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*Article*

# A New Grafting Method for Watermelon to Inhibit Rootstock Regrowth and Enhance Scion Growth

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**Abstract:** Grafting is an effective way to increase plant tolerance to biotic and abiotic stressors, it is widely used in watermelon production. However, grafting is labor intensive due to the additional time is required, such as the management of rootstock regrowth. This study used a new grafting tool to destroy (remove) the epidermis of pumpkin and bottle gourd rootstock cotyledon base during grafting, we called this a new grafting method. Compared with the traditional grafting (100%), the new grafting method had significantly lower rate of rootstock regrowth (2-23%), higher watermelon scion dry weight and leaf area. In addition, the time used for the new hole insertion and one cotyledon grafting method to destroy (remove) the epidermis of rootstock cotyledon base (4.2 s/plant, 4.2 s/plant) is significantly shorter than the time required to remove the rootstock regrowth manually in the traditional grafting (9.3 s/plant, 8.8 s/plant). Thus, this study developed a new grafting method for watermelon to inhibit rootstock regrowth and enhance scion growth, and this new method is cost-effective for grafted watermelon seedlings.

**Keywords:** *Citrullus lanatus*; grafting tool; rootstock regrowth; scion growth; vegetable grafting

## 1. Introduction

Grafting plants permits us to select and combine different rootstock and scion phenotypes, such as soilborne disease resistance and fruit yield traits, independently of genetic introgression [1,2]. Watermelon is an important horticultural crop and grafting is widely used in its production [3,4]. The main purpose of watermelon grafting is to solve fusarium wilt and root-knot nematode problems [5,6], to increase nutrient use efficiency [7], tolerance to salinity [8], cold and drought [9,10], and to increase fruit yield [11]. Zhao [11] reported that the productivity of grafted watermelon can be 13-25% higher than nongrafted plants.

Despite the above advantages of grafting in watermelon, the price of grafted seedlings is much higher than ungrafted ones, which undoubtedly limits the wide use of this technique [4]. Grafting is labor intensive due to the additional time is required, such as the management of rootstock regrowth [12]. Rootstock regrowth can result in graft failure, or a decrease in yield by competing with the scion for water and nutrients [4]. The two most common commercial watermelon grafting methods are the hole insertion and the one cotyledon techniques [13]. Rootstock regrowth is unavoidable for the above two grafting methods, since both grafting methods often leave bud meristem tissue at the base of the rootstock cotyledon, resulting in rootstock regrowth occurring after grafting, which is a major problem inhibiting the use of grafted watermelon plants [4]. To solve this problem, new grafting method was developed, for example, there is no rootstock regrowth with splice grafting (both cotyledons removed from the rootstock) because meristem tissue lies below the axillary bud at the base of the cotyledon and is completely removed [4,14,15]. In addition, chemical compounds such as fatty alcohol was also used to control the rootstock regrowth [16,17].

However, splice grafting often results in a low graft survival rate and poor seeding quality [14,15]. In addition, labor is required for the chemical application, and damage to seedlings can occur [16,17]. Thus, other approach controlling rootstock regrowth should be investigated. This study developed a new grafting method, in which the epidermis of rootstock cotyledon base was destroyed (removed) by a new tool during grafting, as a result, lower rootstock regrowth rate and enhanced scion growth was observed compared with traditional grafting, in addition, this method is labor saving.

## 2. Materials and Methods

### *2.1. Experimental Location and Design*

The experiment was conducted in a plant growth room at the National Center of Vegetable Improvement in Huazhong Agricultural University, Central China (latitude, 30°27′N; longitude, 114°20′E; and altitude 22 m above sea level). The new grafting method was evaluated by comparing with traditional grafting method, with or without rootstock regrowth removal. There were 3 replicates in each treatment, 25 plants for each replicate.

### *2.2. Plant Material and Cultivation*

Watermelon cv. Zaojia 8424 (*Citrullus lanatus,* Shanghai Wells Seed Co., Ltd) was used as the scion, interspecific pumpkin hybrid cv. Qingyanzhen No.1 (*C. maxima* × *C. moschata*, Qingdao Academy of Agricultural Sciences) and bottle gourd cv. Jingxinzhen No.1 (*Lagenaria siceraria*, Jingyan Yinong (Beijing) Seed Sci-Tech Co., Ltd) were used as the rootstocks. The scion and rootstock seeds were sown into 128 and 72-cell trays respectively, one seed in one cell, filled with seedling substrate (Shandong Shangdao Biotech Co., Ltd). During the cultivation, the day and night temperature was 28°C and 18°C, photosynthetic photon flux density was 170 μmol • $m^{-2}$ • $s^{-1}$, with 14/10 h photoperiod, day relative humidity was 65-85%.

### *2.3. Grafting*

One cotyledon grafting was conducted at day 11 after rootstock and scion seeds were sown, while hole insertion grafting was conducted at day 11 after rootstock seeds sowing, and 7 days after scion seeds sowing. Grafting was done as described by Hassell et al. [13]. Immediately after grafting, the plants were placed in the healing chambers. The plants were maintained in complete darkness at day 1 and were maintained under low light (80 μmol • $m^{-2}$ • $s^{-1}$) from day 2 to day 7, the light intensity was kept normal (170 μmol • $m^{-2}$ • $s^{-1}$, 14/10 h photoperiod) from day 7. The day and night temperature were 28°C and 18°C during graft healing. The humidity was kept above 95% during the first 5 days, then decreased to 85% from day 6 to day 10. The plants were removed from the healing chamber at day 10 and were placed in the growth room, following common practice.

### *2.4. New Grafting Tool Development and Usage*

The new grafting tool was developed by Huazhong Agricultural University, we already applied the patent (Application Number: 202110518435.5, China National Intellectual Property Administration). Briefly, one end is composed of conical file sticking with emery grain, which is convenient to destroy (remove) the epidermis of rootstock cotyledon base (Figure 1). The opposite end is grafting needle, the purpose is to make a hole in the hole insertion grafting method (Figure 1).

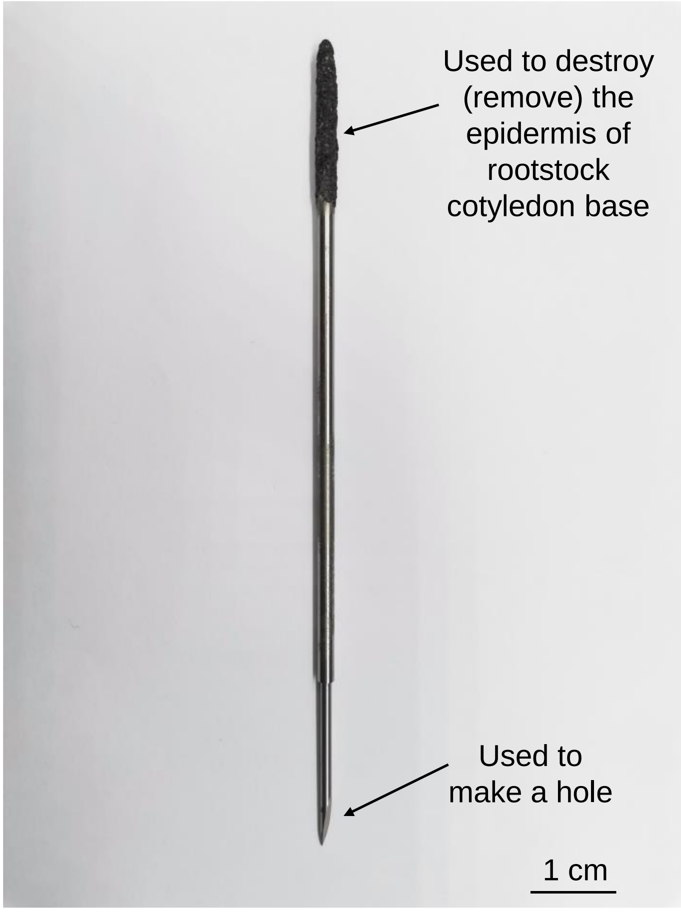


**Figure 1.** New grafting tool used in this study.

The usage of this new grafting tool was shown in Figures 2 and Supplementary Material Video S1. For hole insertion grafting, firstly remove the rootstock true leaf and growing point, then destroy (remove) the epidermis of rootstock cotyledon base using the emery grain end, then make a hole using the grafting needle end, finally insert the cut scion. For one cotyledon grafting, firstly remove the growing point and one cotyledon of rootstock, then destroy (remove) the epidermis of rootstock cotyledon base using the emery grain end, finally hold the rootstock and scion in place with a grafting clip (Figures 2).

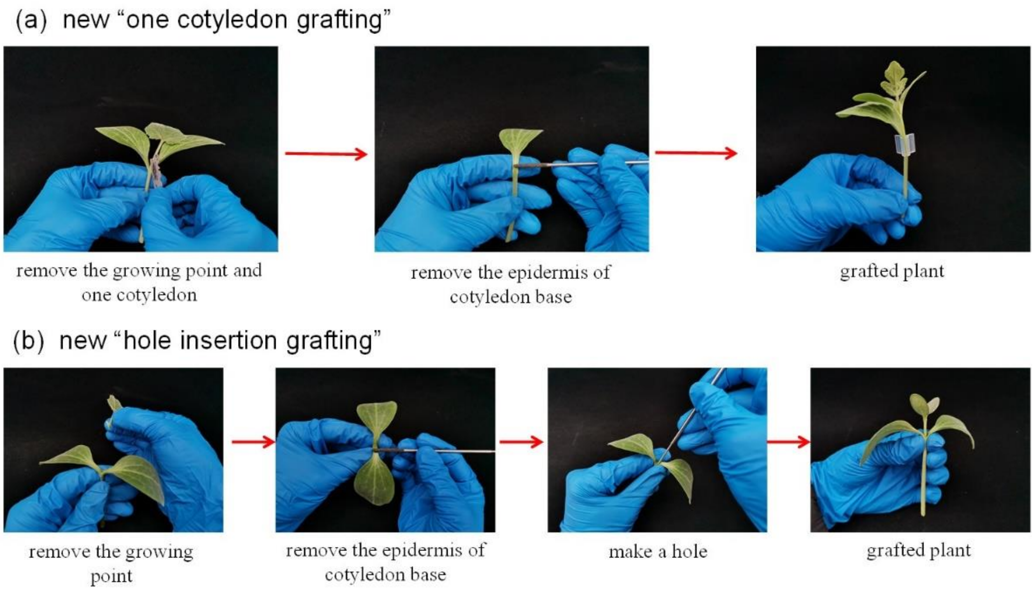


**Figure 2.** Pictures to show how the new grafting method is used.

*2.5. Grafted survival measurement*

The survival of the grafted plants was assessed at day 14 after grafting. The grafted plants were considered alive and survived if the scion leaves and the rootstock stems were turgid, whereas severely wilted scion leaves and stems of both the scion and the rootstock were considered as graft failure. Survival rate = (Survived number/total number of grafted plants) × 100%.

*2.6. Measurement of rootstock regrowth rate*

Rootstock regrowth rate was measured at day 14 after grafting. For this measurement, the rootstock regrowth was not removed. Rootstock regrowth rate = (rootstock regrowth number/total number of grafted plants) × 100%.

*2.7. Anatomical study of rootstock cotyledon base*

To observe the rootstock regrowth earlier, histological study of pumpkin rootstock cotyledon base in the traditional and new hole insertion grafting plants was conducted at 0 h, 12 h, 24 h and 36 h after grafting, using the paraffin section method. Paraffin section and staining were done as described by El-Gazzar et al [18].

*2.8. Measurement of watermelon scion dry weight, leaf area of watermelon scion and rootstock regrowth*

Plants were sampled at day 14 after grafting. The fresh samples of watermelon scion were placed into a forced air oven at 105°C for 15 min, and then at 70°C for 3 days to determine their dry weights. Leaf area of watermelon scion and rootstock regrowth were measured using an area meter LI-3100C (Li-Cor, Inc., Lincoln, NE, USA). Only the rootstock regrowth plants were investigated and calculated for the rootstock regrowth leaf area measurement.

*2.9. Working efficiency measurement*

The experiment was conducted by three skilled master students. The time used to destroy (removal) rootstock cotyledon epidermis in the new grafting method and the removal of rootstock regrowth 3 times in the traditional method was counted.

*2.10. Data analyses*

All data were analyzed by Student's t-test using SPSS 25.0 software (SPSS Inc., Chicago, IL, USA), the figures were made using GraphPad Prism 8.0 (GraphPad Software Inc., San Diego, CA). Significance between new grafting and traditional grafting method was set at $p < 0.05^*$, $p < 0.01^{**}$ or $p < 0.001^{***}$, respectively.

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## 3. Results

*3.1. New grafting method had no significant effect on the graft survival rate*

There was no significant difference on the graft survival rate between new grafting method and traditional grafting method, for pumpkin and bottle gourd, and for hole insertion and one cotyledon grafting methods (Figure 3). The graft survival rate of plants grafted onto pumpkin was 84-98% (Figure 3a,b), while the value was 93-100% for the plants grafted onto bottle gourd rootstock (Figure 3c,d).

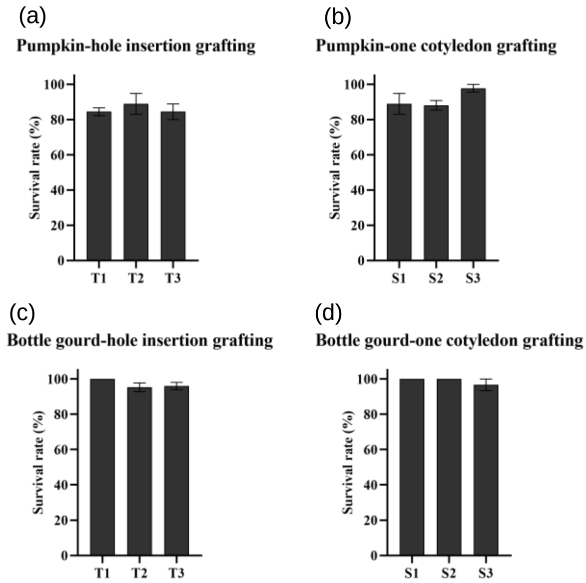


**Figure 3.** Graft survival rate at day 14 after grafting (n=3). T1 and S1, new "hole insertion grafting" and "one cotyledon grafting", respectively, without rootstock regrowth removal; T2 and S2, traditional "hole insertion grafting" and "one cotyledon grafting", respectively, with rootstock regrowth removal manually 3 times; T3 and S3, traditional "hole insertion grafting" and "one cotyledon grafting", respectively, without rootstock regrowth removal.

*3.2. New grafting method largely decreased the rootstock regrowth rate*

Compared with traditional grafting, few rootstock regrowth was seen in the plants using the new method (Figure 4). The rootstock regrowth rate was decreased from 100% to 8% in pumpkin hole insertion using new method as compared with traditional method, and the value was decreased from 100% to 2% in one cotyledon grafting (Figure 5a). For bottle gourd rootstock, the rootstock regrowth rate was decreased from 100% to 23% using hole insertion, and from 100% to 9% using one cotyledon grafting (Figure 5b). Anatomical observation of pumpkin rootstock cotyledon base also showed that new method inhibited the rootstock regrowth at earlier stage (24 and 36 h) after grafting (Figure 6). Among the few rootstock regrowth plants using the new method, the regrowth leaf area was significantly lower than the traditional grafting method, except for the hole insertion of bottle gourd rootstock (Figure 5c,d).

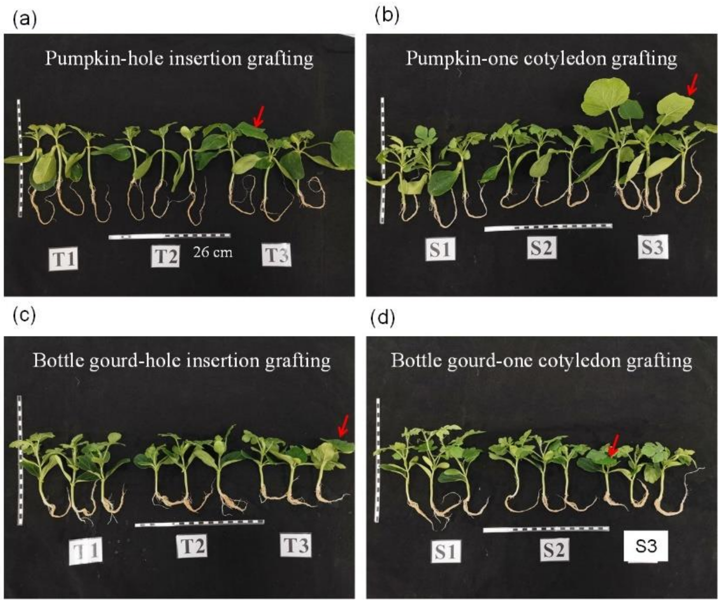


**Figure 4.** Watermelon growth pictures at day 14 after grafting. T1 and S1, new "hole insertion grafting" and "one cotyledon grafting", respectively, without rootstock regrowth removal; T2 and S2, traditional "hole insertion grafting" and "one cotyledon grafting", respectively, with rootstock regrowth removal manually 3 times; T3 and S3, traditional "hole insertion grafting" and "one cotyledon grafting", respectively, without rootstock regrowth removal. Red arrow indicates rootstock regrowth.

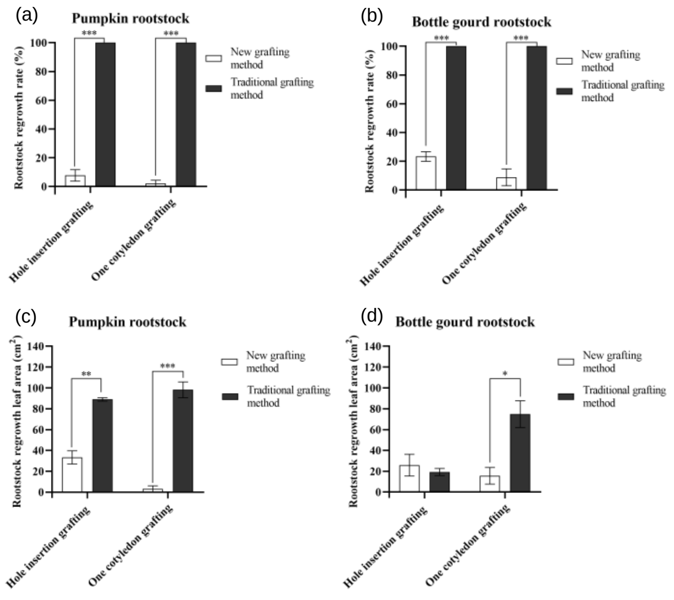


**Figure 5.** Rootstock regrowth rate (a, b) and rootstock regrowth leaf area (c, d) at day 14 after grafting (n=3). The rootstock regrowth was not removed for the new and traditional grafting methods. For the measurement of rootstock regrowth leaf area, only the rootstock regrowth plants were investigated and calculated. *, ** and *** indicates significant difference at $p < 0.05$, 0.01 and 0.001 level.

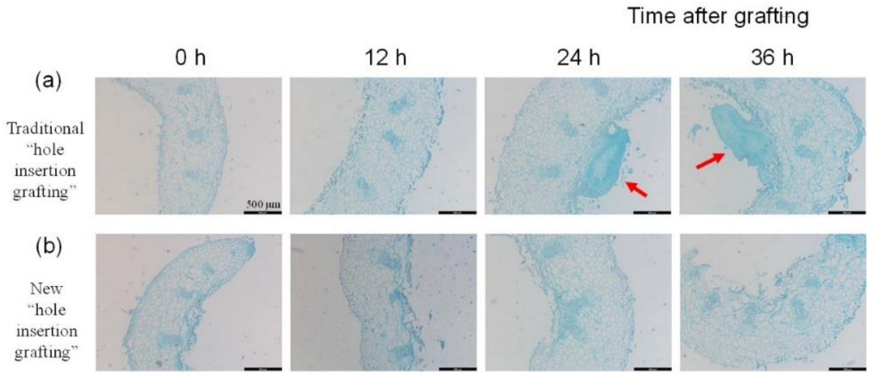


**Figure 6.** Anatomical observation of pumpkin rootstock cotyledon base in the traditional and new "hole insertion grafting" plants. Red arrow indicates rootstock regrowth.

*3.3. New grafting method increased scion growth*

Compared with pumpkin traditional grafting without rootstock regrowth removal, new grafting method significantly increased scion dry weight, increased by 78% and 75% for hole insertion and one cotyledon grafting, respectively (Figure 7a,b). Removal of pumpkin rootstock regrowth is beneficial for the scion dry weight using the traditional method (Figure 7a,b). For bottle gourd roottsock, new grafting method significantly increased scion dry weight, increased by 25% and 24% for hole insertion and one cotyledon grafting, respectively, as compared with traditional grafting with rootstock growth removal (Figure 7c,d).

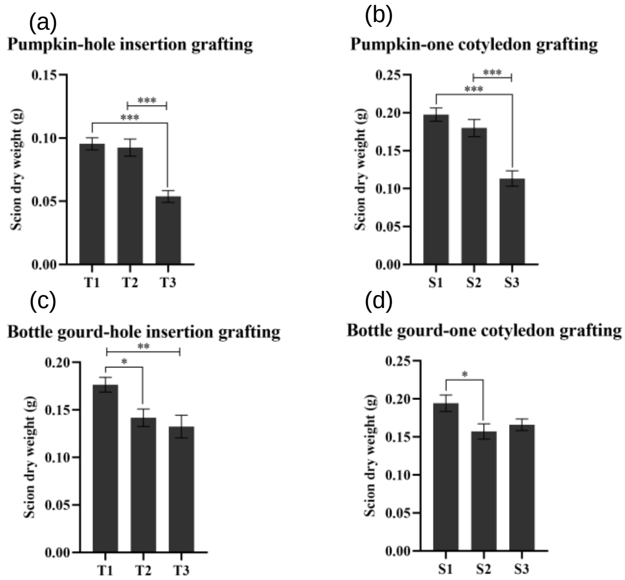


**Figure 7.** Watermelon scion dry weight at day 14 after grafting (n=3). T1 and S1, new "hole insertion grafting" and "one cotyledon grafting", respectively, without rootstock regrowth removal; T2 and S2, traditional "hole insertion grafting" and "one cotyledon grafting", respectively, with rootstock regrowth removal manually 3 times; T3 and S3, traditional "hole insertion grafting" and "one cotyledon grafting", respectively, without rootstock regrowth removal. *, ** and *** indicate significant difference at $p < 0.05$, 0.01 and 0.001 level.

Compared with traditional grafting without rootstock regrowth removal, new grafting method significantly increased scion leaf area for both rootstock, increased by 49% and 67% for pumpkin hole insertion and one cotyledon grafting, respectively (Figure 8a,b), while the value was 26% and 25%, respectively for bottle gourd (Figure 8c,d). Removal of pumpkin rootstock regrowth is beneficial for the scion leaf area using the traditional method (Figure 7a,b).

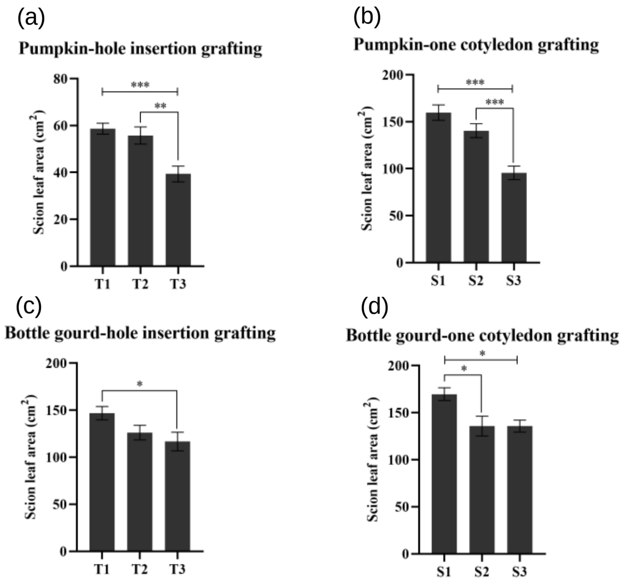


**Figure 8.** Watermelon scion leaf area at day 14 after grafting (n=3). T1 and S1, new "hole insertion grafting" and "one cotyledon grafting", respectively, without rootstock regrowth removal; T2 and S2, traditional "hole insertion grafting" and "one cotyledon grafting", respectively, with rootstock regrowth removal manually 3 times; T3 and S3, traditional "hole insertion grafting" and "one cotyledon grafting", respectively, without rootstock regrowth removal. *, ** and *** indicate significant difference at $p < 0.05$, 0.01 and 0.001 level.

### *3.4. New grafting method is labor saving*

Compared with traditional grafting, new grafting method increase the time to destroy (remove) the epidermis of rootstock cotyledon, but decrease the time used for the removal of rootstock regrowth (Figure 9). By calculation of the working efficiency, the time used for the new method is much shorter than the traditional method, for hole insertion grafting, 4.2 vs 9.3 s/plant; for one cotyledon grafting, 4.2 vs 8.8 s/plant. Considering very few rootstock growth appear using new grafting method (Figure 9), the amount of labour required to remove the rootstock regrowth is very small, thus, new grafting method is labor saving compared with traditional grafting method.

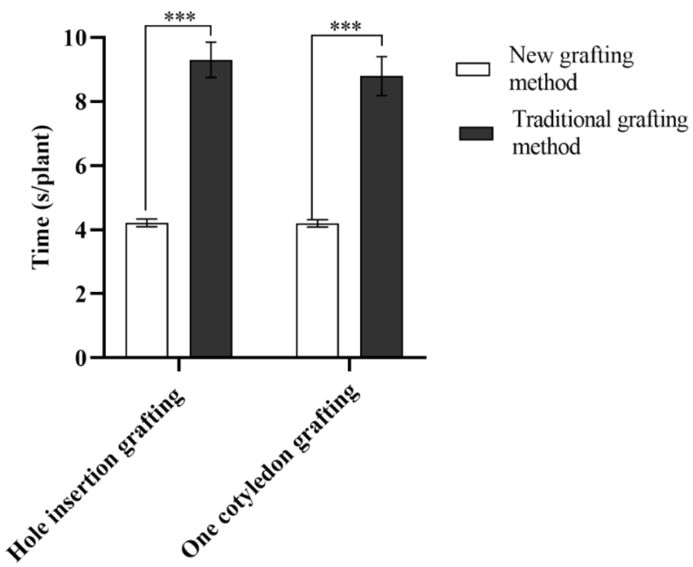


**Figure 9.** Time used to destroy (remove) pumpkin rootstock cotyledon epidermis for the new grafting method, and the time required to remove rootstock regrowth for the traditional grafting method (n=3). *** indicates significant difference at $p < 0.001$ level.

## 4. Discussion

*4.1. New grafting is an effective method to save labor and enhance watermelon scion growth*

Grafting of watermelon scions onto pumpkin or bottle gourd rootstocks is practiced in many of the major watermelon production regions of the world [2,4]. However, grafting needs additional labor, such as the removal of rootstock regrowth. Rootstock regrowth is a major concern for watermelon that are grafted with at least one intact rootstock cotyledon, as it is difficult to remove all the rootstock bud meristem tissue [4]. Many commercial rootstocks are vigorous and will quickly overtake the scion variety if allowed to grow, so scouting for and removal of rootstock regrowth is required. Previous studies [12,16] reported that splice grafting or fatty alcohol treatments can be used to eliminate the meristematic regrowth of rootstock, however, the seedling survival and quality cannot be guaranteed, in addition, the application of exogenous chemicals must be carefully done and more labor work is needed.

In this study, considering the regrowth rate of rootstock, scion growth and labor output, the new grafting has significant advantage compared with traditional grafting (Figures 5, 7-9). Compared with traditional hole insertion and one cotyledon grafting methods, the new method increases the procedure of destroying (removing) the epidermis of rootstock cotyledon base, but largely decreases the procedure of removing the rootstock regrowth manually, which is often conducted three times during seedling stage. The increased scion growth using new grafting method, which could be attributed to the more photosynthate distributed to the scion, rather to the absent rootstock regrowth [12,14,15]. Compared with traditional grafting method, the new grafting can save about 4 s for each grafted seedling (Figure 9). Obviously, labor intensive grafting propagation can potentially benefit from the effective use of this grafting method to minimize labor inputs, thus reduce the overall cost of producing grafted watermelon seedlings and could help increase the adoption of grafted cucurbit plants in the world.

*4.2. Possible mechanism of new grafting tool inhibiting rootstock regrowth*

The rootstock regrowth is a type of axillary bud regeneration, however, the mechanism is unclear. The epidermal cell layer of plants has important functions in regulating plant growth and development [19]. Another factor regulating plant growth and morphogenesis is the hormone cytokinin (CK). Perturbations of CK metabolism and signaling in the epidermis impact shoot development by affecting cell division and differentiation, consistent with the possibility that the epidermis is a source or sink of the hormone [19,20]. Numerous CK metabolism and signaling genes are expressed in the epidermis [21]. Previous study showed that cytokinin signaling precedes *WUS* expression in leaf axils and activates *WUS* expression de novo in the leaf axil to promote axillary meristem initiation [22]. In this study, removal of epidermis of rootstock cotyledon base resulted in a inhibited rootstock regrowth. Speculatively, the removal (destroy) of epidermis of rootstock cotyledon base leads to the deficiency of cytokinin synthesis or signaling, which in turn is incapable of inducing the *WUS* expression and thus the leaf axillary bud development. However, the possible mechanism needs to be proved in future.

## 5. Conclusions

This study developed a new grafting method, in which the epidermis of rootstock cotyledon base was destroyed (removed) by a new grafting tool. Using this new method, the rootstock regrowth rate was largely decreased, and watermelon scion growth was enhanced compared with traditional grafting (Figure 10). Taken together, this new method is labor saving and beneficial for the growth of grafted watermelon seedlings. It has great potential application prospect in practice of cucurbit grafting.

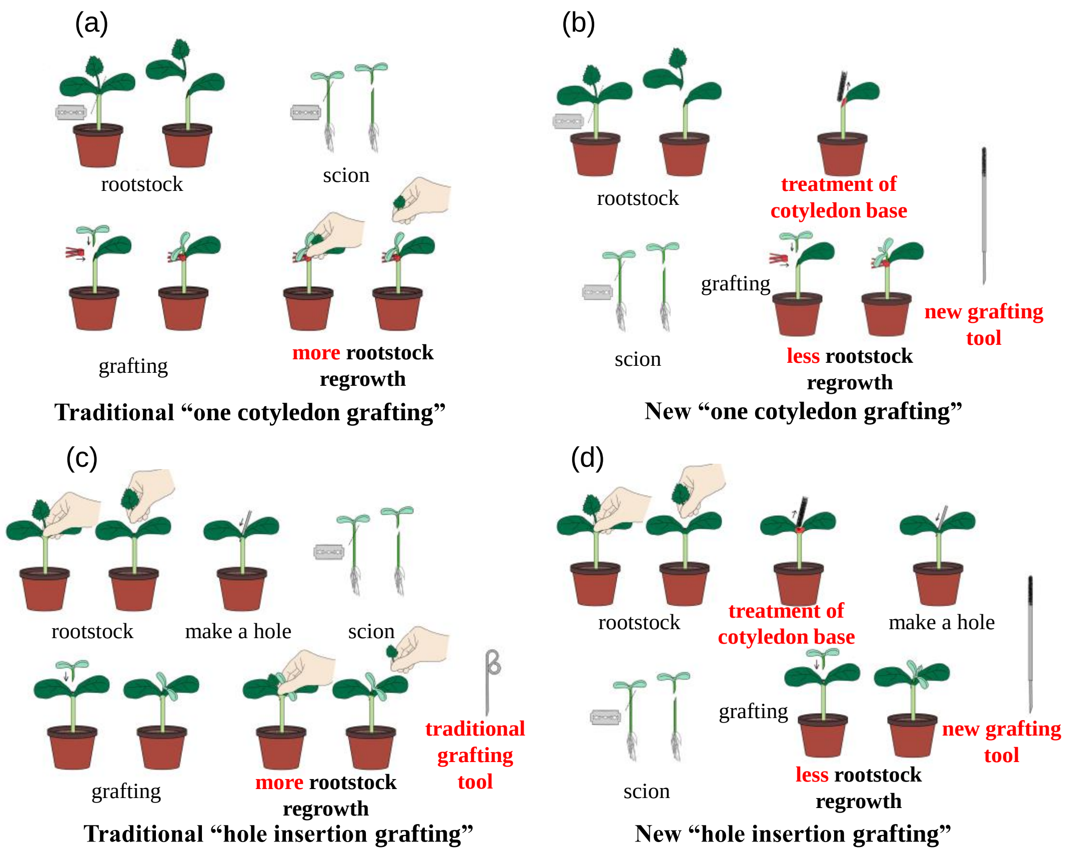


**Figure 10.** A summary diagram for the new grafting method (b, d) compared with traditional grafting method (a, c).

## 6. Patents

Yuan Huang, Changjin Liu, Zhilong Bie, Weiguo Lin, Xiangshuai Wu, Xiaohu Fu. A new cucurbit grafting tool used to inhibit rootstock regrowth and a new grafting method, Application Number: 202110518435.5, China National Intellectual Property Administration.

**Supplementary Material:** The following is available online at www.mdpi.com/xxx/s1, Video S1: Method to show how the new grafting tool is used.

**Author Contributions:** Conceptualization, Y.H. and Z.L.B.; methodology, C.J.L.; formal analysis, C.J.L. and X.S.W.; investigation, C.J.L., X.S.W. and M.X.; new grafting tool development, Y.H., W.G.L. and X.H.F.; pictures drawing, C.R.F.; writing—original draft preparation, Y.H. and C.J.L.; writing—review and editing, Y.H.; supervision, Y.H.; project administration, Y.H.; funding acquisition, Y.H. and Z.L.B. All authors have read and agreed to the published version of the manuscript.

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**Data Availability Statement:** The data presented in this study are available on request from the corresponding author.

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**Conflicts of Interest:** The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results.

## References

1. Mudge, K. A History of Grafting. *Hortic Rev.* **2009**, 35, 437–494.
2. Lee, J.M.; Kubota, C.; Tsao, S.J.; Bie, Z.L.; Hoyos Echevarria, P.; Morra, L.; Oda, M. Current status of vegetable grafting: Diffusion, grafting techniques, automation. *Sci Hortic.* **2010**, 127, 93–105.
3. Bantis, F.; Koukounaras, A.; Siomos, A.S.; Dangitsis, C. Impact of scion and rootstock seedling quality selection on the vigor of watermelon–interspecific squash grafted seedlings. *Agriculture* **2020**, 10(8), 326.
4. Devi, P.; Lukas, S.; Miles, C. Advances in watermelon grafting to increase efficiency and automation. *Horticulturae* **2020**, 6, 88.
5. Yetısir, H.; Sari, N.; Yucel, S. Rootstock resistance to Fusarium wilt and effect on watermelon fruit yield and quality. *Phytoparasitica* **2003**, 31, 163–169.
6. Thies, J.A.; Ariss, J.J.; Kousik, C.S.; Hassell, R.L.; Levi, A. Resistance to southern root-knot nematode (*Meloidogyne incognita*) in wild watermelon (*Citrullus lanatus* var. citroides). *J Nematology*. **2016**, 48(1), 14–19.
7. Nawaz, M.A.; Wang, L.M.; Jiao, Y.Y.; Chen, C.; Zhao, L.; Mei, M.J.; Yu, Y.L.; Bie, Z.L.; Huang, Y. Pumpkin rootstock improves nitrogen use efficiency of watermelon scion by enhancing nutrient uptake, cytokinin content, and expression of nitrate reductase genes. *Plant Growth Regul.* **2017**, 82, 233–246.
8. Yang, Y.J.; Lu, X.M.; Yan, B.; Li, B.; Sun, J.; Guo, S.R.; Tezuka, T. Bottle gourd rootstock-grafting affects nitrogen metabolism in NaCl-stressed watermelon leaves and enhances short-term salt tolerance. *J Plant Physiol*. **2013**, 170, 653–661.
9. Shi, X.F.; Wang, X.B.; Cheng, F.; Cao, H.S.; Liang, H.; Lu, J.Y.; Kong, Q.S.; Bie, Z.L. iTRAQ-based quantitative proteomics analysis of cold stress-induced mechanisms in grafted watermelon seedlings. *J Proteomics*. **2019**, 192, 311–320.
10. Yavuz, D.; Seymen, M.; Suheri S.; Yavuz, N.; Turkmen, O.; Kurtar, E.S. How do rootstocks of citron watermelon (*Citrullus lanatus* var. citroides) affect the yield and quality of watermelon under deficit irrigation? *Agr Water Manage*. **2020**, 241, 106351.
11. Huang, Y.; Zhao, L.Q.; Kong, Q.S.; Cheng, F.; Niu, M.L.; Xie, J.J.; Nawaz, M.A.; Bie, Z.L. Comprehensive mineral nutrition analysis of watermelon grafted onto two different rootstocks. *Hortic Plant J.* **2016**, 2(2), 105–113.
12. Memmott, F.D.; Hassell, R.L. Watermelon (*Citrullus lanatus*) grafting method to reduce labor cost by eliminating rootstock side shoots. *Acta Hortic.* **2010**, 871, 389–394.
13. Hassell, R.L.; Memmott, F.; Liere, D.G. Grafting methods for watermelon production. *HortScience* **2008**, 43, 1677–1679.
14. Devi, P.; DeVetter, L.W.; Lukas, S.; Miles, C. Exogenous treatments to enhance splice-grafted watermelon survival. *Horticulturae* **2021**, 7, 197.
15. Dabirian, S.; Miles, C.A. Increasing survival of splice-grafted watermelon seedlings using a sucrose application. *HortScience* **2017**, 52, 579–583.
16. Daley, S.L.; Hassell, R.L. Fatty alcohol application to control meristematic regrowth in bottle gourd and interspecific hybrid squash rootstocks used for grafting watermelon. *HortScience* **2014**, 49, 260–264.

 doi:10.20944/preprints202107.0693.v1

17. Choi, D.C.; Kwon, S.W.; Ko, B.R.; Choi, J.S. Using chemical controls to inhibit axillary buds of *Lagenaria* as rootstock for grafted watermelon (*Citrullus lanatus*). *Acta Hortic*. **2002**, 588, 43–48.
18. El-Gazzar, T.; Dawa, K.; Ibrahim,E.; El-Banna, M.; Mohamed, A. Anatomical study on watermelon grafting. *J Plant Production*. **2017**, 8, 999–1009.
19. Werner, S.; Bartrina, I.; Novak, O.; Strnad, M.; Werner, T.; Schmülling, T. The cytokinin status of the epidermis regulates aspects of vegetative and reproductive development in Arabidopsis thaliana. *Front Plant Sci.* **2021**, 12, 613488.
20. Chickarmane, V.S.; Gordon, S.P.; Tarr, P.T.; Heisler, M.G.; Meyerowitz, E.M. Cytokinin signaling as a positional cue for patterning the apical–basal axis of the growing Arabidopsis shoot meristem. *Proc Natl Acad Sci USA.* **2012**, 109, 4002–4007.
21. Yadav, R. K.; Tavakkoli, M.; Xie, M.; Girke, T.; Reddy, G.V. A high-resolution gene expression map of the Arabidopsis shoot meristem stem cell niche. *Development* **2014**, 141, 2735–2744.
22. Wang, J.; Tian, C.H.; Zhang, C.; Shi, B.H; Cao, X.W; Zhang, T.Q; Zhao, Z.; Wang, J.W; Jiao, Y.L. Cytokinin signaling activates *WUSCHEL* expression during axillary meristem initiation. *Plant Cell*, **2017**, 29, 1373–1387.