tomato hypocotyl grafting

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plant grafting lab

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1 Tomato (*Solanum lycopersicum*) cultivar Zhongshu No.4 seeds were surface sterilized with 75% ethanol for 1 min, and in sterilization solution containing 35% commercial bleach solution (5.25% [w/v] sodium hypochlorite) and 0.1% Tween-20 (Bio-Rad, Shanghai, China) for 6 min.

2 Plates were tilted using a glass rod of 0.7–1 cm in diameter.

3 The seeds were placed in 1/2strength MS medium containing 0.8% agarose.

4 The germinated seedlings were grown for 5 to 7 d before grafting in growth chambers and maintained under 16 h of light at 25°C and 8 h of dark at 18°C and 60% relative humidity.

5 Seedlings were chosen with long straight hypocotyls and the hypocotyl was cut transversely while on the agar.

6 Cut the petiole to the middle part of the hypocotyl flat as a scion, take another seedling that is relatively straight and the hypocotyl diameter is similar to that of the scion, and cut the upper 1/3 of the hypocotyl as the rootstock.

7 Forceps were used to keep the seedling stable while cutting the hypocotyl with the razor blade under a dissecting microscope,It was important to make the cut quickly and to push the blade rather than pull it, so that the cut surface was clean and smooth.

8 The scion was lifted up to bring it to the stock, and the stock was carefully picked up to approach the scion and connect them together. Note that it is important to be aware of raising the graft union up away from the agar surface (Due to the large volume of tomato seedlings, in order to prevent the interface part from being difficult to heal due to contact with the culture medium, a hole can be dug in the culture medium below the interface part with tweezers to suspend the interface part).

9 The scion or stock was carefully and slightly pushed to adjust the relative position of the two parts, and the graft was inspected from all sides of the graft junction to make sure that the two parts connected and supported each other thoroughly.

10 The plate was returned to the growth room to the same vertical position with the thin side downward and the thick side upward.

11 The grafted tomato seedlings were placed in the growth chambers upright for 0, 0.5, 1, 2, 6, 12 h and inspected regularly.

12 The tomato seedlings grafted at different time points were assessed under a microscope, the plants were gently moved with tweezers, and only those plants that were not separated from the junction were used as samples for successful grafting.

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