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Grafting Manual

Introduction to the Vegetable Grafting Manual

Grafted vegetable plants are ‘physical hybrids’ resulting from combining at least two varieties, a rootstock and at least one scion; first used to provide important traits and the second used to produce fruit. The process is analogous to organ transplantation in th rootstock and scion varieties and seedlings must be compatible, the operating room and patients clean and disease-free, the gra using appropriate methods, and the newly-grafted plants allowed to recover under specific conditions. This collaboratively develo manual describes major aspects of making grafted vegetable plants, emphasizing research-based information and approaches p by experience to be reliable. Still, despite much progress in grafting methodology, there are few absolute truths in vegetable graf and ongoing research and trial and error continue to improve locally-relevant techniques, tools, and knowledge. Therefore, the m is structured and offered as a “living document” that will be updated as new information and improvements become available. Re are encouraged to return to this page periodically to check for new manual components.

Continue to the Grafting Manual

Grafting Manual:
How to Produce Grafted Vegetable Plants

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USDA
United States Department of Agriculture
National Institute of Food and Agriculture

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Chapter 1 **November 2016**

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Synopsis:

Grafting is an old and new technology to enhance the sustainability of vegetable crop production. The history of introducing this practice is summarized.

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History of vegetable grafting

Grafting of vegetable seedlings is a unique horticultural technology that is used worldwide to overcome soil- borne diseases and pests and/or to increase plant vigor under various environmental stress conditions. Today, grafting is used especially when there are limited rotation or soil fumigation options. For example, almost all watermelon produced in Japan, Korea, southern Spain, southern Italy, Turkey, and Greece are grafted, and there are increasing numbers of grafted tomato, eggplant, pepper, cucumber, and melons worldwide. In hydroponic greenhouses, grafting has become a standard practice to increase plant vigor and yield. In high tunnels, heirloom cultivars are grafted onto modern rootstocks to overcome soil-borne disease. Grafting is also used to mitigate environmental stress such as salinity, drought, flooding and low temperature. This article summarizes the history of the development of vegetable grafting and its use worldwide.

The beginning – innovation by a small farmer

The oldest record of grafting vegetable plants is in 500 AD in China (Lee and Oda, 2003) when farmers joined together multiple gourd plants to develop a greater root system to increase the size of gourd fruit (Fig.1). However, vegetable grafting using two different species or cultivars for disease and pest management was not documented until the early 1900s. Koshiro Tateishi published an extension research journal article in Japan (1927) indicating that vegetable grafting can be a revolutionary technology for future vegetable production, and encouraging scientists to further develop the technology so that it would become a common practice among farmers. This article was briefly featured in a Korean extension newsletter (Ashita, 1927), which is often mistakenly cited as the first modern report of vegetable grafting. In his article, Tateishi reported that a small watermelon grower named Ukichi Takenaka in Hyogo, Japan, grafted watermelon onto pumpkin (or squash) and successfully, and easily overcame Fusarium wilt. Based on Takenaka's success, Tateishi set up various experiments to test watermelon grafting, compared grafting methods and recommended cleft grafting over approach grafting. He also noted possible issues of early female flower abortion of grafted plants in the field, as well as impact of grafting on watermelon fruit quality. Tateishi reported that while grafting was theoretically possible between these two species, it was not until farmer Takenaka did it, that grafting was considered to be a viable technology. Many research and extension efforts followed to use grafting at various agricultural experiment stations starting in 1929 (Oda, 1990).

By the 1950s, plastic tunnels had become popular to extend the production season of high value crops in Japan; however, the intensive production using the same land intensified the issue of soil-borne diseases, thereby accelerating the use of grafting (Fig. 2). During this era, seed companies made tremendous gains in breeding rootstocks for disease resistance, and they introduced grafting to many countries through their international marketing efforts.

Israel was one of the early adapters of commercial vegetable grafting. Crops other than cucurbits were grafted in the 1950s and 1960s. For example, eggplant was grafted onto scarlet eggplant (*S. integrefolium*) as a disease management strategy, In the 1960s commercial use of tomato grafting was introduced in Japan and Korea ((Lee and Oda, 2003). During the 1980s and 1990s, milestone grafting innovations were made, including the development of tube grafting, one-cotyledon grafting and healing methods suitable for commercial nursery operations (Itagi, 2009).

These methods integrated vegetable grafting into modern commercial nursery systems that used plug trays introduced from the United States. Tube grafting increased the speed of solanaceous grafting by 2-3 times (Itagi et al., 1990). One-cotyledon grafting was first developed by a watermelon grower (Saito, 1981) and was later introduced as the baseline method to mechanize grafting due to its simplicity compared with other grafting methods used for cucurbit plants (Onoda et al., 1992; Suzuki et al., 1995). Automation for grafting was studied intensively during this era and various grafting ro-

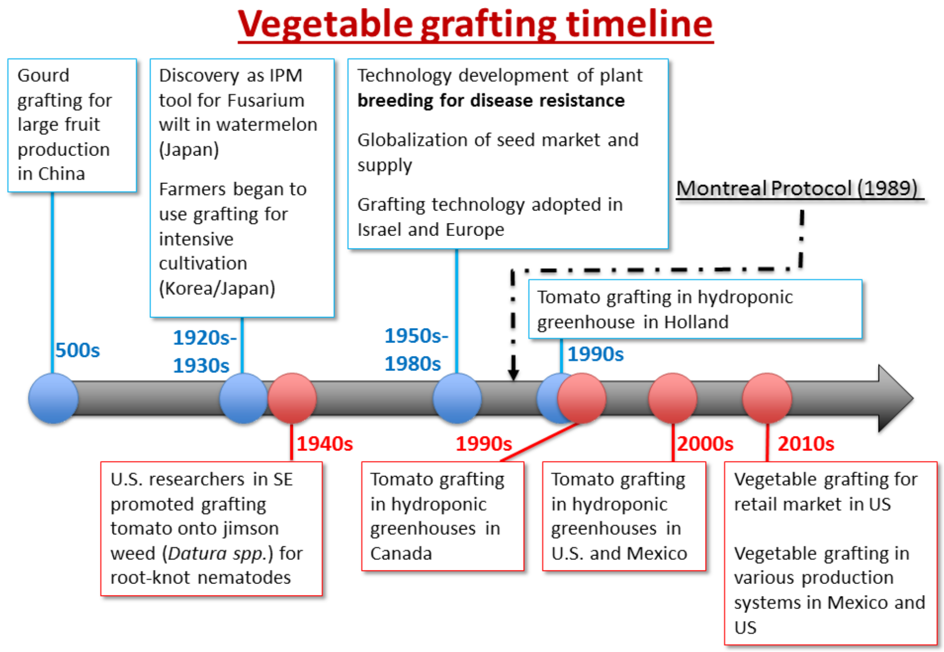

Figure 1. Timeline showing milestones in history of vegetable grafting in the world (upper part, blue) and in North America (lower part, red).

bots were invented and commercialized (Kurata, 1994). However, only a small number of robots were effectively used in commercial operations due to the precision required for preparing scion and rootstock plants suitable for machine use. In the early 1990s, indoor healing systems with environmental control were developed for mid to large-size nurseries (Itagi, 2009). An international symposium focused on transplant production technologies held in Yokohama, Japan (Hayashi et al., 1992) was a showcase of these technologies, and attracted many international scientists and engineers. After the Montreal Protocol identified methyl bromide as one of the substances to phase out and the protocol became effective in 1989, much effort by academia, commercial sectors and international organizations, including rootstock breeding and development, was made to introduce vegetable grafting as an alternative tool for soil-borne disease management in various countries.

History in North America

The U.S. developed one of the earliest innovations in grafting tomato. During the 1930s and 1940s, tomato was grafted onto jimson weed (*Datura stramonium*) and the technique was recommended to gardeners (Isbell, 1944). Southern growers used this technique to overcome root-knot nematodes (Lowman and Kelly, 1946); however, this practice was discontinued partly due to the possible transport of alkaloids to fruits from the rootstock (Lowman and Kelly, 1946). Similarly, germplasm that is considered as weeds have been introduced as rootstocks for other vegetable crops. A good example is Turkey berry weed (*Solanum torvum*) collected from Puerto Rico and Thailand, which are now widely used as rootstock for eggplant.

Commercial grafting used in today's North American vegetable industry was introduced

Figure 2. Bottle gourd rootstock seedlings and watermelon scion seedlings prepared for grafting in a rural village Shimohara in Nagano, Japan in 1967 (dated on April 5, Showa- 42 Year). Bottle gourd seedlings (rootstock) are at the stage of fully unfolding first true leaf inside a bamboo-framed tunnel. Watermelon seedlings (scion) are grown at a high density inside a small wooden tray filled with soil and are just about at the beginning of emerging cotyledons. The particular growth stage of young scion seedlings suggest that grafting would be done with the insertion method. (*Photo credit: Mountainlife, Wikimedia Commons*; Link http://www.wikiwand.com/ja/%E4%B8%8B%E5%8E%9F%E3%82%B9%E3%82%A4%E3%82%AB)

by Dutch growers in the 1990s. In the Netherlands, Dutch greenhouses were using grafting in the 1960s in their intensive cultivation systems; however, in the 1970s and 1980s, their use of grafting declined because other means such as resistant cultivars and chemical fumigants (i.e., methyl bromide) became available (L. Benne, personal communication). In the 1990s, grafting was re-introduced as the increased plant vigor was suitable for the long production cycle in modern greenhouse tomato production. Use of the tube grafting method contributed to making grafting more suitable for the production of large volumes of plants for commercial operations.

Grafting was also introduced in Canada in the 1990s to support the rapidly growing greenhouse industry in Canada and the U.S., and later in Mexico. Canada has been the primary supplier of grafted plants to the U.S. and Mexico, and long distance shipping has been a common practice in distributing grafted plants throughout North America.

In addition to the use of grafted plants in the greenhouse industry, various small growers and extension personnel in land-grant universities have contributed to the transfer of grafting technology. A commercial nursery in Oregon began introducing grafted heirloom tomato plants to retail markets in 2011. Seed retailers started selling small packages of rootstock seeds. This made grafting known to a wider range of stakeholders including home gardeners and professionals.

In Mexico, in the 2000s, grafting was introduced partly through a United Nations effort to reduce the use of methyl bromide in open fields and in tunnels (Martinez, 2015). However, the rapid development of the tomato greenhouse industry was another primary reason for the expanded use of vegetable grafting and its adoption as a standard practice in the greenhouse industry. More recently, grafting watermelon was introduced to mitigate the risk of intensive cultivation.

Vegetable grafting today and tomorrow

In the U.S., the vegetable grafting market has rapidly expanded in recent years despite the concern associated with the cost of grafted plants. More seed companies in the U.S. are now carrying commercial cultivars of solanaceous and cucurbit rootstocks. Today, plants are grafted in commercial nurseries in several states, targeting various markets including retailers, greenhouses, high tunnels, and open fields, of various production scales. New nursery operations are currently being built in collaboration with various sectors of the fruiting vegetable industry. The number of rootstocks available in the U.S. increases each year. Experimentation with grafted processing tomato is underway as a means to overcome issues associated with cultivation in marginal land.

While the exact number of grafted plants used in the U.S. is difficult to document at present, grafting is now recognized as a sustainable cultivation technique in most regions, and use is expected to increase as the grafting industries mature and growers learn how to use grafting technology more effectively and efficiently, with enhanced economic viability.

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Chapter 2.1 September 2017

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Synopsis:

Vegetable grafting creates a new plant by combining two plants with different genetic background, with one (scion) providing the shoots and the other the roots (rootstock), combining the desirable traits of both.

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Why Graft?

The practice of grafting used in vegetable production is similar to fruit tree grafting in that it creates a new plant by physically combining two plants with different genetic background, with one providing the shoots (scion) and the other donating the roots (rootstock). When the two genotypes are compatible, their vascular bundles reconnect at the graft union, where the wounded surfaces of the scion and rootstock meet, without presenting a barrier for water and nutrient translocation (Fig. 1). Despite the relatively high cost of grafted transplants, due to increased labor and input for producing them, as compared to regular (non-grafted) transplants, grafting has evolved into a unique cultural practice that helps reduce pesticide use, enhances yield and production efficiency, and improves economic viability in sustainable vegetable production under both open field and

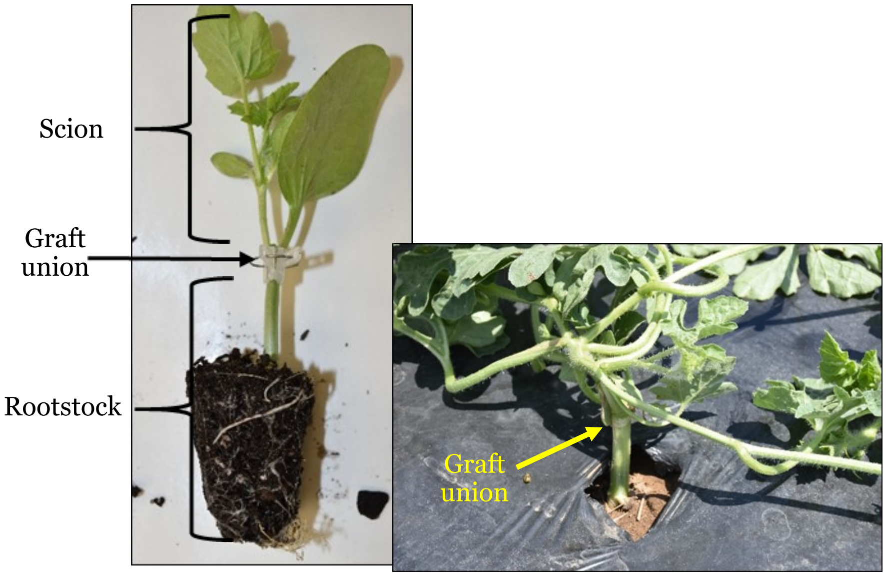
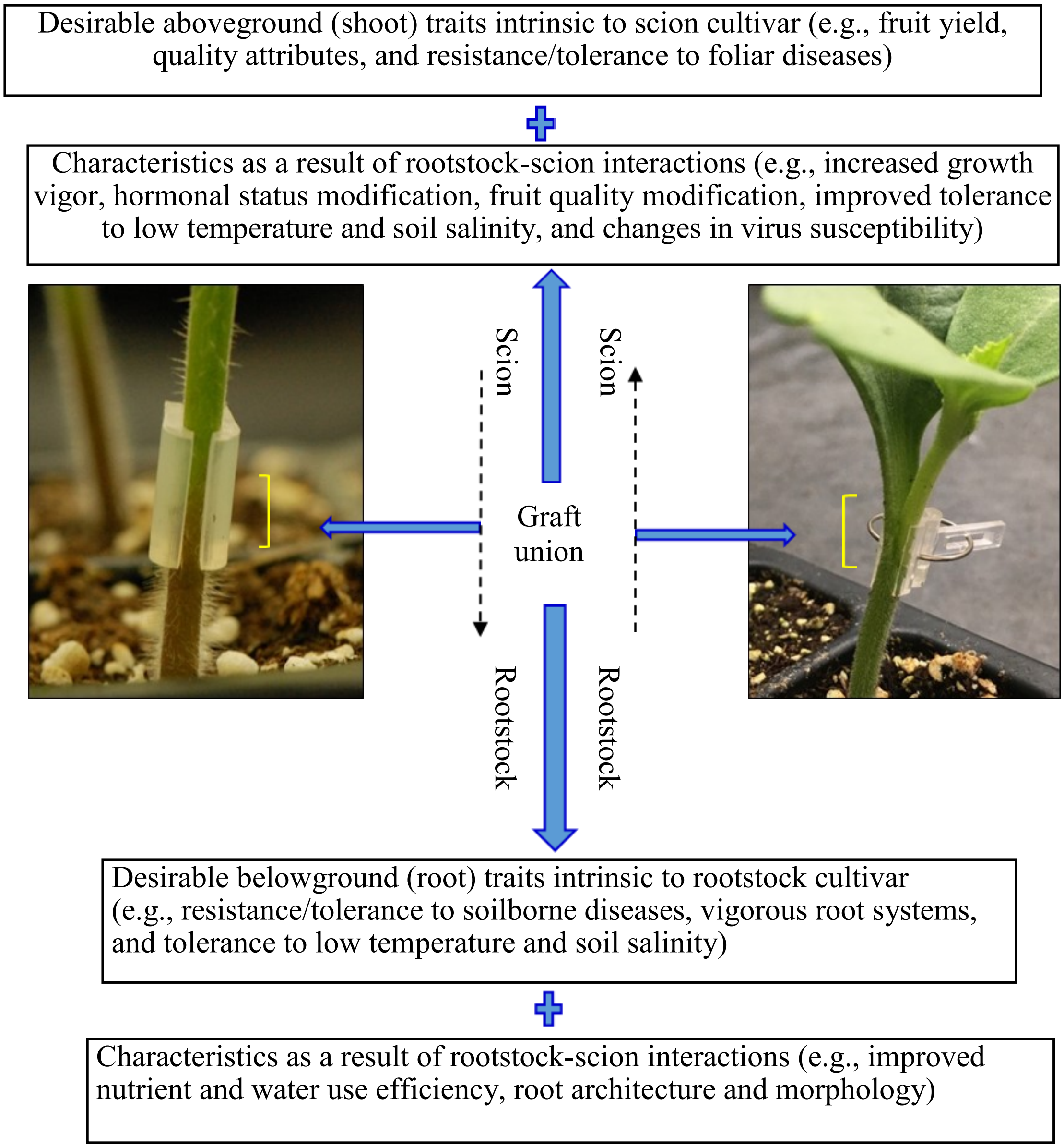


Figure 1. Grafted watermelon transplant ready for transplanting (left) and grafted watermelon grown in plastic-mulched raised bed in an open-field system (right). *(Photos by Michele Colaluce and Xin Zhao)*

protected culture systems (Lee et al., 2010; Kubota et al., 2008). Figure 2 illustrates a general concept of using grafted plants to benefit vegetable production by obtaining desirable traits from scion and rootstock cultivars, which are not only determined by the intrinsic characteristics of scions and rootstocks but also their interactions as well as the environmental conditions.

Grafting as an IPM tool for disease management – complementary to standard disease resistance breeding of new cultivars

At present, vegetable grafting is mainly applied to solanaceous and cucurbitaceous crops, primarily tomato, eggplant, pepper, watermelon, cucumber, and melon. Although some growers may still prefer to graft their own transplants, it is becoming more common to source desirable grafted seedlings from commercial nurseries (Fig. 3). Grafting is an effective IPM (integrated pest management) tool for managing soilborne diseases, which was the primary purpose for the development of vegetable grafting, and continues to be one of the main purposes for its use today (Guan et al., 2012; Louws et al., 2010). In the search for alternatives to methyl bromide, the role of grafting with resistant or tolerant



Desirable aboveground (shoot) traits intrinsic to scion cultivar (e.g., fruit yield, quality attributes, and resistance/tolerance to foliar diseases)

+

Characteristics as a result of rootstock-scion interactions (e.g., increased growth vigor, hormonal status modification, fruit quality modification, improved tolerance to low temperature and soil salinity, and changes in virus susceptibility)

Desirable belowground (root) traits intrinsic to rootstock cultivar (e.g., resistance/tolerance to soilborne diseases, vigorous root systems, and tolerance to low temperature and soil salinity)

+

Characteristics as a result of rootstock-scion interactions (e.g., improved nutrient and water use efficiency, root architecture and morphology)

Figure 2. An illustration of a general concept of vegetable grafting *(Photos by Xin Zhao).*

rootstocks is recognized as an environmentally-friendly approach to control soilborne pests in addition to other cultural strategies that include resistant cultivars, crop rotation, and biocontrol. Compared with traditional long-term breeding efforts of incorporating disease resistance with other desirable traits, rootstock breeding can be more effectively performed as the focus may be mainly on root characteristics including resistance to soilborne diseases. Grafting also allows for better utilization of germplasm resources as rootstock development may make it easier to directly employ related or wild species for deploying new traits as compared to a traditional breeding program. For example, many commercial rootstocks are interspecific hybrids using a wild species as a parent. Hence, grafting offers broader opportunities for optimizing breeding outcomes by encouraging complementary programs of scion and rootstock cultivar development.

With the increase of organic production and other alternative farming systems requiring more restricted use of synthetic pesticides, and in the scenarios of growing heirloom or specialty cultivars lacking effective genetic disease resistance, grafting has become an even more useful tool to manage soilborne disease issues (Fig. 4). The soilborne fungal, oomycete, bacterial, or viral diseases that may be managed effectively by grafting to prevent or minimize fruit yield loss include but are not limited to (as rootstock development continues to advance):

Fungal and oomycete diseases:

Fusarium wilt (caused by *Fusarium oxysporum*) in tomato, pepper, eggplant, cucumber, watermelon, and melon

Fusarium crown and root rot (caused by *Fusarium oxysporum*; *Fusarium solani*) in tomato, pepper, watermelon, and cucumber (Fusarium root and stem rot)

Verticillium wilt (caused by *Verticillium dahliae*) in eggplant, tomato, watermelon, melon, and cucumber

Southern blight (caused by *Sclerotium rolfsii*) in tomato

Phytophthora blight (caused by *Phytophthora capsici*) in tomato and pepper

Monosporascus sudden wilt (caused by *Monosporascus cannonballus*) in melon and watermelon

Corky root (caused by *Pyrenochaeta lycopersici*) in tomato and eggplant

Black root rot (caused by *Phomopsis sclerotioides*) in cucumber

Gummy stem blight (caused by *Didymella bryoniae*) in melon (although not a soilborne disease, it occurs at the lower crown of the plant and may be managed by using resistant rootstocks)

Bacterial diseases:

Bacterial wilt (caused by *Ralstonia solanacearum*) in tomato, eggplant, and pepper

Nematodes:

Root-knot nematodes (*Meloidogyne* spp.) in tomato, eggplant, pepper, melon, watermelon, and cucumber

Figure 3. Large-scale production of grafted watermelon transplants in commercial nurseries in Spain (left) and China (center), and grafted tomato transplants in the U.S (right) *(Photos by Xin Zhao and Carol Miles).*

Viral diseases:

Melon necrotic spot virus in watermelon and melon

Most commercially available rootstocks do not have a complete resistance package. For instance, the interspecific hybrid squash rootstocks are highly resistant to Fusarium wilt but often lack resistance to root-knot nematodes. The rootstock should be selected to address the primary disease problem identified in the production system. Moreover, rootstock resistance to specific pathogen races should be considered; that is, a rootstock may have resistance to one race within a specific disease but not another race. For example, the interspecific tomato hybrid rootstock 'Maxifort' has high resistance to Fusarium races 1 and 2 but not race 3, while another interspecific tomato hybrid rootstock 'Multifort' possesses high resistance to all three races.

Although foliar disease resistance is largely determined by the scion cultivar and grafting has rather limited use in foliar disease management, tolerance of grafted plants to certain foliar diseases might be improved by using selected rootstocks, an area that deserves more research. For example, 'German Johnson' heirloom tomato grafted onto 'GRA 66' tomato rootstock was reported to show lower incidence of tomato spotted wilt virus compared with non-grafted 'German Johnson' (Rivard and Louws, 2008). In general, the overall health of plants may be promoted as plant vigor and growth increases with the use of selected rootstocks.

Grafting as an innovative method for overcoming abiotic stress and improving plant growth, fruit yield and quality

Even under low or no disease pressure, plant growth can be considerably improved especially when vigorous rootstocks are used and water and nutrient uptake becomes more efficient. Increased leaf photosynthetic rate and nitrogen assimilation and endogenous hormonal modification have been observed in grafted plants, and enhancement of plant performance is especially evident when plants are exposed to suboptimal growing conditions (e.g., low and high temperatures) during the production season (Aloni et al., 2010; Martínez-Ballesta et al., 2010).

Desirable root characteristics for nutrient and water uptake can be accomplished through rootstock selection and development. As a result, fruit yield (fruit size and/or fruit number) can be increased and harvest period expanded. For example, the double leader system is commonly used in hydroponic tomato systems to reduce number of transplants while increasing yields by taking advantage of growth vigor

Figure 4. Non-grafted 'Black Cherry' tomato (front) versus 'Black Cherry' grafted onto 'Multifort' rootstock (back) in an organically-managed high tunnel. The plant decline of non-grafted 'Black Cherry' was caused by Fusarium wilt *(Photo by Xin Zhao)*.

promotion by grafting with interspecific hybrid tomato rootstocks (Fig. 5). Rootstocks with excellent tolerance to abiotic stresses such as low temperature, soil salinity, drought, and flooding, are also available to growers for early production and off-season cultivation as well as dealing with environmental constraints. This may add particular value to protected culture with continuous cropping systems that have limited rotation. Reduction in fertilizer and irrigation water use as well as the number of plants needed may be possible. On the other hand, plant vegetative growth and reproduction (fruit set) need to be well balanced through crop management practices (e.g., timing of nutrient application) when vigorous rootstocks are used.

Early flowering and fruit set may be achieved by grafting with certain rootstocks, and some fruit quality attributes may be enhanced, such as fruit size (although increased fruit size might be undesirable sometimes), firmness, and certain nutritional values (e.g., lycopene content),depending on the rootstock-scion combination (Aloni et al., 2010; Lee et al., 2010; Rouphael et al., 2010). However, possible grafting incompatibility might occur occasionally (assuming the decline of plants is not caused by poor quality of grafting and healing), and certain rootstock-scion combinations may also lead to delayed fruit set and ripening and reduced fruit quality such as soluble solids content (Guan et al., 2015). Consumer-perceived sensory properties such as fruit texture, sweetness, and flavor may also be negatively impacted, especially when rootstock and scion cultivars are more distant in their genetic makeup. Rootstock effects and rootstock-scion interactions will need to be examined to avoid negative influence of grafting on overall crop performance.

Advancing rootstock development and optimizing grafted vegetable production systems

While a “super” rootstock may not exist to address every production issue encountered, rootstock selection and breeding will continue to enhance multifaceted benefits of grafting in vegetable production. As new research tools become more readily available to better understand the basis for rootstock-scion “cross talk” at physiological, biochemical, and molecular levels, grafting technology is expected to enter a new era in advancing sustainability of vegetable production. Continued technology improvement of grafting (e.g., automation, suppression of rootstock regrowth) that produces high quality grafted seedlings with increased cost effectiveness will greatly facilitate economical production of grafted vegetables. Meanwhile, the increasing adoption of grafted transplants requires more in-depth research and technology transfer targeting optimization of management practices (e.g., fertilization and irrigation) of grafted vegetable plants in various production systems in order to realize the optimal gains of grafting.

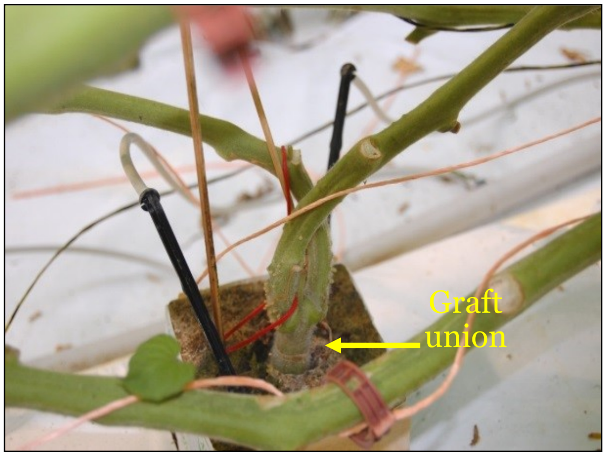


Figure 5. Hydroponic production of grafted tomato plants using a double-leader plant training system. *(Photo by Xin Zhao)*.

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Grafting Manual:

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Chapter 2.2 February 2018

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Synopsis:
Producing seedlings specifically to prepare grafted plants requires particular attention to procuring space and supplies, timing sowings, disease management, and modulating growth through irrigation, lighting, and temperature regimens.

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Preparing Seedlings & Rootstocks to Graft

Most growers are familiar with how to produce seedlings to be used as standard transplants. However, an increasing number of people want to know how to produce seedlings specifically to prepare grafted plants. The two seedling production processes are similar but not identical. Producing seedlings to prepare grafted plants borrows a lot from producing seedlings as standard (non-grafted) transplants. However, specific details about the grafting process demand attention before and during seedling production. These details and steps that seedling and grafted plant suppliers may want to consider in creating 'feedstock' for grafting operations are the subject of this summary.

Numerous research-based resources regarding seedling production for use as standard transplants provide growers with an excellent foundation for producing seedlings for grafting. Growers are encouraged to consult reliable seedling production guides describing seeding methods, promoting germination and stand establishment, and maintaining healthy and disease-free plants (e.g., through fertility and irrigation and crop protection programs) through the point of transplant shipment or use. For grafting, however, growers and nurseries are encouraged to keep the following four major issues in mind.

Inputs increase

Grafting combines two plants into one. Therefore, two seedlings must be produced for each grafted plant to be used in production. In fact, it will be necessary to sow more than one rootstock seed for each grafted plant that is desired since rootstock seed may emerge at lower percentages and less uniformly than scion seed, although seed vigor and germination of many commercial rootstock cultivars have improved recently. In addition, it is necessary to sow more scion seeds than the number of desired grafted plants to ensure that there are enough scion seedlings of the appropriate size and desired quality at grafting time. Further, not all grafted plants will survive, so it is necessary to graft more plants than are needed.

In our laboratory's example, we consistently achieve 95% or greater survival in tomato, but only 80% for cucurbits and 85% for eggplant. Thus, the need for at least two seedlings per plant used in grafted vegetable production can more than double the materials, supplies, labor, and other inputs used to produce a given number of standard (non-grafted) seedlings. An online tool for calculating the number of seeds that must be sown (http://u.osu.edu/vegprolab/seed-to-grafted-plant-calculator/) includes assumptions about germination, and seedling health and selection. In addition to the number of seedlings required, growers are encouraged to verify that all tangible and intangible resources required to produce the desired number of seedlings are available.

Schedules change

Newly-grafted plants must heal before being shipped or set into production areas. Healing periods tend to last 7-14 days, depending on healing conditions and plant species and status. Therefore, if shipping or planting dates remain the same, seedling production may need to begin two or more weeks earlier than for standard transplant production.

Also, it is important for seedling growers to recognize that, for most cases, the stem diameters of rootstock and scion seedlings must be not only similar when grafted but also within a specific range (i.e., 1.5 mm – 3.0 mm in tomato, as measured just below the cotyledonary node). It is also important for growers to recognize that the vigor of rootstock and scion seedlings is often very different. That being true, the different growth rates of rootstock and scion seedlings may result in their reaching, staying in, and passing beyond the period when they are graft-eligible (i.e., 1.5 mm – 3.0 mm stem diameter) on different dates despite being sown on the same date. Therefore, seedling growers are encouraged to become familiar with the relative growth rates/vigor of seedlings of the cultivars to be used as rootstock and scion.
Growers can become familiar with cultivar-specific vigor values through their own experimentation and through the results of experiments completed by others. For example, the Vegetable Production Systems Laboratory at the Ohio State University-Ohio Agricultural Research and Development Center has published relative seedling vigor values for twenty-three tomato varieties (eighteen rootstock, five scion; http://u.osu.edu/vegprolab/files/2016/10/seedling-vigor-table-HT-dec-16-27nsssq.pdf). These results indicate that the difference in vigor can be 575 times greater for the most vigorous varieties compared to the least vigorous varieties.

Rootstock seedlings may emerge less uniformly (and over a longer period) than scion seedlings. However, once emerged, rootstock seedlings may grow more rapidly than some scion varieties. Overall, the different growth rates of seedlings before they are grafted and the need to heal grafted plants lead many seedling and grafted plant suppliers to seed multiple times (i.e., stagger seedings). Differences in growth between rootstock and scion seedlings also lead some growers to adjust the growing conditions and management practices for rootstock and scion seedlings so they reach graft-eligible status simultaneously. This approach allows growers to maximize the use of all seedlings to produce grafted plants.

Clean, disease free seeds and seedlings are essential

All seedling producers and users value clean, disease-free stock. However, for people producing grafted plants, it is even more essential.

Grafting requires wounding that, ordinarily, would be lethal. Also, seedlings, hands, tools, machines, and work surfaces are in contact with wounded seedlings and their exudates. Therefore, the grafting process is incredibly efficient at spreading disease from one to many plants and at exposing weak, newly-grafted plants to seedling diseases that ordinarily may be 'minor' concerns. Assuring that the plant growing, grafting, and healing areas are disease-free is vital. Good sanitation practices including clean hands and grafting tools throughout the grafting process is important.

Special attention needs to be given to common seedling diseases as well as diseases that can be spread by contaminated seed, by contact, and/or transmitted from root to shoot or shoot to root. These include viruses known to be in tobacco products. Therefore, tobacco users should have no contact with tomato scion or rootstock seedlings, or contact only after very specific conditions are met. Additional resources are available (see below) to assist growers in achieving and maintaining disease-free seedling stock.

Seedlings may need specific management, including just prior to grafting

Surgery often leads to better outcomes when patients are prepared beforehand, sometimes beginning days in advance. Likewise, grafted plants may be prepared more effectively and the success rate can be increased when seedling management is altered prior to grafting. This is an active area of research and more details are expected. However, currently, experienced growers often reduce or withhold irrigation and fertigation, especially of rootstock seedlings, for 1-2 days before grafting. This step helps reduce root pressure, allowing firmer contact at the rootstock-scion interface. Figure 1 depicts squash and tomato rootstock seedlings immediately prior to the attachment of the scion section. Note that sap is visible as a droplet at the cut surface of each rootstock seedling.

While all cut surfaces are expected to glisten with moisture, high root pressure in the rootstock may show up as excessive moisture (a droplet) that interferes with setting and maintaining rootstock-scion contact at the graft union. Therefore, steps to lessen root pressure at grafting are needed. However, seedlings should not be stressed to the point of severe wilt prior to grafting.

There is also growing research evidence that altering light intensity or spectral composition

Figure 1. A squash (left) and tomato (right) rootstock seedling containing a sap droplet at the cut surface immediately before grafting due to high root pressure. *(Photos by P. Devi, WSU and K. Chamberlain, OSU-OARDC, respectively)*

before grafting may enhance the success and efficiency of the process. Specific modifications of the light environment before grafting are relatively uncommon in commercial practice. However, growers should pay attention as new information may reveal that such modifications can increase grafting success.

Adjustments beginning earlier in seedling production may also be needed to accommodate the unique needs of grafted plants and different grafting methods. For example, vegetable producers often prefer standard non-grafted transplants to be stocky and thick-stemmed. While grafted plants must also be sturdy, they may need to be taller than standard transplants because grafted plants should be set into production areas so that their graft union remains above the soil line.

In the case of tomato, seedlings are often grafted near the cotyledonary node when seedlings have three or four true leaves. Increasing the length of the rootstock stem (hypocotyl) so that a large portion of it extends upward above the soil-line appears to have two benefits. First, the rootstock stem may resist infection by some bacterial and fungal diseases more reliably than the scion stem. As the transfer of inoculum from soil to plant can occur by rain splash, a taller rootstock stem may provide additional protection. Second, some vegetable producers prefer to set large transplants (i.e., ones with a greater than average number of

Figure 2. Tomato plants grafted immediately below the rootstock cotyledonary node (left) and three and five nodes above the rootstock cotyledons (middle and right, respectively). Yellow circles surround the rootstock-scion graft union. The longer rootstock stem section provides the grower with potentially useful options (e.g., planting deep, increasing the length of stem over which diseases resistances may persist). However, so-called “high-grafted” plants are more time-consuming and may be more difficult to prepare. *(Photo by M. Kleinhenz and M. Soltan)*

nodes and leaves) deep into the soil, especially in high tunnels, to facilitate plant establishment by encouraging adventitious root development. In these situations, longer rootstock stems are required to allow for deeper planting while avoiding contact between scion stem and soil.

Under these circumstances, growers may graft onto rootstocks 3-5 nodes above the cotyledons, as in Figure 2 (plant in middle and at right). The left plant in Figure 2 is grafted immediately below the cotyledonary node of the rootstock seedling (the most common practice) whereas the plants in the middle and at right are grafted three and five nodes above the cotyledons of the rootstock seedling, respectively. Secondary shoots below these nodes were removed when very small and grafting was completed where the rootstock seedling was the correct diameter at the point of grafting.

Regardless of the number of nodes that the rootstock contains, grafted plants should be set into the soil at a depth allowing the graft union to remain above the soil. “High grafting” adds costs to the production of the grafted transplants as suppliers must care for rootstock seedlings for longer periods and adjust grafting areas and methods to accommodate larger rootstock seedlings, and end producers will need to scout for and remove rootstock shoots (suckers). Seedling and grafted plant suppliers may also choose to “stretch” scion seedlings prior to grafting in order to match their stem diameters to rootstock seedlings, as referenced earlier.

Regardless of why vegetable growers desire grafted plants with elongated rootstock stems, it may be necessary to modify pre-grafting seedling production methods or conditions in order to lengthen the rootstock stem before grafting. For example, if grafting is to be done near the cotyledonary node, then temperature, light, fertility, irrigation, and/or other treatments may be required to increase typical rootstock stem (especially hypocotyl) length, and these specific requirements appear to differ among cultivars.

Finally, when double-leader tomato plants are required in production, they are ‘pinched’ after grafting, thereby adding an additional step in grafted plant preparation. For all these reasons, producing seedlings to be grafted requires adjustments in pre-grafting seedling production methods and timelines.

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Grafting Manual:
How to Produce Grafted Vegetable Plants
www.vegetablegrafting.org

Chapter 2.4 **November 2016**

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Synopsis:
Healing is the most critical process of vegetable grafting propagation. Close attention to principles of successful healing is needed for good results.

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Healing and acclimatization methods and design principles

Healing is the most critical process of vegetable grafting propagation. Just like intensive medical care facilities are used after surgery in hospitals, the best results in healing can be obtained in a highly controlled environment as freshly grafted plants are vulnerable to temperature and moisture stress. However, simple tunnel-based systems can also provide good results as long as growers understand the principles for successful healing and pay close attention to the microclimate inside the tunnel.

Critical environmental factors

The three key factors for successful healing are 1) temperature, 2) relative humidity (RH) and 3) light. While there is a narrow optimum window for temperature and RH to achieve the best results, a wider range of light seems to be acceptable as long as the light level is not too high (to induce wilting) or too low (to induce stretching). However, the greater control you have to maintain targets for these three factors, the higher and more consistent the grafting success rate becomes. Grafting methods and scion/rootstock combinations also interact with these environmental factors to influence results. For example, an approach grafting method seems to be more successful than other methods under sub- or super optimum conditions and this is why small growers who have limited resources prefer this rather time-consuming grafting method. Also, cucurbit plants are more sensitive to healing conditions than tomato plants. However, some rootstocks of tomato are more sensitive to substrate moisture content during the healing process, and healing success is best when substrates are not too wet.

Optimum healing temperature should be selected so that the graft union develops a callus bridge (the layer of tissues acting as the interface between the scion and the rootstock) as quickly as possible. Once the callus bridge develops, plants can take up water more easily without the risk of wilting. This process of developing a callus bridge appears to take 3 days for tomato and 4-5 days for watermelon under optimum temperatures.

A study at Osaka Prefecture University, Japan showed that stomata of grafted cucumber plants began opening after 4 days in healing (Fig. 1), about the time the callus bridge is believed to be in place. Based on trials at the University of Arizona, plant temperature of 82-84°F (28-29°C) seems to consistently achieve a fast healing for both tomato and cucurbit species. When temperature is lower, healing takes more days.

While air temperature is controlled in healing, plant temperature is more important than air temperature as plant temperature could be higher than air temperature by a few degrees under solar radiation than under electric lighting especially when plant transpiration is limited (per leaf energy balance). Air temperature inside the healing chamber is often higher than air temperature outside the chamber by a few degrees when the chamber is within a greenhouse or high tunnel, or when structures holding the plants are covered with plastic (such as described below for indoor facilities). This may explain the conflicting temperature recommendations for healing from different scientists and growers, and it would be worthwhile noting the type of healing conditions each used to better understand the plant temperature in each system. A hand-held sensor with a thin thermocouple (0.2-0.5 mm in diameter) can be used for a quick check of plant temperature. An infrared gun is not recommended, as they tend to be inaccurate unless the user fully understands the

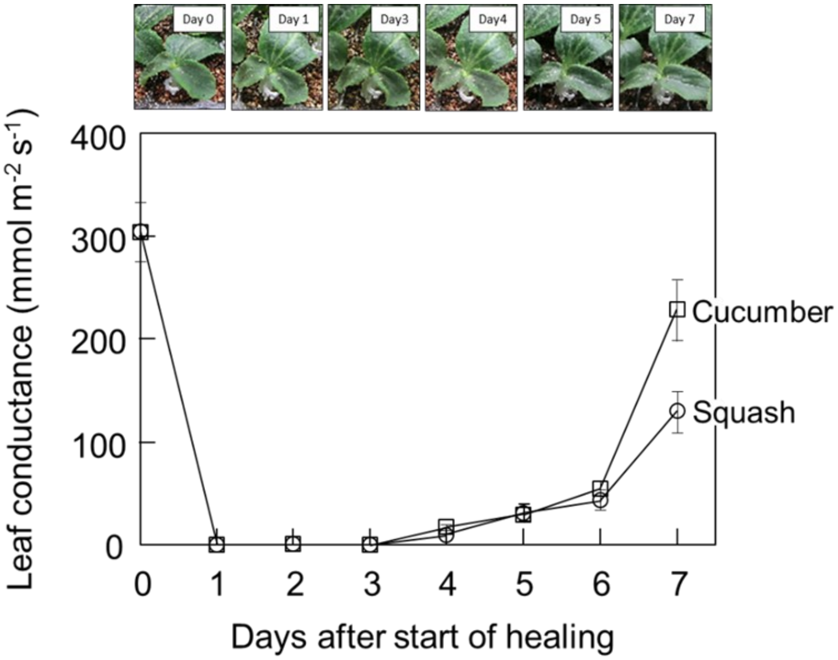

Figure 1. Stomatal conductance of cucumber cotyledons during healing after grafting onto squash rootstock or cucumber (self-grafted). Healing was done at 82°F (28°C) and 95% RH with 12-h per day lighting at $100\ \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ photosynthetic photon flux (T. Shibuya, unpublished data; Shibuya et al., 2015). Stomatal conductance was first measured on day 0 (the day before grafting) as a baseline. The first 3 days after grafting, stomatal conductance was essentially zero, suggesting that stomata are closed during this time. On day 4 after grafting, plants began to transpire, suggesting the functional development of a callus bridge between the scion and rootstock tissues.

physics of infrared measurements and correctly calibrates the sensor.

Relative humidity is most critical during the time when the callus bridge is not yet established. In general, tomato is less sensitive to suboptimal levels of RH than eggplant, and watermelon tends to be very sensitive and thus requires a more controlled environment (Johnson and Miles, 2011). Even though stomata are fully closed, scion cuttings can lose water through cuticular layers and eventually wilt. Maintaining very high RH (nearly saturation) is recommended during this period to minimize cuticular transpiration, and then gradually reducing RH to ambient levels once the critical time is past (~4 days for tomato and ~5 days for cucurbit plants after grafting at optimum plant temperature). Prolonged exposure to 100% RH may lead to problems such as fungal disease, adventitious rooting, and excessive stretching of the grafted plants.

While light is necessary for healthy plant growth, during the time the callus bridge is forming, only minimum light is needed. Some growers place newly grafted plants in darkness or in heavy shade conditions, or allow light only for a short period of time each day for the first 2-3 days following grafting. However prolonged darkness may cause excessive stretching of the plants. Very few growers have access to a light sensor to measure the actual amount of light and therefore it is difficult to find the actual range of light intensity they are using. However, it is unlikely to have a light intensity greater than 100 μ mol·m^{-2}·s^{-1} photosynthetic photon flux (the unit for photosynthetically active radiation) when electric lighting is used. In the greenhouse (or high tunnel), shading over the healing tunnel is often used to avoid wilting and to control temperature. Therefore, the actual range of light intensity during the healing conducted in a greenhouse is not well documented. The human eye is not reliable for judging light intensity inside a shaded structure especially when the structure is placed

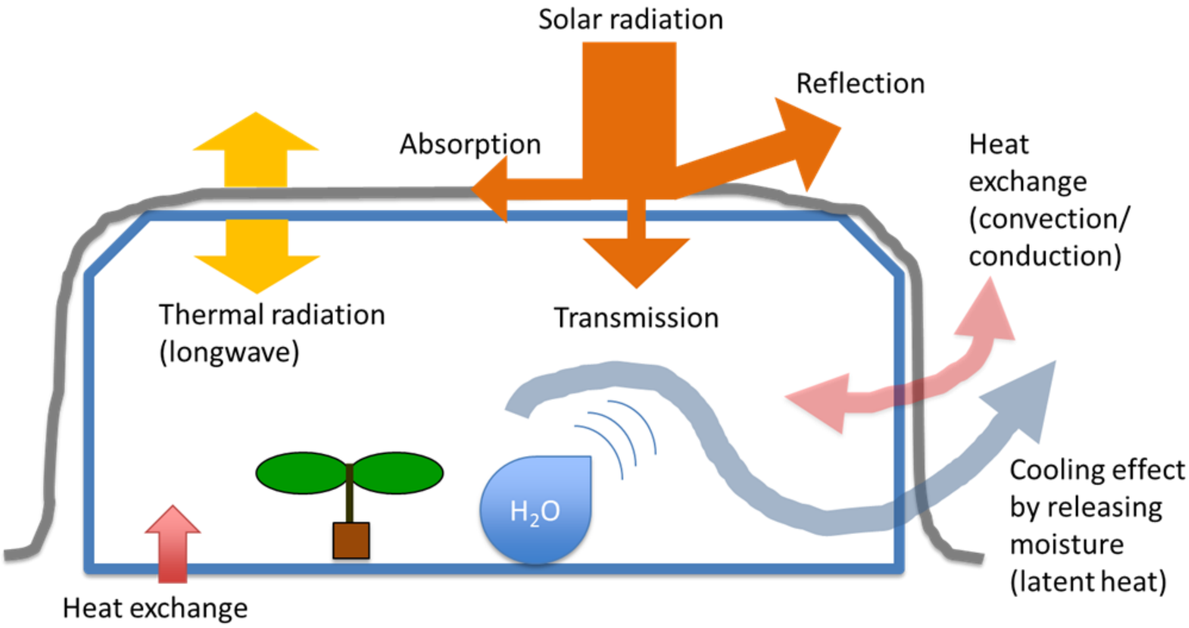

Figure 2. Thermal balance of a low tunnel style healing chamber exposed to solar radiation inside the greenhouse or tunnel. The key components to consider are 1) solar radiation and its transmission and absorption at the surface of the covering material, 2) thermal radiation reemitted from the surface of the covering material, 3) thermal exchange via convection and conduction at the surface of the covering material, 4) ventilation and associated evaporative cooling by releasing latent heat, and 5) heat exchange via the floor of the chamber.

within a bright greenhouse environment, and so observational information has limited value.

Understanding the energy balance of a healing chamber placed in a greenhouse or high tunnel (Fig. 2). would help growers to design a better healing system and to manage it to achieve the optimum healing conditions. Solar radiation affects the air temperature of the chamber and the plant temperature. Venting humid air from the chamber can effectively reduce the air temperature within the chamber (i.e., evaporative cooling) but ventilation must be managed to maintain a high level of RH inside the chamber. The internal conditions of a healing chamber that is exposed to solar radiation will fluctuate greatly due to the changing weather conditions (solar radiation, air circulation, air temperature and RH inside and outside the greenhouse or high tunnel). This is why standard operating procedures for healing chambers are site-specific or grower-specific. However, the key principles described here will provide growers and extension personnel the basis for developing their own successful protocols.

Design principles per healing system type

Low tunnel or tent inside a greenhouse or high tunnel: The key components for building a healing structure inside a greenhouse or high tunnel are:

1) Hoops or a frame to hold plastic and shade cloth
2) Clear or semi-transparent plastic cover to increase RH
3) Shade cloth to reduce light intensity
4) Fogging or misting system, or standing water for humidification
5) Heating system and thermostat control to maintain temperature within the desirable range when the ambient temperature is not optimal

Fig. 3 and Fig. 4 show some examples of low tunnel and tent healing structures placed inside a greenhouse or high tunnel. When this type of healing system is used, growers need to closely monitor and maintain the microclimate within the healing structure and not just within the greenhouse or high tunnel.

Figure 3. A bench-top healing system inside a greenhouse or high tunnel, commonly used by commercial nurseries. Overhead shade inside the greenhouse is used for better temperature control of the greenhouse and black shade cloth over the healing system is used for reducing light intensity. An under-bench misting system provides humidification without directly wetting the plants and hot-water bench heating keeps the plant temperature at an optimum range. Wetting the floor of the healing chamber is another technique commonly used to provide high RH without directly wetting the plants. (*Photos by Chieri Kubota*)

Indoor systems using electric lighting: Key components for designing an indoor healing chamber that can be placed inside a warehouse are:

1) Thermally well-insulated walk-in structure
2) Multi-tiered shelving units (movable or permanent)
3) Lamps
4) Humidification system
5) Cooling and heating system

Although consistency of healing conditions is achieved with this system, the capital investment and operational electricity costs are often discouraging. For example, a walk-in healing system with a capacity to hold 176 trays in multi-tiered shelving units was estimated as $13,193 to build and $0.0052 per plant per week to operate (at a density of 200 plants per tray, 35,200 plants total) when electricity was $0.09/kWh (Lewis et al., 2014). Lamps are a significant portion of the capital costs as well as the main user of electricity in this system. Therefore, selecting an efficient lamp system such as LEDs can reduce the operational costs but likely will increase the capital costs as the market price of LEDs is currently higher than conventional fluorescent lamps.

A typical design challenge is operation of electrical appliances and light in a wet environment. One solution commonly used in commercial operations is to cover the shelving units with a clear plastic film for humidification instead of fogging and misting inside the healing facility (Fig's. 5 and 6). Use of dry fogging is also preferred to standard fogging or misting to minimize surface wetness. Further optimization of the healing environment may improve the quality of plants coming out of the healing system. It may be worth considering a sophisticated computer-based algorithm for controlling the environment. Such an algorithm was developed to acclimatize vulnerable tissue cultured plants, where temperature and RH were controlled following a cyclically oscillating sine-curve function and increasing amplitudes over time from the starting set point and ending set point (Kozai et al., 1987).

Small-scale chambers: The key principles to follow to manage a healing system for

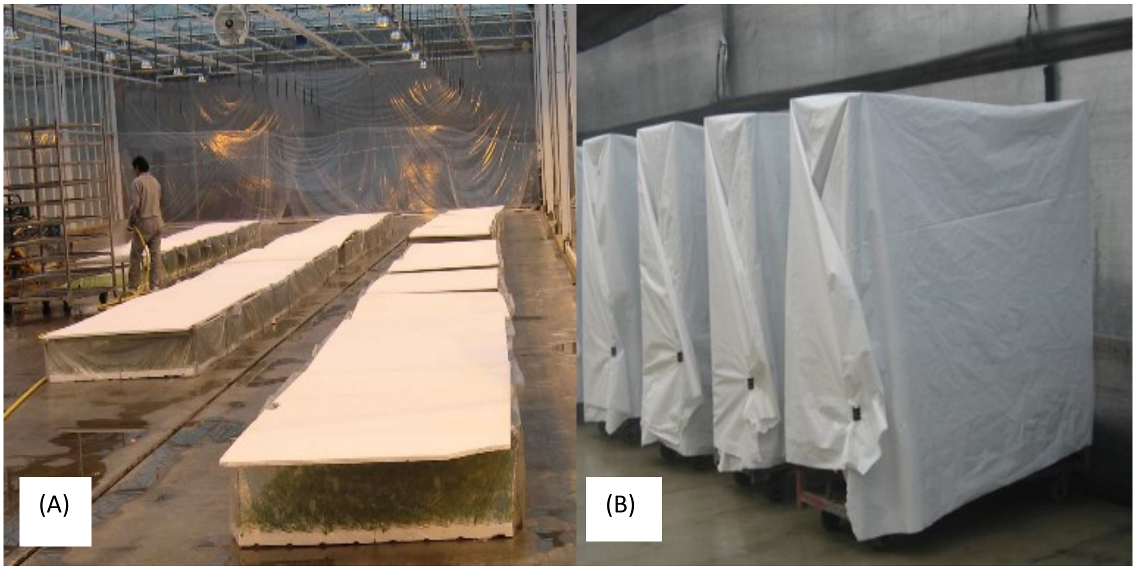

Figure 4. Various healing systems designed to place inside a greenhouse. (A) Plastic "tent" system to maintain the high humidity. Styrofoam panels are placed over the plastic tent for the first few days to reduce the solar radiation; (B) Nursery carts covered with white plastic sheets to reduce the light and increase the humidity. *(Photos by Carol Miles)*

very small applications are the same as those mentioned above. When the primary goal is to produce a small number of plants at low cost, a slightly lower success rate is not as critical. Examples of healing systems for small applications are shown in Fig. 7.

Hygiene practices

Regardless of the type of healing system used, hygiene practices including preventive measures to manage the risk of fungal diseases during healing are critical for successful healing. In some commercial grafting nurseries, steam is applied to sterilize the healing facility

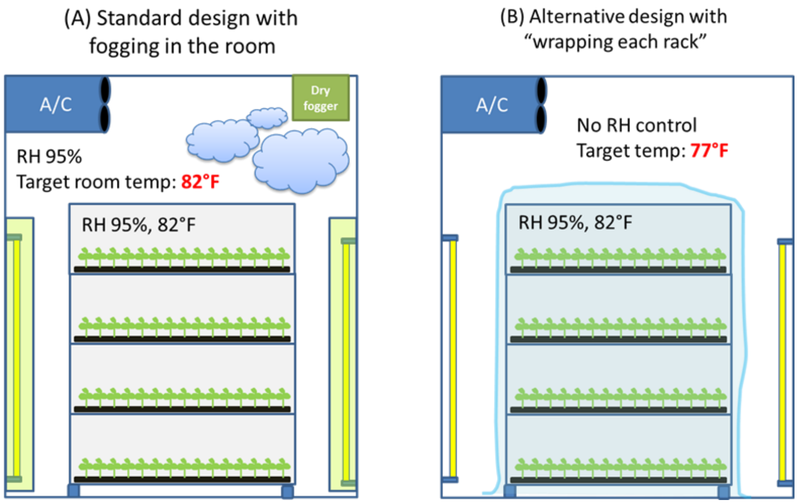

Figure 5. Two methods used to create high RH environment inside an indoor walk-in healing chamber. (A) Use of dry-fogging with lamps protected from moisture; (B) Use of plastic sheeting to wrap around the shelving units to increase the humidity without using fogging. The air temperature inside the chamber is lower for System B as the additional containment (plastic wrapped shelving units) would cause the temperature surrounding the plants to increase.

Figure 6. A healing facility developed in a commercial grafting nursery, with lamps protected with water-proof sleeves. *(Photo by Chieri Kubota)*

before and after use, and fungicides may be applied prior to healing if disease incidence is high. A 10% bleach solution is also used to sterilize benches, plastic, trays, grafting clips, and other supplies used for grafting. Minimizing the time that high RH is used in healing can minimize the likelihood of disease occurring in the healing facility.

Acclimatization after healing

Plants coming out of a high RH and low light environment need a few more days to fully acclimatize to the ambient environment inside the greenhouse or high tunnel. Grafted tomato plants tend to acclimatize more quickly than grafted eggplant and pepper (2 to 4 days, respectively), and grafted watermelon and other cucurbit crops tend to take the longest to acclimatize (up to 5-6 days) (C. Miles unpublished data). Following acclimatization, plants need to be hardened (2-3 days of reduced watering and exposure to outside temperature) before shipping or transplanting to the open field.

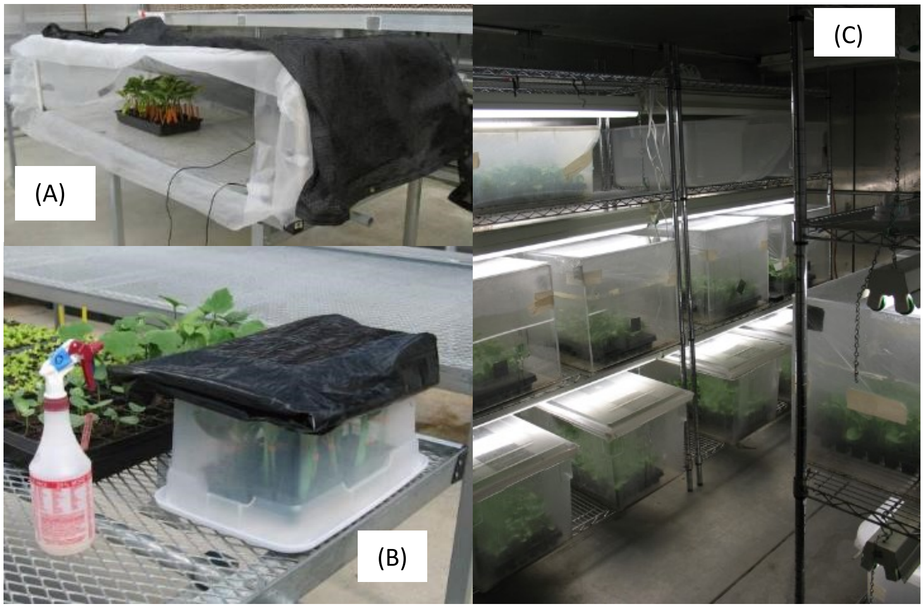

Figure 7. Healing chambers for small scale production. (A) A bench-top system developed at Washington State University (photo by Carol Miles); (B) A small upside down plastic container with a black plastic bag for home gardeners (photo by Carol Miles); (C) Small containers under fluorescent lamps used at the University of Arizona. If necessary, a thermostat-controlled heating mat can be placed under each container to maintain temperature inside the container. *(Photos by Chieri Kubota)*

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Grafting Manual:
How to Produce Grafted Vegetable Plants

www.vegetablegrafting.org

Chapter 2.5 October 2017

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Synopsis:
After healing, grafted vegetable plants need to be prepared for transplanting to the growing area. This is especially important when planting in open fields. Best practices will vary with conditions.

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Transplanting Grafted Plants

Acclimatization of grafted plants to the production environment

Following successful graft healing, grafted vegetable plants need to be acclimatized to prepare them for transplanting to growing conditions. If the grafted plants are produced in a different climate region from where they will be grown, or if they are to be grown in an open field environment, acclimatization is especially important. If grafted plants are to be grown in protected culture systems such as greenhouses or high tunnels, acclimatization may not be as critical because the growing environment is more stable and plants are unlikely to be exposed to extreme temperatures, rainfall, solar radiation, and high wind.

To acclimatize the plants for open field production, place them in an outside protected area for up to 3 days where they will be somewhat exposed to the outside environment. An open-ended high tunnel or an open shade structure can work well for this purpose. During this time, plants should be protected from heavy rains and direct sunlight but exposed to diurnal temperature patterns similar to the open field environment. Water plants lightly to further prepare them for transplanting. Adjust the amount of time and the acclimatization environment as needed based on your environmental conditions so that plants are not stressed when they are placed in the field. To acclimatize plants to a greenhouse or high tunnel, place the trays of plants on the floor of the structure overnight.

Transplanting grafted plants

Plants are ready to transplant when they are 6-10 inches tall and ideally do not have any flower buds, flowers, or fruits. Keep the graft union above the soil line when planting (Fig. 1) so the scion will not root into the soil and negate any advantages of disease resistance that would have been provided by the rootstock. When the rootstock is selected solely for vigor and not for soil-borne disease, deep-planting is acceptable.

For open field production in temperate climates, transplant after the last frost danger has passed. Soil temperature should be between 70-90°F (21-32°C) for planting most fruiting vegetable crops. Plastic mulch will help warm the soil if needed. Some rootstocks (such as interspecific squash rootstock for cucurbit crops) are known to be cold-tolerant (King et al, 2010) and grafted plants can establish when the soil temperature is lower than optimal. If conditions are breezy, leave the grafting clip and small plastic stick (optional) on the plant for a week or so to provide support. Do not transplant grafted seedlings under extremely windy conditions.

Whether transplanting in the open field or in protected culture, consider transplanting in the late afternoon so that plants are not immediately exposed to strong solar radiation. Solar radiation can cause plants to wilt, and this stress may slightly delay establishment. Planting density, and irrigation and nutrient management practices may be adjusted to maximize grafting benefits and economic returns (Djidonou et al., 2013; 2015). Optimal practices may vary for each rootstock-scion combination and production system, and growers are encouraged to experiment to find the best practices for their conditions.

Field maintenance of grafted plants

Most silicone grafting clips (commonly called tubes) used for solanaceous crops will fall off as the stem increases in diameter (Fig. 2), and therefore removal of grafting clips may not be needed. Spring plastic clips commonly used for cucurbit crops may not fall off, and if they are not removed they will severely restrict plant growth. For both solanaceous and cucurbit crops, remove any remaining grafting clips 2 weeks after transplanting. Grafting clips are typically re-usable after appropriate sanitation (e.g., washing in 10% hypochlorite solution and rinsing in water).

Figure 1. Grafted watermelon transplanted in the field with graft union well above the soil line. (*Photo by Xin Zhao)*

Figure 2. Silicone Grafting clip falls off the graft union as the grafted tomato plant grows in the field. *(Photo by Xin Zhao)*

Many commercial rootstocks are extremely vigorous and may generate regrowth (suckers) after grafting (Fig. 3). For solanaceous crops, suckers may develop from adventitious buds on the stem; or if the plant is grafted above the rootstock cotyledons, suckers will grow from the axil of the cotyledon. For cucurbit crops, the meristem tissue at the base (axil) of the cotyledons may not be completely removed at the time of grafting and regrowth can occur at this point. Rootstock suckers can quickly overtake the scion variety and reduce fruit yield (Fig. 4). Within 1-4 weeks after transplanting, check grafted plants once a week for rootstock regrowth. Remove rootstock suckers immediately; suckers can reoccur on the same plant, so it will be necessary to keep checking all plants regularly.

Figure 3. ‘Brandywine’ tomato grafted onto ‘Maxifort’ rootstock above the rootstock cotyledons (left; *photo by Xin Zhao*). ‘Crimson Sweet’ watermelon grafted onto ‘Emphasis’ (right; *photo by Carol Miles*).

Figure 4. Growth of tomato scion plant (left) and watermelon (right) is suppressed by rootstock suckers which were overlooked earlier in the season. (*Photos by Xin Zhao and Carol Miles, respectively*)

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Grafting Manual:
How to Produce Grafted Vegetable Plants

www.vegetablegrafting.org

Chapter 3.1 August 2017

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Synopsis:
Tomato (*Solanum lycopersicum*) is one of the most important and widely cultivated vegetable crops in the world. Grafting gained popularity as a method to manage soil-borne plant diseases, and to ameliorate certain abiotic stressors affecting tomato.

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USDA United States Department of Agriculture National Institute of Food and Agriculture

Tomato

General information

Tomato (*Solanum lycopersicum*) is one of the most important and widely cultivated vegetable crops in the world. Grafting onto resistant rootstocks was first introduced to vegetable production in Japan and Korea in the late 1920s as a strategy against Fusarium wilt and other diseases (Tateishi, 1927). In North America, innovations in tomato grafting occurred during the 1930s and 1940s when tomato was grafted onto jimson weed (*Datura stramonium* L.) as a method for root-knot nematode control. However, the practice was discontinued due to evidence of the transport of alkaloids to the fruit from the rootstock (Lowman and Kelly, 1946; Peacock et al., 1944).

Grafting has gained popularity as a method to manage plant diseases previously controlled by soil fumigation with methyl bromide. Some of the most significant soil-borne pest problems for which resistant rootstocks may be beneficial include root-knot nematodes (*Meloidogyne* spp.; Kokalis-Burelle and Rosskopf, 2011; Barrett et al., 2012a), Verticillium wilt (*Verticillium albo-atrum* and *V. dahliae*), and southern blight (*Athelia rolfsii* anamorph *Sclerotium rolfsii*) (Rivard et al., 2010).

Bacterial wilt caused by *Ralstonia solanacearum* is a significant concern for tomato producers in many regions. Rivard and Louws (2008) have reported inadequate control of bacterial wilt with soil fumigants such as methyl bromide, chloropicrin, and a combination of chloropicrin and 1,3-dichloropropene. The pathogen has a wide host range and persists in the soil for long periods of time, making crop rotation ineffective, and forcing farmers to abandon fields as losses can rapidly reach 100%. Additionally, tomato varieties with resistance to bacterial wilt often produce small, unmarketable fruit. Thus, some resistant tomato cultivars are used as rootstocks, making grafting an important option to manage bacterial wilt for the tomato industry. For some diseases, rootstocks may have to be selected with resistance to a specific race of the pathogen. This is the case for rootstocks resistant to *Fusarium oxysporum* f. sp. *lycopersici* (Fol), the cause of Fusarium wilt in tomato. Many of the rootstocks that are commercially available have resistance to races 1 and 2 of this pathogen, but few have resistance to race 3 of Fol, which is more limited in its distribution but is becoming increasingly more common, particularly in the southeastern United States. One commonly available rootstock, interspecific hybrid 'Maxifort' (*S. lycopersicum* × *S. habrochaites*), has resistance to Fol races 1 and 2 as well as crown rot, *F. o.* f. sp. *radici-lycoperisici*.

Tomato can be impacted by a number of abiotic stressors that grafting can ameliorate. Salinity, for example, has become a worldwide agricultural concern. Salt tolerant rootstocks have the potential to overcome osmotic stress, ion toxicity, and nutrient imbalances under high salinity (Colla et al., 2010). Under experimental conditions, grafted tomato plants resulted in 40-80% increase in yield compared to non-grafted or self-grafted plants at 50 mM NaCl (Santa-Cruz et al., 2002; Estañ et al., 2005; Martinez-Rodrigues et al. 2008). Interestingly, the scion and rootstock selection both have an influence on tolerance to salinity as well as on the mechanism of tolerance (Di Gioia et al., 2013; Giuffrida et al., 2014). In addition to salinity, the use of tomato rootstocks that are interspecific hybrids or wild accessions of *S. habrochaites*, a high-altitude wild tomato relative, has the potential to improve root growth and yield under sub-optimal (15 °C) temperatures (Venema et al., 2008). Similarly, but not as extensively studied, is the development of tomato rootstocks that maintain fruit yield under drought conditions and high temperature (Schwarz et al., 2010). Commonly, grafted tomato is used to increase plant vigor and yields and to extend harvest periods in protected cultivation (Kubota et al., 2008; Lee, 1994).

Grafting tomato has become essential for many heirloom tomato growers. Heirloom tomatoes lack resistance to soil-borne diseases and are typically heterogeneous because they are open-pollinated. Grafting allows for disease resistant rootstocks with improved plant vigor and disease resistance to be paired with scions that maintain high fruit quality and the desirable flavor associated with heirloom varieties (Rivard

Table 1. Commonly used rootstock cultivars for tomato, and their disease resistance/susceptibility.

Rootstock	Major Tomato Diseases						
	Tomato Mosaic Virus	Fusarium Crown Rot (*Fusarium oxysporum* f.sp. *radici-lycoperisici*)	Fusarium Wilt (*Fusarium oxysporum* f. sp. *lycopersici*)		Verticillium Wilt (*Verticillium albo-atrum* & *V. dahliae*)	Bacterial Wilt (*Ralstonia solanacearum*)	Root-knot Nematodes (*Meloidogyne* spp.)
			Race 1	Race 2			
Anchor-T	R	N/A	R	R	R	R	R
Arnold	R	R	R	R	N/A	N/A	R
Beaufort	R	R	R	R	R	S	R
Body	R	N/A	S	R	R	S	R
Estamino	R	R	R	R	R	S	R
Maxifort	R	R	R	R	R	S	R
Multifort	S	S	R	R	S	S	R
RT-04-105T	S	R	R	R	R	R	R
RT-04-106T	R	S	R	R	S	R	R

R = Resistant, S = Susceptible, N/A = not characterized and Louws, 2008, 2012; Di Gioia et al., 2010). Some commonly used rootstock cultivars and their disease resistance/susceptibility are listed in Table 1. There are approximately 60 rootstocks commercially available for tomato grafting, consisting of primarily hybrid tomato rootstocks and interspecific hybrid tomato rootstocks. Availability of rootstocks changes regularly, and an extensive list is available on the vegetable grafting research-based information portal www.vegetablegrafting.org, under Resources (http://www.vegetablegrafting.org/wp/wp-content/uploads/2015/02/usda-scri-tomato-rootstock-table-feb-15.pdf).

Choosing the rootstock and scion

When grafting tomato, careful rootstock selection, timing, and attention to healing and planting in the field are essential for successful production of healthy transplants. Rootstocks that are specifically chosen for disease resistance should be selected based on disease pressure at the planting location. Assistance in the identification of diseases can be obtained by contacting your county extension agent or contacting a local plant disease diagnostic facility (https://www.apsnet.org/members/directories/Documents/SoilLabsandPlantClinics.pdf). Scions are chosen based on desired fruit characteristics and quality. Scion and rootstock compatibility is important when selecting plant material. One important consideration, particularly when using heirloom tomato cultivars as the scion, is the presence or absence of genes for resistance to Tomato mosaic virus (ToMV).

Many hybrid rootstocks have Tm-2 or Tm-2^2 resistance genes that provide resistance to most known ToMV isolates, while heirloom scions have none or "low" resistance conferred from the Tm-1 gene. When scion and rootstock do not have the same genes for resistance, grafted tomato are more likely to fail when infected with ToMV (Yamakawa, 1982). This is a critical consideration for selecting rootstocks for heirloom tomato, and more information can be found in Rosskopf et al. (2013).

Grafting method, equipment, and procedures

Splice grafting, also known as "Japanese top-grafting" or "tube-grafting," is the most commonly used technique when grafting Solanaceous crops. Cleft and side grafting are the other two main grafting techniques used to graft tomato.

Before starting to graft, it is important to arrange a clean and functional grafting area and make sure you have all the required equipment and tools:

- Healing chamber
- Disposable razor blades or scalpels to cut scion and rootstock plants.
- Silicone grafting clips to secure the rootstock and scion together and minimize water loss at the graft union.
- Antibacterial soap to sanitize hands.
- Spray bottles to mist plants with water during the grafting process.

Sanitation is extremely important for successful grafting, as plant pathogenic bacteria and viruses can be passed plant-to-plant from hands, cutting surfaces, and tools. Therefore, particular care is required in cleaning the

Figure 1. Tomato seedlings ready for grafting. *(Photo by Erin Rosskopf)*

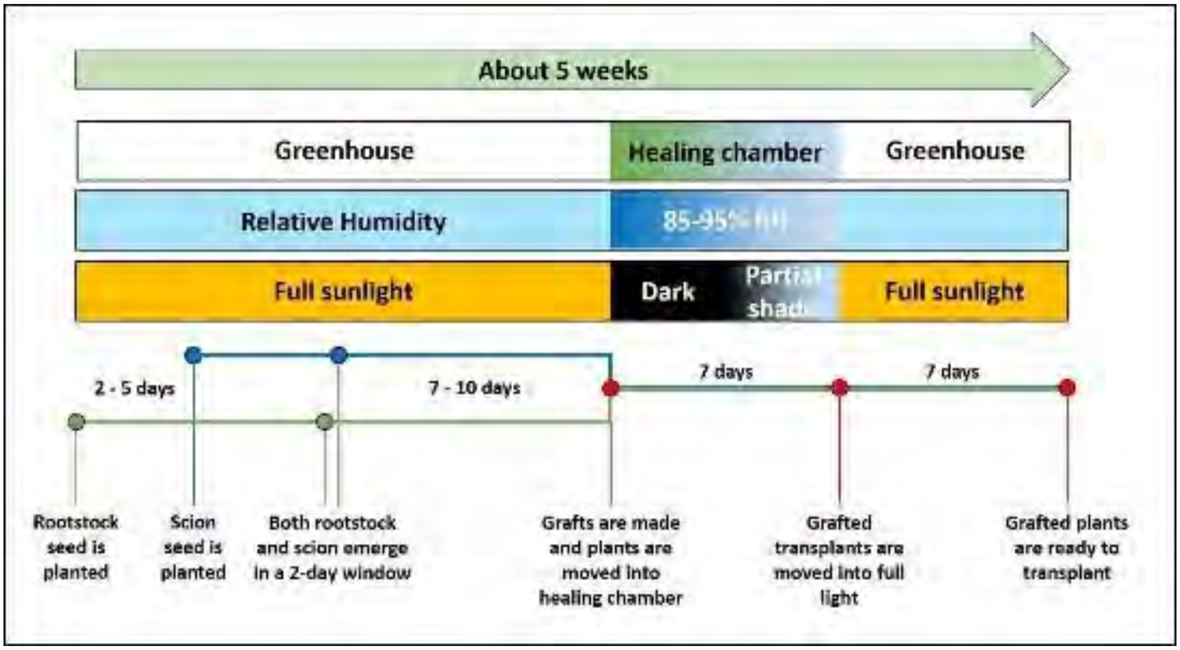

Figure 2. Grafting timeline. Adapted from Hartmann and Kester's Plant Propagation: Principles and Practices. 8th Edition.

grafting area and cleaning or changing razor blades frequently.

When grafting, regardless of the method used, it is critically important to have good contact between the scion and rootstock vascular systems. This is accomplished by grafting plants with similar stem diameters. Plants should be grafted when plants have 2-4 true leaves and stem diameters between 1.5-2.5 mm (Fig. 1). For this purpose, is important to define the grafting timeline (Fig. 2).

Conduct a germination test several weeks prior to grafting in order to determine how long both the scion and the rootstock will take to reach the optimal size, and time seeding accordingly (Hu et al., 2016). Keep plants in a shaded area prior to grafting, and cease watering rootstock plants 12-24 hours prior to graft-

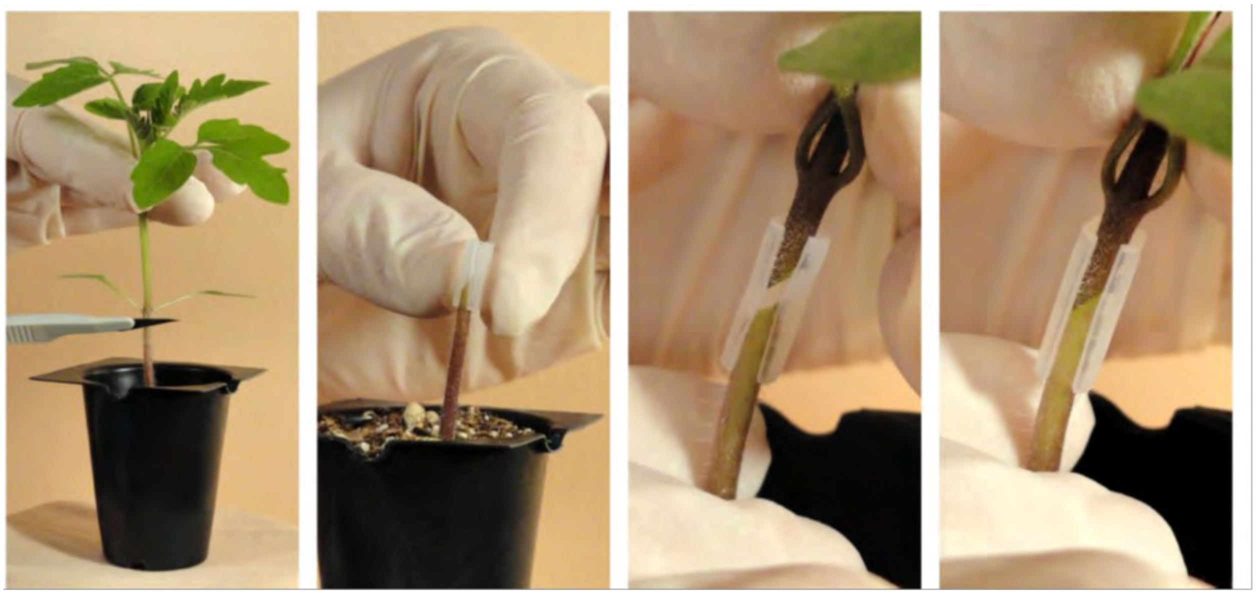

Figure 3. Splice grafting tomato. *(Photos by Cary Rivard)*

ing. This will prevent water from pushing the scion away from the rootstock.

Splice grafting is achieved by cutting the rootstock and scion stems at a 45° angle, putting the cut edges of the two plants together, and securing with a grafting clip (Fig. 3). It is important to cut the rootstock below the cotyledons to avoid unwanted rootstock regrowth, which would require additional pruning. The scion can be cut below or above the cotyledon, wherever the stem diameter best matches the rootstock.

Once the rootstock is cut, slip the silicon grafting clip onto the rootstock, followed by the scion, making sure that both stems are in tight contact with each other and no air is visible at the graft union. Mist plants frequently with water during the grafting process and place them in the healing chamber in the dark immediately after grafting (Fig. 4).

When adventitious roots from a non-resistant scion come into contact with infested soil, the plant can become infected. Recent research shows that growth of adventitious roots (Fig. 5) can be reduced by removing scion leaves prior to grafting (Meyer et al., 2016). At low humidity (<70%), removing 50% of the leaves reduced scion adventitious root formation; but at 95% humidity, removal of 90% of leaves was required to reduce adventitious root formation. However, when a less vigorous rootstock such as 'Trooper Lite' is used, and all true leaves are removed and only the cotyledons remain during grafting, tomato marketable yield is significantly reduced (Masterson et al., 2016).

Healing chamber

Construction of a healing chamber is essential when grafting any type of vegetable, and should be done in advance of grafting. The chamber's main purpose is to allow the vascular tissue of the newly cut seedlings to reconnect for water and nutrient transport, and to gradually acclimate the plants to greenhouse conditions. To keep the scion from becoming water stressed, the scion's transpiration rate is slowed during the healing process by increasing the humidity and decreasing the light for several days after grafting. Temperature, humidity and light levels in the healing chamber should remain constant to avoid stress on the plants. The chamber should be maintained be-

Figure 4. Example of a healing chamber. *(Photo by Carol Miles)*

tween 21 and 27 °C and 80-95% humidity, and light level should be low for 3-5 days (Fig. 2).

The lower the healing temperature, the longer the period of time required for the graft union to heal. Add water to the floor of the chamber or in pans to keep high relative humidity. Do not irrigate or mist plants, as excess water may pull the scion away from the rootstock due to high root pressure, and water on the leaves may accumulate in the grafting clip leading to rot. A cool-mist vaporizer can be used to provide humidity without increasing temperature, but is an added cost. Cover the chamber with plastic sheeting to maintain high humidity, and cover the plastic with dense shade cloth to block light from newly grafted plants.

Gradually acclimate plants to greenhouse conditions. Three days after grafting, open the healing chamber and mist the walls and floor with water. The healing chamber should be left closed and undisturbed for the fourth day, but remove the black cover from the front of the chamber. Increase light levels in the chamber each day so that on day 6 after grafting, the black cover is completely removed. Plants require at least two days at medium light and humidity before they can be moved to the greenhouse, where humidity is low and light levels are high.

In walk-in healing chambers, use fluorescent lights to increase light levels. On days 5, 6 and 7, open the plastic of the healing chamber for 30 minutes, 1 hour, and 6-8 hours, respectively; each day, add water to the floor before closing the chamber again. On day 8, remove plants from the healing chamber and place in the greenhouse. Although plants are fully acclimated, it may take an additional 5-6 days for the graft union to fully heal. It is therefore important to water plants from below to prevent any damage to the recently healed graft union. The grafting clip will gradually open as the stem grows, and will naturally fall off.

Figure 5. Comparison between a robust and high quality grafting union (left) and a poor quality tomato transplant with the development of adventitious roots from the scion (right). *(Photos by Francesco Di Gioia)*

Figure 6. Double-stem grafted tomato plants grown in a greenhouse soilless system in a year-round crop cycle. *(Photo by Francesco Di Gioia)*

About 14 days after grafting, plants are ready to be hardened and then transplanted to the field.

Using grafted plants

Grafted tomato can be grown in the open field or under "protected" agricultural systems, including greenhouses and high tunnels. The use of high tunnels has increased considerably and allows farmers with small acreage to produce high-quality produce without the large investment of a greenhouse structure. High tunnels also allow for extended production seasons, allowing for earlier plantings and additional harvests and increased economic returns (Galinato and Miles, 2013). While high tunnels can protect tomato from wind and rain, which

Figure 7. A double-stem grafted tomato plant (left), correctly transplanted with the grafting union above the growing medium (right). *(Photos by Carol Miles and Francesco Di Gioia)*

reduces exposure and infection by foliar pathogens, soil-borne diseases remain a problem, which can be alleviated through the use of a disease-resistant rootstock. Grafting an indeterminate tomato scion onto a vigorous rootstock makes it possible to extend the harvest period when environmental conditions are adequate. Grafted tomato plants are increasingly used also in soilless cultivation systems, where, under controlled conditions and with crop cycles extended up to one year, grafted plants can reach their full potential (Fig. 6).

An important factor limiting the extensive adoption of grafted plants is the high cost of grafted seedlings. A solution commonly adopted in commercial tomato production is the use of double-leader grafted seedlings (Fig. 7). The use of double-leader plants halves the plant number, substantially reducing the cost of the plant material.

To produce a double-stem plant, the healed and acclimated grafted seedling is pinched

Figure 8. Open-field fresh-market tomato production with a determinate cultivar and the “stake and weave” training system. *(Photo courtesy of Francesco Di Gioia)*

Figure 9. Grafted tomato plants with indeterminate scion grafted onto a vigorous rootstock raised vertically in a multi-tunnel greenhouse with a simple trellis. *(Photo by Francesco Di Gioia)*

Figure 10. Grafted tomato plants with indeterminate scion grafted onto a vigorous rootstock in a greenhouse with a woven-stem system. *(Photos by Erin Rosskopf and Carol Miles)*

back to the cotyledonary leaves, to induce the development of two lateral shoots. Plants are then maintained in the nursery for an additional 1 to 3 weeks (Kubota, 2008). Alternatively, a lateral shoot along with the main stem is maintained to form the two main producing shoots.

Cultural practices used in both high tunnels and open-field plantings with indeterminate scions may require modification to maximize productivity. For example, vigorous rootstocks may have increased nitrogen assimilation as well as reduced water usage (Djidonou et al., 2013). While grafting with determinate scions will still require 1-2 “suckering” events, season-long pruning is required with vigorous rootstocks coupled with indeterminate scions. The type of trellis system may also need to be modified.

While a “stake and weave” system is often used in open-field production with determinate varieties (Fig. 8), trellising is needed with indeterminate scions grafted onto a vigorous rootstock (Fig. 9). Trellis systems are attached to a secondary structure, not to the high tunnel hoops, because the trellis system will need to support a significant amount of weight. The trellis can be a single line (Fig. 9) or a woven support system (Fig. 10).

Regardless of the growing environment and system, it is critical that grafted plants are set high enough in the planting hole that the graft union is above the soil line (Fig. 7). Deep planting or having soil in contact with the graft union can negate the benefit of a resistant rootstock by allowing infection of the susceptible scion by soilborne pathogen propagules.

Success stories

Several soilborne diseases can result in complete crop loss when susceptible tomato cultivars are grown in infested fields. Two of the most devastating diseases, Fusarium wilt and bacterial wilt, can be successfully managed using resistant rootstocks. In a Florida study using a susceptible scion grafted onto multiple rootstocks with resistance to *R. solanacearum*, the incidence of bacterial wilt was significantly reduced when compared to the non-grafted and self-grafted plantings (Rivard et al., 2012). Total marketable tomato fruit yields using grafted plants increased by 25-99% depending on the severity of the disease (McAvoy et al., 2012). For growers who produce heirloom tomato, either in high

Figure 11. Fusarium wilt resulting in mortality of non-grafted ‘Prudence Purple’ (left) and no disease when grafted onto ‘TD-2’ in the same field (right). *(Photos by Erin Rosskopf)*

tunnels or open-field, the lack of resistance to soilborne diseases can result in a failed crop. Losses to Fusarium wilt in non-grafted ‘German Johnson’ (GJ) tomato in one field reached 50%, whereas GJ grafted onto ‘Maxifort’ plants experienced no symptoms of disease (Rivard and Louws, 2008). On three farms in Florida, where heirloom tomatoes are grown in both high tunnels and open-field, the incidence of Fusarium wilt had caused the growers to abandon these desirable varieties. Grafting ‘Amana’, ‘Black Cherry’, ‘Prudence Purple’, and ‘Cherokee Purple’ scions onto ‘Maxifort’, ‘Multifort’, and ‘TD-2’ rootstocks made the production and sale of these highly sought-after cultivars possible again (Fig. 11; Rosskopf unpublished).

The ability to grow heirloom tomato with high marketable yields has significantly contributed to farm sustainability. While increases in tomato fruit yield resulting from grafting are relatively consistent, the impact of grafting on tomato fruit quality is variable. Soluble solids, sugar content, and flavor components, as well as taste panel preferences appear to be dependent upon which scion and rootstock combinations are used (Di Gioia et al., 2010; Flores et al., 2010; Barrett et al., 2012b).

Grafting is a valuable tool for tomato producers worldwide. As breeding efforts continue and result in desirable rootstocks, more grafting facilities become available in the United States, and growers learn how to successfully graft their own transplants, grafted vegetable plants will continue to gain acceptance as an effective technique and an economically viable tool to reduce disease incidence and increase yields.

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Grafting Manual:

How to Produce Grafted Vegetable Plants

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Chapter 3.2.1 **November 2017**

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Synopsis:

Watermelon grafting became a standard practice to manage disease as early as the 1930s. In addition, grafted plants are also used to manage salinity in soil and irrigation water.

Editors:
Chieri Kubota (The Ohio State University)
Carol Miles (Washington State University)
Xin Zhao (University of Florida)

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USDA United States Department of Agriculture National Institute of Food and Agriculture

Watermelon and Melon Grafting

The use of grafted watermelon (*Citrullus lanatus*) became a standard production practice in Japan in the 1930s as a means to manage Fusarium wilt, a soil-borne disease, and grafting was soon thereafter applied to melons (*Cucumis melo*). The benefits of using grafted plants have been increasingly recognized to address production challenges with the ban of the broad-spectrum soil fumigant methyl bromide (Davis et al., 2008; Lee et al., 2010). Today, commercial use of grafted watermelon plants in some regions of Asia and Europe accounts for up to 95% of total watermelon production, while about 30% of melons grown in Japan, 90% in Korea, and 5% in China are grafted (Lee et al., 2010). In addition to disease management, grafted plants are also used to manage salinity in soil and irrigation water. Beginning in the 1990s, commercial growers and home gardeners in the U.S. have gradually become more aware of the advantages of using grafted plants. Grafted watermelon and melon plants are used in the U.S. primarily to manage soil-borne diseases (e.g., Fusarium wilt, Verticillium wilt) by providing more vigorous, disease-resistant root system.

There are three methods commonly used to graft watermelon and melons: one cotyledon splice, hole insertion, and tongue-approach (Davis et al., 2008; Guan and Zhao, 2014; Zhao et al., 2016). The tongue-approach method was developed in the Netherlands, and is still popular in some European countries such as Spain and also in Japan. Some nurseries prefer to cut rootstock seedlings at the soil line (referred to as root excision) prior to grafting with the one cotyledon or hole insertion method, as this allows grafted plants to regenerate roots during the graft healing process (Guan and Zhao, 2015). Additionally, it is easier to handle the cuttings rather than whole plants, the work surface tends to be cleaner, and the seedlings are more suitable for automated grafting operations.

Some propagators believe that adventitious roots from the rootstock may develop a stronger root system than the original root system. We will provide a summary of the advantages and disadvantages of each method, but plant propagators often develop specific techniques for each method to optimize production of grafted transplants in their operation.

Grafting preparation

In general, watermelon plants are grafted when they are 14–21 days old. Each grafting method has specific requirements regarding seedling size (see below), but seedlings are usually ready to graft when the rootstock seedling has 1 fully emerged leaf and the scion seedling has 2.

In the one-cotyledon and tongue approach grafting methods, the scion and rootstock plants should have similar stem diameters at the time of grafting so their vascular bundles can be aligned and in complete contact with one another. In the hole-insertion grafting method, scion plants need to have a smaller stem size than rootstock plants. The scion and rootstock seedlings may not germinate or grow at the same rates, so it is important to conduct a preliminary test to determine their growth rates in your growing environment.

Figure 2. Supplies commonly used for grafting. (*Photo by Pinki Devi*)

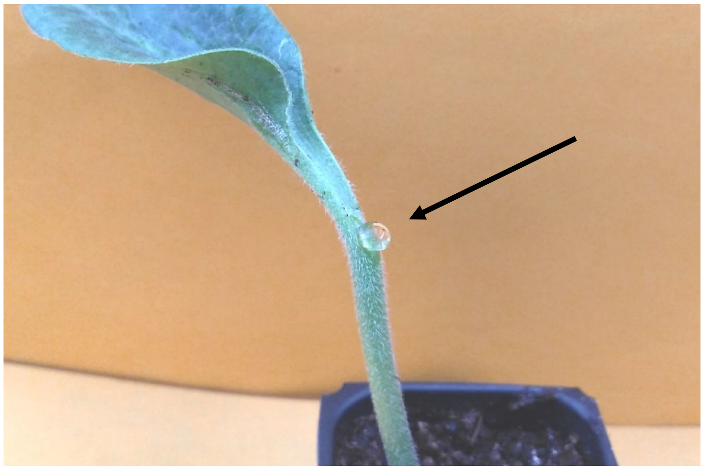

Figure 1. A water bubble will form from the cut plant stem if the seedling is too turgid at the time of grafting. (*Photo by Pinki Devi*)

To achieve uniformity in both germination and growth, planting dates must be optimized for both the scion and the rootstock. Seed more plants than necessary so you have a greater selection when matching stem diameters. Also, it is rare to get 100% graft survival for watermelon, so graft at least 20% more plants than needed. Adjust this rate as appropriate for graft survival in your production system.

Water both rootstock and scion plants 12–24 hours before grafting. Do not water plants immediately before grafting, unless they are wilted. If seedlings are too turgid, a water bubble will form when the plants are cut (Fig. 1), and this bubble will prevent a tight graft union from forming. Use only clean, sharp razor blades for grafting, and wash your hands with anti-bacterial soap or hand gel before working on the plants, or use latex-type surgical gloves.

While there are many tools that can be used for cutting vegetable plants for grafting, the double-edged razor blade snapped in half is most commonly used as it has the thinnest sharpest blade and is the least expensive (Fig. 2). If reusing grafting clips, wash them in warm, soapy water, sterilize them by soaking for 1 minute in a 10% bleach solution, and rinse them under tap water. Allow the clips to air-dry before reuse. Fill a spray bottle with tap water in order to mist plants frequently during grafting.

A clean area such as work bench, with no direct sunlight is required for grafting. Grafting is commonly done in a greenhouse, but a shaded area may be needed so that temperature is 21-23 °C (70-73 °F). Do not graft near a fan to protect the plants from water loss and avoid disturbance to the graft union. If you are using a healing chamber, spray the inner surfaces of the chamber with water and add a thin layer of water to the chamber floor a few hours before grafting to raise the relative humidity to about 95% within the chamber. Do your grafting early in the morning or late in the day to avoid water stress and drying of the cut surfaces.

Grafting techniques

1. One-cotyledon grafting

The one cotyledon grafting method is by far is the most popular method used for watermelon and melons in Korea, Europe and North America. This method is also known as the splice graft, and was originally developed by Japanese engineers for use with automated grafting. Due to the procedure's simplicity and speed as well as the relatively low rate of rootstock regrowth (2-3%), it has become the most commonly used manual grafting method in Eu-

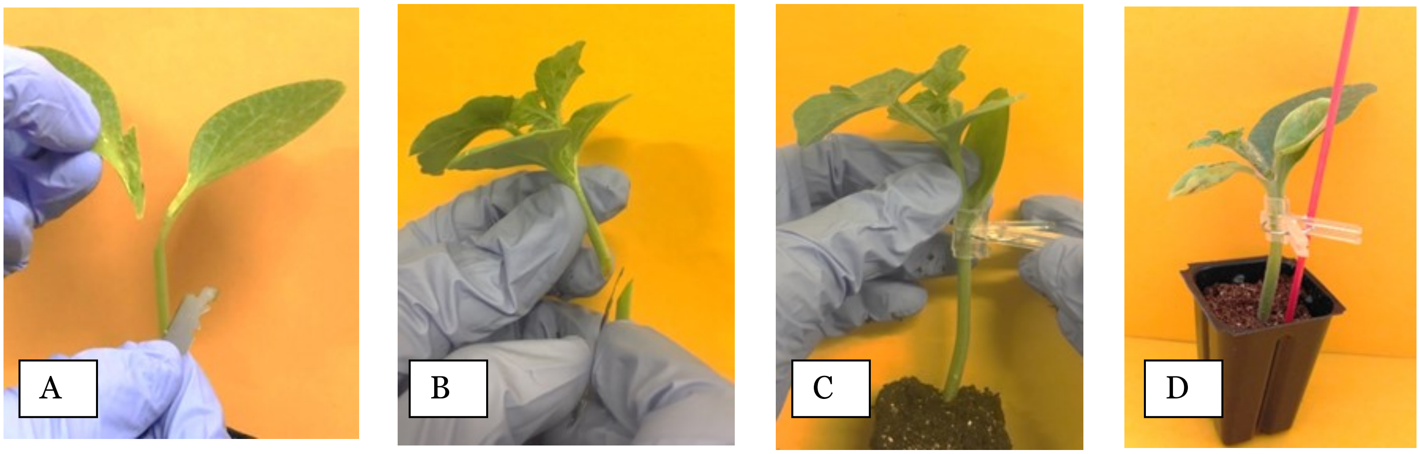

Figure 3. One cotyledon grafting: (A) cut the rootstock at a 60° angle with one cotyledon remaining on the plant; (B) cut the scion at a 60° angle below the cotyledons, where its diameter matches that of the rootstock; (C) join the two cut stems together; and (D) secure with a grafting clip. (*Photos by Pinki Devi*)

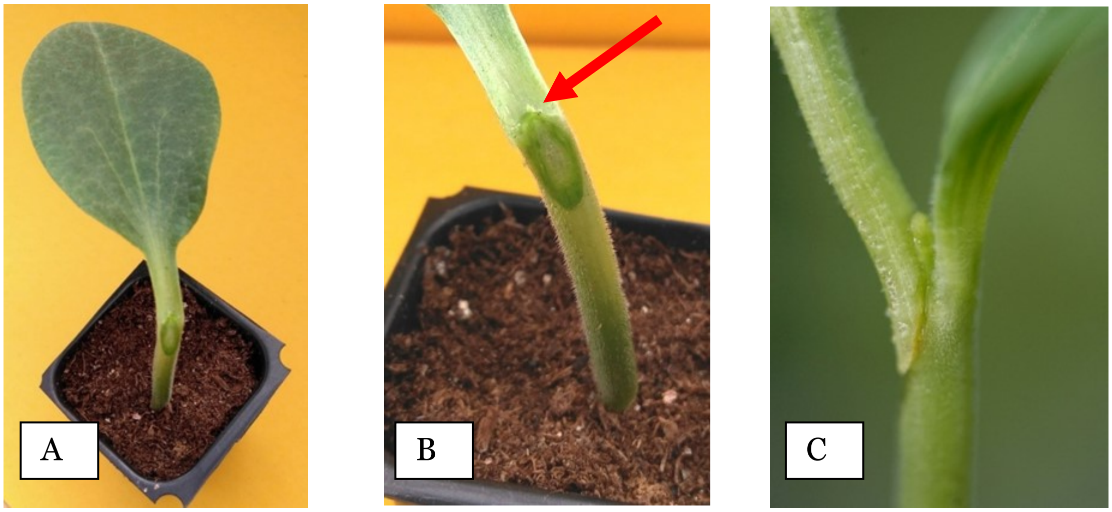

Figure 4. (A) & (B) The meristem tissue lies below the axillary bud at the base of the cotyledon; (C) if it is not completely removed, the rootstock will regrow. (*Photos by Pinki Devi*)

rope. Grafting machines can be used to graft watermelon and melons using this method, but high initial cost for equipment and strict requirement for uniformity of seedlings present obstacles for wider adoption of grafting automation of cucurbit crops.

Rootstock seedlings should have one true leaf, and scion seedlings should have two true leaves. Cut the rootstock at a 60° angle (Miles et al., 2013) so one cotyledon remains and one is removed (Fig. 3). Cut carefully so as to keep the remaining cotyledon firmly attached to the rootstock stem. The angled cut should also remove the apical meri-stem in the remaining cotyledon (these are undifferentiated cells at the base of the axillary bud) (Fig. 4).

It is important to remove all of the apical meristem to prevent future shoot growth of the rootstock. To better ensure that all of the axillary bud tissue has been removed, swipe the corner of the razor blade back and forth two times over the meristem area. Cut the scion at a 60° angle below the cotyledons, where its diameter matches that of the rootstock. Bring the two cut stem surfaces together, and hold them in place with a grafting clip. Place a small plastic straw through the clip to provide support as needed (Fig. 5); do not use wood/bamboo as it will mold in the healing chamber. Mist the grafted plants with water before placing them in the healing chamber.

Advantages:

- The most simple and rapid technique for grafting watermelon.
- Grafting automation can be conveniently accomplished.

Disadvantages:

- Requires careful control of humidity, light, and temperature after grafting.
- High losses and possible diseases or physiological disorders may occur if the healing environment is not optimal.
- Some meristem tissue may remain in the rootstock, requiring removal later in the production cycle.

2. Hole insertion grafting

The hole insertion method is the most widely used method for watermelon and melon grafting in China and Japan because it tends to have a high success rate with relatively minimal management during the healing period. Rootstock seedlings should have one small true leaf, and scion seedlings should have just the cotyledons or the first true leaf just emerging. The diameter of the scion stem must be smaller than

Figure 5. Place a small plastic straw through the clip to provide support. *(Photo by Pinki Devi)*

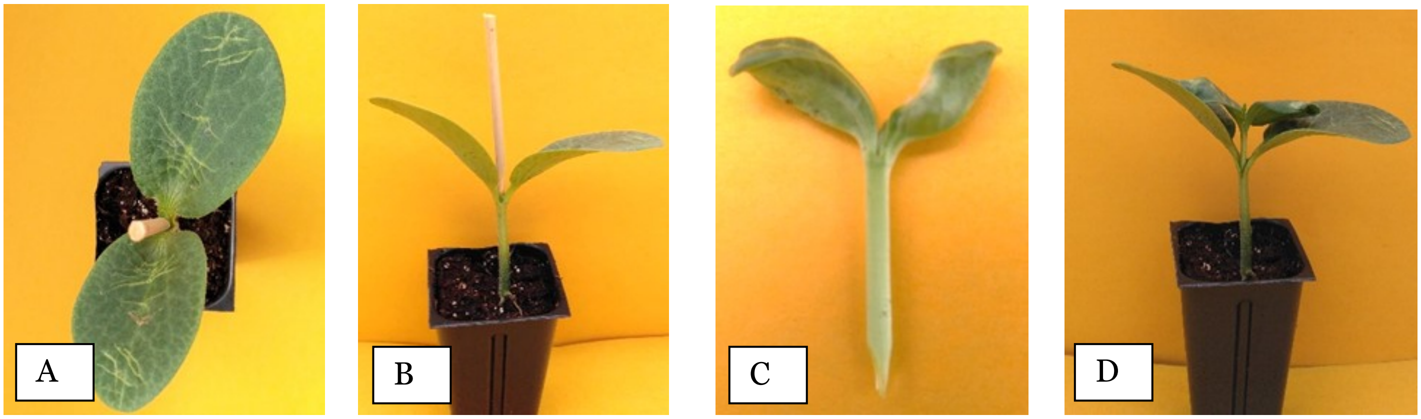

Figure 6. (A) & (B) Create a hole in the rootstock that removes the cotyledons and damages the meristem tissue; (C) cut the scion at a 45° angle on two sides to form a wedge; (D) insert the scion into the rootstock. (*Photos by Pinki Devi*)

the diameter of the rootstock stem so that the scion can be inserted into a hole made between the two cotyledons of the rootstock. Because seedlings can grow quickly in a day or two at this young growth stage, this grafting method has a short grafting window and timing of planting rootstock and scion seeds is critical.

With a pointed probe, remove the true leaf, the apical meristem, and the axillary buds from the topmost growing point of the rootstock plant (Fig. 6). It is important to remove all of the apical meristem and the axillary buds to prevent future shoot growth of the rootstock. Japanese grafters commonly use a bamboo probe as it is more effective at creating the hole and removing the apical meristem (Fig. 7). Use the probe to create a hole in the top of the rootstock where the tissue was removed; leave the probe inserted in the growing point while cutting the scion.

Figure 7. Wooden probe commonly used by Japanese farmers for hole insertion grafting.

(*Photo by P. Devi*)

Cut the scion below the cotyledons at a 45° angle on two sides to form a wedge and insert it into the rootstock as the probe is removed. Oftentimes, the scion cotyledons are positioned to avoid overlap with the rootstock cotyledons, however, this is not necessary as long as the cut surfaces are oriented so that they are in good contact. It is not necessary to use a grafting clip to hold the two plants together. Mist the grafted plants with water and place in healing chamber.

Advantages:

- A grafting clip is not essential, which saves time and labor involved in collecting grafting clips after healing.
- Tends to have a high success rate.
- Maximizes the contacting surface area between rootstock and scion which helps create a strong graft union.

Disadvantages:

- Requires slightly more skill than most other grafting techniques.
- It may require more time to graft than some of the other grafting techniques depending on the grafter's skill and the grafting operation.
- Regrowth of the rootstock will occur if not all the meristem tissue has been removed.
- In some cases, internal rooting of the scion through the pith of rootstock may occur, especially when the scion stem is not oriented in the rootstock stem properly.

3. Approach (tongue) grafting

The approach grafting method is popular for small-scale grafters who are grafting up to a few hundred plants. It is slower than other methods but it has a high success rate. However, when grafting melon plants using melon (*C. melo*) rootstocks, it may be challenging to use the approach grafting method because melon rootstocks tend to have thin stems; therefore, it is easier to use squash rootstocks that have thicker stems for melon grafting using this method.

Both rootstock and scion should have one or two true leaves and similar stem diameter. Remove the rootstock and scion seedlings from the seedling trays and cut a 45° downward slit

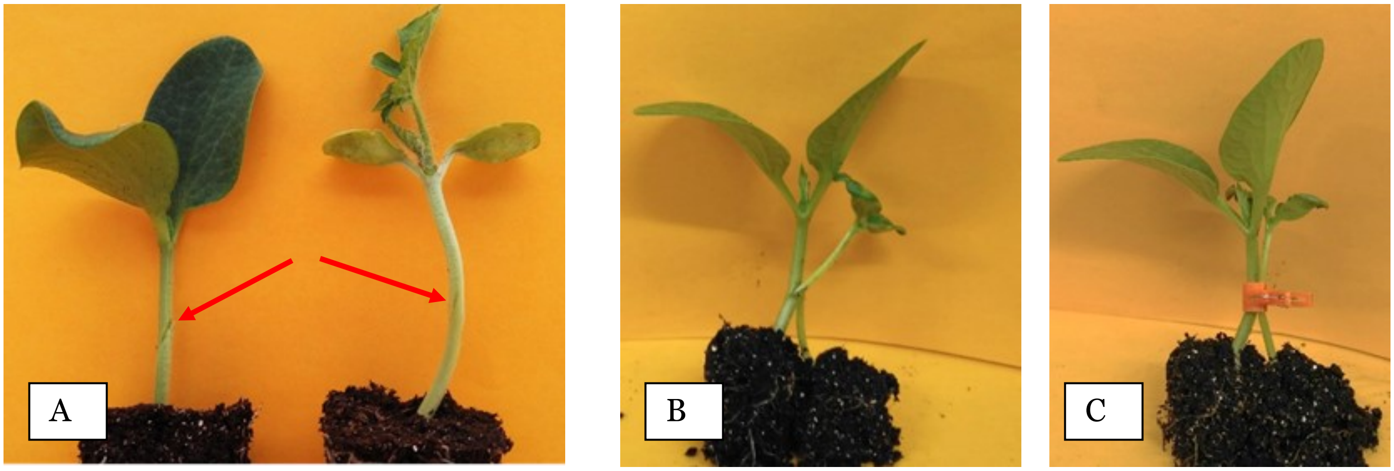

Figure 8. (A) Cut a 45° downward slit halfway through the rootstock stem below the cotyledons, and cut an identically angled upward slit in the scion stem; (B) bring the two cut stems together so the plants are joined; (C) attach a clip to secure the plants. (*Photos by Pinki Devi*)

Figure 9. A larger (left) and smaller (right) chamber used to heal grafted vegetables. (*Photos by Carol Miles*)

halfway through the rootstock stem below the cotyledons, and cut an identi-cally angled upward slit in the scion stem (Fig. 8). The angle and location of the cuts must be relatively precise so the scion can be placed on top of the rootstock. Join the two cut stems together and attach a clip or securely wrap the joined stems in plastic wrap or parafilm. Place the joined plant in a transplant tray or small pot that is just large enough to fit both rootballs. Mist the plant with water and place it on a greenhouse bench. Water the plant as needed. Cut off the top of the rootstock 5 days after grafting. Wait 7 days (12 days after grafting), then cut off the bot-tom portion of the scion.

Advantages:

- A relatively simple technique.
- A high humidity and low light environment is not required for successful healing of the graft union; a shaded greenhouse environment is sufficient.
- There is no shoot regrowth from the rootstock.

Disadvantages:

- Relatively slow grafting process.
- Requires more space for grafted transplants on the greenhouse bench.
- Not suitable for rootstock with hollow stems.

Figure 10. Healed watermelon plant with regenerated roots. (*Photos by Yuan Liu*)

Figure 11. Rootstock regrowth (squash-like leaves) from grafted watermelon (left) and melon (right) plants. (*Photos by Carol Miles and Xin Zhao*)

- It can be challenging when rootstock and scion plants look similar, making it difficult to distinguish between rootstock and scion seedling after grafting.

Healing chamber regime

Following grafting, it is critical to follow a successful graft healing regime (Johnson and Miles, 2011). Whatever healing chamber structure you use, it is necessary to control the humidity and temperature during the healing period, especially during the first 48-72 hours after grafting. After the critical 48-72 hours, gradual reduction of humidity and an increase in the amount of light is important for graft healing and disease prevention.

The following healing schedule is based on the greenhouse grafting environment at Washington State University Northwestern Washington Research and Extension Center in Mount Vernon (Johnson et al., 2011), where the average greenhouse temperature is 78/70 oF day/night (26/21 oC), and the relative humidity is 45%-60% in the late spring, when grafting usually occurs. Your green-house or grafting environment may be different (higher or lower temperature and humidity), and you may need to adjust the exposure times for grafted plants so they are not stressed when introducing them back into the greenhouse environment. The key is to slowly acclimatize the grafted plants without causing permanent wilting, which will lead to plant death.

Place grafted plants in a pre-misted humidity chamber (Fig. 9). In the following schedule, Day 1 is the day of grafting, and all chamber openings are during the day.

- Day 1. Close plastic of healing chamber; cover chamber with black fabric.
- Day 2. Keep chamber closed and covered with black fabric.
- Day 3. Open the chamber add water to chamber floor if needed (should be wet) close the chamber, and fold the black fabric up and away from the front of the chamber.
- Day 4. Open chamber for 15 minutes, wet floor of chamber if needed, reclose chamber, fold black fabric half way up all sides of the chamber.
- Day 5. Open chamber for 30 minutes, wet floor of chamber if needed, reclose chamber, remove black fabric from sides but keep top covered.
- Day 6. Open chamber for 1 hour, wet floor of chamber if needed, reclose chamber, remove black fabric entirely.
- Day 7. Open the chamber for 3 hours, wet floor of chamber if needed, reclose the chamber.
- Day 8. Open chamber for 6 hours, wet floor of chamber if needed, reclose chamber
- Day 9. Remove plants from the chamber and water them.

If greenhouse conditions are very bright and hot (greater than 90°F / 32 °C), place plants in a shaded area in the greenhouse for a few days to transition them before fully exposing them to regular greenhouse conditions.

If the root excision method was used for rootstock seedlings, the grafted plants should have regenerated roots by the time they are removed from the healing chamber (Fig. 10).

Remove rootstock regrowth

For plants grafted with the one cotyledon or the hole insertion methods, scout plants in the greenhouse during and after the healing process, and remove rootstock regrowth as soon as it appears. It is more time efficient and effective to remove rootstock regrowth when grafted plants are very young, before they are placed in the field. As plants get older, rootstock regrowth can cause graft failure. Shoot regrowth from the rootstock competes with the scion for water and nutrients, and reduces yield.

Preparation for transplanting into the field

Although vascular connection is established between scion and rootstock at approximately 7 days after grafting, it takes at least 14 days after grafting for the graft union to fully heal. After removing plants from the healing cham-ber, allow them to rest in the greenhouse for 5 days, and then move them outside for 3 days, so they can harden off before transplanting. Adjust this schedule as needed if plants appear stressed when they are introduced into each new environment.

Leave the clips on after transplanting to provide extra support, especially in windy conditions. Do not place grafted transplants into the field under very windy conditions. Grafting clips generally do not fall off the stem as the stem increases in diameter, so be sure to remove them 1-2 weeks after transplanting. Clean and sterilize grafting clips before you use them again (see Grafting Preparation above).
When transplanting, make sure the graft union remains above the soil line. If the graft union is buried, the scion will root into the soil and any advantages provided by the rootstock, such as resistance to soil-borne diseases, will be lost.

Field maintenance of grafted plants

Check the plants once a week the first month after transplanting to see if the rootstock has regrown (Fig. 11), and remove rootstock shoots immediately. It is very challenging later in the season to go through the field and remove rootstock suckers as the vines will be covering the field. However, many commercial rootstocks are extremely vigorous and will quickly overtake the scion variety if allowed to grow, so continued diligence for rootstock regrowth removal is recommended.

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Grafting Manual:
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Chapter 3.2.2 **March 2018**

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Synopsis:
Discusses rootstock selection and other important aspects of grafted melon and watermelon production for improving disease management, fruit yield, and crop performance.

Editors:
Chieri Kubota (The Ohio State University)
Carol Miles (Washington State University)
Xin Zhao (University of Florida)

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USDA United States Department of Agriculture National Institute of Food and Agriculture

Rootstock selections and important considerations in melon and watermelon grafting

General information

Chapter 3.22 in this grafting manual discusses the major methods used for melon (*Cucumis melo*) and watermelon (*Citrullus lanatus*) grafting, specific advantages and disadvantages of each method, the graft healing process, as well as preparation for field transplanting and maintenance of grafted plants. This chapter addresses rootstock selection and other important aspects of grafted melon and watermelon production towards improving disease management, fruit yield, and crop performance.

Major rootstocks used for grafted melon and watermelon production

Grafting is used primarily for managing soil-borne diseases including Fusarium wilt (caused by *Fusarium oxysporum*) and Verticillium wilt (caused by *Verticillium dahliae*) in melon and watermelon production. Moreover, improved cold tolerance of grafted cucurbits by using selected rootstocks also makes grafting a viable tool for early planting and harvest especially under protected culture. Currently, *Lagenaria siceraria* bottle gourd, *Cucurbita moschata* squash, and *C. maxima* × *C. moschata* interspecific hybrid squash are the most commonly used commercial rootstocks for melons and watermelons. *Lagenaria siceraria* is among the early generation of cucurbit rootstocks, and its popularity declined over the years due to broken resistance to Fusarium, as well as shallow root system. More interspecific *Cucurbita* hybrids with vigorous root systems are now being developed for watermelon for their high resistance to Fusarium wilt, tolerance to heat and drought conditions, and for cold tolerance.

The early work of rootstock evaluation among cucurbit species also confirmed the grafting compatibility of *C. maxima*, *C. pepo*, *Benincasa hispida*, and *Luffa cylindrica* with melons (Zhao et al., 2016), while commercial use of these rootstocks is rather limited. However, *C. maxima* rootstocks are sometimes used to manage Monosporascus vine decline (caused by *Monosporascus cannonballus*) in infested fields (Davis et al., 2008). The lack of resistance/tolerance to root-knot nematodes (*Meloidogyne* spp.) in interspecific hybrid squash rootstocks (Fig. 1) also stimulated the selection of *Cucumis metulifer* as an effective rootstock to control root-knot nematodes in melon production (Guan et al., 2014). However, given the weaker growth vigor of *C. metulifer* compared with squash rootstocks, more breeding work is needed to develop more vigorous *C. metulifer* rootstocks with greater potential for improving melon yield.

Although *C. moschata* rootstocks may possess a certain level of tolerance to nematodes, the relatively high susceptibility of *Cucurbita* rootstocks, particularly the interspecific hybrids, has led to the search for wild watermelon (*C. lanatus* var. *citroides*) as a new source of rootstocks for managing both Fusarium wilt and root-knot nematodes in watermelon production.

Despite their effectiveness in dealing with biotic and abiotic stresses, interspecific squash hybrid rootstocks have been found to negatively affect melon fruit quality of certain genotypes, especially specialty cultivars with aromatic flavors (Guan et al., 2015b). Such decline of fruit quality as a result of rootstock-scion interactions associated with use of interspecific hybrid squash rootstocks resulted in breeding effort to seek promising rootstock candidates in *Cucumis* species. *Cucumis melo* rootstock cultivars are commercially available, and most recently interspecific *Cucumis* hybrid rootstocks including *C. ficifolius* × *C. myriocarpus* 'UPV-FMy' and *C. ficifolius* × *C. anguria* 'UPV-FA' were developed in Spain (Caceres et al., 2017).

Figure 1. Root-knot nematode infestation in the roots of a melon plant grafted with an interspecific hybrid squash rootstock. (*Photo by Yufan Tang*)

Table 1 lists the major types of rootstock species used for grafted melon and watermelon production and their key characteristics. If the melon fruit are targeted to a premium market, the rootstock-scion combination needs to be tested to determine potential rootstock effects on fruit quality attributes such as soluble solids content before large-scale adoption occurs.

Important considerations for using grafted plants to improve disease management

By using resistant/tolerant rootstocks, soilborne fungal diseases including Fusarium wilt, Verticillium wilt, and Monosporascus root rot and vine decline can be successfully controlled in grafted melon and watermelon production (Guan et al., 2012; Louws et al., 2010). Both bottle gourd and interspecific hybrid squash rootstocks for watermelon grafting are considered susceptible to gummy stem blight (caused by *Didymella bryoniae*), and managing gummy stem blight on watermelon and rootstock seedlings with fungicides during transplant production may be necessary to ensure grafted watermelon health and quality prior to field planting (Keinath, 2013). On the other hand, grafting with certain *C. maxima* × *C. moschata* rootstocks has been shown to effectively manage gummy stem blight that survives in crop residues and initiates infection in the lower crown

Table 1. Major types of rootstocks used for melon and watermelon grafting and their key characteristics.

Type of rootstock	Rootstock cultivar example	Major beneficial characteristic	Limitation
Lagenaria siceraria (bottle gourd)	'Emphasis' (Syngenta), 'Macis' (Nunhems)	Resistant to Fusarium wilt; tolerant to low temperature	Possible susceptibility to new Fusarium races; shallow root system
Cucurbita moschata (squash)	'Marvel' (American Takii)	Resistant to Fusarium wilt; possibly tolerant to root-knot nematodes; tolerant to low temperature	Possible susceptibility to *Phytophthora*; possible adverse effect on fruit quality
Cucubita maxima × *C. moschata* (interspecific hybrid squash)	'Carnivor', 'Super Shintosa' (Syngenta); 'Cobalt' (Rijk Zwaan); 'RST-04-109- MW' (DP Seeds); 'Flexifort' (Enza Zaden)	Resistant to Fusarium wilt; possibly resistant to melon necrotic spot virus; tolerant to low and high temperatures; vigorous growth	Susceptibility to root-knot nematodes; possible susceptibility to *Phytophthora*; possible delay in fruit set and reduction in fruit quality
Citrullus lanatus var. citroides (wild watermelon)	'Ojakkyo' (Syngenta)	Resistant to Fusarium wilt and root-knot nematodes	For watermelon grafting. Lack of good tolerance to low temperature
Cucumis melo (melon)	'Dinero' (Syngenta)	Resistant or tolerant to Fusarium wilt; maintaining fruit quality	For melon grafting. Possible susceptibility to *Phytophthora*; less vigorous compared to *Cucurbita* rootstocks
Cucumis metuliferus (African horned cucumber)	No commercial rootstocks available	Resistant to Fusarium wilt; resistant to root-knot nematodes	For melon grafting. Less vigorous compared to *Cucurbita* rootstocks; lack of good tolerance to low temperature

Adapted from Davis et al. (2008) and Louws et al. (2010).

of melon plants (Crinò et al., 2007). In addition, some *C. maxima* × *C. moschata* rootstocks bred for resistance to melon necrotic spot virus, a soil-borne virus, can be employed to limit virus infection. When virus disease is prevalent, growers need to be well aware of possible virus resistance or susceptibility of rootstocks, because if the rootstock is more susceptible than the scion, the virus issue can become worse with grafted plants.

It needs to be noted that most of the current rootstocks used for melon and watermelon grafting may be susceptible to *Phytophthora*. Growers are advised to identify the production constraints on site, including soil-borne disease problems and environmental stress conditions, in order to choose the appropriate rootstocks to best suit their needs. Few commercial rootstock cultivars with high resistance to root-knot nematodes are currently available, although *C. moschata* rootstocks may show tolerance to root-knot nematodes under intermediate levels of field infestation.

While their capability of coping with low temperatures and other abiotic stress is yet to be fully determined, wild watermelon rootstocks can be used as an effective management tool for both Fusarium wilt and root-knot nematodes. It is important to understand the complete disease resistance package of the rootstock for successful use of grafted plants in addressing specific soil-borne disease problems. Rotation of rootstocks is also recommended to

Figure 2. Commercial greenhouse operations of grafted melon production in Italy. (*Photos by Xin Zhao*)

minimize new pathogen emergence and possible shifts in the host specificity of the pathogen population (Louws et al., 2010).

Optimizing improvement of crop performance of grafted plants beyond disease resistance

In addition to soil-borne disease management, resistant rootstocks, especially the squash rootstocks, often possess good to excellent tolerance to abiotic stress particularly low temperature conditions. Almost all the melons and watermelon grown in South Korea are grafted. For watermelon, disease resistance is the main consideration in Korea as well as other countries. In contrast, for melons, cold tolerance is the main consideration because melons are primarily cultivated under winter greenhouse production conditions.

Grafting melon onto low-temperature-tolerant rootstocks reduces the risk of severe growth inhibition caused by low soil temperatures in winter greenhouses (Lee et al., 2010). Using

Figure 3. Commercial production of grafted melon in low-cost tunnel systems in Sicily, Italy for improved earliness. (*Photos by Xin Zhao*)

grafted plants under protected culture for enhancing earliness of melon fruit is also widely practiced in Italy (Fig. 2 and 3). Research trials are taking place in the southeastern U.S. and other regions to explore the use of grafted plants for early spring planting of seedless watermelons in open field systems. Selected *C. maxima* × *C. moschata* rootstocks may also hold promise for improving plant tolerance to soil salinity (Rouphael et al., 2012).

Even though the yield improvement potential varies with the genotype of rootstock, modification of nutrient and water management as well as plant spacing is suggested to maximize the benefits of using grafted transplants. Without appropriate management programs, the excessive vegetative growth resulting from the use of vigorous interspecific rootstocks could cause delayed appearance of female flowers and fruit set. Meanwhile, more vegetative growth observed in grafted watermelons could potentially create a microclimate condition that is more favorable for foliar disease development.

Typically, nitrogen application needs to be reduced during the vegetative phase to minimize the potential adverse impact of vigorous rootstocks on flowering and fruit set. Wider plant spacings can also be considered to better utilize the rootstock vigor. Such management considerations will not only assist with optimizing crop performance of grafted plants but also help justify the high price of grafted transplants by reducing production input costs.

Dealing with fruit quality concerns

As described earlier, especially in melon, fruit off-flavor was noticed when vigorous hybrid squash rootstocks are used (Davis et al., 2008). This may be a reason that introduction of grafting is more advanced in watermelon than in melon worldwide. Melons (including muskmelon and honeydew type) belong to four groups of two subspecies of *Cucumis melo*, including the groups of cantalupensis, reticulatus, and inodorus within the subspecies *melo*, and the group of makuwa within the subspecies *agrestis* (Guan, et al., 2013). The complexity of various melon cultivars makes melon a good crop to study rootstock-scion interaction effects in terms of physiological and biochemical changes in grafted plants, but also results in complicated scenarios for examining fruit quality.

For example, some interspecific hybrid squash rootstocks could reduce soluble solids content of aromatic galia melons and sensory attributes perceived by consumers (Guan et al., 2015a). The delay in fruit set together with accelerated ripening by grafting with the vigorous interspecific squash rootstock contributed to the fruit quality decline at harvest. Because of the fruit quality concerns, Fusarium wilt-susceptible melon cultivars are grafted onto Fusarium wilt-resistant *C. melo* rootstocks in Japan to maintain fruit quality while accomplishing disease management (King et al., 2010).

As honeydew melons in the inodorus group tended to be less influenced by squash rootstocks in contrast to melons in the reticulatus and cantalupensis groups (Guan et al., 2015b; Traka-Mavrona et al., 2000), it has been speculated that melon cultivars producing climacteric fruit with aromatic flavors might be more likely to respond to grafting in terms of fruit quality attribute changes as opposed to nonclimacteric, less aromatic melons.

Interspecific hybrid squash rootstocks generally have good grafting compatibility with melons and watermelons. Nevertheless, a nonpathological vine decline specifically associated with certain combinations of melon scion and interspecific hybrid squash rootstock has been reported in Israel, U.S. (Fig. 4) and Cyprus (Aloni et al., 2008; Guan et al., 2015a; Soteriou et al., 2016). The crop failure observed later during the melon planting season is likely caused by increased water stress as a result of heavy fruit load and high temperature, while more in-depth investigations are expected to shed light on the fundamental mechanisms at plant physiological and hormonal levels.

Since fruit size increase is often found in grafted melon and watermelon plants when interspecific squash rootstocks are used, growers are advised to consider market acceptability of certain fruit size categories to avoid unexpected loss of marketable yield. In general, the adverse impact of grafting with squash rootstocks on watermelon fruit quality appears to be less extensive in contrast to the situation with melon grafting. Fruit soluble solids content, pH, titratable acidity, lycopene content, and consumer perceived sensory attributes may be well maintained in grafted watermelon (Liu et al., 2017). The *C. lanatus* var. *citroides* wild watermelon rootstock has been found to have rather limited influence on watermelon fruit quality and aroma profile besides yielding larger fruit with thicker rind (Fredes et al., 2016). On the other hand, increase in watermelon flesh firmness, reduction in hollow heart incidence and severity, and higher level of lycopene observed in certain rootstock-scion combinations could be the added value of using grafted plants.

As more studies are needed to fully understand the intrinsic impacts of rootstock and rootstock-scion interaction on fruit quality modification, environmental conditions during crop production and fruit maturity at harvest also deserve attention in assessing fruit quality of grafted plants (Rouphael et al., 2010). Rootstock breeding and development for melon and watermelon production will continue to address the fruit quality concerns. Ultimately, grower's decision of integrating grafting with selected rootstocks into melon and watermelon production will be driven by their needs to overcome production challenges towards longer-term environmental and economic sustainability.

Figure 4. Grafted muskmelon plants (with an interspecific hybrid squash rootstock) collapse during fruit maturation stage later in the spring production season in north Florida. This non-pathological disorder might be caused by increased water stress as a result of heavy fruit load and high temperature; however, this physiological disorder is not well understood. (*Photo by Wenjing Guan*)

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Grafting Manual:

How to Produce Grafted Vegetable Plants

www.vegetablegrafting.org

Chapter 3.3 May 2017

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Synopsis:

Grafting eggplant (*Solanum melongena*) to disease resistant rootstocks is commonly used for disease and abiotic stress management in commercial production. Grafting methods can be adapted to various systems of plant production.

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USDA United States Department of Agriculture National Institute of Food and Agriculture

Eggplant

Splice grafting is the most common method used to graft eggplant (*Solanum melongena*) because it has a high success rate (≥85%), is relatively simple, and can be used to graft a large number of plants in a short amount of time. For larger plants, the cleft grafting technique is commonly used.

Common rootstocks

Eggplant is commonly grafted onto either eggplant rootstocks or tomato rootstocks (Bletos and Olympios, 2008). When soil conditions are disease-free, eggplant rootstocks provide more consistent plant growth and yield (King et al., 2010), while tomato rootstocks generally have been more developed for disease resistance. Growers typically have more experience with tomato rootstocks, thus, tomato rootstocks are most commonly used for grafting eggplant in North America. Table 1 is a list of commercial eggplant rootstocks, and tomato rootstocks that have been investigated by university researchers for eggplant grafting. For a more complete list of tomato rootstocks, see the chapter on tomato grafting (NA, 2015). Local tomato cultivars are also commonly used as rootstock for eggplant; however, only commercial rootstocks are included here.

In addition to rootstocks that have been bred for eggplant grafting, there are several closely related wild species that are commonly used or marketed as rootstocks for eggplant. Most common of these is *Solanum torvum*. However, *S. torvum* seeds tend to have poor shelf-life and uneven germination, seedlings can be slow growing, and in the U.S. it is listed as a noxious weed (USDA-NRCS, 2016). *Solanum aethiopicum*, formerly called *Solanum integrifolium* Poir, is another wild species that was first used for grafting eggplant in the 1950s. Seed of these and other *Solanum* wild species are available through seed catalogs and through seed banks. They can be crossed with closely related species, including eggplant (*S. melongena*), to create interspecific hybrid rootstocks. Wild species that have been used in eggplant rootstock breeding programs include S. *sisymbriifolium*, S. *aculeatissumum*, S. *capsicoides*, S. *incanum*, S. *linnaeanum*, and S. *viarum* in addition to those mentioned above (Bletos and Olympios, 2008).

Advantages of rootstocks

Rootstocks can provide added vigor under favorable growing conditions, while under unfavorable conditions they may provide a level of resistance to abiotic stress, insect pests or disease. The common insect pests and diseases impacting eggplant production are root-knot nematode, bacterial wilt, Fusarium wilt and Verticillium wilt. While seed companies may state a particular rootstock may be resistant to a particular insect pest or disease, research studies have shown variable results (Bletos, et al, 2003; Johnson et al, 2011). Table 2 is a summary of research studies and findings for eggplant grafted onto various rootstocks.

Preparing for splice grafting

Graft plants when they have 2–3 true leaves, which typically takes 3–5 weeks from seeding. For a successful graft union to form, the cambium must be aligned when the cut stem surfaces of the scion and rootstock are placed together. To accomplish this, the scion and rootstock plants must have similar stem diameters at the time of grafting. However, the scion and rootstock may not germinate or grow at the same rate, and eggplant typically grows slower than tomato.

Both eggplant and tomato germinate best when soil temperature is 85°F (29°C), but will germinate well when soil temperature is in the range of 75–90°F (24–32°C) (Dupont 2012; Kemble et al, 1998). Conduct a preliminary trial to determine the growth rates of scion and rootstock plants in your growing environment in order to

Table 1. Rootstocks for eggplant grafting and disease resistance for each rootstock cultivar.

Rootstock type	Variety	Bacterial Wilt	Fusarium Wilt Race 1	Fusarium Wilt Race 2	Verticillium Wilt
Eggplant	Java[1]		HR[2]	HR	HR
	Red Scorpion[1]	IR	HR	HR	
	Zippy[1]	HR	HR	HR	
Tomato	Meet				
	Estamino		HR	HR	HR
	Survivor	IR	HR	HR	HR
	Beaufort		R[3]	R	R
	Maxifort		R	R	R
	E16R.4040F1				
	Multifort		R	R	
	RST-04-106-T	HR			
Wild type	*Solanum aethiopicum*	IR	HR	HR	IR
	Solanum torvum	HR	HR	HR	HR

[1] Information provided by rootstock seed companies.
[2] HR represents high resistance, IR represents intermediate or partial resistance.
[3] R represents resistance in cases where seed companies or literature did not rate for partial resistance.

Table 2. Research studies that have investigated eggplant grafting in North America.

Rootstock type	Variety	Source	Research Location[1]	Focus of Research[2]	Year(s) Researched
Eggplant	Java	Takii	WSU	VW	2013, 2014, 2015, 2016
	Red Scorpion	Takii	WSU, UA	VW, cold tolerance	2013, 2014
	Zippy	Takii			
Tomato	Meet	Takii	WSU	VW	2013, 2014, 2015, 2016
	Estamino	Johnny's	WSU	VW	2014, 2015, 2016
	Survivor	Takii	WSU	VW	2016
	Beaufort	DeRuiter Seeds	WSU	VW	2011
	Maxifort	DeRuiter Seeds	WSU, Cornell, NCSU	VW, herbicdes	2010, 2011, 2012, 2016
	E16R.4040F1	Enza Zaden	WSU	VW	2015
	Multifort	DeRuiter Seeds	WSU	VW	2009
	RST-04-106-T	DP Seeds	NCSU	BW	2016
Wild Spp.	*Solanum aethiopicum*		WSU	VW	2010

[1] WSU=Washington State University; UA=University of Arizona; NCSU=North Carolina State University.

2VW=Verticillium wilt; BW= bacterial wilt.

determine proper seeding dates for both. Seed more plants than necessary to have a selection for matching stem diameters, and also to account for some graft failure. Overall, seed 15-20% more scion and rootstock plants than the final number needed.

The day before grafting, sort scion and rootstock plants into small, medium and large size categories based on stem diameter. Stem diameter range will be 1.5-3.0 mm overall. Water both scion and rootstock plants 12–24 hours before grafting. Unless absolutely necessary, do not water rootstock plants immediately before grafting, as this will cause the plants to exude water when they are cut, which will prevent the cut surfaces from being in tight contact. If reusing grafting clips, make sure they have been cleaned and sterilized. Use only clean, sharp razor blades, and sanitize hands with antibacterial soap or hand gel. Fill spray bottles with tap water to mist plants as needed during grafting.

Splice grafting

Graft during a time of day when plant transpiration is lowest, such as early morning, to minimize water stress in the newly grafted plants. If needed, shade the grafting area to reduce temperature and water stress of plants (Zhao, 2010). Graft plants in groups based on stem diameter category (small, medium, large). It is critical to match stem diameter so that the entire cut surface of both scion and rootstock stems are in close contact, with no air between them. If the cut surface of the scion or the rootstock dries out, the graft will fail.

1. Cut the rootstock stem at a 45° angle below the cotyledons to prevent the rootstock from producing new shoot growth; immediately discard the rootstock tops.

2. Place a grafting clip over the cut rootstock stem.
3. Cut the scion at a matching 45° angle and slip the plant into the grafting clip (Fig. 1).
4. Cut rootstock and scion seedlings one flat (72-cell trays) at a time to attain more rapid grafting time.
5. If needed, place a small plastic stick into the potting media to hold plant straight and upright.
6. Lightly mist plant leaves with water as needed to reduce water stress, but do apply too much water

Place each flat of grafted plants into the healing chamber as soon as the flat is grafted.

Cleft grafting

The cleft graft technique is a good choice for grafting larger plants (5-6 true leaves), and is also known as apical grafting and wedge grafting. The cleft cut holds the scion more tightly than splice grafting. However, cleft grafting takes more time than splice grafting, and the rootstock stem may split if the scion wedge is too large.

1. Cut the rootstock stem horizontally to remove the top of the plant and discard the top.
2. Cut a ~¼ inch (0.5 cm) long vertical incision into the center of the rootstock stem.
3. Where the scion stem is the same diameter or slightly smaller than the rootstock stem diameter, cut into a ~¼ inch long wedge.
4. Insert the scion stem into the vertical incision in the rootstock. (Fig. 2).
5. Place a plastic clip around the graft union to hold the plant tightly together.
6. Place small plastic stick through the clip into the potting media to hold plant straight and upright.

For graft survival, it is more essential to maintain a high humidity for eggplant than for tomato (Johnson and Miles, 2001).

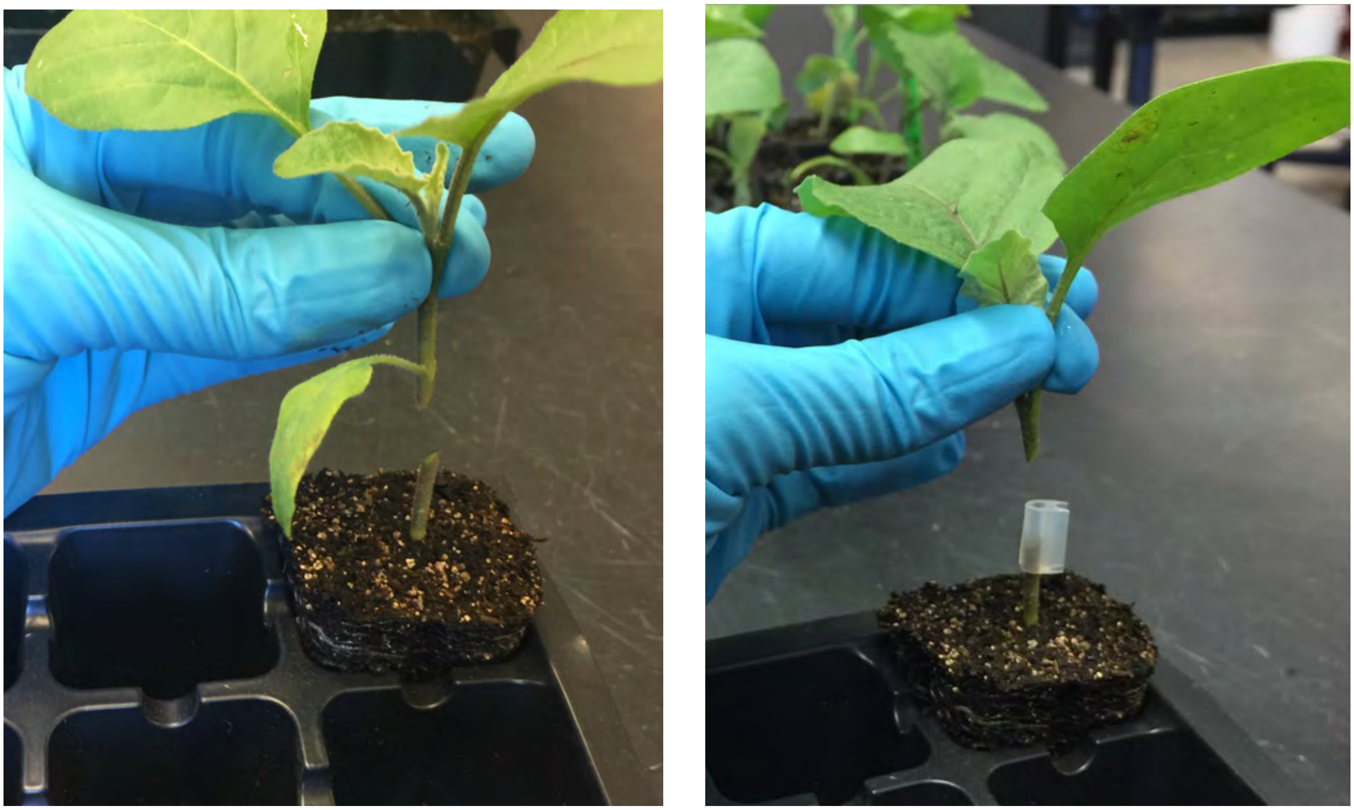

Figure 1. Splice Grafting: Cut the rootstock and scion stems at a 45° angle below the cotyledons (L) and slip the scion into the grafting clip on the rootstock. (*Photos by Ed Scheenstra and Patti Kreider*)

Figure 2. Cleft Grafting: Cut the rootstock horizontally below the cotyledons and make an incision in the stem, cut the scion into a wedge below the cotyledons and insert into the rootstock; attach with a grafting clip, and add a plastic stick to keep the plant upright. (*Photos by Ed Scheenstra and Patti Kreider*)

Depending on the greenhouse and healing chamber temperature, humidity, and solar radiation, this healing schedule may need to be adjusted so that plants are not stressed. For example, decrease the number of hours each day that plants are exposed to the greenhouse environment so they are not too wilted. The key is to slowly acclimate the grafted plants without causing permanent wilting, which will lead to plant death. Refer to Chapter 4, “Healing and acclimatization methods and design principles,” for a more detailed description on plant healing.

Although the scion and rootstock establish vascular connection at approximately 4-7 days, it takes at least 10-14 days from grafting for the graft union to fully heal. After removing plants from the healing chamber, leave plants in the greenhouse for an additional 4 days. Then, move plants outside to the hardening off area for 3 days before transplanting them into the field. Adjust this schedule if needed so plants are not stressed when they are placed in the field (Spalholz, 2013).

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Rootstocks can provide added vigor under favorable growing conditions, while under unfavorable conditions they may provide a level of resistance to abiotic stress, insect pests or disease. The common insect pests and diseases impacting eggplant production are root-knot nematode, bacterial wilt, Fusarium wilt and Verticillium wilt. While seed companies may state a particular rootstock may be resistant to a particular insect pest or disease, research studies have shown variable results (Bletos, et al, 2003; Johnson et al, 2011). Table 2 is a summary of research studies and findings for eggplant grafted onto various rootstocks.

Preparing for splice grafting

Graft plants when they have 2–3 true leaves, which typically takes 3–5 weeks from seeding. For a successful graft union to form, the cambium must be aligned when the cut stem surfaces of the scion and rootstock are placed together. To accomplish this, the scion and rootstock plants must have similar stem diameters at the time of grafting. However, the scion and rootstock may not germinate or grow at the same rate, and eggplant typically grows slower than tomato.

Both eggplant and tomato germinate best when soil temperature is 85°F (29°C), but will germinate well when soil temperature is in the range of 75–90°F (24–32°C) (Dupont 2012; Kemble et al, 1998). Conduct a preliminary trial to determine the growth rates of scion and rootstock plants in your growing environment in order to

Table 1. Rootstocks for eggplant grafting and disease resistance for each rootstock cultivar.

Rootstock type	Variety	Bacterial Wilt	Fusarium Wilt Race 1	Fusarium Wilt Race 2	Verticillium Wilt
Eggplant	Java[1]		HR[2]	HR	HR
	Red Scorpion[1]	IR	HR	HR	
	Zippy[1]	HR	HR	HR	
Tomato	Meet				
	Estamino		HR	HR	HR
	Survivor	IR	HR	HR	HR
	Beaufort		R[3]	R	R
	Maxifort		R	R	R
	E16R.4040F1				
	Multifort		R	R	
	RST-04-106-T	HR			
Wild type	*Solanum aethiopicum*	IR	HR	HR	IR
	Solanum torvum	HR	HR	HR	HR

[1] Information provided by rootstock seed companies.
[2] HR represents high resistance, IR represents intermediate or partial resistance.
[3] R represents resistance in cases where seed companies or literature did not rate for partial resistance.

Table 2. Research studies that have investigated eggplant grafting in North America.

Rootstock type	Variety	Source	Research Location[1]	Focus of Research[2]	Year(s) Researched
Eggplant	Java	Takii	WSU	VW	2013, 2014, 2015, 2016
	Red Scorpion	Takii	WSU, UA	VW, cold tolerance	2013, 2014
	Zippy	Takii			
Tomato	Meet	Takii	WSU	VW	2013, 2014, 2015, 2016
	Estamino	Johnny's	WSU	VW	2014, 2015, 2016
	Survivor	Takii	WSU	VW	2016
	Beaufort	DeRuiter Seeds	WSU	VW	2011
	Maxifort	DeRuiter Seeds	WSU, Cornell, NCSU	VW, herbicdes	2010, 2011, 2012, 2016
	E16R.4040F1	Enza Zaden	WSU	VW	2015
	Multifort	DeRuiter Seeds	WSU	VW	2009
	RST-04-106-T	DP Seeds	NCSU	BW	2016
Wild Spp.	*Solanum aethiopicum*		WSU	VW	2010

[1] WSU=Washington State University; UA=University of Arizona; NCSU=North Carolina State University.

2VW=Verticillium wilt; BW= bacterial wilt.

determine proper seeding dates for both. Seed more plants than necessary to have a selection for matching stem diameters, and also to account for some graft failure. Overall, seed 15-20% more scion and rootstock plants than the final number needed.

The day before grafting, sort scion and rootstock plants into small, medium and large size categories based on stem diameter. Stem diameter range will be 1.5-3.0 mm overall. Water both scion and rootstock plants 12–24 hours before grafting. Unless absolutely necessary, do not water rootstock plants immediately before grafting, as this will cause the plants to exude water when they are cut, which will prevent the cut surfaces from being in tight contact. If reusing grafting clips, make sure they have been cleaned and sterilized. Use only clean, sharp razor blades, and sanitize hands with antibacterial soap or hand gel. Fill spray bottles with tap water to mist plants as needed during grafting.

Splice grafting

Graft during a time of day when plant transpiration is lowest, such as early morning, to minimize water stress in the newly grafted plants. If needed, shade the grafting area to reduce temperature and water stress of plants (Zhao, 2010). Graft plants in groups based on stem diameter category (small, medium, large). It is critical to match stem diameter so that the entire cut surface of both scion and rootstock stems are in close contact, with no air between them. If the cut surface of the scion or the rootstock dries out, the graft will fail.

1. Cut the rootstock stem at a 45° angle below the cotyledons to prevent the rootstock from producing new shoot growth; immediately discard the rootstock tops.

2. Place a grafting clip over the cut rootstock stem.
3. Cut the scion at a matching 45° angle and slip the plant into the grafting clip (Fig. 1).
4. Cut rootstock and scion seedlings one flat (72-cell trays) at a time to attain more rapid grafting time.
5. If needed, place a small plastic stick into the potting media to hold plant straight and upright.
6. Lightly mist plant leaves with water as needed to reduce water stress, but do apply too much water

Place each flat of grafted plants into the healing chamber as soon as the flat is grafted.

Cleft grafting

The cleft graft technique is a good choice for grafting larger plants (5-6 true leaves), and is also known as apical grafting and wedge grafting. The cleft cut holds the scion more tightly than splice grafting. However, cleft grafting takes more time than splice grafting, and the rootstock stem may split if the scion wedge is too large.

1. Cut the rootstock stem horizontally to remove the top of the plant and discard the top.
2. Cut a ~¼ inch (0.5 cm) long vertical incision into the center of the rootstock stem.
3. Where the scion stem is the same diameter or slightly smaller than the rootstock stem diameter, cut into a ~¼ inch long wedge.
4. Insert the scion stem into the vertical incision in the rootstock. (Fig. 2).
5. Place a plastic clip around the graft union to hold the plant tightly together.
6. Place small plastic stick through the clip into the potting media to hold plant straight and upright.

For graft survival, it is more essential to maintain a high humidity for eggplant than for tomato (Johnson and Miles, 2001).

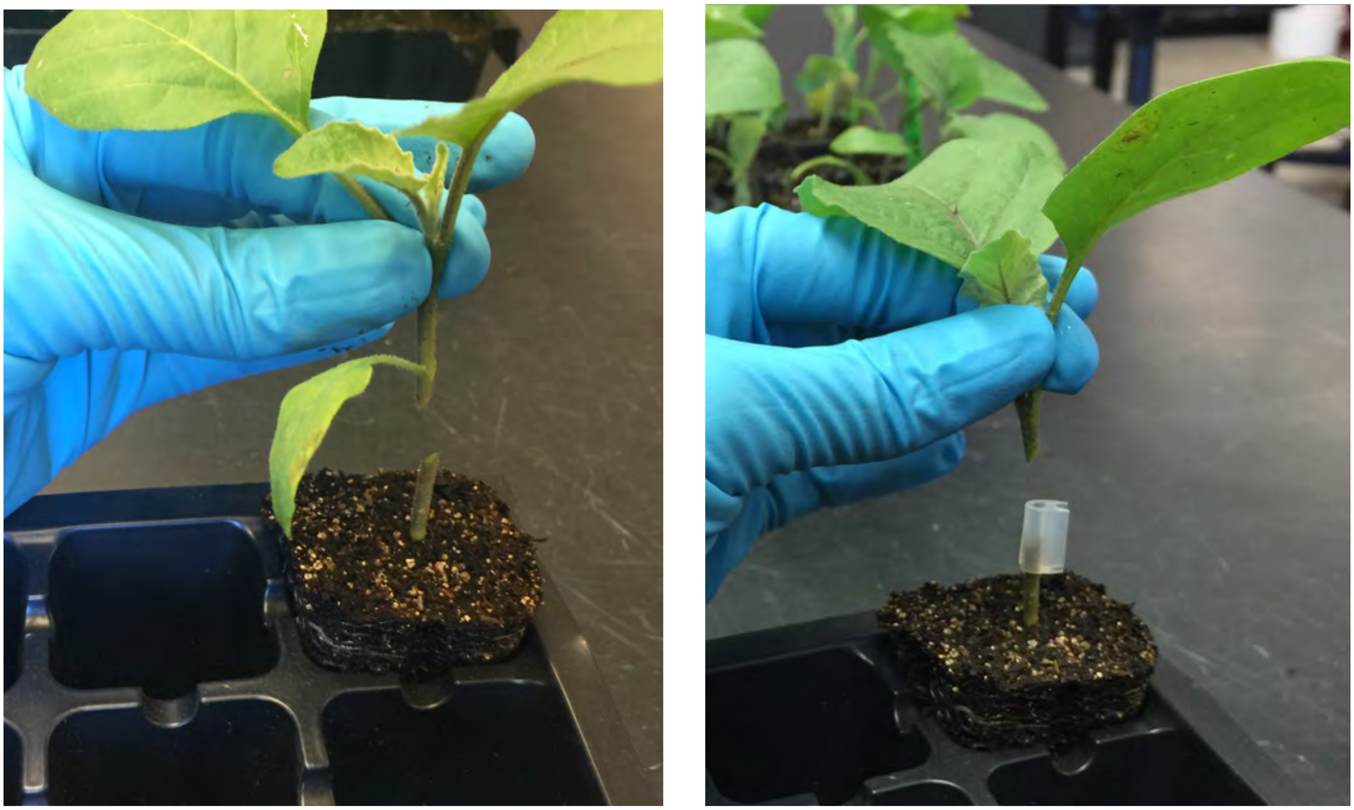

Figure 1. Splice Grafting: Cut the rootstock and scion stems at a 45° angle below the cotyledons (L) and slip the scion into the grafting clip on the rootstock. (*Photos by Ed Scheenstra and Patti Kreider*)

Figure 2. Cleft Grafting: Cut the rootstock horizontally below the cotyledons and make an incision in the stem, cut the scion into a wedge below the cotyledons and insert into the rootstock; attach with a grafting clip, and add a plastic stick to keep the plant upright. (*Photos by Ed Scheenstra and Patti Kreider*)

Depending on the greenhouse and healing chamber temperature, humidity, and solar radiation, this healing schedule may need to be adjusted so that plants are not stressed. For example, decrease the number of hours each day that plants are exposed to the greenhouse environment so they are not too wilted. The key is to slowly acclimate the grafted plants without causing permanent wilting, which will lead to plant death. Refer to Chapter 4, "Healing and acclimatization methods and design principles," for a more detailed description on plant healing.

Although the scion and rootstock establish vascular connection at approximately 4-7 days, it takes at least 10-14 days from grafting for the graft union to fully heal. After removing plants from the healing chamber, leave plants in the greenhouse for an additional 4 days. Then, move plants outside to the hardening off area for 3 days before transplanting them into the field. Adjust this schedule if needed so plants are not stressed when they are placed in the field (Spalholz, 2013).

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Chapter 4 **November 2016**

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Synopsis:
Technologies for nursery automation have been developed to address high labor cost issues in commercial operations. Types of automation include machines to handle most aspects of plant production.

Editors:
Chieri Kubota (The Ohio State University)
Carol Miles (Washington State University.)
Xin Zhao (University of Florida)

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USDA United States Department of Agriculture National Institute of Food and Agriculture

Automation in vegetable grafting nurseries

Healing Technologies for nursery automation have been developed to address high labor cost issues in commercial operations. Attempt to automate vegetable grafting has been made since the 1990s (Kurata, 1994) to contribute to efficiencies in labor at various stages of the grafting nursery operations. This article summarizes currently available automation technologies specific to vegetable grafting nurseries.

Types of automation

In the U.S. commercial vegetable nursery industry, various types of automation have been used for many years. These include machines that mix substrate, fill trays, seed trays, transport trays within the facility, irrigate trays, and control environmental conditions throughout the various stages of seed germination and plant growth. Grafting nurseries may consider additional automation in two categories: 1) sorting plants for uniformity prior to grafting, and 2) grafting (cutting and joining together scion and rootstock).

Sorting machines rearrange seedlings to create trays with uniform stands (Fig. 1). Having uniform seedlings improves grafting speed especially when the grafting method requires matching stem size between scion and rootstock. Uniformity of seedling growth is largely affected by the uniformities of seed germination and cotyledon emergence. Therefore, sorting is typically done when seedlings have fully expanded cotyledons and are about to develop the first true leaf. A sorting machine developed in the Netherlands can sort as many as 7,000-8,000 tomato seedlings per hour into three classes (Table 1). After sorting, nursery growers would manage irrigation, temperature and/or light to accelerate growth of small plants and restrain growth of large plants so that all plants would, ideally, be about the same size at grafting time. One major drawback of sorting machines is that types of substrate (media) and trays compatible with machines are currently limited to specific products. More flexible sorting machines need to be developed to accommodate the variety of trays and substrates used by the nursery industry. Machines or tools that would sort a smaller number of trays would also be useful for small to mid-size nurseries.

Grafting machines have been developed over the years by several manufacturers. Initially automation was targeted mainly towards grafting cucurbit seedlings as the manual grafting speed for these crops is relatively slow (K. Kobayashi, personal communication). For example, an experienced grafter with good work logistics may reach up to only 150 grafts per hour for cucurbits, while 300 grafts per hour can be achieved for tomato.

Recent efforts, however, focus on developing the automated grafting capacity for tomato in addition to cucurbit plants, presumably due to the large increase in the demand for grafted tomato plants compared with 20-30 years ago. Most widely used type of grafting machines are ‘semi-automated’ machines that typically require at least one worker to manage the operation of each machine (Fig. 2). The specifications for plant size, tray type, and substrate type are less restrictive for semi-automated machines and therefore they are relatively easier to be managed in nurseries than fully automated machines. This specification of the materials compatible with grafting machines was shown as a significant cost factor in an economic sensitivity analysis (Lewis et al., 2014). Table 1 summarizes the grafting machines currently available in the North American market.

Use of automation

The level of automation that is considered adequate for each nursery depends on the size of the individual nursery as well as availability of labor. Grafting machines appear to take a vital role for nurseries that are quickly developing their grafting capacity with limited experience in grafted seedling production. One of the largest operations of automated grafting is now located in the U.S. and it utilizes five semi-automated grafting machines with two shifts of assisting workers (Rootility, 2015).

However, nurseries need to be aware that effective use of automation always requires a foundation of good nursery practices that includes the ability to grow uniform plants of specified size. Quality control by well experienced nursery personnel is essential to achieve the expected plant quality with automated operations. For

Figure 1. A sorting machine that creates uniform stands of seedlings in each tray. (*Photo by Chieri Kubota*)

this reason, most automated grafting nursery operations have at least one or more quality control personnel who examine the quality of each graft. A unique effort being developed recently is the combination of automated grafting with a highly controlled environment for growing scion and rootstock seedlings (Zhao and Kubota, 2015). Indoor transplant production technologies with vertically stacked multi-tiered growing systems and electric lighting such as LEDs were developed in the late 1990s (Kozai et al., 2000; Ohyama et al., 2000) and recently introduced to the U.S. (Hortalizas, 2015) to produce high quality uniform transplants with efficient resource use. Because of the high seedling density that can be produced in indoor growing systems, the increased production cost due to the electricity necessary for lighting and cooling was reportedly only 1.2 U.S. cents per plant as compared to traditional greenhouse nursery seedling production (Kozai, 2016). Indoor transplant production technology can fulfill the need for highly uniform scion and root-

Table 1. Grafting automation available in North America.

Model, make	Type	Usage[1] as of July, 2016
Sorting (grading) machines		
ISO Grade 7000, ISO Group, The Netherlands (http://www.isogroepmachinebouw.nl)	Fully automated grading machine that sorts young tomato seedlings into 3 different sizes with preset spacing between plants. Hourly operation capacity is 7,000 plants.	One machine used in North America; 9 machines used in other regions (Europe and Asia)
Select-O-Mat series, Visser Horti Systems (https://www.visser.eu)	Fully automated grading machine that sorts young tomato seedlings by size. Hourly operation capacity is 4,000-8,000 plants, depending on the model.	Three machines used commercially in North America; 9 machines used in other regions (Europe etc.)
Grafting machines		
ISO Graft series, ISO Group, The Netherlands (http://www.isogroepmachinebouw.nl)	Semi-automated grafting machines for tomato with 1-2 workers assisting each machine, depending on the model. Speed is adjustable and up to 1,050 grafts per hour.	Six machines used in North America; 14 machines used in other regions (Europe and Asia)
GR-series, Helper Robotech, Korea (http://www.helpersys.co.kr/)	Semi-automated grafting machines compatible with both tomato and cucurbit seedlings; 2 workers needed per machine to feed scion and rootstock seedlings. Speed with 2 workers with good coordination is up to 800 grafts per hour, but average is 625 grafts per hour.	Ten machines used in North America; 73 used in other regions (Europe, Asia, and Middle East)
EMP-300, Conic System, Spain (http://www.conic-system.com)	Semi-automated grafting machine for tomato or other solanaceous plants. One machine assists each worker to achieve 400-600 grafts per hour.	Four machines used in North America; 14 used in other regions (Europe, Asia, and Middle East)

[1] Each manufacturer provided information for their machines.

stock seedlings consistently throughout the year, which is needed for automated grafting (Zhao and Kubota, 2015).

These technologies enable nursery operations to address the increasing difficulty of securing the needed number of grafting workers. However, while both technologies reduce labor costs and loss of plants, they both require large capital investments.

Synergistic automation—New concept sought for future nursery automation

Increasing uncertainty regarding agricultural labor availability during the production peak has created a pressing need to develop sustainable solutions. Automation and mechanization are traditional solutions to increase production efficiency for industries where large numbers of workers are needed. One common approach has been to develop reliable automation that can conduct routine tasks currently done by humans. This is by far the exact approach that most efforts towards grafting automation took.

An alternative approach while providing more skilled job opportunities is to develop tools, logistics and automation that can significantly increase worker efficiency. The key concept of this alternative approach is to assist humans instead of replacing them. For example, a machine developed for tomato grafting in Spain (Conic System, Table 1) is designed to make unskilled workers achieve the grafting speed of skilled workers.

A similar type of approach is being pursued for grafting cucurbit plants as the grafting methods most commonly used for these crops is generally more complex and therefore slower than methods used for tomato grafting. Another approach may be to develop a tool or an expert system to enhance the work efficiency of a team of many workers, rather than of each individual. Computational optimization may assist managers of a grafting department to develop a weekly plan of efficient use of machines as well as task distribution among available workers according to their skill levels. A team of scientists and engineers are currently working in the area of expert system development at the University of Arizona.

Figure 2. A grafting operation in Mexico with semi-automated grafting machines. (*Photo by Helper Robotech, Korea*)

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Chapter 5 **October 2017**

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Synopsis:
As grafting is primarily an assembly operation, logistics and workflow in the context of labor management are critical considerations for commercial grafting nurseries. These operations need to be optimized to reduce time spent on each step while assuring quality of the end product.

Editors:
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Carol Miles (Washington State University)
Xin Zhao (University of Florida)

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USDA United States Department of Agriculture National Institute of Food and Agriculture

Designing Logistics & Workflow of Grafting Nurseries

General information

The grafting process typically involves multiple steps (e.g., cutting scion and rootstock plants, placing a grafting clip on rootstock, etc.) as described in other chapters of this grafting manual. As grafting is primarily an assembly operation, logistics and workflow in the context of labor management are critical considerations for commercial grafting nurseries. Logistics and workflow need to be optimized in order to reduce the time spent for individual grafting operational steps (cutting, placing clips, etc.), while assuring the quality of the end product, grafted plants. Labor is the highest cost constituent in vegetable grafting (e.g., Lewis et al., 2014), and worker performance throughout the whole workflow of the grafting department is a critical control point affecting the costs and quality of the end product. The author visited several commercial grafting operations where grafting was done manually, and following is a summary of typical successful workflows observed in these facilities along with suggested approaches to further improve the efficiency in a given workflow design. This chapter focuses on manual grafting operation design; please refer to the 'Automation' chapter for grafting with machines or robots.

Workflow designs in grafting operations

There are two basic workflow designs employed in manual grafting operations. Each design has pros and cons, and propagation nurseries must select a workflow design that works best for their labor and production conditions.

1.Cellular manufacturing

This workflow design is also known as 'one-person assembly' where each worker performs every step/task of the grafting process. Workers typically sit in front of a work table where trays of scion and rootstock seedlings as well as grafting tools (knives, cutting boards, a spray bottle of disinfestation solution, etc.) are placed (Fig. 1). A large table could be shared by many workers, or multiple smaller tables shared by a few workers can be used. In either case, each worker needs to have a complete set of tools. This is the most common workflow design observed, and is advantageous for tracing individual worker performance and work quality (e.g., success rate, disease introduction).

Large-scale grafting operations often have a separate supervisory crew, who watch individual's performance and respond to their needs such as providing new trays and replenishing consumable materials as needed so that the grafting worker's work performance is not interrupted. Use of a conveyer system to move the trays in or out of individual workspaces is effective to improve work efficiency.

2. Assembly line

This workflow design is also known as 'line operation' where each worker is assigned to perform one or a few specific tasks of the grafting process (Fig. 2). In the workflow design shown in Fig. 2, rootstock preparation consists of rearranging the plants, cutting the stem at a consistent angle (e.g., 45 degree), and placing a grafting tube (clip) on the cut end of each rootstock plant. The scion preparation process consists of cutting shoots under the cotyledons at the same angle. Excised scion cuttings and trays of rootstock plants are brought to the final stage of assembling, where workers are picking up a scion cutting and inserting it into the tube to join these two plants. An additional worker is assigned to move the finished trays with grafted plants to healing chambers.

This workflow design is more suitable for grafting operations where the order lot size is relatively large, or when a fewer number of scion-rootstock cultivar combinations are used. When the lot size is small and consists of several different scion and/or rootstock graft combinations, there tends to be idle time between switching from one order to the next, which can slow down the whole team's grafting efficiency. This is the reason this workflow design is not used in Asian countries where lot size is typically very small (e.g., sometimes only a few trays) and these propagation nurseries tend to graft a wide range of scion and rootstock combinations.

This assembly line operation is effectively used in Canada where a relatively limited number of scion and rootstock combinations are used. One advantage of this type of workflow is that it allows a worker to be assigned tasks based on his/her skill set, and may be suitable in an operation where the majority of workers are new and un-

Figure 1. An example of a cellular workflow design in a grafting nursery where each worker performs all the steps of "assembling" grafted plants.

trained.

Cutting scion and rootstock seedlings in a consistent sharp angle typically requires some training, while placing grafting tubes on the cut end of rootstock plants is an easy task for beginners. Placing an experienced worker as inspector at the end of the workflow can enhance the quality of grafted plants. Another inspector or an experienced worker in the assembly line can observe the performance of other workers and change work-assignment as needed to improve the whole grafting assembly line efficiency.

Some considerations in grafting workflow design

Horticultural operations consist of individual processes highly dependent on each other. Improving grafting efficiency in a given workflow design often requires optimizing ergonomic time-motion at the individual worker's level. Additionally, the selection of a grafting method that is best suited for a particular grafting propagation nursery should be evaluated in the context of the whole nursery operation. The following are two examples of how to select a grafting method and a workflow design for a grafting propagation nursery.

1. Grafting plants in or out of the tray

Fig. 3 shows two nurseries grafting tomato plants using the splice (tube) grafting method. However, the first nursery (Fig. 3-left) performs grafting by removing the plants from the tray and lining them up to assure an accurate angle cut for rootstock and scion plants. Note that plants are lined up on a cutting board with X-Y grids to assist workers to make consistent, accurate cut angles.

This process might achieve the highest quality of grafted plants by assuring the quality of the cut (correct angle and clean sharp cut) for each plant. However, the extra steps of pulling plants from the tray, laying on the cutting board, and placing the grafted plants back in the tray add time to the grafting process and increase the labor cost per plant. Workers can pull one whole row of plants (8 plants in this case) at a time to minimize the additional motion-time needed in the process, but overall grafting speed still is slower than when plants are not removed from the tray.

Another notable issue is that the work area tends to become messy with substrate materials, and workers need to wipe or clean the work surface regularly, an additional step in the grafting process. In contrast, grafting

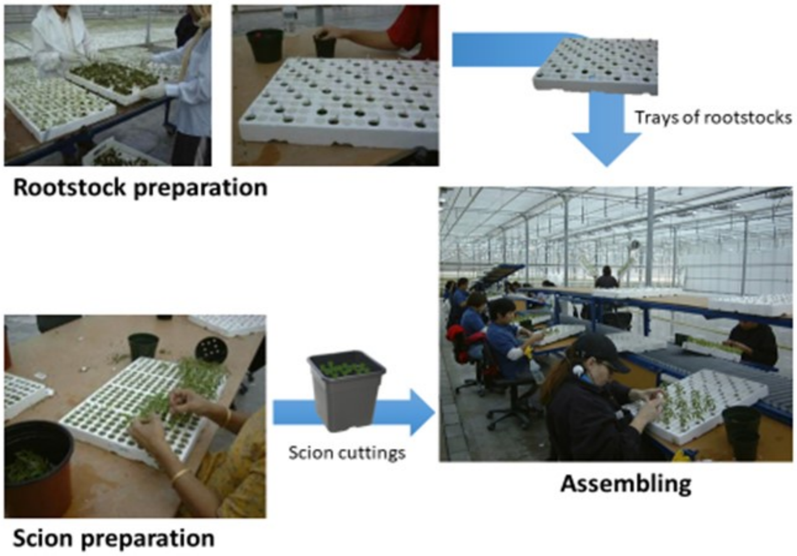

Figure 2. An example of an assembly line workflow design, where rootstock and scion plants are prepared separately by different sets of workers.

Figure 3. Examples of two different cutting methods: pulling out plants and cutting them on a cutting board assures the highest quality and consistency of the cut (left), while cutting plants in the tray reduces the time needed to process scion and rootstock plants for grafting.

plants in the tray is faster because it minimizes the time spent on each grafted plant (Fig. 3-right and Fig. 4). Nurseries that graft plants in the tray typically grow rootstock plants at lower density so that there is ample space around each plant to work. This will increase the costs of producing rootstocks and limit the production capacity, as more space is required for growing the same number of plants at lower densities. Some growers re-space and sort rootstock plants prior to grafting to address the space issue, despite the increase of additional labor. Cutting plants at a consistent angle in the tray without removing the plants requires training, and a simple tool to assist workers to make consistent angle cuts is useful (Fig. 4).

2. Timing to remove rootstock meristems

In cucurbit grafting, one process critical for producing high quality grafted plants is removal of cotyledonary axillary meristems/buds from rootstock seedlings. The cotyledonary axillary buds of most rootstocks used for cucurbit are not visible at the time of grafting but these buds become problematic as they grow after grafting.

There is a need to develop effective methods to completely remove the axillary meristematic tissue from cucurbit rootstocks (Daley et al., 2014; Dabirian and Miles, 2017). One process that is commonly used to prevent rootstock grow-out is to remove the meristematic layer of

Figure 4. A grafting cutter is designed to cut plants in the tray at a consistent angle.

tissue in the axil region using a sharp scalpel or razor blade (Table 1).

This process can be difficult for workers with limited experience. While complete removal of meristematic tissue during grafting will eliminate the grow-out of rootstock axillary shoots, this additional step slows down the grafting speed and increases the grafting costs.

As another approach, in commercial nurseries in China and Taiwan, grafters intentionally leave the invisible axillary buds. These buds grow out when plants are going through the healing and greenhouse finishing processes, and workers remove the extended axillary shoots in the greenhouse (sometimes more than once). At this growth stage, shoots are extended, visible, and easily snapped off. This approach is perhaps more advantageous than trying to remove invisible meristematic tissue with a scalpel or razor blade at the time of grafting, and is a good example of a whole system approach to improve overall efficiency and thereby reduce the overall costs.

Integration of information technology—Smart grafting operation

Workflow design is typically created by trial and error processes driven by an experienced production manager. Successful operation of grafting relies on the skills and experience-based wisdom of such personnel. Recent developments in information technology such as real time data collection for optimization should assist personnel to make decisions regarding labor management and assignment of workers with various skill levels to selected tasks to meet production timelines and demand patterns. In the U.S., while migrant temporary workers are the key workforce during peak production seasons of agricultural and horticultural operations, the costs and regulatory issues relating to migrant workers can be problematic for propagation nurseries. Utilizing computer algorithms to optimize the production process, logistics, and task assignment among workers with different skill sets and wage scales could provide helpful solutions in future horticultural operations as demonstrated by Masoud et al. (2017) and Kubota et al. (2017).

Axil removal methods	Axillary bud extension (%) after 7 days	Note
Non-treated control	92%	
Three light scratches with scalpel over the axil region	25%	The incidence can be further reduced by increasing extent of scratching depth.
Cutting off a cotyledon deep enough to completely remove the remaining axil region	0%	Too deep of cut could damage rootstock plants.

Table 1. Incidence of ‘Strong Tosa’ interspecific hybrid rootstock axillary bud extension (%) as affected by the method of removing axillary buds at the time of grafting (preliminary unpublished data obtained at the University of Arizona; n=12 per treatment.

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Grafting Manual:
How to Produce Grafted Vegetable Plants

www.vegetablegrafting.org

Chapter 5 **October 2017**

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Synopsis:
As grafting is primarily an assembly operation, logistics and workflow in the context of labor management are critical considerations for commercial grafting nurseries. These operations need to be optimized to reduce time spent on each step while assuring quality of the end product.

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USDA United States Department of Agriculture National Institute of Food and Agriculture

Designing Logistics & Workflow of Grafting Nurseries

General information

The grafting process typically involves multiple steps (e.g., cutting scion and rootstock plants, placing a grafting clip on rootstock, etc.) as described in other chapters of this grafting manual. As grafting is primarily an assembly operation, logistics and workflow in the context of labor management are critical considerations for commercial grafting nurseries. Logistics and workflow need to be optimized in order to reduce the time spent for individual grafting operational steps (cutting, placing clips, etc.), while assuring the quality of the end product, grafted plants. Labor is the highest cost constituent in vegetable grafting (e.g., Lewis et al., 2014), and worker performance throughout the whole workflow of the grafting department is a critical control point affecting the costs and quality of the end product. The author visited several commercial grafting operations where grafting was done manually, and following is a summary of typical successful workflows observed in these facilities along with suggested approaches to further improve the efficiency in a given workflow design. This chapter focuses on manual grafting operation design; please refer to the 'Automation' chapter for grafting with machines or robots.

Workflow designs in grafting operations

There are two basic workflow designs employed in manual grafting operations. Each design has pros and cons, and propagation nurseries must select a workflow design that works best for their labor and production conditions.

1.Cellular manufacturing

This workflow design is also known as 'one-person assembly' where each worker performs every step/task of the grafting process. Workers typically sit in front of a work table where trays of scion and rootstock seedlings as well as grafting tools (knives, cutting boards, a spray bottle of disinfestation solution, etc.) are placed (Fig. 1). A large table could be shared by many workers, or multiple smaller tables shared by a few workers can be used. In either case, each worker needs to have a complete set of tools. This is the most common workflow design observed, and is advantageous for tracing individual worker performance and work quality (e.g., success rate, disease introduction).

Large-scale grafting operations often have a separate supervisory crew, who watch individual's performance and respond to their needs such as providing new trays and replenishing consumable materials as needed so that the grafting worker's work performance is not interrupted. Use of a conveyer system to move the trays in or out of individual workspaces is effective to improve work efficiency.

2. Assembly line

This workflow design is also known as 'line operation' where each worker is assigned to perform one or a few specific tasks of the grafting process (Fig. 2). In the workflow design shown in Fig. 2, rootstock preparation consists of rearranging the plants, cutting the stem at a consistent angle (e.g., 45 degree), and placing a grafting tube (clip) on the cut end of each rootstock plant. The scion preparation process consists of cutting shoots under the cotyledons at the same angle. Excised scion cuttings and trays of rootstock plants are brought to the final stage of assembling, where workers are picking up a scion cutting and inserting it into the tube to join these two plants. An additional worker is assigned to move the finished trays with grafted plants to healing chambers.

This workflow design is more suitable for grafting operations where the order lot size is relatively large, or when a fewer number of scion-rootstock cultivar combinations are used. When the lot size is small and consists of several different scion and/or rootstock graft combinations, there tends to be idle time between switching from one order to the next, which can slow down the whole team's grafting efficiency. This is the reason this workflow design is not used in Asian countries where lot size is typically very small (e.g., sometimes only a few trays) and these propagation nurseries tend to graft a wide range of scion and rootstock combinations.

This assembly line operation is effectively used in Canada where a relatively limited number of scion and rootstock combinations are used. One advantage of this type of workflow is that it allows a worker to be assigned tasks based on his/her skill set, and may be suitable in an operation where the majority of workers are new and un-

Figure 1. An example of a cellular workflow design in a grafting nursery where each worker performs all the steps of "assembling" grafted plants.

trained.

Cutting scion and rootstock seedlings in a consistent sharp angle typically requires some training, while placing grafting tubes on the cut end of rootstock plants is an easy task for beginners. Placing an experienced worker as inspector at the end of the workflow can enhance the quality of grafted plants. Another inspector or an experienced worker in the assembly line can observe the performance of other workers and change work-assignment as needed to improve the whole grafting assembly line efficiency.

Some considerations in grafting workflow design

Horticultural operations consist of individual processes highly dependent on each other. Improving grafting efficiency in a given workflow design often requires optimizing ergonomic time-motion at the individual worker's level. Additionally, the selection of a grafting method that is best suited for a particular grafting propagation nursery should be evaluated in the context of the whole nursery operation. The following are two examples of how to select a grafting method and a workflow design for a grafting propagation nursery.

1. Grafting plants in or out of the tray

Fig. 3 shows two nurseries grafting tomato plants using the splice (tube) grafting method. However, the first nursery (Fig. 3-left) performs grafting by removing the plants from the tray and lining them up to assure an accurate angle cut for rootstock and scion plants. Note that plants are lined up on a cutting board with X-Y grids to assist workers to make consistent, accurate cut angles.

This process might achieve the highest quality of grafted plants by assuring the quality of the cut (correct angle and clean sharp cut) for each plant. However, the extra steps of pulling plants from the tray, laying on the cutting board, and placing the grafted plants back in the tray add time to the grafting process and increase the labor cost per plant. Workers can pull one whole row of plants (8 plants in this case) at a time to minimize the additional motion-time needed in the process, but overall grafting speed still is slower than when plants are not removed from the tray.
Another notable issue is that the work area tends to become messy with substrate materials, and workers need to wipe or clean the work surface regularly, an additional step in the grafting process. In contrast, grafting

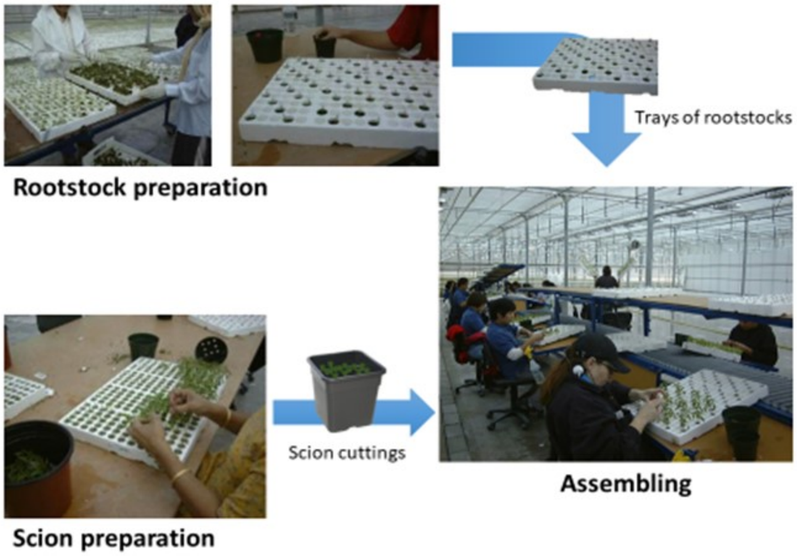

Figure 2. An example of an assembly line workflow design, where rootstock and scion plants are prepared separately by different sets of workers.

Figure 3. Examples of two different cutting methods: pulling out plants and cutting them on a cutting board assures the highest quality and consistency of the cut (left), while cutting plants in the tray reduces the time needed to process scion and rootstock plants for grafting.

plants in the tray is faster because it minimizes the time spent on each grafted plant (Fig. 3-right and Fig. 4). Nurseries that graft plants in the tray typically grow rootstock plants at lower density so that there is ample space around each plant to work. This will increase the costs of producing rootstocks and limit the production capacity, as more space is required for growing the same number of plants at lower densities. Some growers re-space and sort rootstock plants prior to grafting to address the space issue, despite the increase of additional labor. Cutting plants at a consistent angle in the tray without removing the plants requires training, and a simple tool to assist workers to make consistent angle cuts is useful (Fig. 4).

2. Timing to remove rootstock meristems

In cucurbit grafting, one process critical for producing high quality grafted plants is removal of cotyledonary axillary meristems/buds from rootstock seedlings. The cotyledonary axillary buds of most rootstocks used for cucurbit are not visible at the time of grafting but these buds become problematic as they grow after grafting.

There is a need to develop effective methods to completely remove the axillary meristematic tissue from cucurbit rootstocks (Daley et al., 2014; Dabirian and Miles, 2017). One process that is commonly used to prevent rootstock grow-out is to remove the meristematic layer of

Figure 4. A grafting cutter is designed to cut plants in the tray at a consistent angle.

tissue in the axil region using a sharp scalpel or razor blade (Table 1).

This process can be difficult for workers with limited experience. While complete removal of meristematic tissue during grafting will eliminate the grow-out of rootstock axillary shoots, this additional step slows down the grafting speed and increases the grafting costs.

As another approach, in commercial nurseries in China and Taiwan, grafters intentionally leave the invisible axillary buds. These buds grow out when plants are going through the healing and greenhouse finishing processes, and workers remove the extended axillary shoots in the greenhouse (sometimes more than once). At this growth stage, shoots are extended, visible, and easily snapped off. This approach is perhaps more advantageous than trying to remove invisible meristematic tissue with a scalpel or razor blade at the time of grafting, and is a good example of a whole system approach to improve overall efficiency and thereby reduce the overall costs.

Integration of information technology—Smart grafting operation

Workflow design is typically created by trial and error processes driven by an experienced production manager. Successful operation of grafting relies on the skills and experience-based wisdom of such personnel. Recent developments in information technology such as real time data collection for optimization should assist personnel to make decisions regarding labor management and assignment of workers with various skill levels to selected tasks to meet production timelines and demand patterns. In the U.S., while migrant temporary workers are the key workforce during peak production seasons of agricultural and horticultural operations, the costs and regulatory issues relating to migrant workers can be problematic for propagation nurseries. Utilizing computer algorithms to optimize the production process, logistics, and task assignment among workers with different skill sets and wage scales could provide helpful solutions in future horticultural operations as demonstrated by Masoud et al. (2017) and Kubota et al. (2017).

Axil removal methods	Axillary bud extension (%) after 7 days	Note
Non-treated control	92%	
Three light scratches with scalpel over the axil region	25%	The incidence can be further reduced by increasing extent of scratching depth.
Cutting off a cotyledon deep enough to completely remove the remaining axil region	0%	Too deep of cut could damage rootstock plants.

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