Mar 25, 2022

Solanum pimpinellifolium seed propagation protocol

DOI
https://dx.doi.org/10.17504/protocols.io.j8nlk4dexg5r/v1

Hayley Sussman[1], Magdalena M Julkowska[1]

[1]Boyce Thompson Institute

Stress Architecture and RNA Biology Lab BTI
Tech. support phone: +1 (607) 279-6002 email: mmj55@cornell.edu

Hayley Sussman
Boyce Thompson Institute

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Protocol Citation: Hayley Sussman, Magdalena M Julkowska 2022. Solanum pimpinellifolium seed propagation protocol . **protocols.io** https://dx.doi.org/10.17504/protocols.io.j8nlk4dexg5r/v1

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Protocol status: Working
We use this protocol and it's working

Created: January 27, 2021

Last Modified: March 25, 2022

Protocol Integer ID: 46725

Keywords: solanum pimpinellifolium seed propagation protocol, seed germination, seed storage, protocol, seed, seed cleaning, fruit harvesting, plant

Abstract

This protocol describes S. pimpinellifolium propagation from seed to seed. It describes seed germination, plant care and growth. It also covers pollination, fruit harvesting, seed cleaning and seed storage.

1 - Place 2-4 seeds (more if you know the seeds are old) per genotype in a 1.5 ml tube filled with tap water and stratify at 4C overnight

1.1 If the accession has difficulty germinating, nick the seed coat by lightly slicing the seed coat along its margin with a razor blade, being careful not to cut the embryo.

2 Pre-moisten soil (Cornell/Cornell plus osmocote or Giovannoni for lycopersicum; Sunshine mix or Metro Mix 360 for pimpinellifolium) in 4×8 inch inserts by putting the pots in the tray filled with tap water for 1h and spray the soil from the top using the spray bottle.

3 Place seeds in a hole ~2-4 cm deep. Cover lightly with soil.

4 Place a cover over the pots and place on a heated mat in the greenhouse (BTI Greenhouse #18 - conditions:). The trays are remaining to be covered with plastic transparent lid until the seedlings germinate (1-2 weeks in most cases)

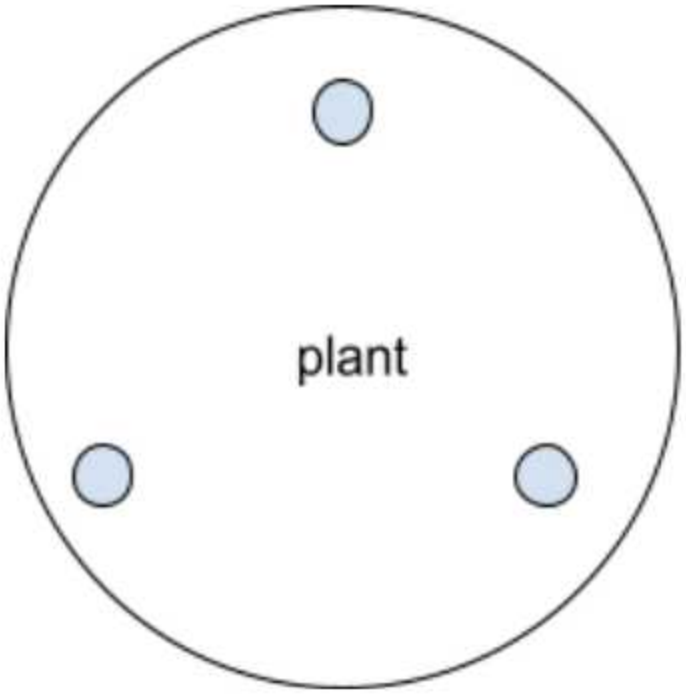
5 Remove the plastic cover when seedlings begin to grow (~2-3 cm tall).

NOTE: Some seeds take 2-4 weeks longer to germinate than the majority of the seeds.

NOTE: In some cases - we observed seedlings showing signs of oedyma / cancer-like growth. We noticed that these effects subside when these specific genotypes are grown under UVB lights (up to 2h exposure every morning - the UVB bulbs that one can buy in the pet-store for reptiles are sufficient - hang at approximately 1 - 1.5 m distance from the plant). The UVB exposure should continue throughout the lifecycle of the plant for best results.

6 When seedlings are ~20-30 cm tall, transplant to 3 gallon pots filled with pre-moistened soil (Cornell/Cornell plus osmocote for lycopersicum; Sunshine Mix or Metro Mix 360 for pimpinellifolium)

7 Once the plants are starting to gain height - we set up three large stakes in a triangle around the plant tp support up to three side branches. The remaining branches (pimpinellifolium tends to grow a lot of them) are removed twice a week. The position of the stakes in the pot is as on the picture below:



8 When the plant begins to flower, pollinate each flower in the clusters with the VegiBee wand while wearing gloves. You can see successful pollination events when you can see the pollen being released from the flowers. Clean your hands and the wand with 70% ethanol between plants to prevent cross pollination. The pollination rounds are being executed three times a week during the flowering period: Monday, Wednesday, and Friday



NOTE: pollination is the most effective during the mid-day hours.

Attach a mesh bag as close to the flower cluster as possible and make a tally mark on the wooden stake to keep track of the number of clusters that have been pollinated. Continue adding clusters until there are 20. More flowers may need to be pollinated if the clusters are small or if the plant is difficult to successfully pollinate.



Mesh bag tied by the base of the growing fruit cluster

9 Once all of the flowers in a cluster are pollinated (at least 5 fruits in a cluster) untie the mesh bag that was used to mark the cluster and place it over the fruit. You may need to use a large mesh bag or multiple bags to fit them all.



Mesh bag over developing fruit

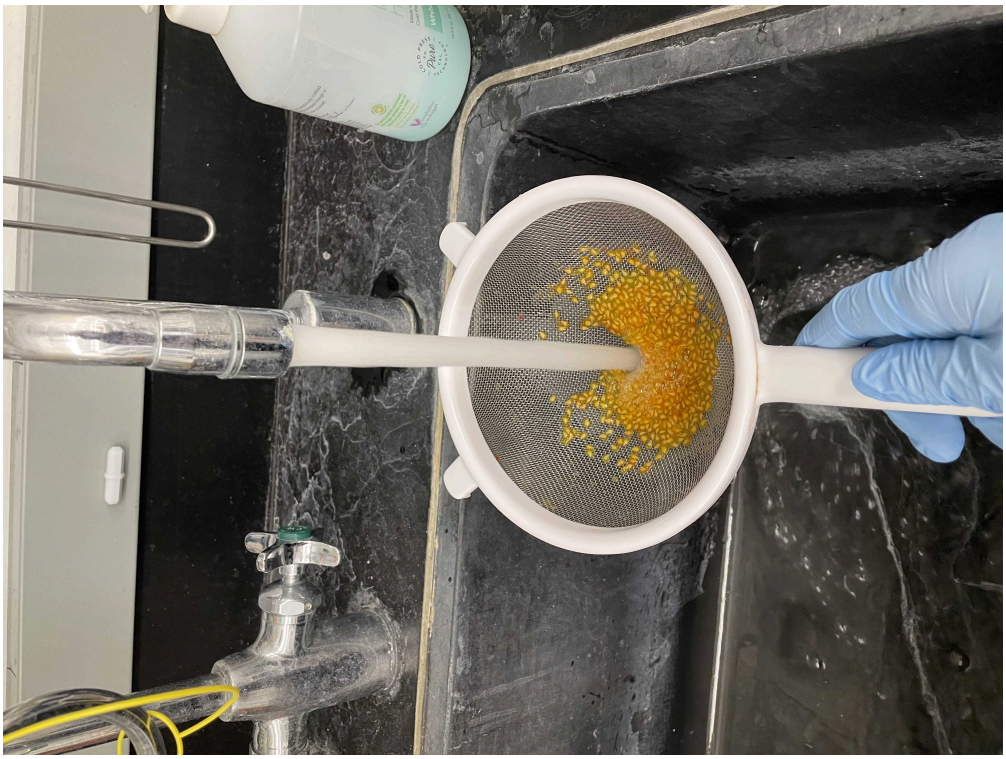
10 About twice a week, prune the plants to prevent overgrowth and blossom end rot (although pimpinellifolliums are not very susceptible for blossom end rot). This can be excess leaves, branches where fruit are not being pollinated, areas where clusters have already been harvested, off of the top of the plant to keep from getting too tall, etc. If a cluster that is being pollinated is accidentally pruned off, find another, newly emerging flower cluster, tie a mesh bag near it, and begin to pollinate that one instead.

11 When all or most of the fruit are red on a given cluster, remove the cluster and place it in a Ziploc bag labeled with the accession identifier. If the cluster has very uneven ripening, close the Ziploc bag and leave on the lab benchtop for a night or two until the remaining fruits are red.

NOTE: Ripe fruits can be stored at 4°C for up to two weeks.

12 Extract the seeds by either cutting the fruits open with a knife / scalpel or pressing the seeds into a large container with water (approximately 2L). the fruit skins will be floating on top of the water - so you can easily remove them by hand.

For separating the seeds from the pulp - use the fine strainer to wash all of the flesh pulp under running water.



13 Put the cleaned seeds harvested off one plant into a jewelry mesh bag, to keep them together, and put them in 50% bleach solution for 10 minutes, to remove the remaining flesh.

Clean the seeds in the mesh bag under the running tap water thoroughly to remove all of the bleach



14 Dry the seeds in the mesh bag on the paper towel on the benchtop for over 2-3 days. If necessary - change the paper towel.

The seeds will form a clump and might stick together in the process



15 Un-clump the seeds by rubbing the mesh bag between your fingers so that majority of the seeds do not clump together. Put all of the seeds into a 5 ml cryovial and label it accordingly with 1) Accession ID 2) harvest date.

Prior to storage - leave the cryovial open in the dessicator connected to a vacuum for over-night to remove the remaining humidity. Afterwards - close the cryovials and store at room temperature.