**PREVIOUS WORM** : review the previous worm

**NEXT WORM** : score the next worm

**SAVE SCORES** : save a .csv file of your scores (can be clicked multiple times to create a backup)

**EXIT** : save a .csv file of your scores and quit the GUI

Instructions

1. Run simple\_nictation\_GUI\_4.py or nictation\_scoring\_GUI.exe

2. Choose a folder containing videos of individual tracked worms to score.

3. Score worm behavior by using the “TOGGLE” button to select the behavioral score of the worm in the current frame. Scores are recorded when playback moves in the forward direction (>, >>, or >>>), and previous scores, if any, are overwritten. The *pause* ||, *step backward* <, and *step forward* > buttons should be used to go back and ensure that each frame is scored correctly when there is a change in behavior, and to examine difficult-to-score behavior. The *fast forward* button >>> can be used to quickly review long periods of consistent behavior. Use the following definitions for scoring behavior:

**quiescent:** The worm is recumbent and not moving.

**crawling:** The worm is recumbent and moving

**waving:** The worm is nictating and moving

**standing:** The worm is nictating and not moving

**censored:** The behavior cannot be scored

Further explanation: “recumbent” means that greater than 4/5 of the worm is in contact with the substrate. “nictating” means that at least 1/5 of the worm (*e.g.* at least the head) is lifted off the substrate. “moving” means that you can see movement if you flip between two consecutive frames. Sometimes a nictating worm will settle or sway slowly over many frames. If this motion is not perceptible by looking at two consecutive frames, the worm should be scored as *standing* rather than *waving*. Frames where the object being tracked is not a worm or is more than one worm, or wherein the worm is partly obscured from view or otherwise cannot be scored, should be scored as *censored*.

4. When you reach the end of a worm video and are satisfied with the scores, click “NEXT WORM” to score the next worm, or “PREVIOUS WORM” to revisit a worm you scored earlier.

5. Click “SAVE SCORES” periodically to save your scores in case the computer crashes.

6. Click “EXIT” when you are done. This will also save your scores.