



CARE AND USE OF ARABIDOPSIS SEED LOADER

1. A video CD has been included with the Arabidopsis Seed Loader which demonstrates the process of loading the seeds into the loader and then transferring the seeds to a microplate. Please watch this demonstration.
2. The VP 725ZB-3 is made from anodized aluminum and contains 96 seed collector holes that are 0.7 mm in diameter at the top and taper to 0.4 mm.
3. The 96 seed collector holes each have a stainless steel pin 0.229 mm diameter in the center of each hole. These pins are mounted on a spring-loaded plate and the distance the pins project into the hole can be raised and lowered with the four adjustment screws (see Figure 1).

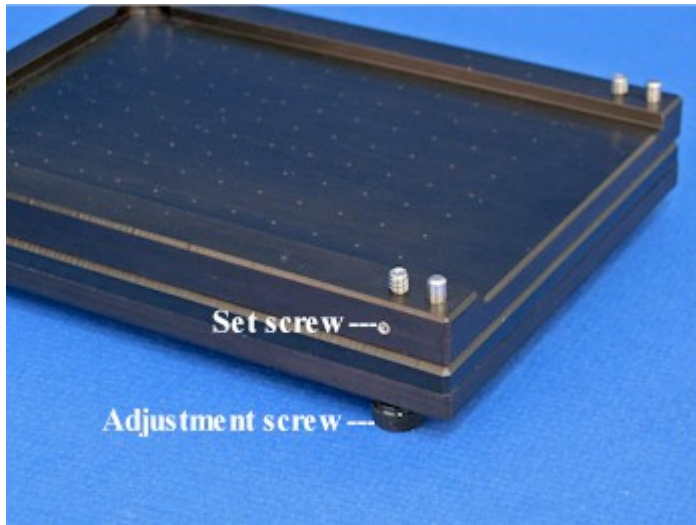


Figure 1

4. The four adjustment screws are adjusted up and down by turning the plastic knobs on the bottom of the VP 725ZB-3. Once the proper height adjustment is reached they are locked in place using the set screws on the side. See Figure 1.
5. The purpose of the pins is two-fold: first, to adjust the depth of the hole and therefore allow for only one seed/hole. With different varieties or sources of Arabidopsis seeds, the size of the seeds may vary. This will allow you to adjust the dispenser accordingly. The second purpose is to push the seeds out of the holes by pressing down on the VP 725ZB-3 when it is inverted over the microplate.

6. **Application:** Several techniques may be employed to load the Arabidopsis seeds in the holes:

A. Place an excess of seeds in the VP 725ZB-3 and swirl them around until all the holes are loaded then incline the VP 725ZB-3 over a catch dish and continue swirling until the unloaded seeds fall into the catch dish.

B. Place an excess of seeds in the VP 725ZB-3 and use a soft artist's paint brush to move the seeds over the holes and load them. See Figure 2. Then incline the VP 725ZB-3 over the catch dish and use the brush as shown in the video to remove the excess seeds.

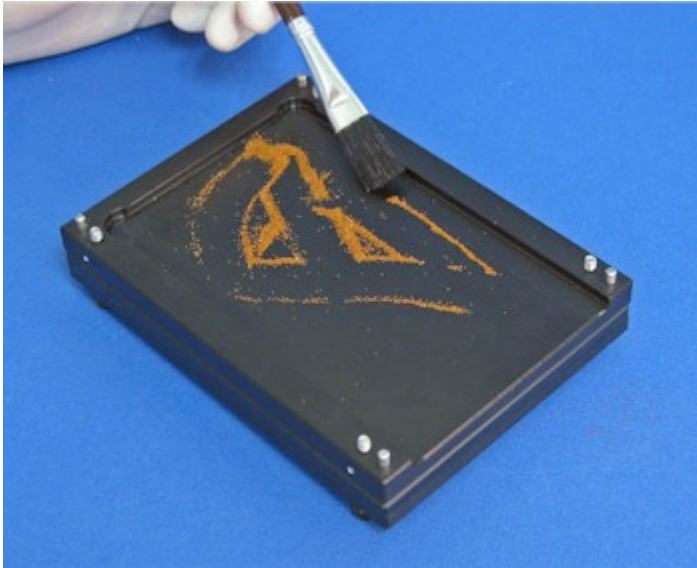


Figure 2

7. Once the VP 725ZB-3 is loaded with Arabidopsis seeds place a Nunc 1 ml filter plate without frit (part Number 278012) but with nutrient agar (See: Patrick Krysan, (2004) Ice-Cap. A High-Throughput Method for Capturing Plant Tissue Samples for Genotype Analysis. Plant Physiology, July 2004, Vol 135, pp. 1162-1169). See Figure 3.

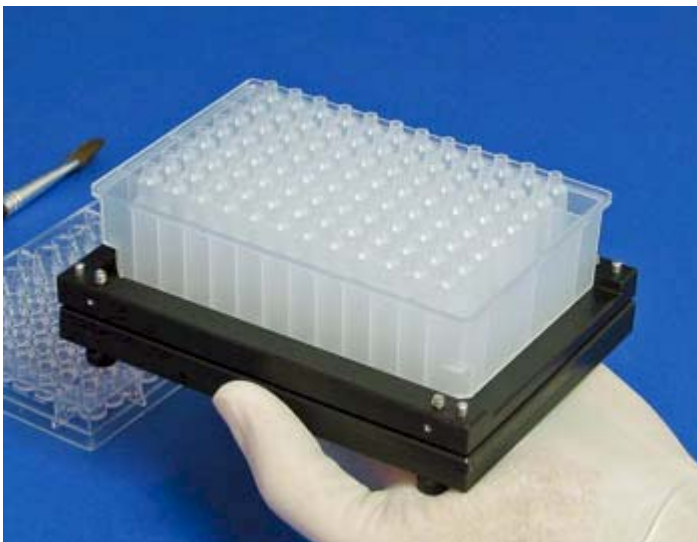


Figure 3.

8. Note that the VP 725ZB-3 is made specifically for the Nunc #278012 plate which has narrower top than most microplates. Consequently you cannot directly load most other microplates with the VP 725ZB-3. However, you can use the Nunc #278012 as an intermediary plate to load other microplates as shown in the video. Also see Figure 4, which illustrates this technique.
9. The VP 725ZB-3 can be cleaned by wiping with alcohol.



Figure 4.