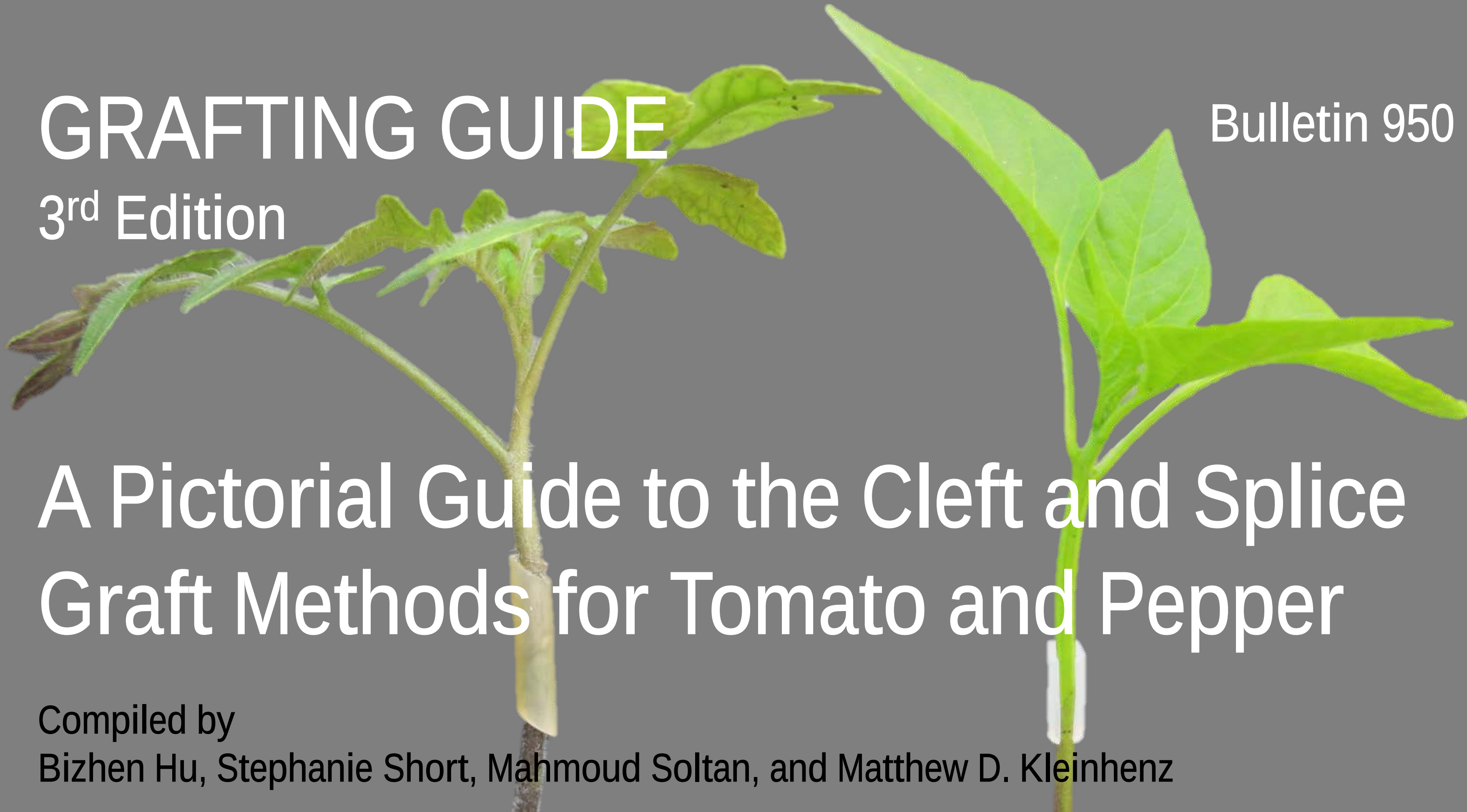

GRAFTING GUIDE

3rd Edition

Bulletin 950

A Pictorial Guide to the Cleft and Splice Graft Methods for Tomato and Pepper

Compiled by
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THE OHIO STATE UNIVERSITY
COLLEGE OF FOOD, AGRICULTURAL, AND ENVIRONMENTAL SCIENCES

OHIO STATE UNIVERSITY EXTENSION
OHIO AGRICULTURAL RESEARCH AND DEVELOPMENT CENTER
UNITED STATES DEPARTMENT OF AGRICULTURE

Acknowledgments

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The substantial technical contributions of Ken Chamberlain, Aaron Cochran, Dr. Sally Miller, Jennifer Moyseenko, M.S., Sonia Walker throughout all phases of the development of this document are appreciated.

About the cover. Grafted tomato (left) and pepper (right) plants showing a portion of the scion section and the use of a homemade latex (tomato, left) and manufactured silicon (pepper, right) clip to secure the rootstock-scion union while it heals (i.e., while root and stem sections unite to re-form a fully functional root-shoot axis).

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Introduction and Overview

Parts of a Grafted Plant and Benefits of Using Grafted Plants in Fruit Production

Grafting combines two or more seedlings into one plant, creating a type of physical hybrid in the process. The simplest and most common type of grafted plant contains two varieties (cultivars, genotypes). The top (fruiting) variety is the scion. The bottom variety, used for its roots, is the rootstock. Rootstock varieties typically DO NOT produce marketable fruit. However, their root systems are superior to those of the scion in at least one way that makes creating and using grafted plants more appealing than using ungrafted scion plants. Grafted vegetable plants have long been used to limit the effects of soil pathogens (e.g., Rivard and Louws, 2008) on cucurbit and solanaceous crops and to enhance plant vigor. However, new reasons to consider using grafted plants continue to be reported.

Compared to ungrafted plants of the same scion variety, research has shown that grafted plants:
1) may display greater tolerance to low and high temperatures (Rivero et al., 2003; Venema et al., 2008) and saline or flooded soils (Colla et al., 2010a; He et al., 2009; Martinez-Rodriguez et al., 2008; Yetisir et al., 2006);
2) may enhance crop nutrient and water uptake efficiency during production (Colla et al., 2010b; Djidonou et al., 2013, 2015; Rouphael et al., 2008a; Ruiz et al., 1997);
3) may limit the uptake of organic pollutants from soils (Otani and Seike, 2006; Otani and Seike, 2007); and
4) may show fewer negative effects after being exposed to excessive levels of boron, copper, cadmium and manganese (Edelstein et al., 2005; Rouphael et al., 2008b; Savvas et al., 2009).

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Most research also suggests that the characteristics of tomato fruit are largely unchanged by grafting. This allows growers to employ grafted plants with few concerns about buyers' potential reactions. However, given the nature of some varieties (especially rootstocks) and the potential to create many rootstock-scion variety combinations, growers looking to use grafted tomato plants should verify that the characteristics of scion fruit they harvest will not be changed by grafting.

Challenges of Making Grafted Plants and Using them in Fruit Production

The previous page outlined some of the recognized benefits of using grafted tomato and pepper plants. It is also important to be aware of its possible drawbacks, beginning with the making of grafted plants. For example, the grafting process can spread disease very effectively. So, proper techniques, and clean stock, spaces, and materials are required. Also, grafting requires growing two seedlings to make each grafted plant, meaning more greenhouse space and other materials are required. In addition, the price of rootstock seed can be high. Therefore, the costs of each plant used in production and the resources required to prepare them are greater than when standard seedlings are used.

Similarly, given their genetics, grafted plants tend to respond to growing conditions differently than standard transplants. Grafted plants may benefit from wider spacing, more fertilizer and water, different pruning and trellising systems, and other management. Conversely, they may tolerate drier and less fertile soils better than standard plants. Regardless, the pros and cons of making and using grafted plants should be considered carefully before proceeding and the proper materials and methods should be used when grafting.

Introduction and Overview

This guide is designed to provide comprehensive information on tomato and pepper grafting techniques, focusing on the ones used to graft by hand. Principles and approaches described in the guide apply to many operations, regardless of size (i.e., the goal/need to produce a few grafted plants or many thousands). The guide contains information on grafting economic feasibility (p. 7), rootstock and scion variety selection (pp. 12-24), grafting methods (pp. 54-85), grafting tool (pp.36-40) and space preparation before grafting (pp. 30-31), seedling production before grafting (pp. 33-34), sanitation during grafting (pp. 51-53), step by step grafting procedures (pp. 55, 66, 72), and grafted plant management after grafting (pp. 79-85).

This guide focuses on principles and proven approaches. However, there are few absolute truths in vegetable grafting. Some are included in the guide but ongoing research and trial and error continue to improve locally-relevant techniques, tools, and knowledge. For example, most of the images included in the guide were obtained in an environmentally controlled greenhouse at The Ohio Agricultural Research and Development Center (Wooster, Ohio) of The Ohio State University. Although the same general principles apply everywhere, spaces, materials, and techniques can be adjusted according to scale of operation, budget, local environmental conditions, and available resources.

Introduction and Overview

Economic Analysis and On-Farm Testing

Different models have been used to analyze the economic feasibility of producing grafted plants as a business and of using them in commercial fruit production. Models and experience agree on two points: (1) that producing grafted plants is more costly than producing standard seedlings (transplants) (e.g., Rivard et al., 2010), and (2) that increases in production costs depend on the number of plants produced (e.g., Lewis et al., 2014). In Rivard et. al. (2010), producing grafted plants increased total transplant production costs 64% to 354%.

Most agree that the costs of producing grafted plants can be estimated more reliably than the economic benefits of using grafted plants in commercial fruit production. Advantages that superior genetics can provide to a farm, including advantages given by grafted plants, are difficult to quantify. Although benefits are real, they must be determined one farm, one season, one crop and, maybe, one field at a time. In an experiment focused on organic heirloom tomato production (Barrett et al., 2012), economic risk of crop loss due to root-knot nematode damage exceeded the 46% higher cost of grafted transplants.

More research and grower testing are needed. Season after season, crop after crop, as growers experiment with grafted plants, they and others will come to understand more completely how grafted plants fit and benefit (if at all) individual farm operations. Consider taking three steps. First, evaluate how grafted plants may benefit your operation. Second, experiment responsibly with grafted plants and, using careful records, evaluate your return on investment in them. Third, use trusted tools, advisors, and local information in steps 1 and 2.

As you plan to make grafted plants, note that your costs will probably be influenced by the following and other factors:

- Location and environmental conditions
- Type of seedling growing facility
- Seed costs
- Labor costs
- Type, availability and cost of grafting materials
- Post-grafting facility and care
- Utility costs
- Automation level (grafting capacity)
- Scale of operation

Introduction and Overview

As you plan to market or use grafted plants, note that some fruit-growing operations do not appear to benefit from using grafted plants. For others, the risk of crop loss due to disease or some other stress makes investing in grafted plants worthwhile. Other growers prefer to use grafted plants because of their increased production potential in good growing conditions.

Currently, people familiar with vegetable grafting agree that grafted plants tend to offer vegetable growers in the U.S. the greatest return on investment when one or more of the following conditions are true.

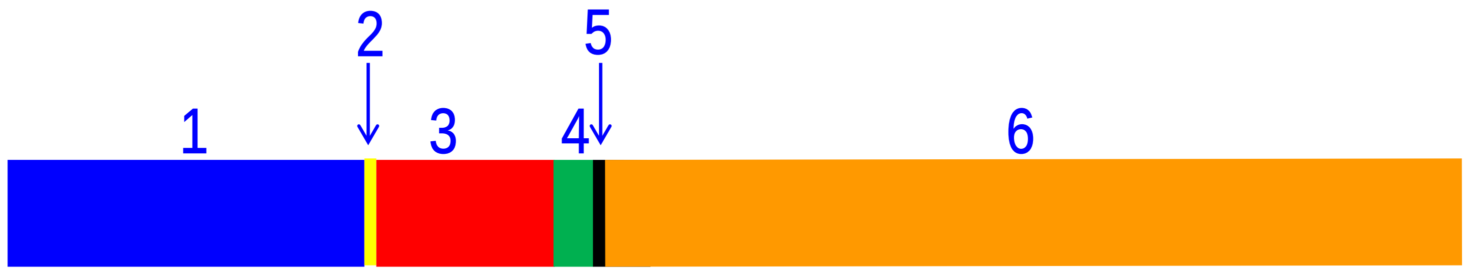
1. Their operation is being hurt or is likely to be hurt by a serious soilborne disease that can be tolerated by at least one rootstock variety.
2. Their preferred scion variety is susceptible to that disease and using another scion variety is unappealing or impossible.
3. The farm has a small land base and few other disease management options will bring the needed level of (economic) control.
4. Crop vigor and yield are very important, especially to enhance or maintain the farm's presence in the market.
5. Single plantings will be picked as many times as conditions allow; in other words, total seasonal yield is important and the grower may be willing to tolerate having a smaller early yield in exchange for a greater total seasonal yield.

Introduction and Overview

Overall, data and grower experience make clear that grafted plants can be made and used and are effective at any scale. Grafted plants can benefit many types of growers, but only when used properly. Obtaining a high return on investment in grafted plants requires knowing that grafting: (a) complicates variety selection, (b) increases transplant costs and may complicate transplant supply chains, (c) raises the potential to transmit disease during transplant production, and (d) raises questions, regardless of grower experience.

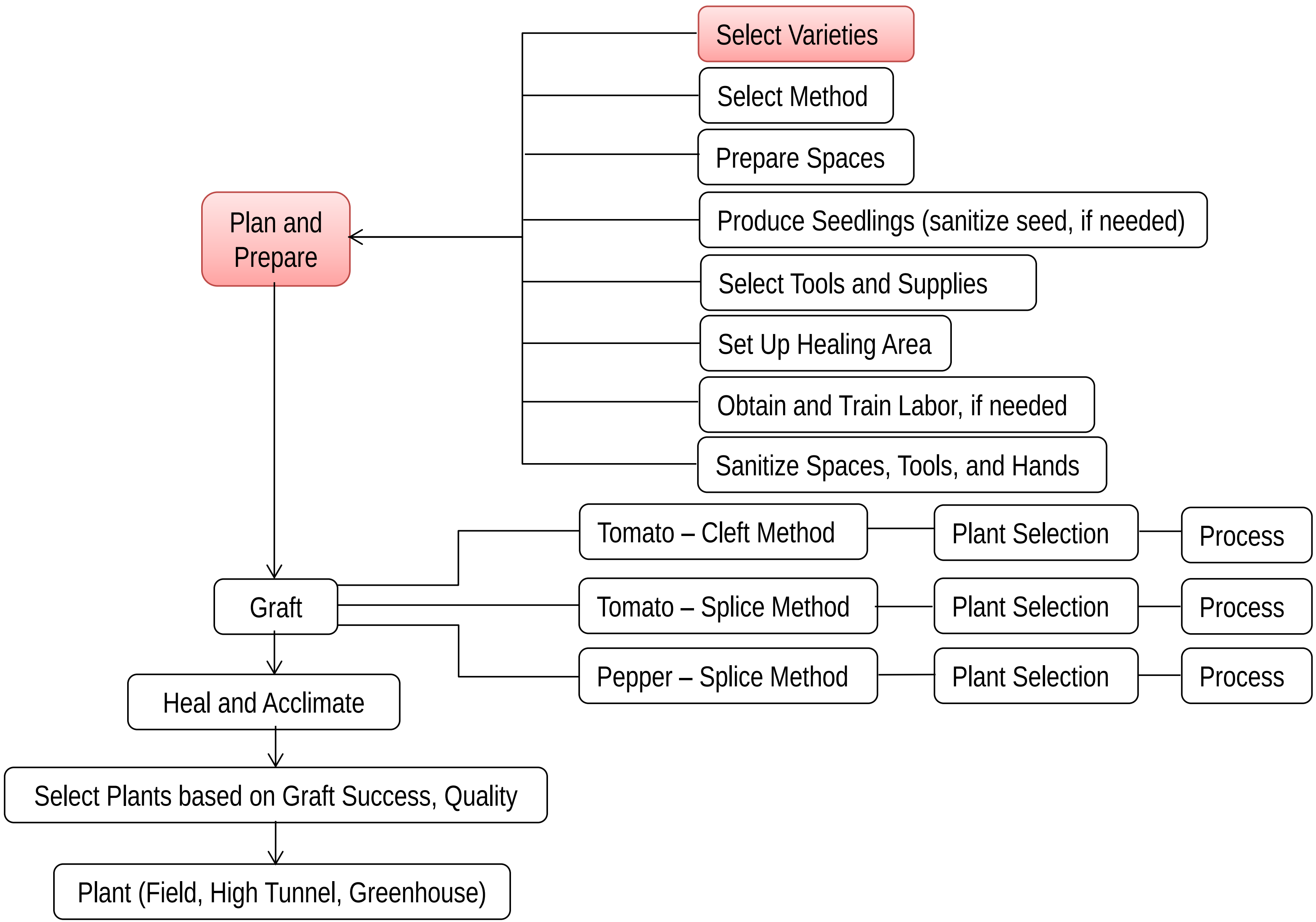
Therefore, it is prudent to consider and test the potential value of grafting to your business. Patience, caution, and careful testing are required to fully understand the influence of grafting on your business.

Approximate Timeline for Production of Grafted Tomato or Pepper Plants from Seeding to First Harvest



1 - grow seedlings (4 weeks)
2 - graft (0.25-1.0 minute)
3 - heal (2 weeks)
4 - ship, if needed (2 days)
5 - transplant (0.25-1.0 minute)
6 - time to first harvest (6-8 weeks)

This guides focuses on stages 1-3 above.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Pepper – Splice Method
Plant Selection
Process
Heal and Acclimate
Select Plants based on Graft Success, Quality
Plant (Field, High Tunnel, Greenhouse)

Rootstock Variety Selection

In the U.S., the number of commercially available tomato and pepper rootstock (RS) varieties has changed each year since 2009. The trend is upward, with more rootstock varieties available in 2016 than in 2009. However, it is important to note that, like scion (SC) varieties, rootstock varieties come and go.

It is also important to recognize that the number of rootstock varieties is increasing faster than most users can become familiar with them. So, while potential users should continue to look to trusted sources for information about rootstock varieties (including Cooperative Extension), users may also need to experiment on their farm. Again, it may be useful to involve Cooperative Extension or other personnel in tests for advice, assistance, etc.

Together, rootstock varieties contain an array of potentially useful traits with the clear majority of traits being tolerant of/resistant to a specific set of nematodes, soilborne diseases, or viruses (see Table 1 — p. 15 and Table 2 — p. 16). Some rootstock varieties are also marketed based on their vigor or tolerance of/resistance to abiotic stresses (e.g., salinity, temperature, flooding). In fact, each rootstock variety has a specific set of traits. In 2016, the 60 tomato rootstock varieties commercially available in the U.S. were listed as having 41 different sets of traits (see Tables 3 and 4 — p. 17).

The changing availability of rootstock varieties, the relative lack of information on most of them, and the different sets of traits they contain makes selecting rootstock varieties difficult for commercial plant suppliers and fruit growers. This is unfortunate because rootstock selection is a key early indicator of the success of the fruit production that will be done with grafted plants. Selecting rootstock varieties takes diligence and, so far, there are few clear favorites. Three suggestions follow.

First, consider your (or your customers') production goals and history, and growing conditions and resources, then select rootstock that fit your (or your customers') needs. Ask these key questions: 1) why do I intend to prepare and use grafted plants?; 2) What benefits will grafted plants offer to me?; 3) How much time, money, effort and space can I devote to preparing and using grafted plants?; and 4) What is my expected return on investment? Grafted plant performance depends on growing conditions and rootstock-scion combinations. In selecting rootstock, do your homework to determine, as reliably as

Rootstock Variety Selection

possible what you may gain from specific rootstock-scion pairings. The most common example of matching rootstock traits with production needs involves selecting specific rootstock to limit the damage ordinarily caused by certain diseases. Rootstock variety tables (http://www.vegetablegrafting.org/rootstock-tables/) list the traits of available rootstock. Refer to the tables to help select one or more rootstock varieties to address a production need. Also, get input from seed suppliers, extension-research and industry personnel, and other growers.

Second, track the success of your grafting operation and the performance of the grafted plants. Again, rootstock numbers and diversity are increasing and the number of scion varieties is large. Therefore, many rootstock-scion combinations are possible. Their performance in grafting operations and on farms is likely to differ. So, having clear and complete records from grafting through harvest can help select varieties going forward. Include ungrafted plants for comparison.

Finally, note that new information helping to optimize the various applications of grafting becomes available each day. Watch for it and experiment in areas that matter to you. For example, grafting shoots of heirloom varieties as scions to roots of standard disease-tolerant/resistant fruiting hybrids as rootstock may benefit some growers. Many heirloom varieties are highly susceptible to various soilborne diseases while standard hybrids may be resistant. Using a standard hybrid as a rootstock may be beneficial. That approach is untested but it may be worth exploring on a limited scale and with caution. Look for descriptions of other applications of grafting.

Table 1. Number of tomato rootstock varieties advertised in 2016 as resistant to the diseases listed.

# of Rootstock Varieties	Bacterial Wilt	Corky Root Rot	Fusarium Wilt Race 1	Fusarium Wilt Race 2	Fusarium Wilt Race 3	Fusarium Crown and Root Rot	Southern Blight	Verticillium Wilt	Nematode	Tomato Mosaic Virus	Tomato Spotted Wilt Virus
1	■	■	■	■		■		■	■	■	■
1	■	■	■	■		■		■	■	■	
3	■	■	■	■		■		■	■	■	
1	■	■	■	■		■		■	■		
1	■	■	■	■					■	■	
1	■	■	■				■	■	■	■	
1	■	■		■		■			■	■	
1	■		■	■	■	■		■	■	■	
1	■		■	■	■	■			■	■	
1	■		■	■	■			■	■		
6	■		■	■		■		■	■	■	
1	■		■	■		■			■	■	
2	■		■	■					■		
1	■		■								
1		■	■	■	■	■				■	
1			■	■	■	■		■	■	■	
1		■	■	■		■		■	■	■	■
7		■	■	■		■		■	■	■	
1		■	■	■		■		■		■	
1		■	■	■		■			■	■	
1		■	■			■		■	■	■	
1		■		■		■			■	■	
1		■						■		■	
3			■	■	■	■		■	■	■	
1			■	■	■	■		■		■	
2			■	■	■	■		■	■	■	
1			■	■	■			■	■	■	
1			■	■		■		■	■	■	■
1			■	■		■		■	■	■	
1			■	■		■		■	■		
1			■	■		■		■		■	
1			■	■		■			■	■	
1			■	■				■	■	■	
2			■	■				■	■		
1			■	■					■		
1											
1			■			■			■		
1				■		■		■	■	■	
2				■		■			■	■	
1										■	
1											
Column sum 60	22	23	51	52	12	42	1	39	48	44	3

■ = full or partial resistance

Table 2. Number of pepper rootstock varieties advertised in 2016 as resistant to the diseases listed.

	# of Rootstock Varieties	Bacterial Wilt	Corky Root Rot	Fusarium Wilt Race 1	Fusarium Wilt Race 2	Fusarium Wilt Race 3	Fusarium Crown and Root Rot	Phytopthora	Potato Y Virus	Verticillium Wilt	Nematode	Tomato Mosaic Virus	Tomato Spotted Wilt Virus
	1												
	1												
	1												
	1												
Column sum	4	1						3	1		3	1	

= full or partial resistance

Table 3. Number of tomato rootstock varieties commercially available in the U.S. by year.

Year	Number of Rootstock Varieties
2009	17
2013	61
2014	72
2015	53
2016	60

Table 4. Number of pepper rootstock varieties commercially available in the U.S. by year.

Year	Number of Rootstock Varieties
2014	7
2015	7
2016	4

Variety	February 27-March 17, 2014	March 28-April 15, 2014
Aiboh	19	15
Akaoni	20	13
Aooni	21	15
Armada	19	13
Arnold	16	13
BB	18	12
Beaufort	17	14
Better Boy*	20	14
Brandywine*	20	13
Celebrity*	19	13
Cheong Gang	20	14
Cherokee Purple*	19	14
Estamino	24	15
Kaiser	20	10
Maxifort	18	12
Resistar	18	13
RST-04-105	23	15
RST-04-106	20	14
San Marzano2*	17	12
Shield	21	14
Stallone	19	13
Supernatural	19	14
Trooper	30	19

Table 5. Estimated number of days from seeding to graft-readiness for tomato varieties.

The growth rates, including the stem diameter, of twenty-three tomato varieties (eighteen rootstock, five scion) were monitored in a greenhouse experiment in Wooster, Ohio. The table at left displays the predicted number of days from sowing to a stem diameter of 1.5 mm for each variety. A stem diameter of 1.5 mm is generally thought to be the minimum required for grafting.

* Scion varieties

Tomato and pepper seedling growth rates, including stem expansion, differ among rootstock and scion varieties.
Rootstock and scion seedlings sown on the same day are likely to differ in stem diameter on any day thereafter. This creates scheduling challenges since successful grafts are produced more efficiently when rootstock and scion seedling stem diameters are similar. Grafting efficiency is also greatest when seedlings are neither too young nor too woody when grafted. So, the challenge is to schedule sowings and manage seedlings so that rootstock and scion seedlings have similar stem diameters when they are the proper age (at the proper stage) to be grafted.

Choosing seeding dates carefully and sowing both varieties multiple times over approximately 1-1.5 weeks can increase the number of well-matched seedlings of the proper age available to graft on any one day. Sowing several times over 7-10 days to reach the total target number of seedlings (instead of one sowing) reduces the number of seedlings with mismatched stem diameters (reducing plant waste) and the effort required by small teams of grafters to keep pace with plant availability. A drawback to this approach is that grafted plants may differ in age when set into the field or high tunnel. Regardless, growers are encouraged to take notes on variety growth rates under their conditions to assist in scheduling sowing and grafting operations in the future.

Differences in the Growth of Tomato Varieties Over Time

Days after Seeding

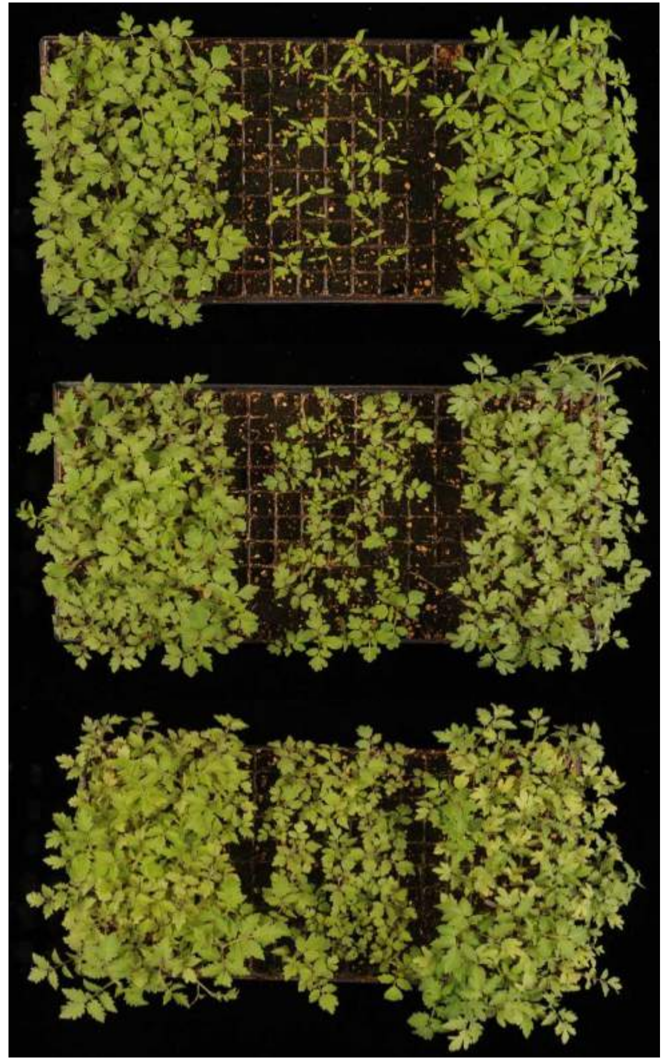
Scion

Rootstock

14

21

28



Variety | Better Boy | San Marzano 2 | Cherokee Purple | Maxifort | Trooper | DP-106

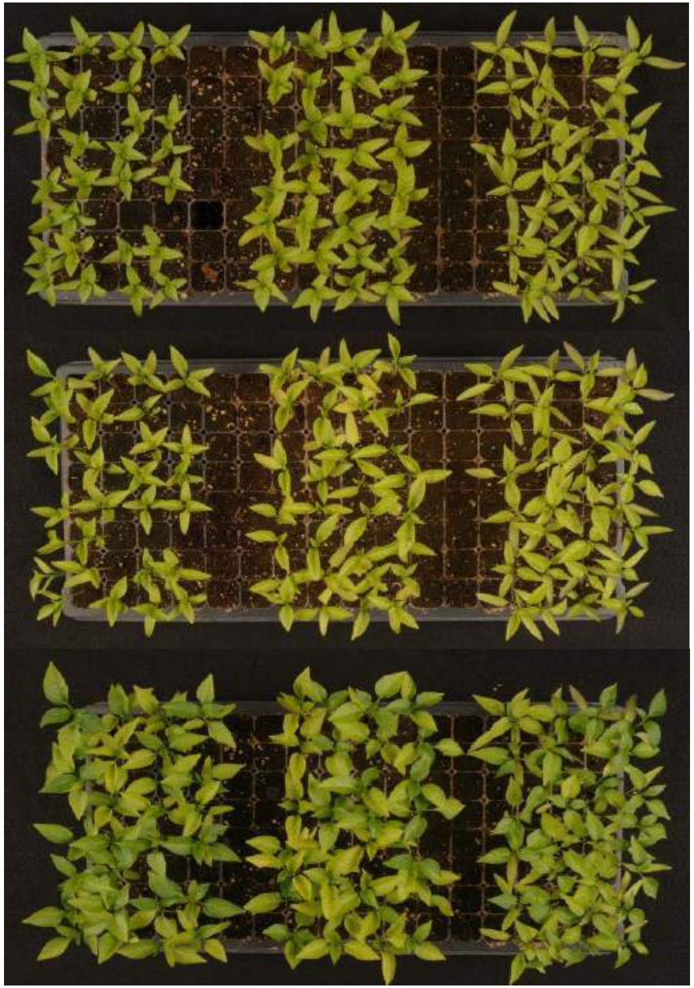
Differences in the Growth of Pepper Scion Varieties Over Time

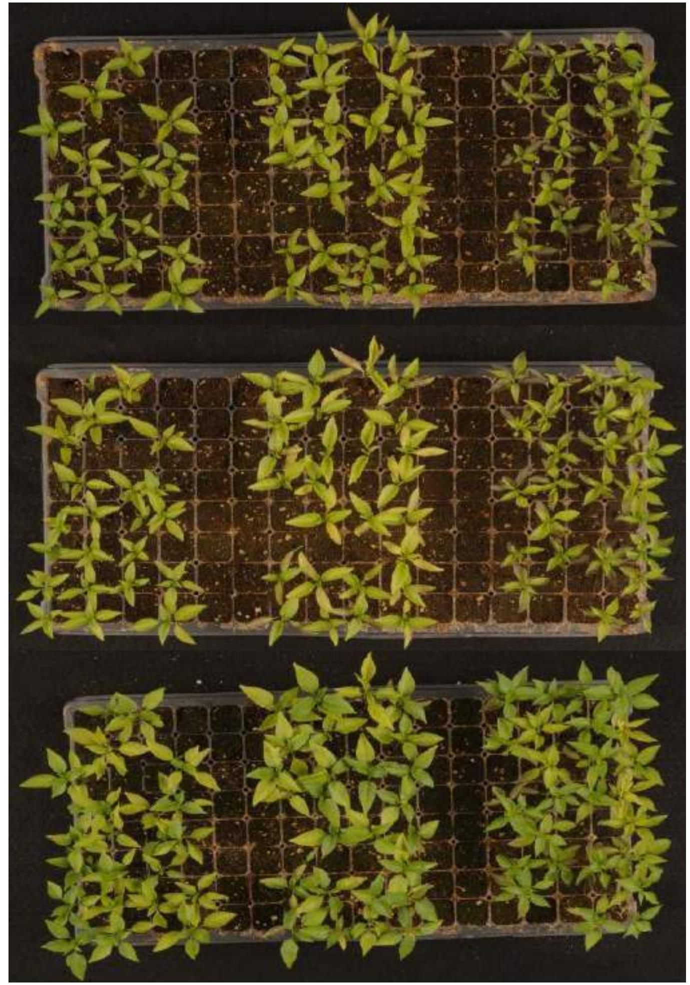
Days after Seeding

26

31

38





Variety	Jalapeño M	Early Sunsation	Aristotle	Thai Hot	Hungarian Hot Wax	Cayenne Large Thick

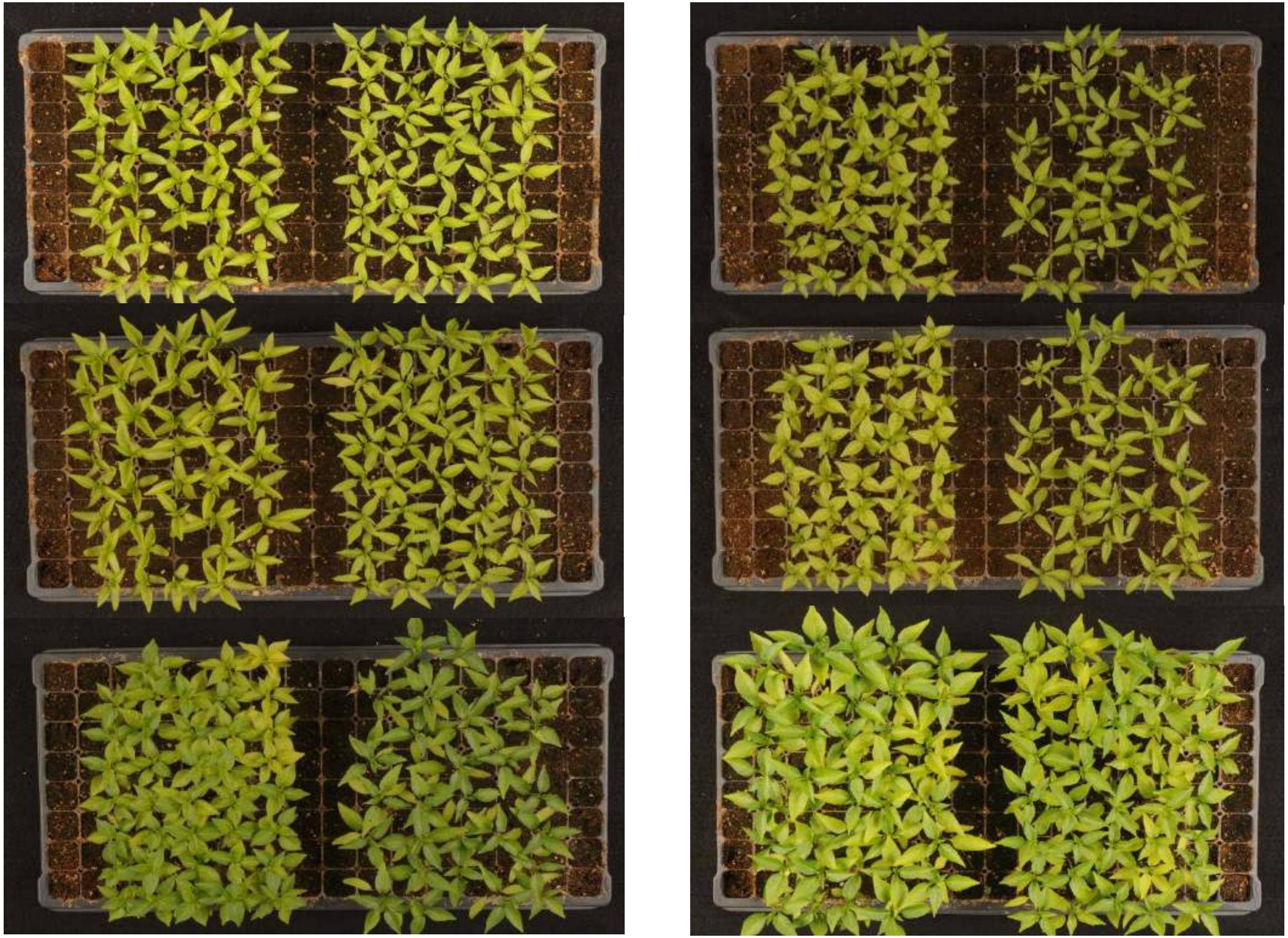
Differences in the Growth of Pepper Rootstock Varieties Over Time

Days after Seeding

26

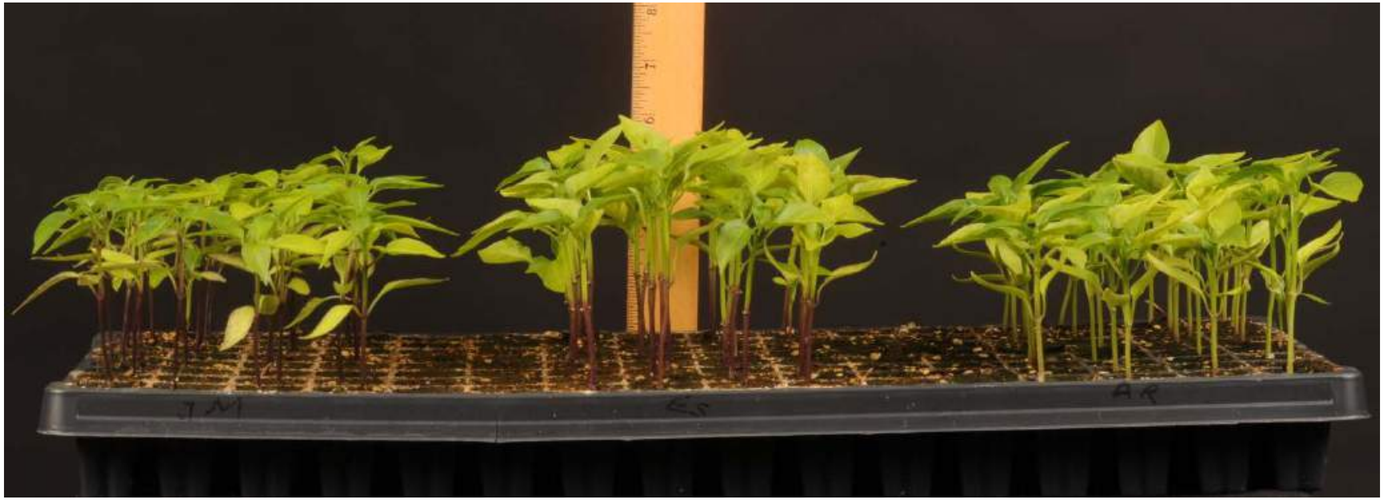
31

38



Variety | Foundation | TI-135 | Scarface | Dorado

Pepper Scion Seedlings at 38 Days after Seeding



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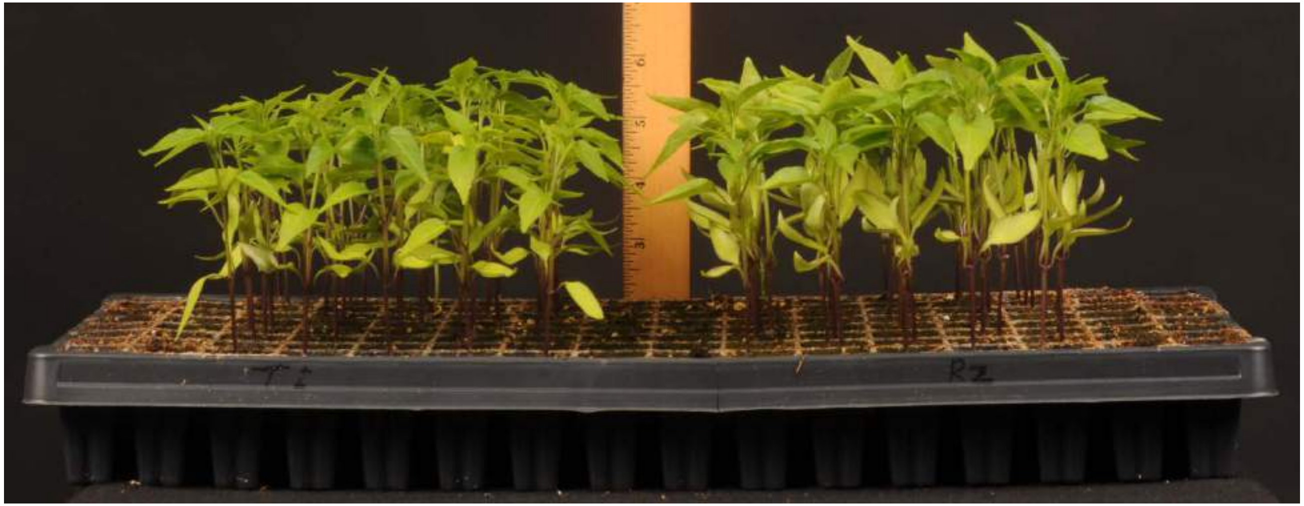
Thai Hot Hungarian Hot Wax Cayenne Long Thick

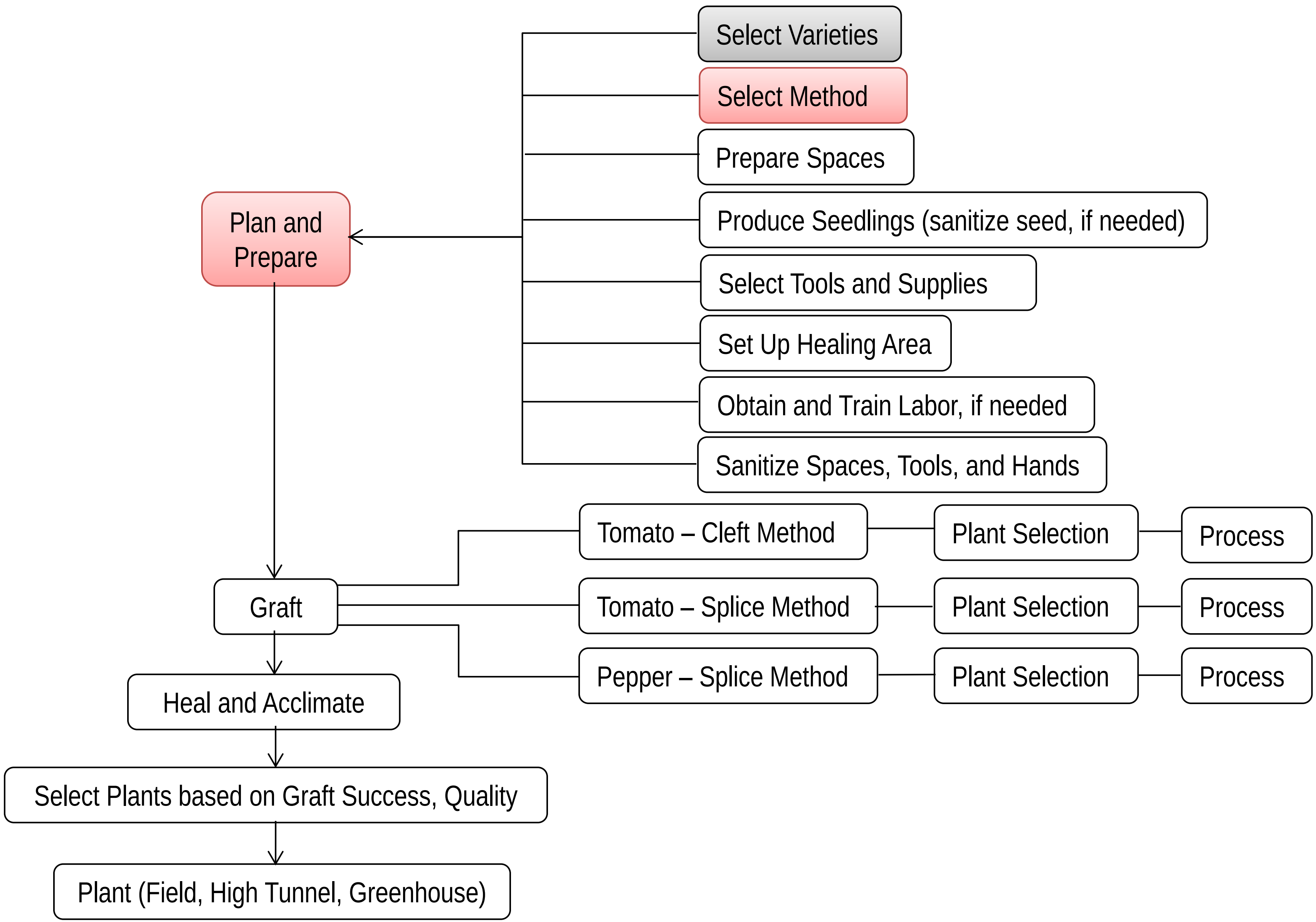
Pepper Rootstock Seedlings at 38 Days after Seeding



TI-135 Foundation

Dorado Scarface

Pepper varieties also differ in overall growth rate, including height and hypocotyl length (i.e., soil line to cotyledons). Hypocotyl length matters since most plants are grafted near the cotyledonary node. Therefore, grafting can be more difficult when rootstock seedlings have short hypocotyls. It may be possible to encourage rootstock hypocotyl lengthening while maintaining seedling quality for grafting.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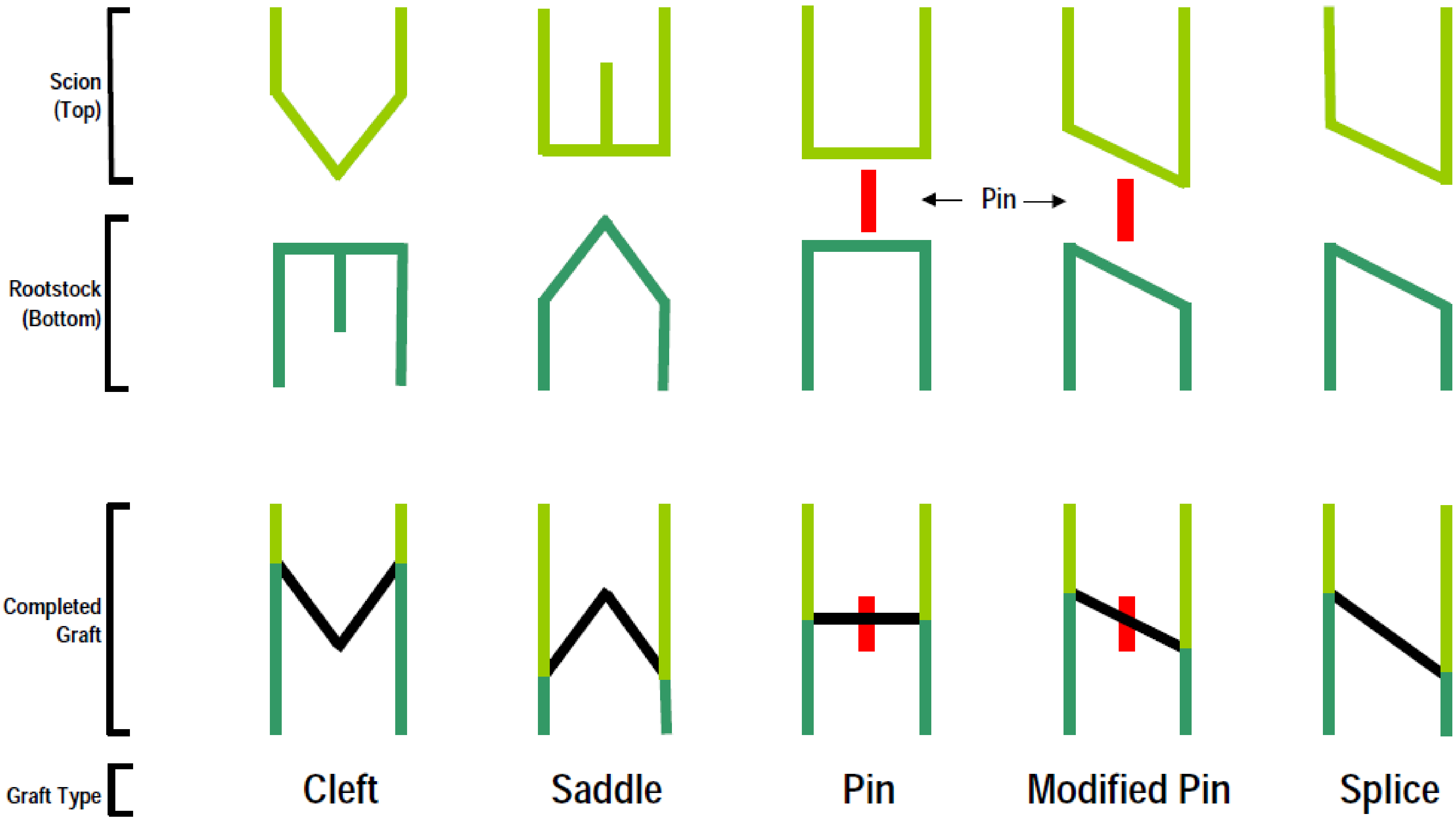
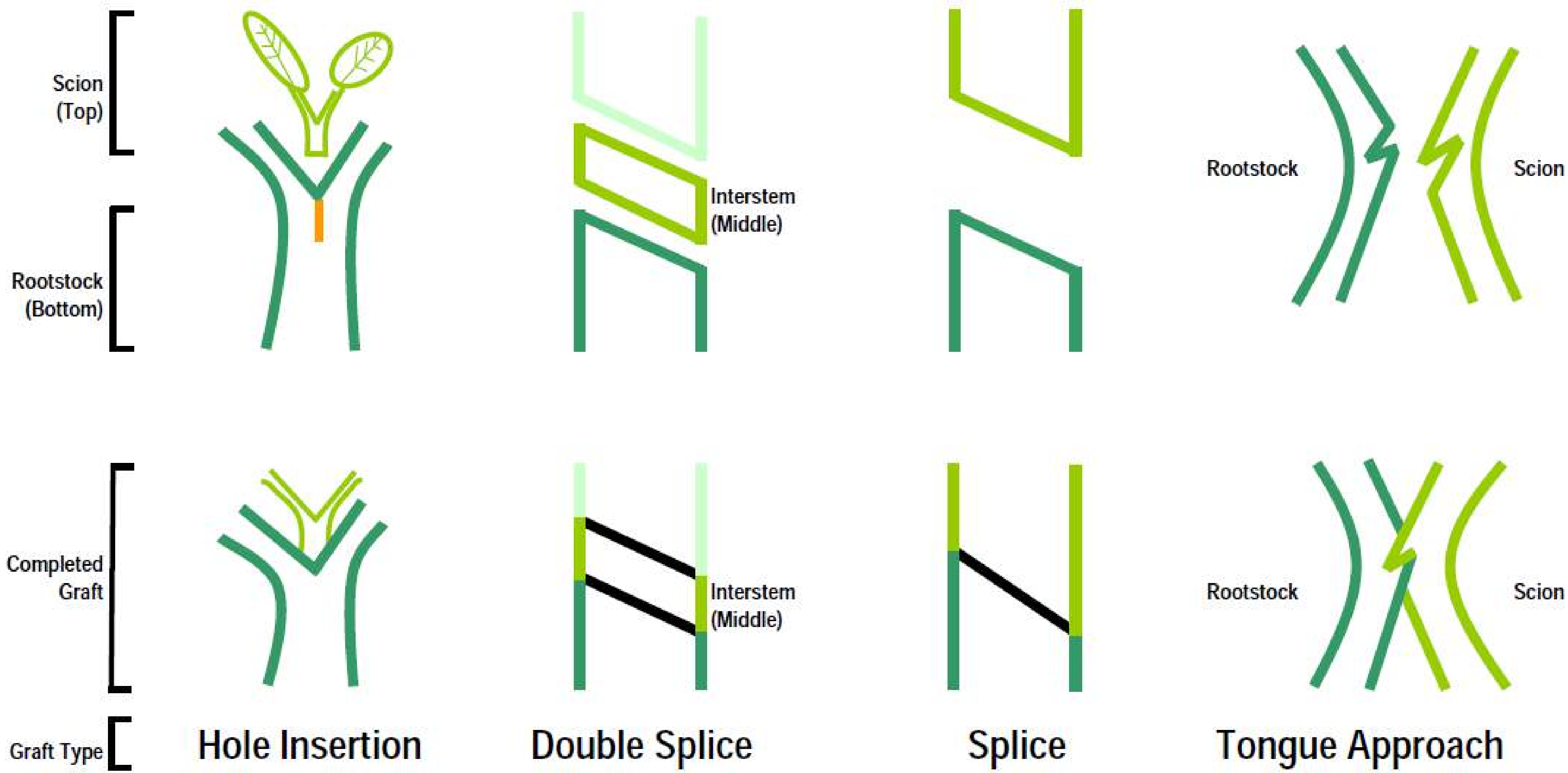
Types of Grafts for Solanaceous Crops (tomato, pepper, eggplant)



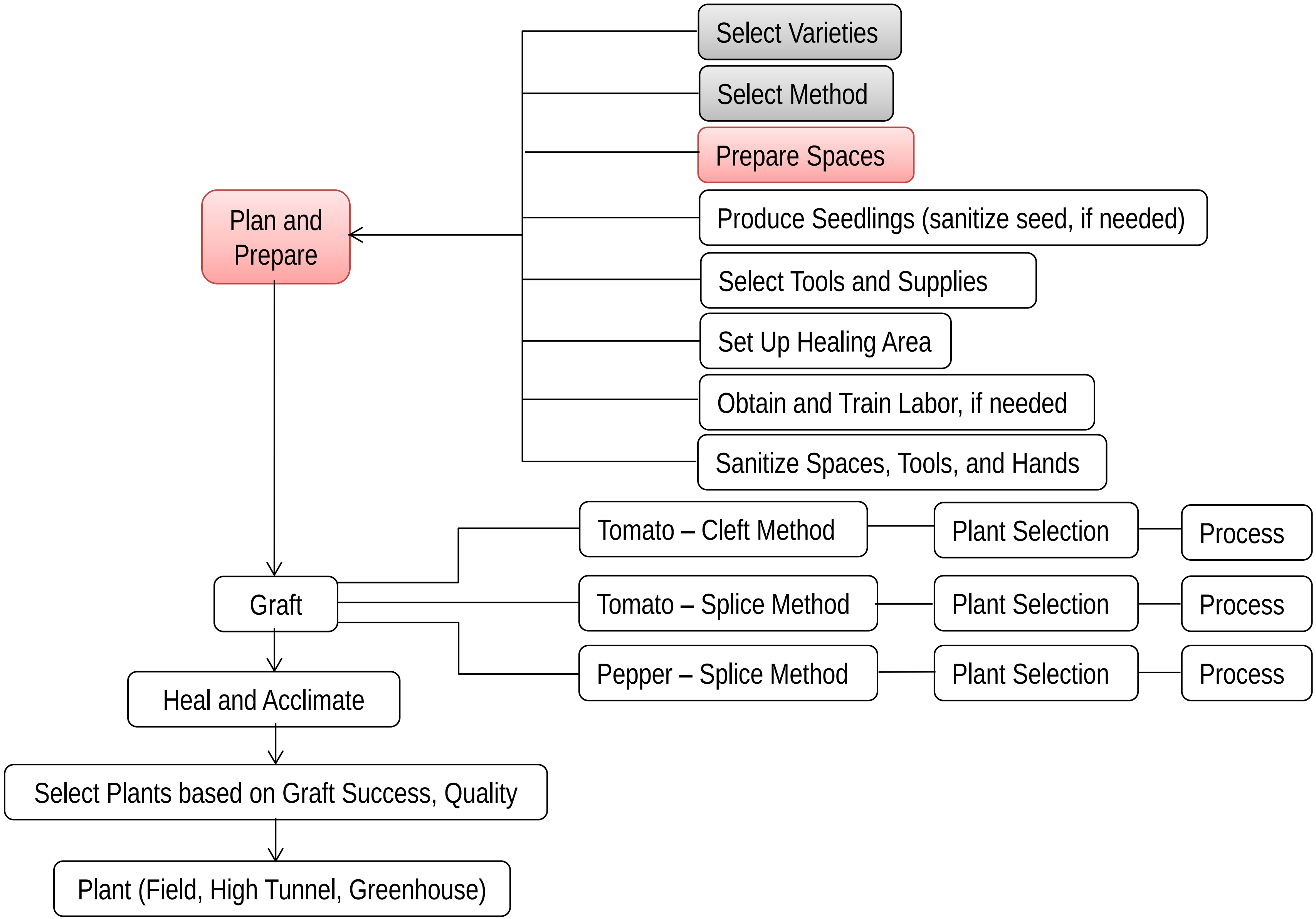
Drawings after Lee (2003).

Types of Grafts for Cucurbits (melon, cucumber)



Drawings after Lee (2003)

While not considered in this guide, other techniques are used to graft other fruiting vegetables (e.g., cucurbits). Overall, grafting is similar in cucurbit and solanaceous crops and requires attention to detail throughout. For more information on cucurbit grafting methods, see Hassell, et al., 2008 and Miles, et al., 2013.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Seed-to-Grafted-Plant Calculator

The number of high-quality grafted plants needed to produce fruit does not equal the number of seeds that are initially sown. In fact, the number of seeds sown must exceed the number of plants to go to the field or high tunnel.

So, how many seeds must be sown?

The calculator at http://u.osu.edu/vegprolab/research-areas/grafting/resources/seed-to-grafted-plant-calculator/ can be used to assist in this determination.

The calculator is based on the below equations. These equations can be used to calculate seed requirements manually.

Step 1.

$$\frac{\text{\# of grafted plants desired for planting in field or high tunnel (A)}}{\text{\% of grafted plants of high quality (suitable for fruit production) (B)}} = \text{\# of surviving plants available after 14 days}$$

Step 2.

$$\frac{\text{\# of surviving grafted pants available after 14 days}}{\text{\% of grafted plants surviving after 14 days (C)}} = \text{\# of grafted plants to attempt}$$

Step 3.

$$\frac{\text{\# of grafted plants to attempt}}{\text{\% seedlings suitable to graft (D)}} = \text{\# of seedlings suitable to graft}$$

Step 4.

$$\frac{\text{\# of seedlings suitable to graft}}{\text{\% mergence after 21 days (E)}} = \text{\# of seeds to sow}$$

(Helpful hint: Use decimals when calculating with percentages. For example use .90 for 90%)

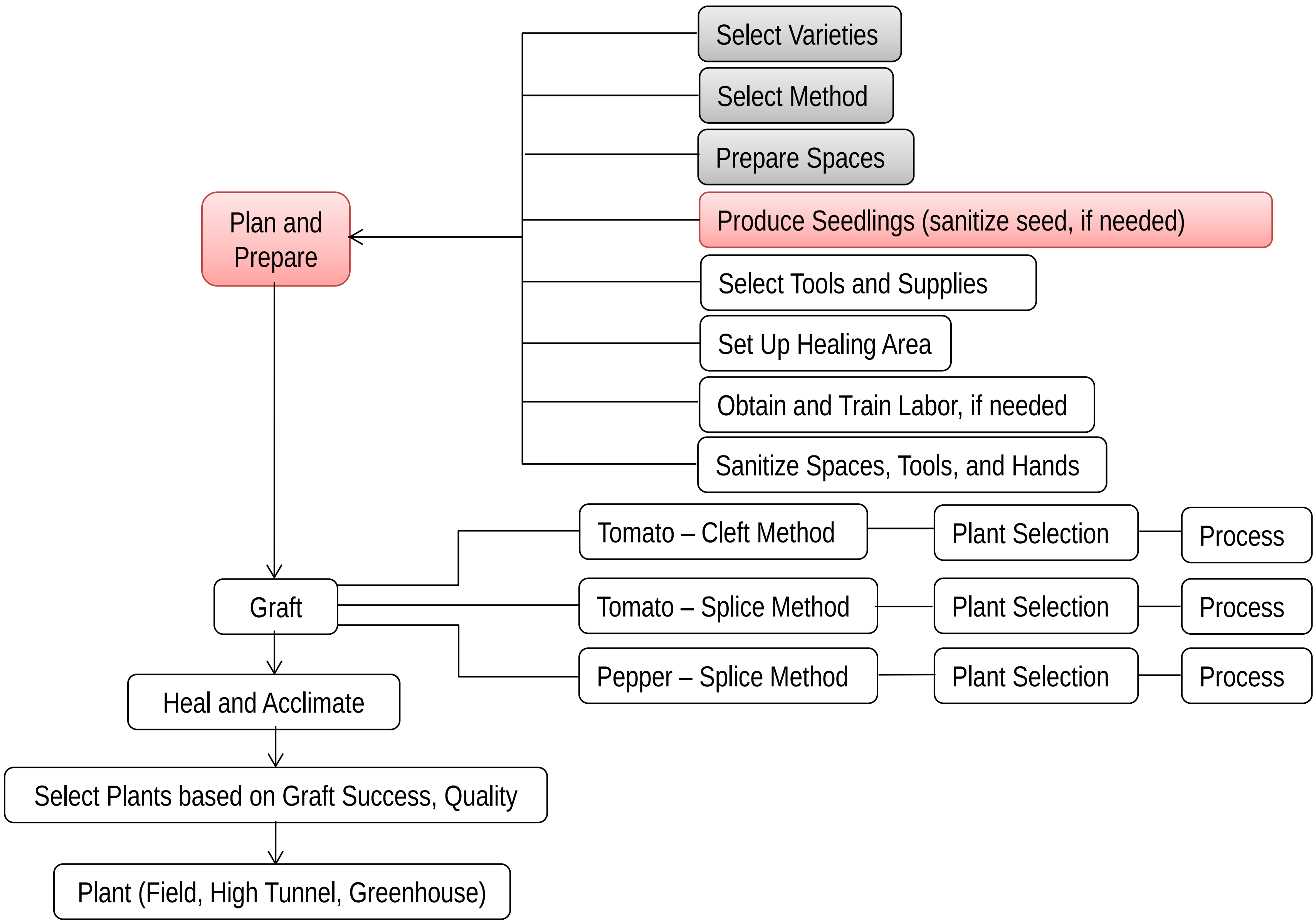
Vegetable Production Systems Laboratory
Ohio Agricultural Research and Development Center, Department of Horticulture and Crop Science
1680 Madison Avenue, Wooster, Ohio 44691 Phone 330.263.3810 Fax 330.263.3887

Prepare Adequate Space for Rootstock and Scion Seedling Production

Recall that two seedlings are needed to create a grafted plant. So, roughly double the space is needed to produce seedlings to graft than to produce standard transplants.

Prepare The Grafting Area

Steps required to prepare the grafting space will differ by location and operation. Regardless, the work space for grafting should be clean, well-lit and at moderate temperature. Also, seedling health must be maintained in the grafting area. Do not allow seedlings to experience significant stress, including desiccation. Chairs, tables and clean water should also be available for grafters.



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Select Tools and Supplies
Set Up Healing Area
Obtain and Train Labor, if needed
Sanitize Spaces, Tools, and Hands
Plan and Prepare
Graft
Tomato – Cleft Method
Plant Selection
Process
Tomato – Splice Method
Plant Selection
Process
Pepper – Splice Method
Plant Selection
Process
Heal and Acclimate
Select Plants based on Graft Success, Quality
Plant (Field, High Tunnel, Greenhouse)

Seedling Production for Grafting

1. Scheduling

Determine your reason for grafting, and select scion and rootstock seed accordingly. In general, begin sowing tomato and pepper seed 6-8 weeks before the target date of transplanting. Sowing 6-8 weeks before transplanting provides time to produce and graft seedlings, and to allow them to heal and be hardened-off, if needed. Sowings can be staggered depending on the expected rootstock and scion germination and growth rates and grafting method that will be used.

2. Sowing, germination, and seedling management

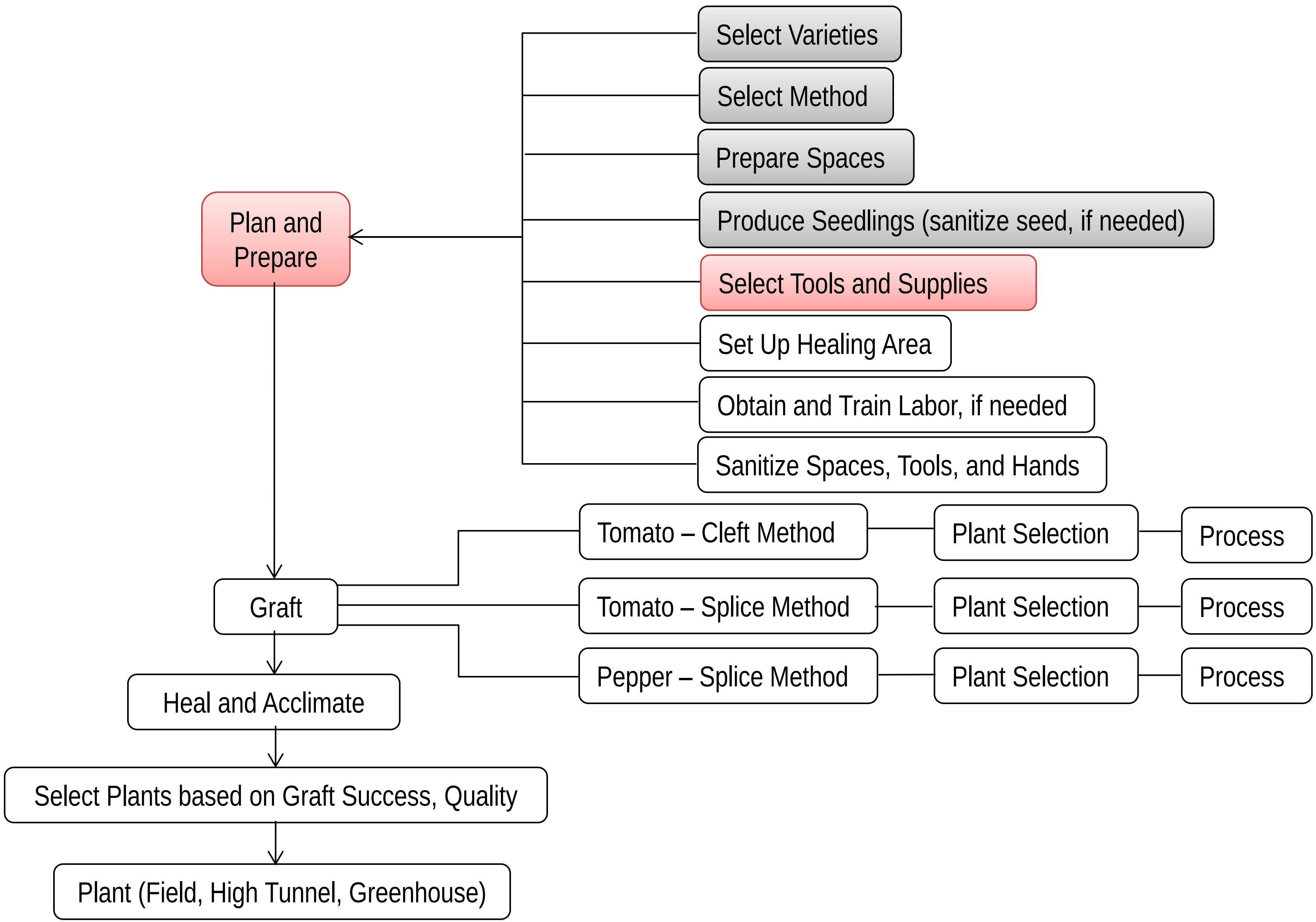
Overall, the process for producing seedlings to graft is very similar to producing standard, ungrafted transplants. Proven methods appropriate for local conditions should be used in both cases. Resources to guide light, temperature, humidity, irrigation, pest, disease, and other aspects of seedling management are available (e.g., Fernández-García et al., 2004; Nguyen et al., 2014). Overall, it is important to keep a number of points in mind when producing seedlings to graft.

For example, rootstock and scion varieties can behave differently during seedling production. Rootstock seedlings may emerge less uniformly (over a longer period), especially if the scion variety is a hybrid. Also, the ratio of seedlings obtained to seeds sown may be lower for some rootstock varieties than scion varieties. However, rootstock seedlings may grow more rapidly than some scion varieties (see images on pp. 20-24)

The particular emergence and growth characteristics of rootstock and scion varieties require specific steps to be taken during seedling production for two reasons. First, these steps help insure that all seedlings can be grafted when they are eligible, thereby wasting few seed and other resources used in seedling production. As described in the next section, the diameter of rootstock and scion seedlings must be similar and within a specific range at grafting. Seedlings, especially rootstock, can become too large to graft by standard methods if sowing is too early or growing conditions are mismanaged. Second, these steps also help ensure that the targeted number of grafted plants are available for planting. Having fewer plants than needed lowers fruit production and profit potential.

Seedling Production for Grafting

Collectively, rootstock and scion variety differences may require: 1) rootstocks to be sown more times and at higher rates than scion varieties; 2) numbers used in the seed-to-grafted plant calculation (p. 29) to differ for rootstock and scion varieties; and 3) light, temperature, irrigation, fertility, and other regimes to be individualized to control rootstock and scion seedling growth. Recall that matching stem diameters, controlling the length of the hypocotyl and distance between nodes (i.e., internode length), and maintaining seedling health are essential. Also, seedling management soon before grafting can affect graft success. For example, withholding irrigation and fertigation for 1-2 days before grafting, especially to rootstock seedlings, can be beneficial. Excess root zone moisture at grafting increases root pressure and can disrupt graft union formation, reducing the success of the operation. Roots transfer water to the cut surface of the rootstock, pushing back the scion and loosening its contact with the rootstock, perhaps lowering the probability of plant survival or strength at the graft union.


Plan and Prepare
Select Varieties
Select Method
Prepare Spaces
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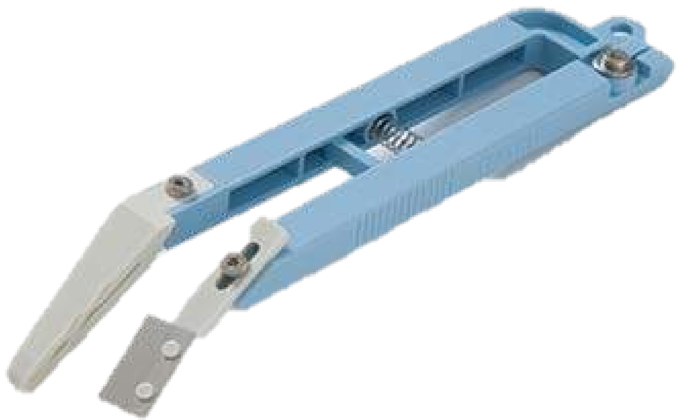
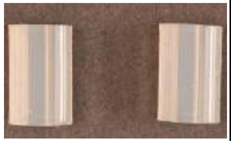
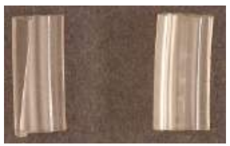
Tools

Razor blades can be used for both cleft and splice grafting.



Japanese-made grafting clippers designed for use in splice grafting cut at a consistent angle.

Clips and Fasteners

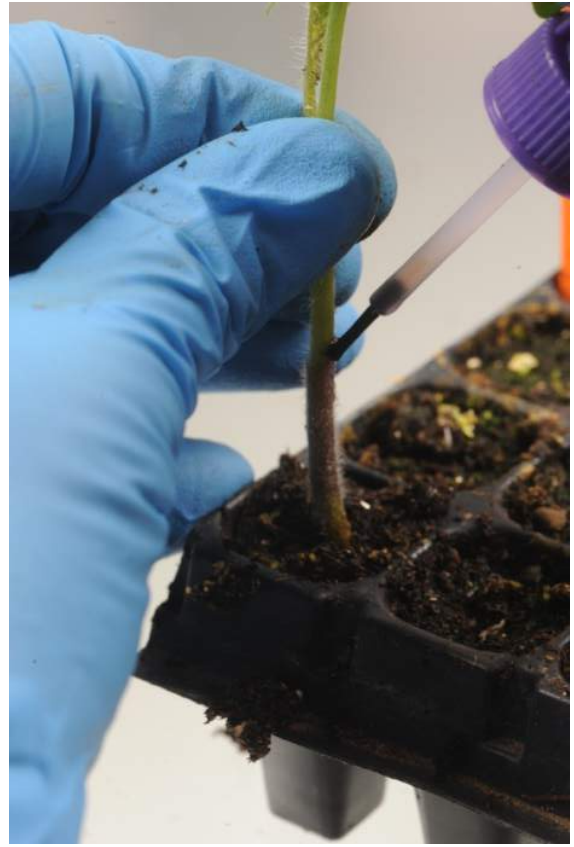
Fastener	Type	Quantity	Approximate Price (January 2016)
	Silicone Grafting Clip	200	$14.95 - $15.95
	Spring Loaded Grafting Clip	200	$44.50
	Self-made Grafting Clip	200	≥ $0.90*
	Latex tubing	200	~$2.00
	Glue (cyanoacrylate), brush on	.18 oz	$4.00

The graft union where rootstock and scion seedlings are joined must be secured with a type of 'splint' until plant cells complete the job. The fastener must provide firm support while allowing some stem expansion. Commercial clips are available for this purpose and others can be self-made. The type used depends on budget, grafting method, and other considerations. For example, in concept, all clips, except for ones made from latex tubing, can be reused. However, the incentive and ease may be greatest with commercial spring-type clips. Spring-type clips also tend to accommodate a wider range of stem diameters. When using self-made clips on a range of stem diameters, tubing ranging in internal diameter is needed. Various fasteners are shown in the table at left and featured throughout this guide. In our experience, self-made clips have been very useful in cleft grafting (described in later pages). Glue is an interesting option but, currently, it is not used widely. Page 39 of the guide describes the use of glue to secure graft unions. Otherwise, the guide focuses on the use of clips.

*Note that the price of self-made clips varies with material and labor costs.

Self-Made Clips

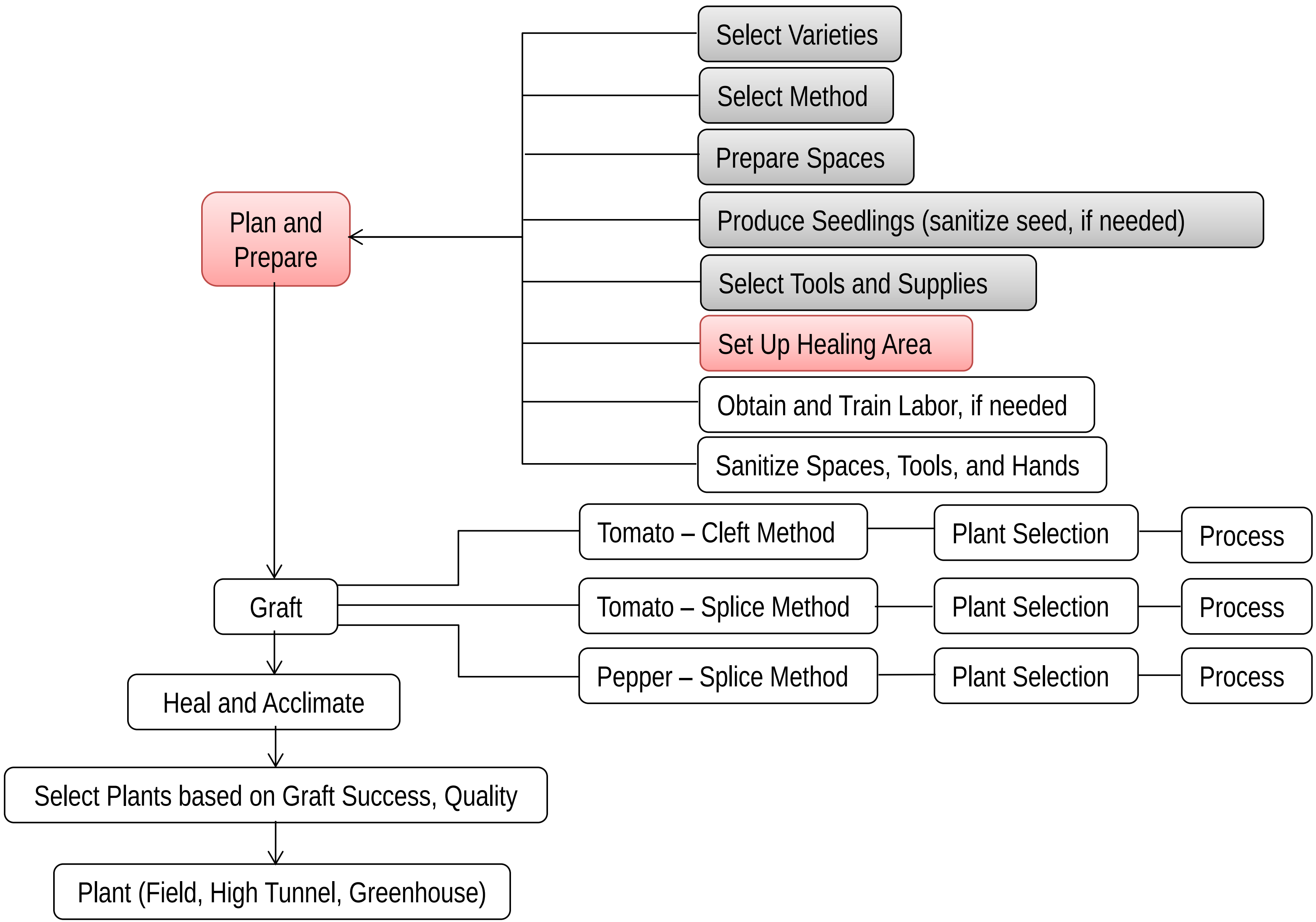
Rolls of plastic tubing are available from various sources. Each roll contains tubing of a specific internal diameter. The desired stem diameter(s) must be known when buying tubing. We use tubing of three internal diameters (3.2, 3.8, 4.0 mm). Clips cut from different diameters are used on plants having small, medium, or large stem diameters at grafting. Regardless, each clip is cut to a length of approximately 20 mm. The device shown at right above helps us make clips from tubing. Tubing is pulled past the vertically-mounted razor blade, thereby opening the tubing lengthwise. Then, the tubing is cut to clip-length using the horizontally-mounted blade (far end of wood block).

Self-made (left) and commercial (center) clips are not the only devices that can be used to secure new grafts, particularly of the splice grafts. Using glue to secure graft unions (right) is also being studied. If utilizing glue, grafting proceeds as described in terms of preparing the rootstock and scion. However, instead of using a clip to secure the graft, “instant bond” (cyanoacrylate or “super” glue) is brushed around the EXTERIOR of the graft union, and the grafter holds the union in place for 3-4 seconds until the glue sets. Graft success rates have been acceptable but lower with glue. Also, securing the graft union with glue applied by hand takes more time than when using a clip. Glue is toxic and ineffective when applied to the INTERIOR tissues (cut surfaces).

Silicon clip used to secure pepper graft union.

Glue used to secure pepper graft union.


Select Varieties
Select Method
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Select Plants based on Graft Success, Quality
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Set Up the Healing Area

Grafting requires wounding seedlings severely. Also, immediately after grafting, the scion portion of the grafted plant has little access to water and the rootstock portion cannot photosynthesize. Therefore, conditions around newly grafted plants must be maintained carefully to promote rapid healing – like around patients recovering from operations. Target conditions are described in later pages and emphasize light, temperature, and humidity levels. Healing areas or chambers worldwide vary greatly given local environmental conditions, the availability of materials, scale of operation, budget, and other factors. Regardless, all are intended to maintain light, temperature, and humidity near the grafted plants within target ranges.

Examples of healing chambers are shown in following pages.

Simple healing chambers without a greenhouse

dallasgardenbuzz.com

VPSL, The OSU-OARDC

www.blackdogled.com

AVRDC

Healing chambers in the greenhouse with natural light

Healing areas or chambers can be setup on greenhouse benches to provide proper conditions for the post-grafting environment. A polyvinyl chloride (PVC) frame covered with plastic and shade cloth is sufficient. Cool-mist foggers/humidifiers can help humidify. Capillary mats with drip irrigation under plant trays moisten the rooting medium and humidify the chamber, a plastic covering helps stabilize humidity levels, and shade cloth reduces incoming light levels 30-50% depending on its weave.

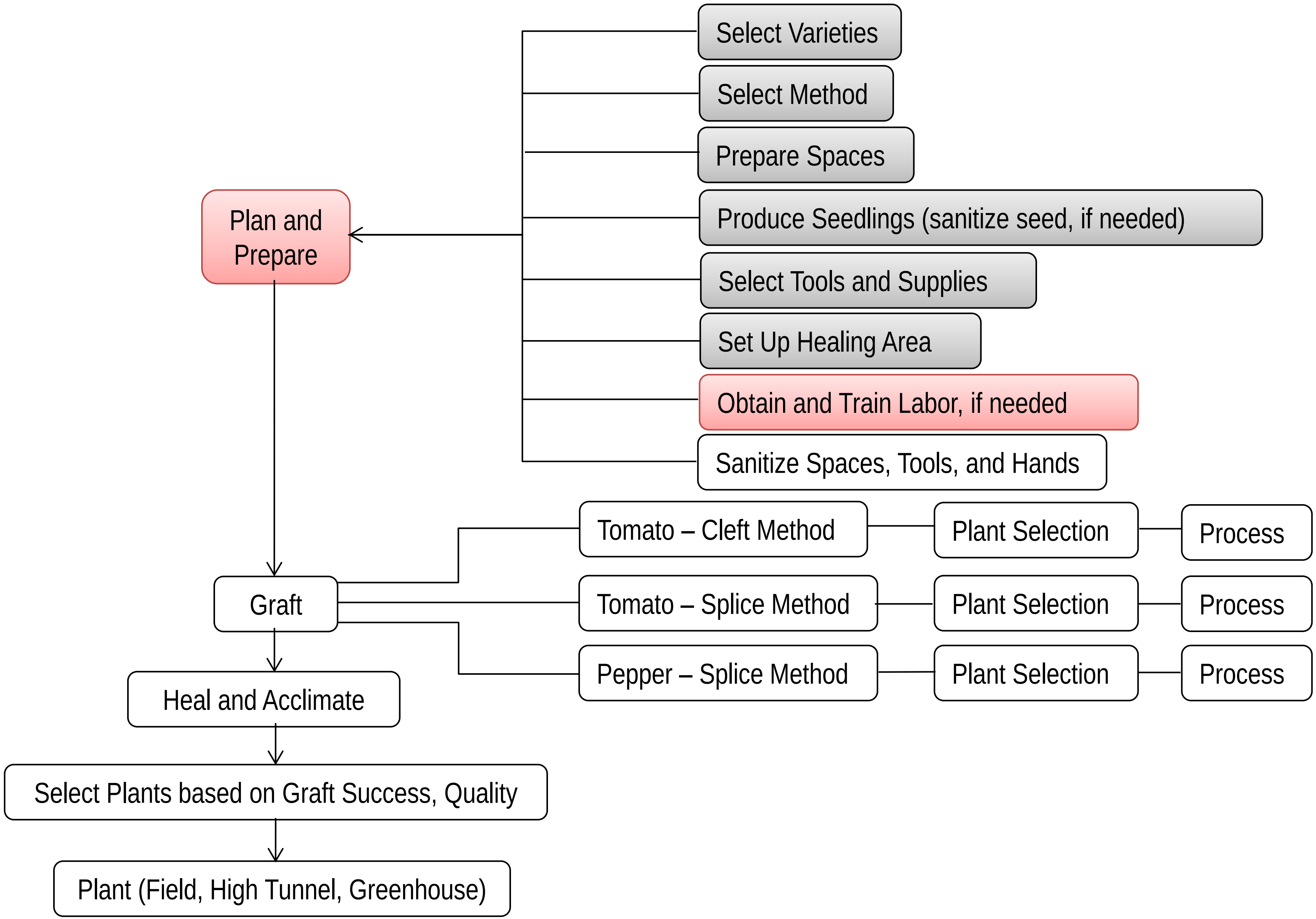
Highly controlled growth chambers with artificial light

Highly controlled environments such as plant growth chambers are expensive and may be most suitable for research or larger scaled operations. The pictures are growth chambers used for research on graft healing at The Ohio State University, Ohio Agricultural Research and Development Center.

Additional options for controlled growing environments



While these technologies are still being developed, they may provide an effective, affordable for housing mobile propagation systems independent from outside climates.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Labor

Grafting is easy to learn, but experienced grafters often are more efficient. One person can complete the process, although some people are more adept at certain stages of grafting. Therefore, a team approach can speed the process and increase success, especially if a large number of plants are to be grafted.

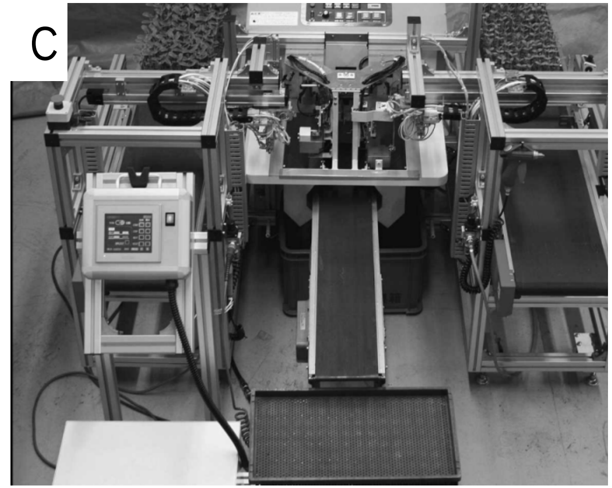
Well Designed Logistics Can Increase Grafting Efficiency

Different logistic designs can be optimized based on automation levels, facilities, labors and other available resources.

Images from Kubota, et al., 2008.

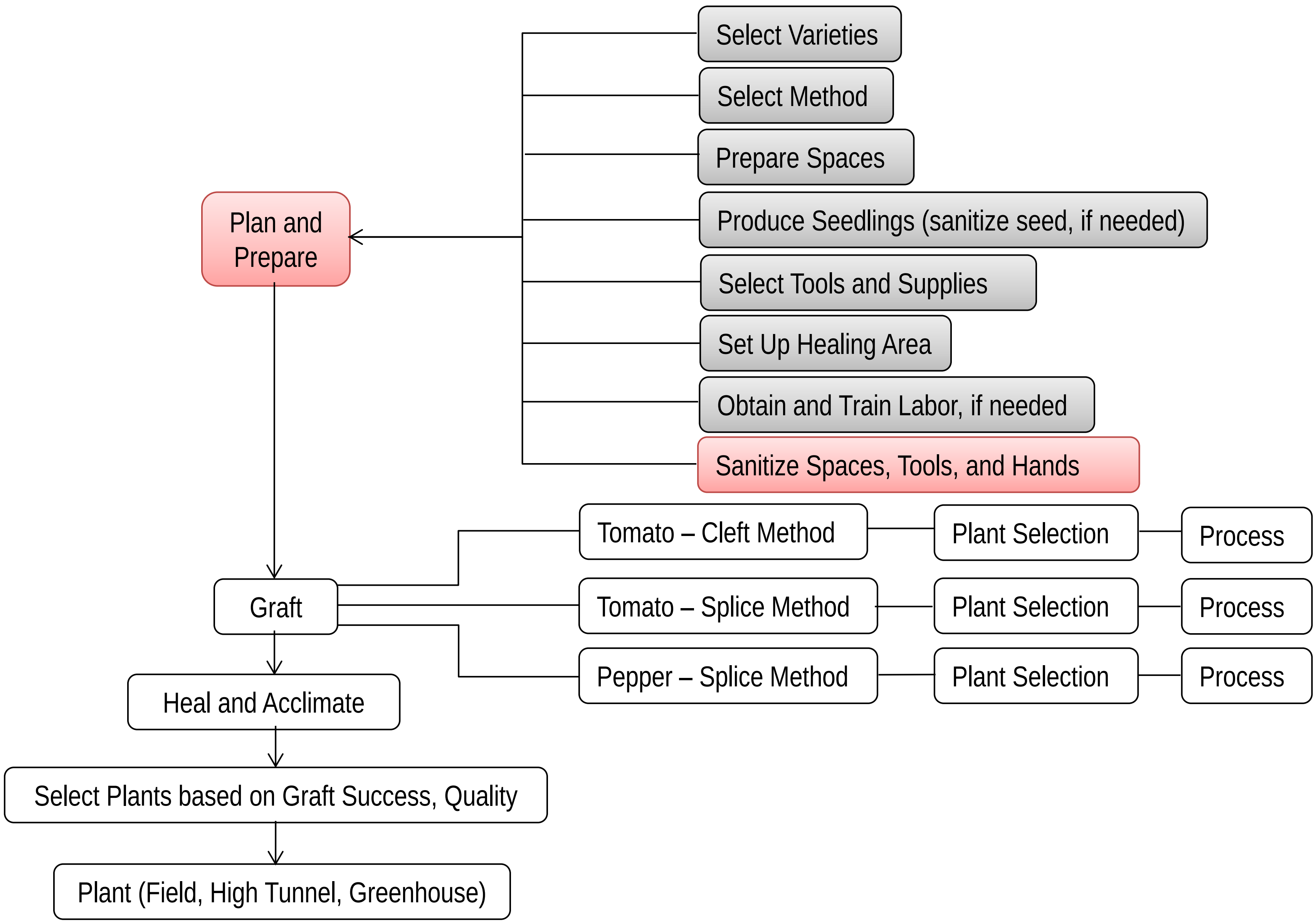
A. Each grafter finishes all steps.

B. Each grafter finishes one step as a line operation.



C. Automatic grafting with minimum grafter assistance.

An analytic Hierarchy Process approach was developed to help propagators design grafting operations, as published in Meng, et al., 2014.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Sanitation

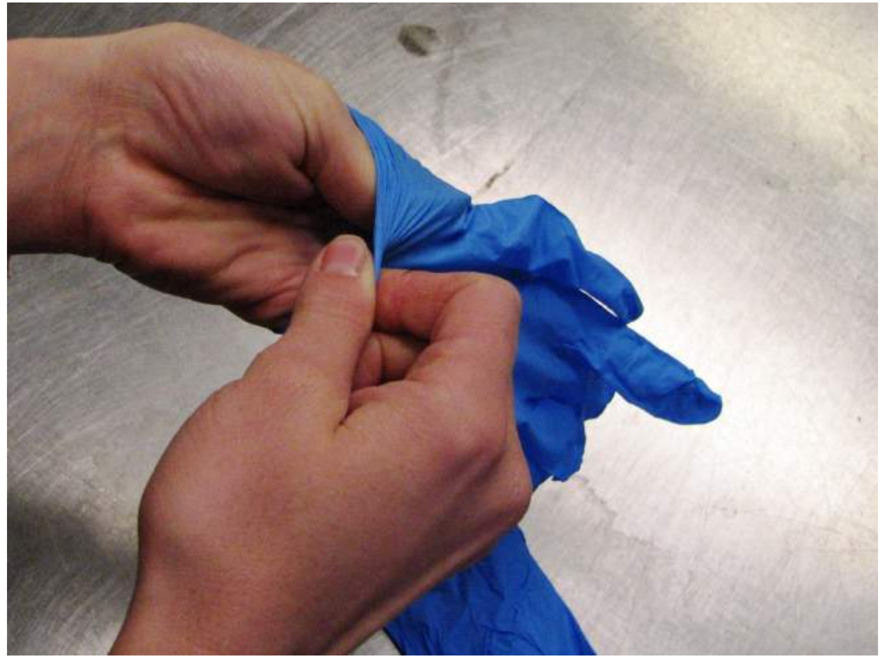
Grafting is a type of organ transplantation. If sanitation is not carefully maintained, mechanically transmitted diseases can be spread during the grafting process

Tobacco mosaic virus (TMV) is a major disease that can be transmitted by grafting. It can be seed-borne, and readily transmitted mechanically during grafting operations. Therefore, washing hands and replacing tools with new or clean ones frequently are important to minimize possible transmission of TMV. Additionally, TMV may be present in tobacco products. Therefore, workers should NOT be involved in the grafting operation until after taking appropriate steps (including careful sanitation) to limit TMV transmission.

Other tomato virus and viroid diseases that may be mechanically transmitted during the grafting process include *Tomato mosaic virus* (ToMV), *Pepino mosaic virus* (PepMV), *Potato spindle tuber viroid* (PSTVd), *Tomato chlorotic dwarf viroid* (TCDVd), *Mexican papita viroid* (MPVd), *Citrus exocortis viroid* (CEVd), *Columnea latent viroid* (CLVd), *and Tomato apical stunt viroid* (TASVd). Fungal diseases such as Damping off, Early blight, Fusarium crown rot, Fusarium wilt, Late blight, Powdery mildew, Southern blight, and Verticillium wilt, and bacterial diseases such as Bacterial wilt can also be transmitted or spread during the grafting process if sanitation procedures are not followed.

For further information on seed sanitation, please see Appendix A.

Hand Sanitation



Grafting requires clean hands, clean surroundings, and no tobacco use. Disposable gloves help maintain a sanitary work space.

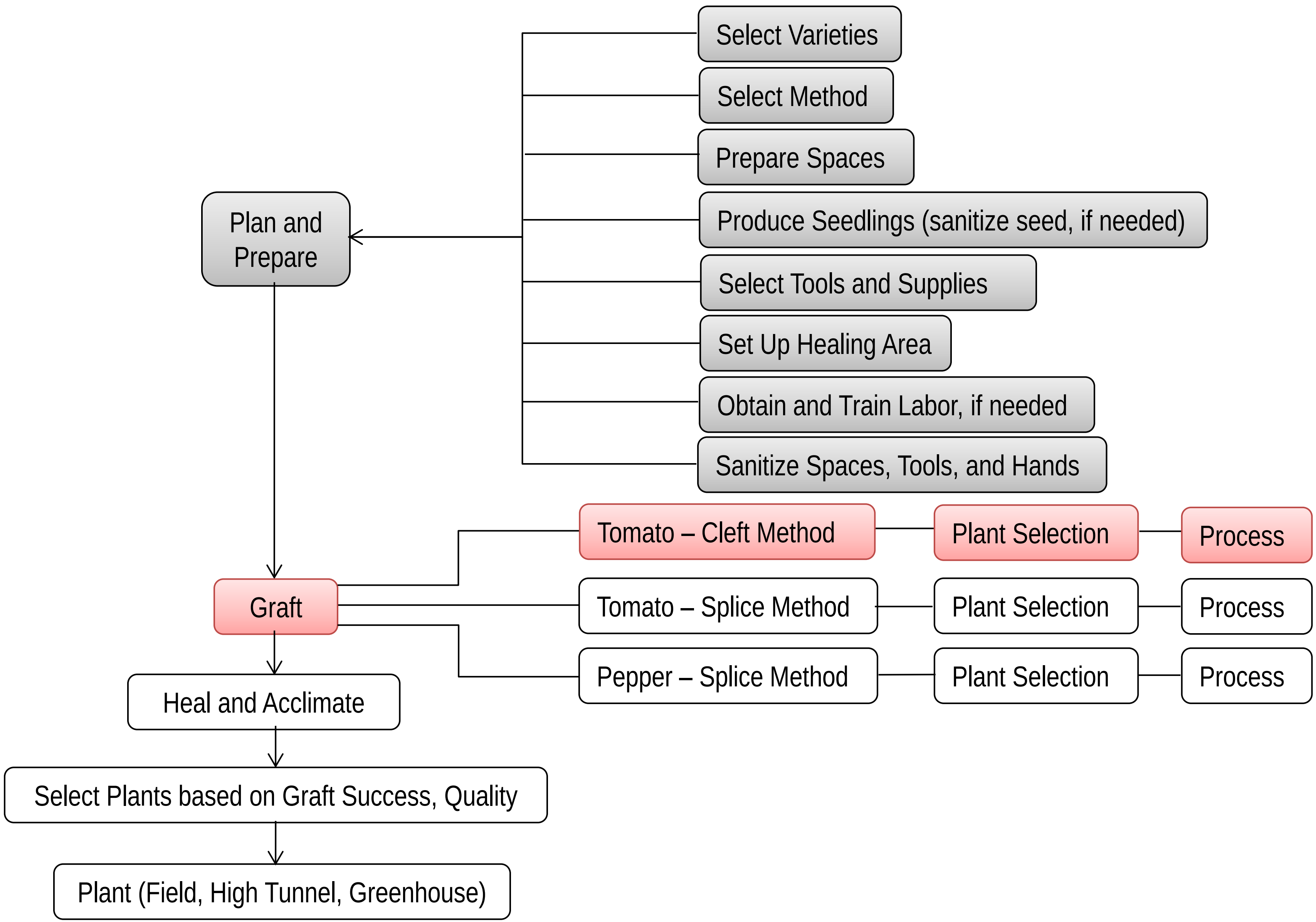
Those who use tobacco should be excluded from participation in the grafting process or included only with special care since tobacco is a source of disease inoculum.

Tool Sanitation

Proper sanitation will greatly improve your success with grafting. Use a clean work space, clean cutting tools, and detergent and alcohol to kill and limit the spread of pathogens.

Options for sanitizing grafting tools—Dr. S.A. Miller, The Ohio State University, OARDC				
Options	Step 1	Step 2	Step 3	Step 4
1	Dip tools in 33% bleach	Rinse tools in clean water	Soak tools in 70% ethanol for 15 minutes	Allow tools to dry on a clean surface
2	Soak tools in 10% bleach for 30 minutes	Rinse tools in clean water	Soak tools in 70% ethanol for 15 minutes	Allow tools to dry on a clean surface
3	Soak tools in Physan 20 or similar disinfectant for 10 minutes	Rinse tools in clean water	Soak tools in 70% ethanol for 15 minutes	Allow tools to dry on a clean surface

In addition to the sanitation methods in the chart, tools may be sanitized by flaming. Dip the tool (e.g. blade) in alcohol, expose to flame, and allow to cool in air before using. Take proper steps to limit fire and fume hazards.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Cleft Grafting Overview—Tomato

1) Remove the shoot of the rootstock by making a cut below the cotyledons.

2) If using a self-made plastic tube clip, place it on and around the rootstock stump.

3) Create a vertical cut in the rootstock stump that bisects it.

4) Remove the roots of the scion by making a cut above or below the cotyledons.

5) **Optional:** Remove leaves from scion; do NOT damage the youngest leaf or apical meristem.

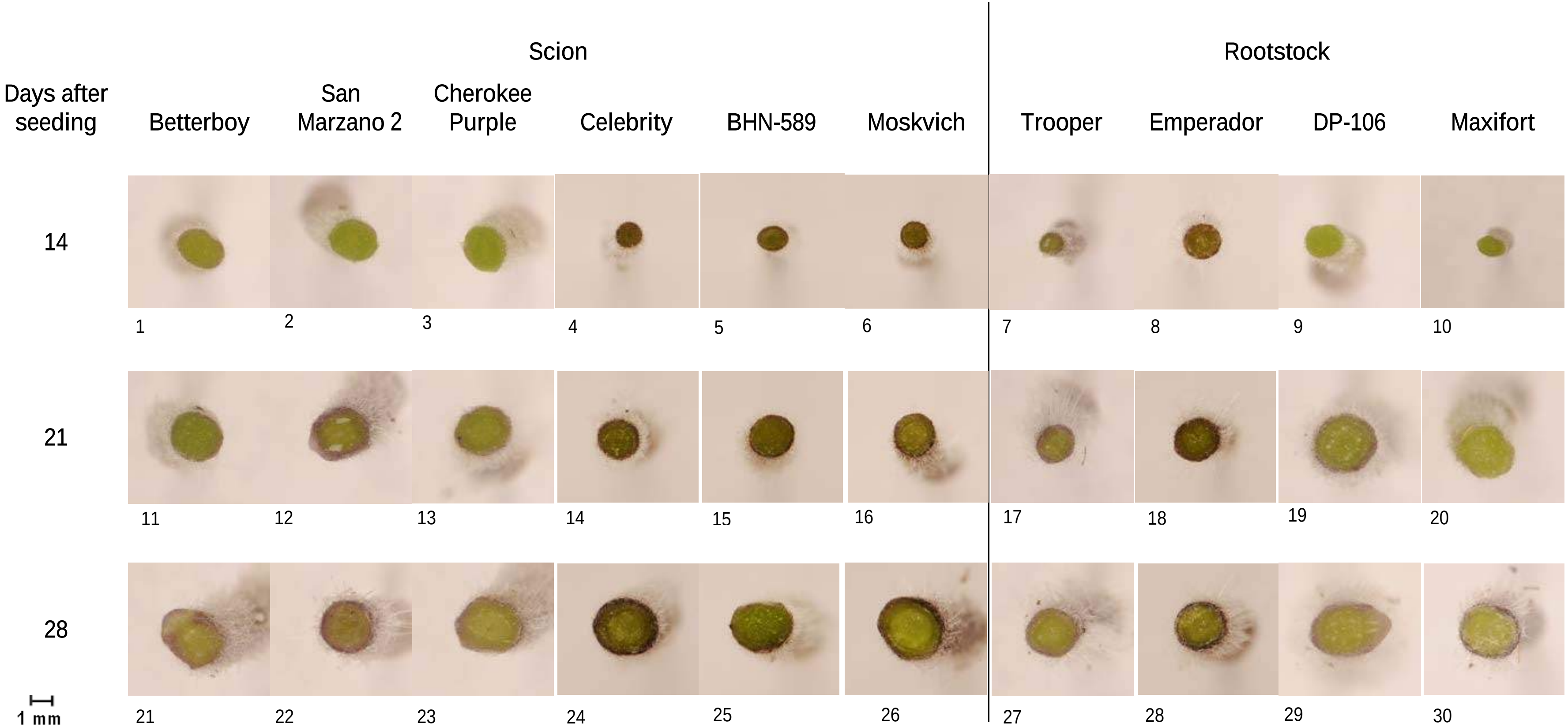
6) Trim scion stem into a wedge shape with two diagonal cuts.

7) Place the prepared scion shoot into the bisected rootstock stump.

8) Pull the self-made clip upward to secure the graft union or attach a commercial clip.

Experienced grafters cut and join seedlings using individualized versions of the major steps above. Their goals are always: 1) to complete the steps carefully and quickly and 2) to insure that the two cut surfaces are joined firmly and in the correct orientation. They also clean tools or change to a clean, new razor blade frequently. Once plants and other materials are in place, experienced grafters can prepare a grafted plant in much less than one minute.

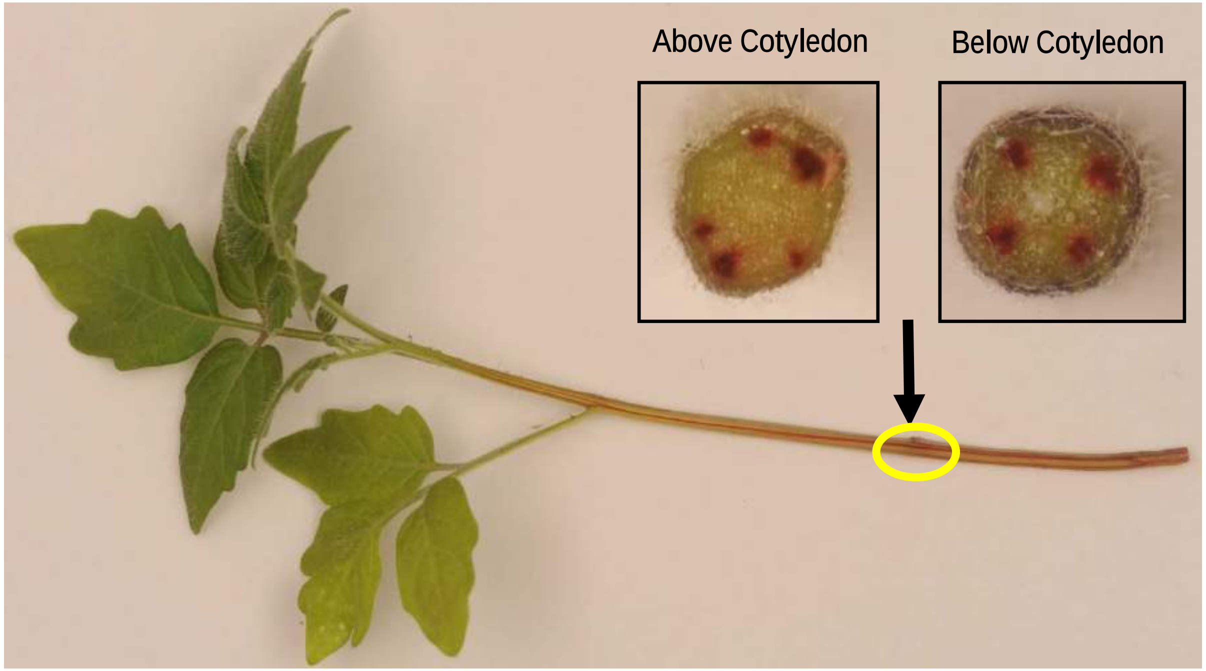
Grafting methods can be chosen based on the stem diameters of scion and rootstock seedlings. Splice grafting requires rootstock and scion stem diameters to be nearly identical. Cleft grafting tends to allow slightly greater variance in rootstock and scion stem diameters. Therefore, cleft grafting may be ideal for new grafters or when rootstock and scion seedling size differences are intolerably high for splice grafting. Also, the location of the graft on the stem of the scion can be adjusted to obtain the closest match with the rootstock stem diameter where the rootstock cut was made.



	Cleft					
Examples of matching rootstock/scion combinations	1×18	2×20	3×20	11×19	14×17	22×30

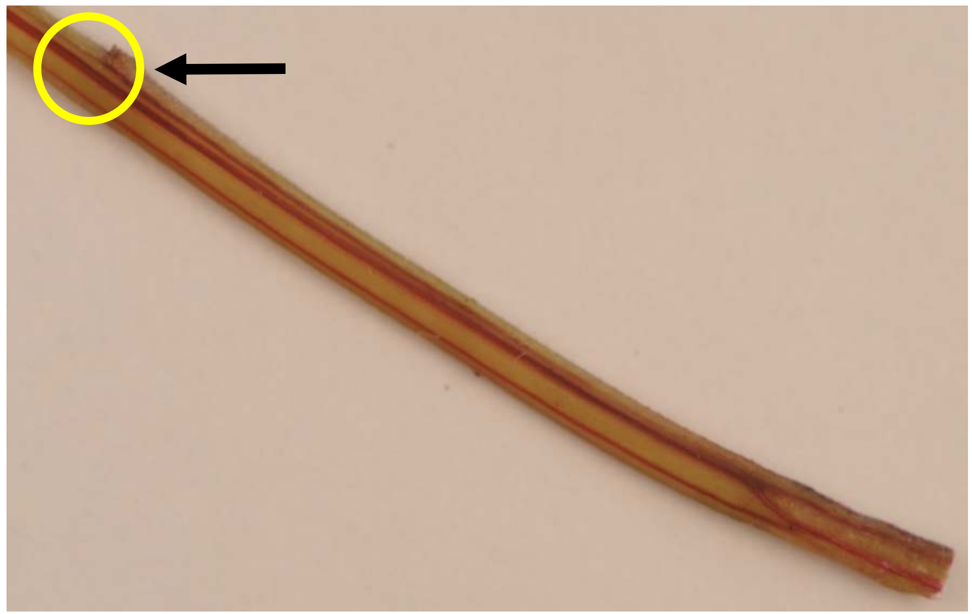
Trimming scion leaves

Removing some of the older true leaves prior to grafting can be useful. Water loss from the scion must be minimized until the vasculature of the rootstock and scion seedlings connect. Trimming some scion leaves is one way to minimize water loss. Trimming reduces leaf area, lowering evapotranspiration. Also, some grafters find it easier to graft semi-leafless scions. Leafy scions also may be top-heavy, complicating management in the healing area or slowing healing. In one study, removing about 75% of the leaf surface area from the scion increased graft success (Masterson et al., 2016). However, in another study, trimming did not affect plant survival in pepper. It is clear that leaf removal can reduce the photosynthetic potential of the plant when returning to greenhouse conditions after healing and it creates additional wounds. Trimming leaves may also slow down the grafting process. You may wish to experiment on your own to determine if trimming works for you. Balance the benefits and drawbacks of trimming the scion.



The red stain highlights the anatomical differences in the vasculature above and below the cotyledons.

Water and other materials move in preset pathways (vasculature) within plant leaves, stems, and roots. These pathways are depicted in red in the figures here and they must align, at least to a minimal extent, for a graft to be successful. Therefore, matching stem diameters of rootstock and scion makes it more likely that the graft will be successful.



Healthy, uniform tomato seedlings with 2-4 true leaves and a stem diameter of about 1.5 mm (0.06 in.) to 2.5 mm (0.1 in) are best for grafting. Some grafters prefer to sort their rootstock and scion seedlings by size before grafting. Using rootstock and scion seedlings with a similar stem diameter will help align the pathways (vasculature) at the graft union.

Using calipers can provide accurate measures of stem diameter. If calipers are not available, graftability can also be determined by how tight fitting a grafting clip is on the plant stem. If the clip fits tight then the plant is a suitable size for grafting. Further, stem diameters can also be compared to thicknesses of wire from a hardware store. American Wire Gauge (AWG) standards list the diameter of gauge 15 and 10 wire as 1.45 mm and 2.58 mm, respectively.

Remove the upper portion of the rootstock seedling with one horizontal cut 5 mm below the cotyledons.

The top of the rootstock has been removed and only the stem remains.

Clips are used to secure grafts. If using a self-made plastic tube clip, place it on the remaining portion of the rootstock stem immediately after cutting and slide it down to the soil line. Commercial clips are often positioned after graft assembly. Self-made and commercial silicon clips are removed naturally by plant growth. Commercial clips, if spring-loaded, may need to be removed manually.

Create a vertical cut in the center of the rootstock stump. The cut should be the same depth as the length of the scion section that will be placed in it (see next pages). In this image and our regular practice, a cut approximately 4 mm deep is used. This depth corresponds to the location of the notch in the center of the razor blade.

Remove the lower portion of the scion seedling with two or three cuts. In the three-cut process, de-root the scion with a single cut. The cut can be made either above or below the cotyledons; regardless of its position relative to the cotyledons, the cut should be at a point where the scion stem diameter matches the rootstock stem diameter.

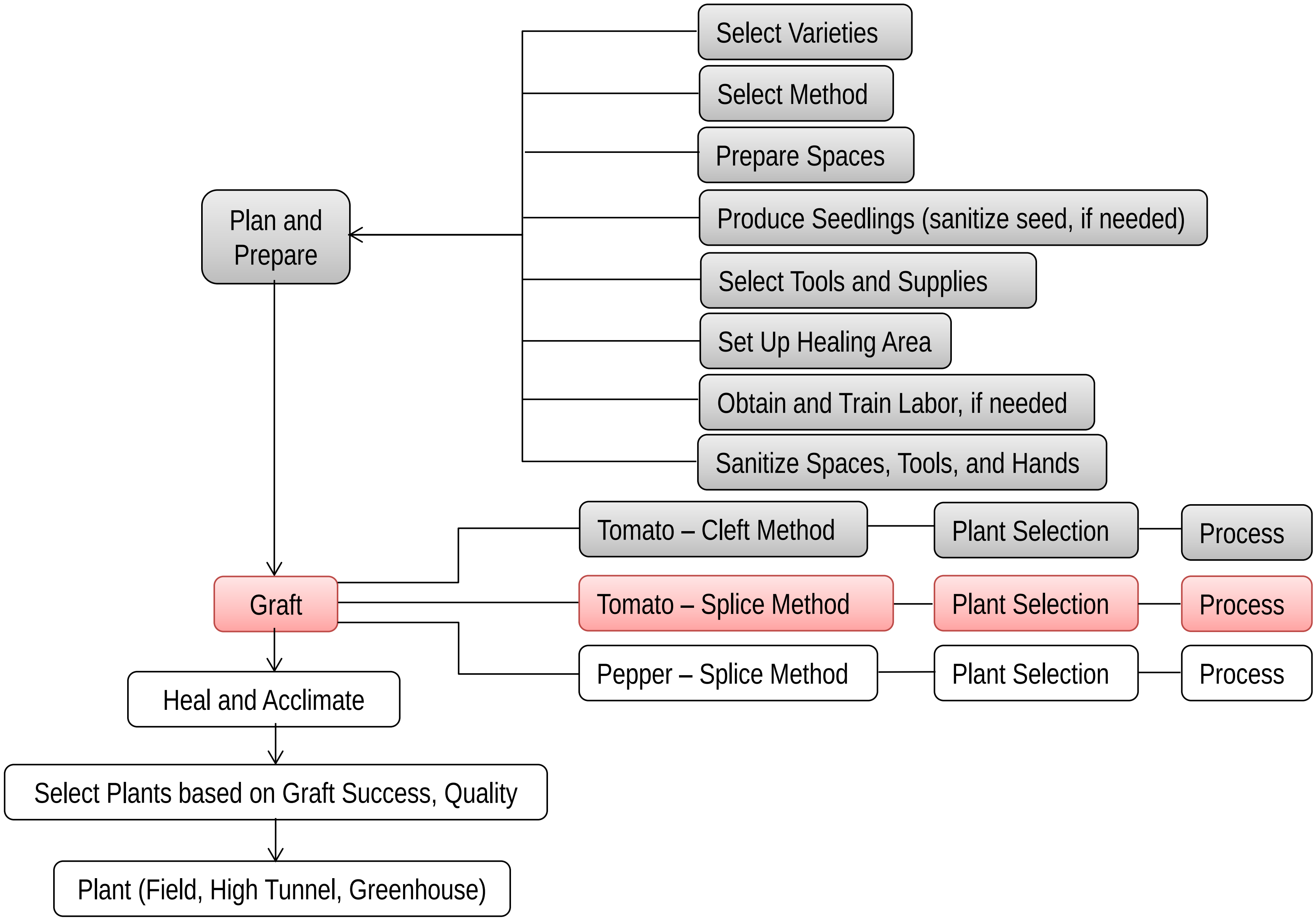
In the three-cut process, trim the lower section of the scion seedling stem to form a blunt wedge.

A scion is ready for insertion into the rootstock. The scion in this example was created using a three-cut process (a horizontal cut followed by two diagonal cuts). As mentioned, it is also possible to prepare the scion using only two diagonal cuts, simultaneously separating the scion from its roots and forming the wedge.

In the two-cut process, make two diagonal (65 degree) cuts to de-root the scion, creating the wedge to be placed in the bisected rootstock. A two-cut process can save time. Trial and error will help determine which process is best for you.

Insert the wedge end of the scion into the bisected rootstock.

Position the clip around the graft union. A self-made tube clip is shown.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Heal and Acclimate
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Plant (Field, High Tunnel, Greenhouse)

Splice Grafting Overview—Tomato

1) Remove the shoot of the rootstock by making a cut at an angle of about 45-75° below the cotyledons.

2) If using a self-made plastic tube clip, place it on and around the rootstock stump.

3) Remove the roots of the scion by making a cut at the same angle as the rootstock cut was made above or below the cotyledons where the stem is similar in diameter to that of the rootstock stump.

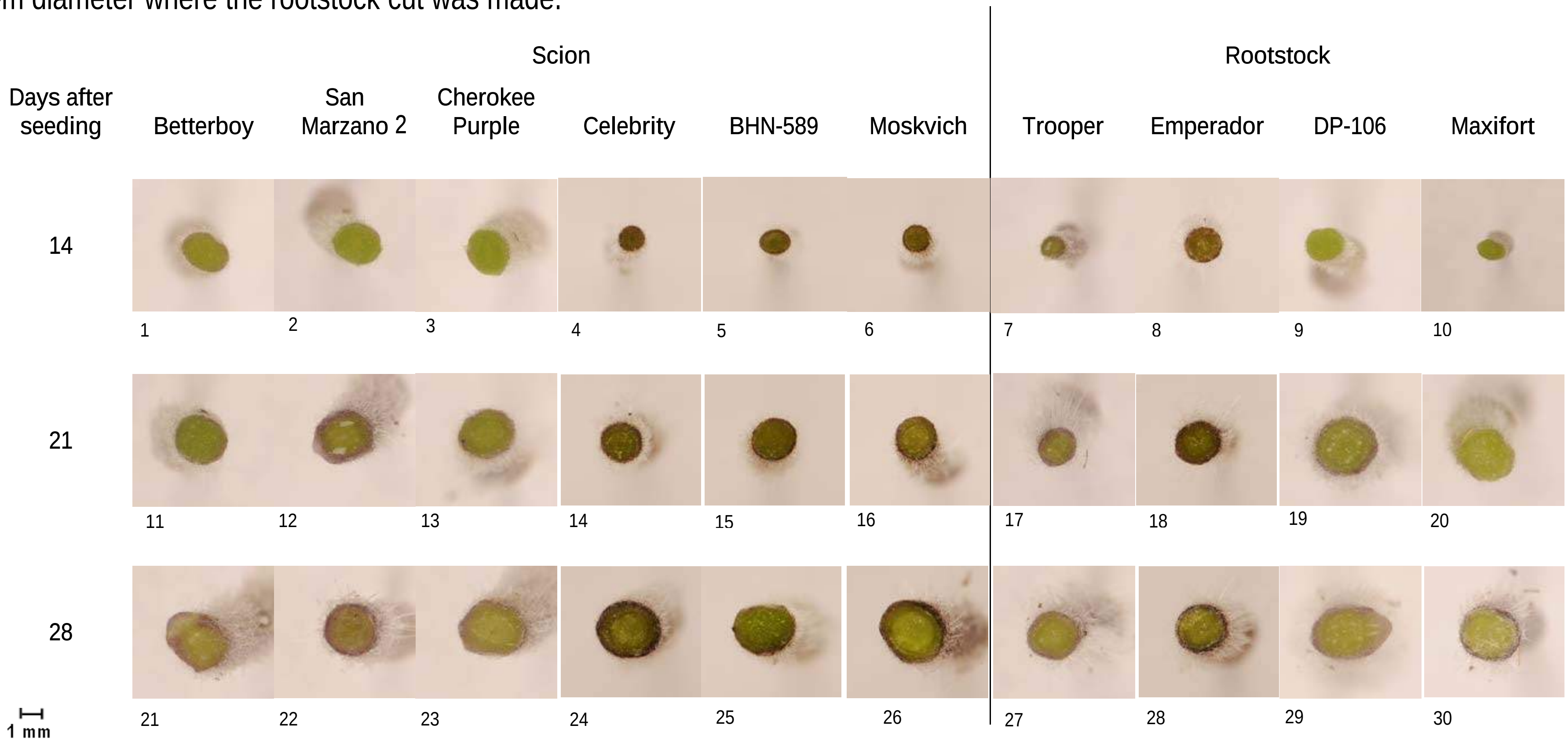
4) **Optional:** Remove leaves from scion; do NOT damage the youngest leaf or apical meristem.

5) Place the prepared scion shoot on the cut end of the rootstock stump to allow the two cut edges to match as closely as possible.

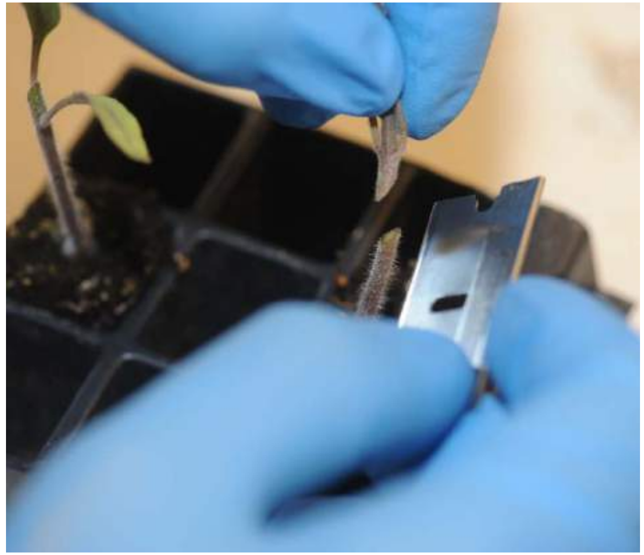
6) Pull the self-made clip upward to secure the graft union or attach a commercial clip or use glue.

Experienced grafters cut and join seedlings using individualized versions of the major steps above. Their goals are always: 1) to complete the steps carefully and quickly and 2) to insure that the two cut surfaces are joined firmly and in the correct orientation. They also clean tools or change to a clean, new razor blade frequently. Once plants and other materials are in place, experienced grafters can prepare a grafted plant in much less than one minute.

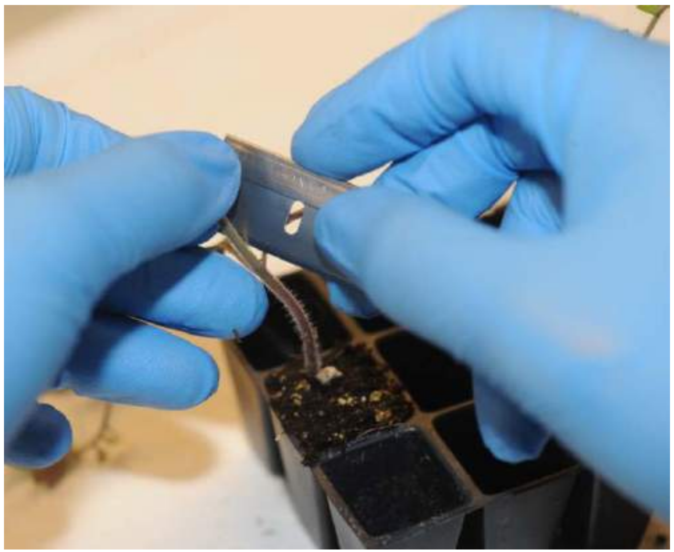
Grafting methods can be chosen based on the stem diameters of scion and rootstock seedlings. Splice grafting requires rootstock and scion stem diameters to be nearly identical. Cleft grafting tends to allow slightly greater variance in rootstock and scion stem diameters. Therefore, cleft grafting may be ideal for new grafters or when rootstock and scion seedling size differences are intolerably high for splice grafting. Also, the location of the graft on the stem of the scion can be adjusted to obtain the closest match with the rootstock stem diameter where the rootstock cut was made.



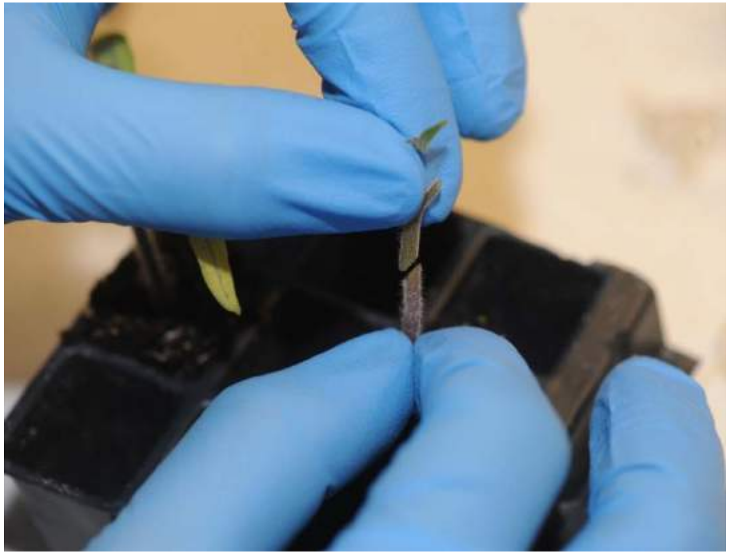
	Splice					
Examples of matching rootstock/scion combinations	3×18	11×20	11×27	15×18	23×29	24×30

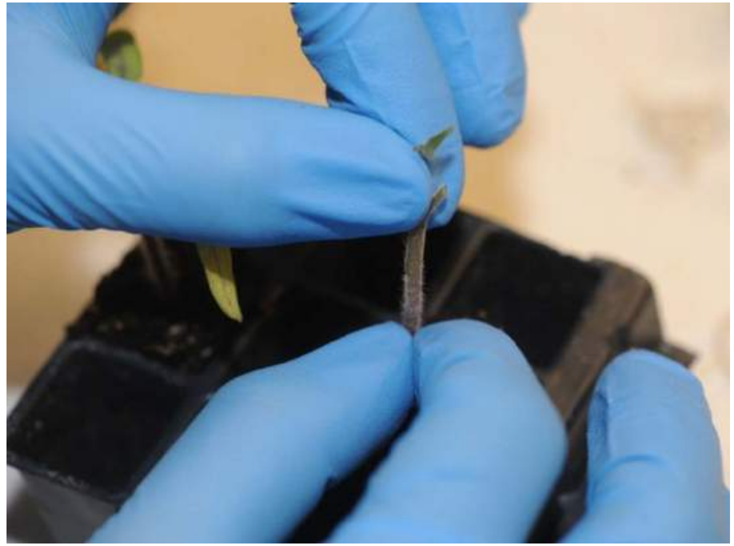


Remove the shoot of the rootstock seedling with one cut approximately 5 mm below the cotyledons. The angle of the cut can vary according to your preference and experience. However, the surface area of the graft union increases with the angle of the cuts made on both seedlings. A cut of approximately 45 degrees may be best.



Prepare the scion by separating it from its roots with a diagonal cut across the stem. The cut can be above or below the cotyledons but it needs to be at a point where the scion stem diameter will match the rootstock stem diameter. Likewise, for healing and optimum graft quality, the cut made to de-root the scion should be of a similar angle to the cut made to de-foliate the rootstock.

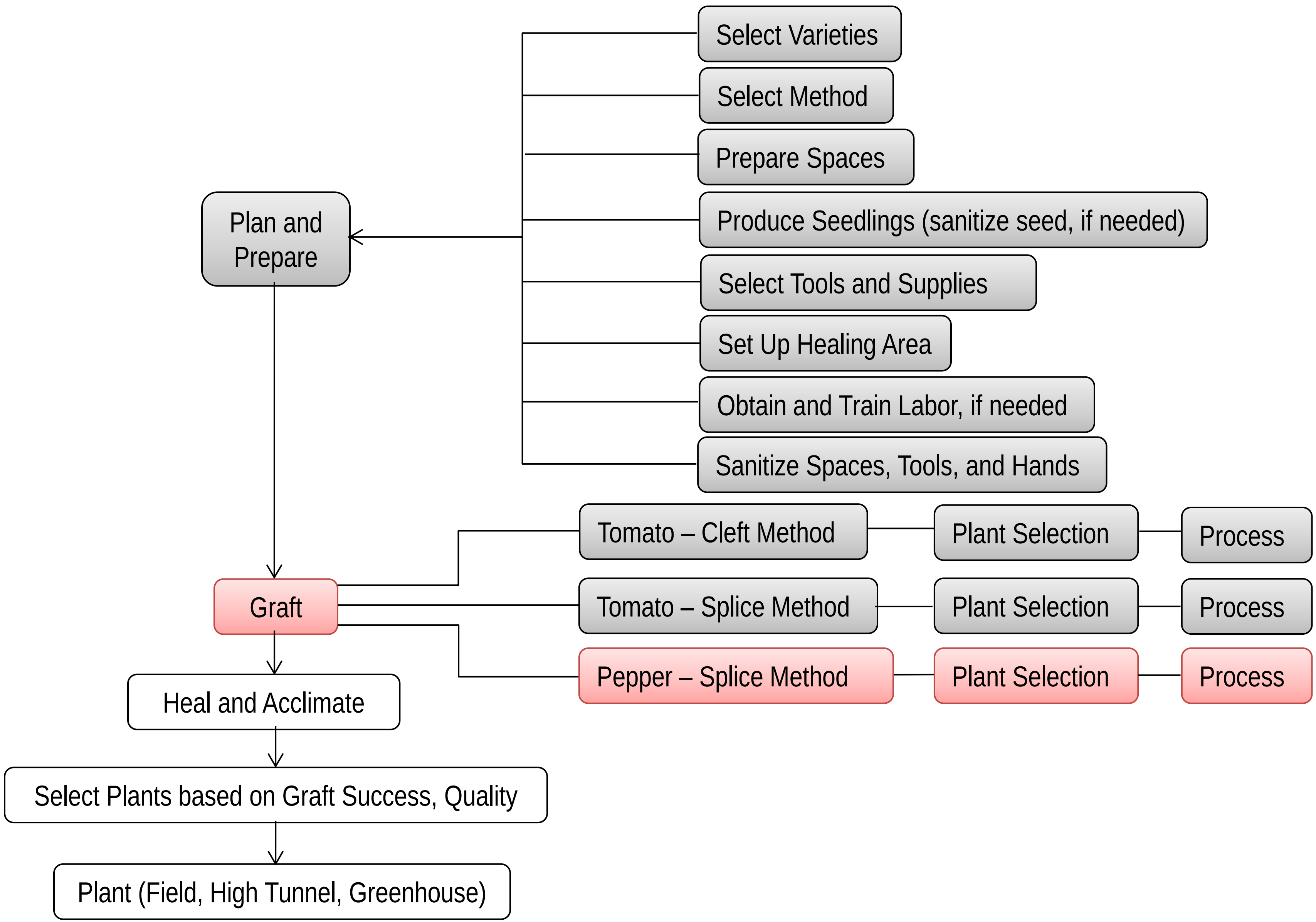




Carefully place the cut edge of the scion on the cut edge of the rootstock in a manner that allows the angles of the cut to match as closely as possible.

Position the clip around the graft union in a manner that securely connects the scion and rootstock. A spring-loaded clip with two possible diameter positions (1.5 and 4 mm) is shown here. The clip as shown is used on the 1.5 mm diameter position on the clip.

Newly splice-grafted and clipped tomato plants.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Select Method
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Graft
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Plant Selection
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Pepper – Splice Method
Plant Selection
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Heal and Acclimate
Select Plants based on Graft Success, Quality
Plant (Field, High Tunnel, Greenhouse)

Splice Grafting Overview—Pepper

1) Remove the shoot of the rootstock by making a cut at an angle of about 45-75° below the cotyledons.

2) If using a self-made plastic tube clip, place it on and around the rootstock stump.

3) Remove the roots of the scion by making a cut at the same angle as the rootstock cut was made above or below the cotyledons where the stem is similar in diameter to that of the rootstock stump.

4) Optional: Remove leaves from scion; do NOT damage the youngest leaf or apical meristem.

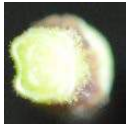
5) Place the prepared scion shoot on the cut end of the rootstock stump to allow the two cut edges to match as closely as possible.

6) Pull the self-made clip upward to secure the graft union or attach a commercial clip or use glue.

Experienced grafters cut and join seedlings using individualized versions of the major steps above. Their goals are always: 1) to complete the steps carefully and quickly and 2) to insure that the two cut surfaces are joined firmly and in the correct orientation. They also clean tools or change to a clean, new razor blade frequently. Once plants and other materials are in place, experienced grafters can prepare a grafted plant in much less than one minute.

Scion

Jalapeño M



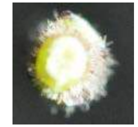
Early Sunsation

Aristotle

Cayenne

Hungarian Hot Wax

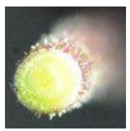
Thai Hot



1 mm

Rootstock

Foundation



TI-135

Scarface

Dorado

Both tomato and pepper can be grafted effectively when stem diameters are 1.5 mm – 2.5 mm. However, pepper can be grafted effectively at the lower end of this range, when plants are younger. In fact, some recommend pepper to be grafted when stem diameters are approx. 1.8 mm. Regardless, pepper rootstock and scion stem diameters must be matched at grafting. Pictures shown here were taken 38 days after seeding.

Healthy, uniform scion (left) and rootstock (right) seedlings with 2-4 true leaves are ready for grafting.

Pepper seedlings with stem diameters about 1.5-2.5 mm can be grafted, although a diameter of 1.8 mm may be best. Regardless, as with tomato, rootstock and scion stem diameters should be similar.

Remove the shoot of the rootstock seedling with one cut approximately 5 mm below the cotyledons. The angle of the cut can be about 45-75°. It is important for the angle of cut on the rootstock and scion to be similar. A Japanese-made grafting clipper is used here to make the angled cut.

Prepare the scion by trimming a portion of the plant stem with a single cut above or below the cotyledons.

Leaves trimmed, self-made clips.

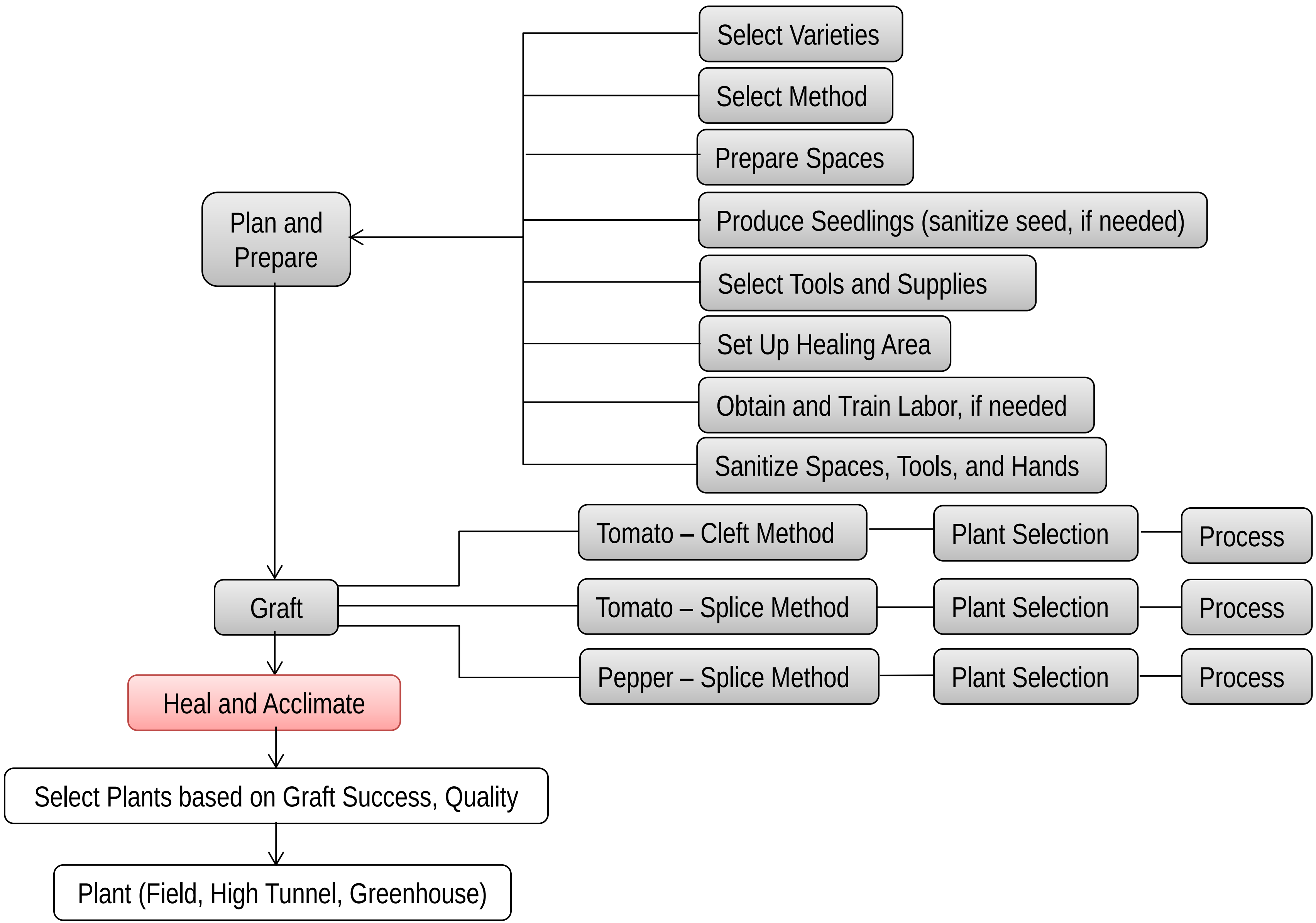
Carefully place the cut edge of the scion on the cut edge of the rootstock in a manner that allows the angles of the cut to match as closely as possible. Pepper splice grafts may require very careful clip selection. Self-made and commercial clips can be used; however, commercial clips may be preferable.

Leaves not trimmed, commercial clips.

Newly splice-grafted pepper plants

Leaves trimmed, self-made clips

Leaves not trimmed, commercial clips



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Healing Area/Chamber

Place newly grafted plants into healing chambers as soon as possible after grafting. There is no difference in construction and conditions of the healing chamber for grafted pepper and tomato plants. However, tomato plants tend to heal more quickly, requiring 7-10 days instead of about 15 days for pepper. During the first week of healing, maintaining relative humidity above 90% and temperature at approximately 21-27/29 °C, 70-80/84 °F night/day appears to be best. The effects of light level are direct and indirect and must be considered carefully. Higher light intensity (e.g., about 300 µmol/m^2/s) can benefit the healing process IF humidity and temperature can be maintained at target levels. If the capacity to control humidity and temperature under high light intensity is limited, light may need to be lowered (e.g., using shade cloth) to moderate the temperature and relative humidity. From the second week of healing, humidity can be lowered gradually, while light should be increased if shading is applied in the first week.

During the second week of healing, humidity can be gradually decreased by opening the chamber and light can be increased by removing the shade cloth. Temperature remains at 21-27/29 °C, 70-80/84 °F night/day. Graft failure tends to become evident at this stage.

Misters, cool-air humidifiers, and irrigation on the capillary mats can help maintain humidity levels in healing chambers and are recommended instead of overhead hand-watering. Regardless, do not saturate the rooting medium as doing so may increase “root pressure” (pressure exerted by the upward flow of water from root to growing tip) weakening the graft union.

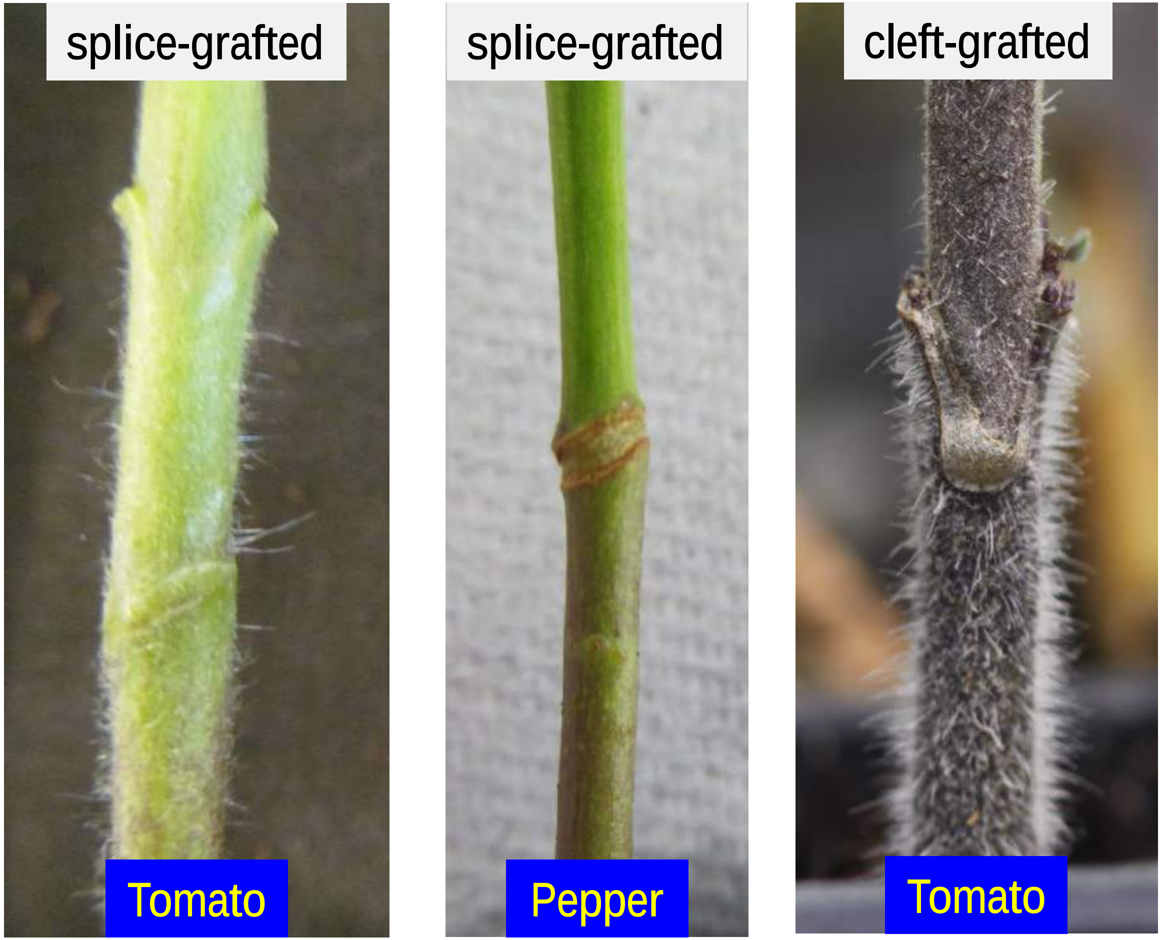
Take care to NOT disrupt the graft union in any way, including by applying water. Monitor light, temperature, and humidity levels in all healing areas because these factors strongly influence grafted plant survival.

Plants 1 hour after grafting.

Healthy grafted plants 1-2 weeks after grafting.

Note new growth on the scion, which becomes evident beginning about 1 week after grafting.

High-quality grafts after healing. Vascular connections appear to be strong. Callus formation on the graft is associated with a healthy grafted plant suitable for field use. Plants are typically ready for transfer to the field 2–3 weeks after grafting.



More time is required to produce grafted plants ready for transplanting than to produce field ready ungrafted seedlings. Therefore, consider adjusting typical seeding schedules and other practices to offset the greater amount of time required to prepare field ready grafted plants. Otherwise, field planting may be delayed.

High Grafting

Graft position is important because of planting depth and its effects on the handling and performance of plants in the field.

Growers are routinely encouraged to set grafted plants in the field so that graft unions remain above the soil line, especially in disease-infested soil. Keeping graft unions above the soil line is difficult for growers who prefer to use large transplants and plant them deep, burying three or more nodes to encourage additional rooting. Keeping graft unions above the soil line may also be difficult when mechanical transplanters are used. Overall, increasing the length of the rootstock stem can expand the number of production situations in which grafted plants could be used reliably. How to increase the length of the rootstock stem?

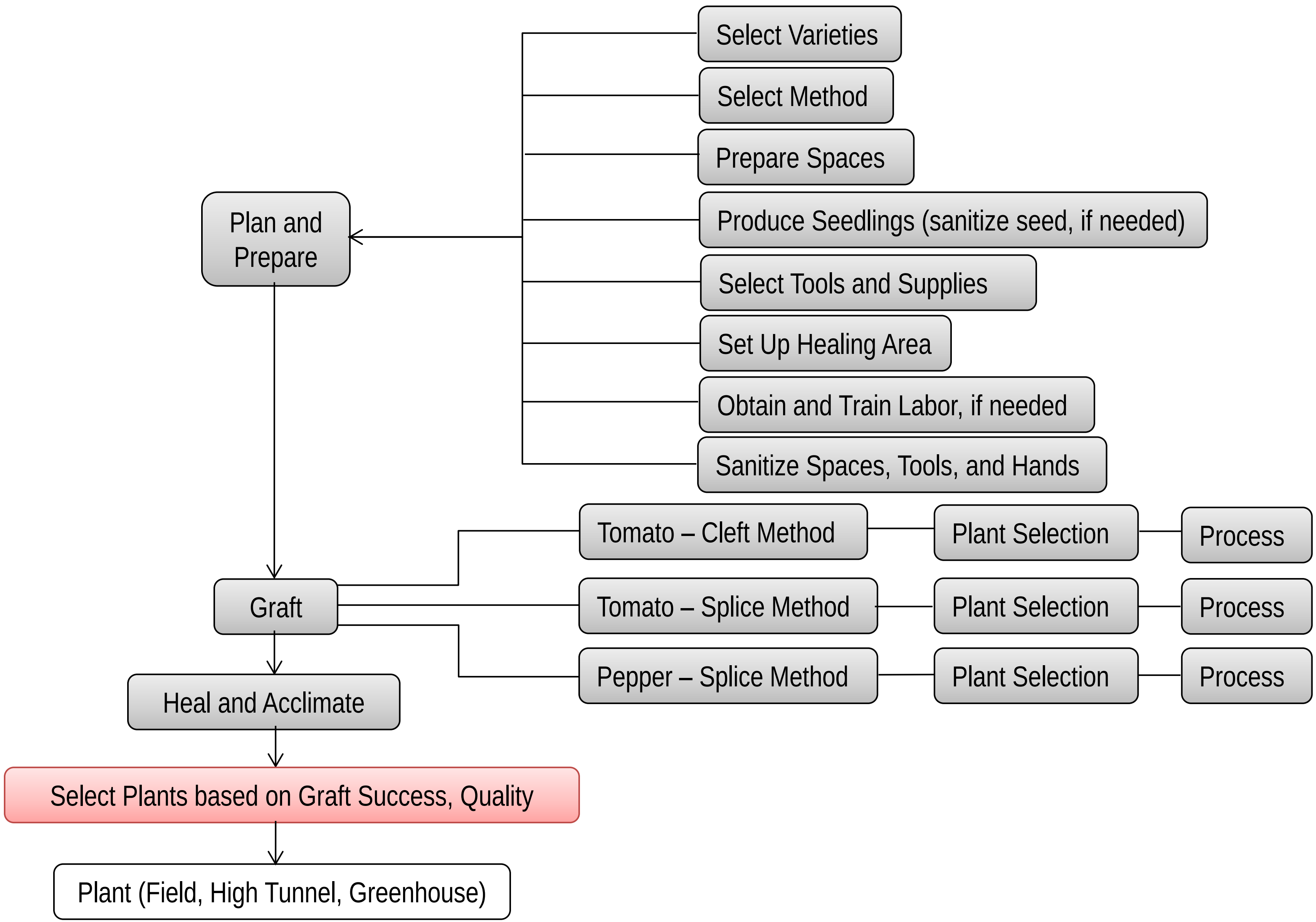
Encouraging the rootstock hypocotyl to ‘stretch’ during seedling production (before grafting) is one option. Specific light, temperature, irrigation, and nutrient level conditions may be required to achieve this goal.

Grafting at a node on the rootstock high above its cotyledonary node (i.e., “high grafting”) is a second option. The high grafting technique is the same as standard cleft of splice grafting. However, high grafting differs from standard grafting in two ways. First, the rootstock seedling is much older and taller than the scion seedling. Second, instead of grafting at the rootstock’s cotyledonary node, the graft is made at a node above it. Patience and practice can allow a grafter to complete high grafts as effectively as they prepare standard grafts, for both tomato and pepper.

High Grafting

High grafting employs the same technique as standard grafting at the cotyledonary node. However, in high grafting, the rootstock seedling is older and taller than the scion seedling. Also, the rootstock seedling is cut at a node above the cotyledons. In these images, the rootstock is cut 3-4 nodes above the cotyledons.

Grafted pepper (left) and tomato (right) plants that differ in the position of the graft union on the rootstock stem: (A) standard position below cotyledons, (B) high position two nodes above the cotyledons, and (C) three or more nodes above the cotyledons. Recall that nodes are sites at which leaves attach to the stem. These plants were grown to be high-grafted. Leaves were removed during seedling production. Removing leaves soon after they arise from the stem results in nodal scars being minimal and nodes being more difficult to identify, as in tomato plants B and C above .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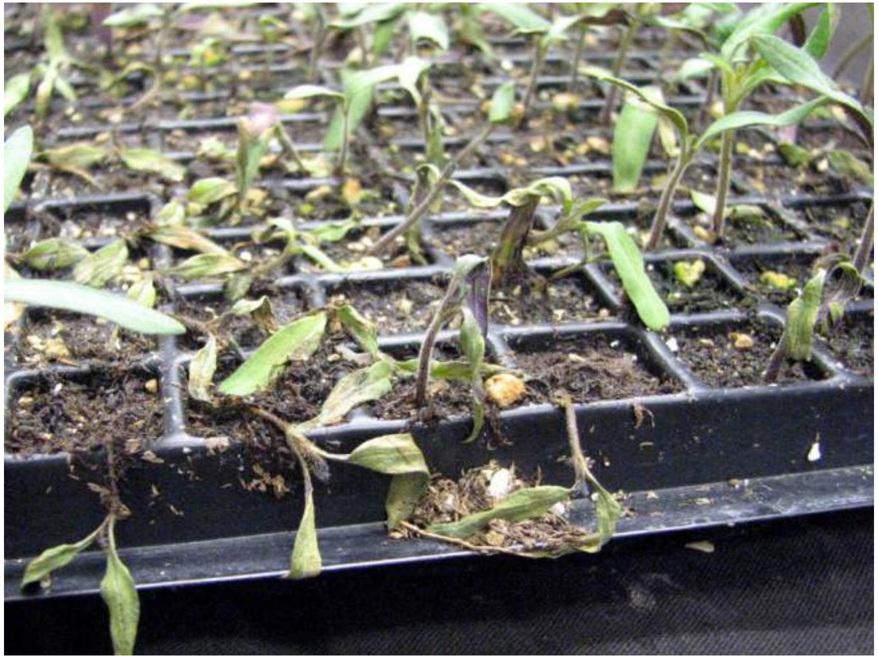
Reasons for Graft Failure

1. Rootstock and scion varieties that are genetically incompatible.
2. Insufficient sanitation at any stage from seed sowing through grafted plant healing. Disease inoculum can be spread easily by grafting, especially mechanically when materials or hands are contaminated. Conditions during seedling production and healing can promote disease, if inoculum is present.
3. Selected seedlings that are unhealthy, have dissimilar stem diameters, or of the improper age (e.g., too woody).
4. Poor grafting technique.
5. Improper management of or conditions in the healing area/chamber, including:
    a. extreme temperatures that desiccate plants or slow rootstock-scion connection;
    b. extreme humidity levels that soften and break down the graft union or allow it to dry;
    c. extreme root-zone moisture levels that weaken the rootstock, at minimum, or create excessive “root pressure” (pressure exerted by the movement of water from the root to shoot), especially early in healing, that strains the graft union;
    d. very low light levels during the later stages of healing; and
    e. mechanical (bending) stress on the graft union (e.g., by contacting the plants or using forceful overhead watering).
6. Insufficient healing period.

Some varieties tend to either produce shoots from the rootstock or roots from the scion. Neither are a form of graft failure but both conditions are undesirable and should be monitored and corrected.

Examples of Undesirable Grafting Outcomes



Poor sanitation at any stage from sowing through healing can lead to serious disease problems and reduce yield and quality.

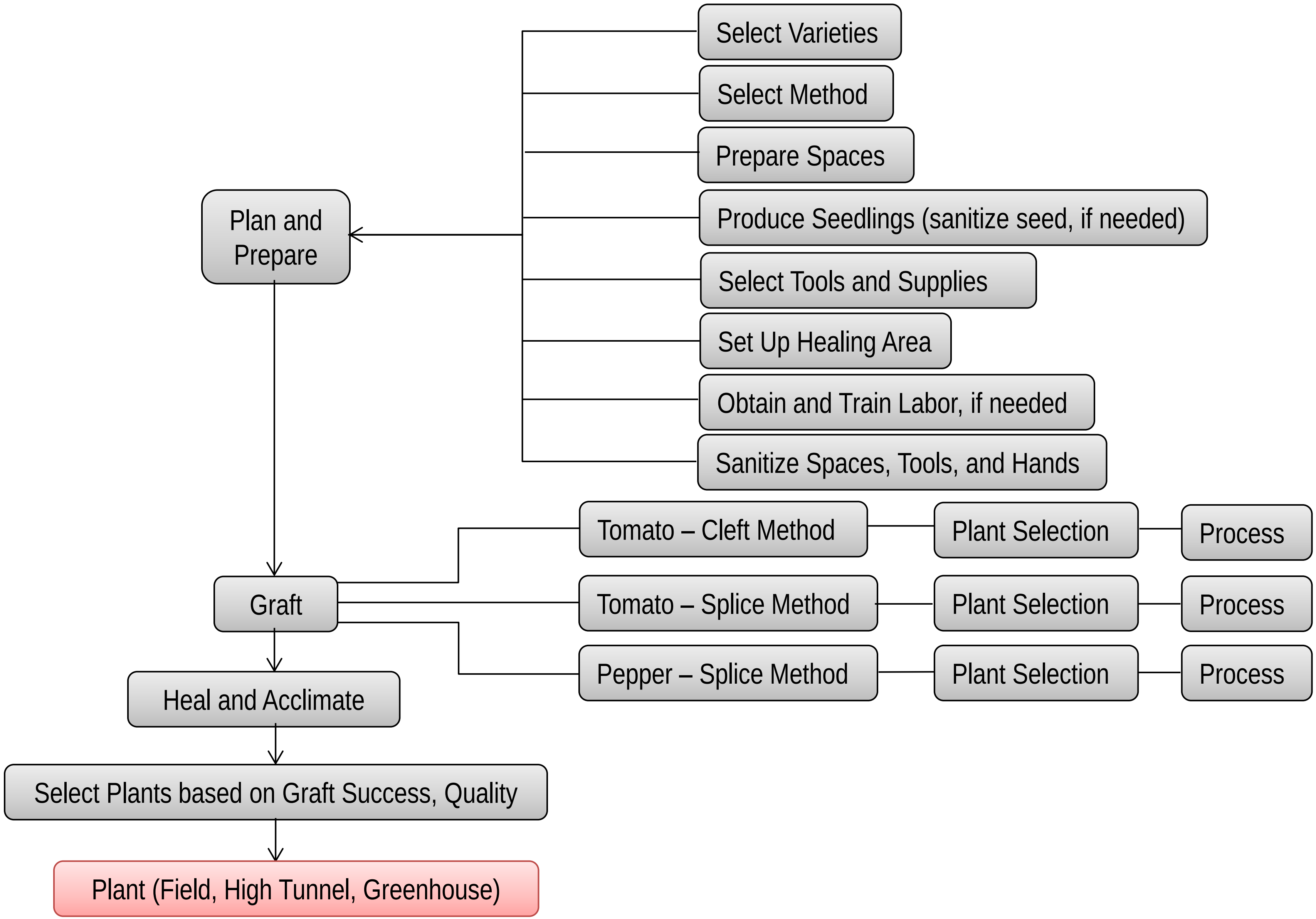
Unwanted environmental conditions in the healing area stresses plants, lowers the number of successful grafts, increases costs, and decreases profit potential.

Low quality grafts resulting from improper grafting technique and/or sub-optimal conditions during healing.

Adventitious roots forming on the scion or shoots forming on the rootstock are undesirable. Both conditions can be the characteristic of the varieties or the result of improper grafting technique or sub-optimal conditions during healing (Bausher, 2011).

Adventitious shoots may develop on the rootstock when the graft union is above the rootstock cotyledons.

Adventitious roots may develop on the scion when the graft union is below the scion cotyledons or when humidity in the healing area is excessive and grafted plants are well-healed.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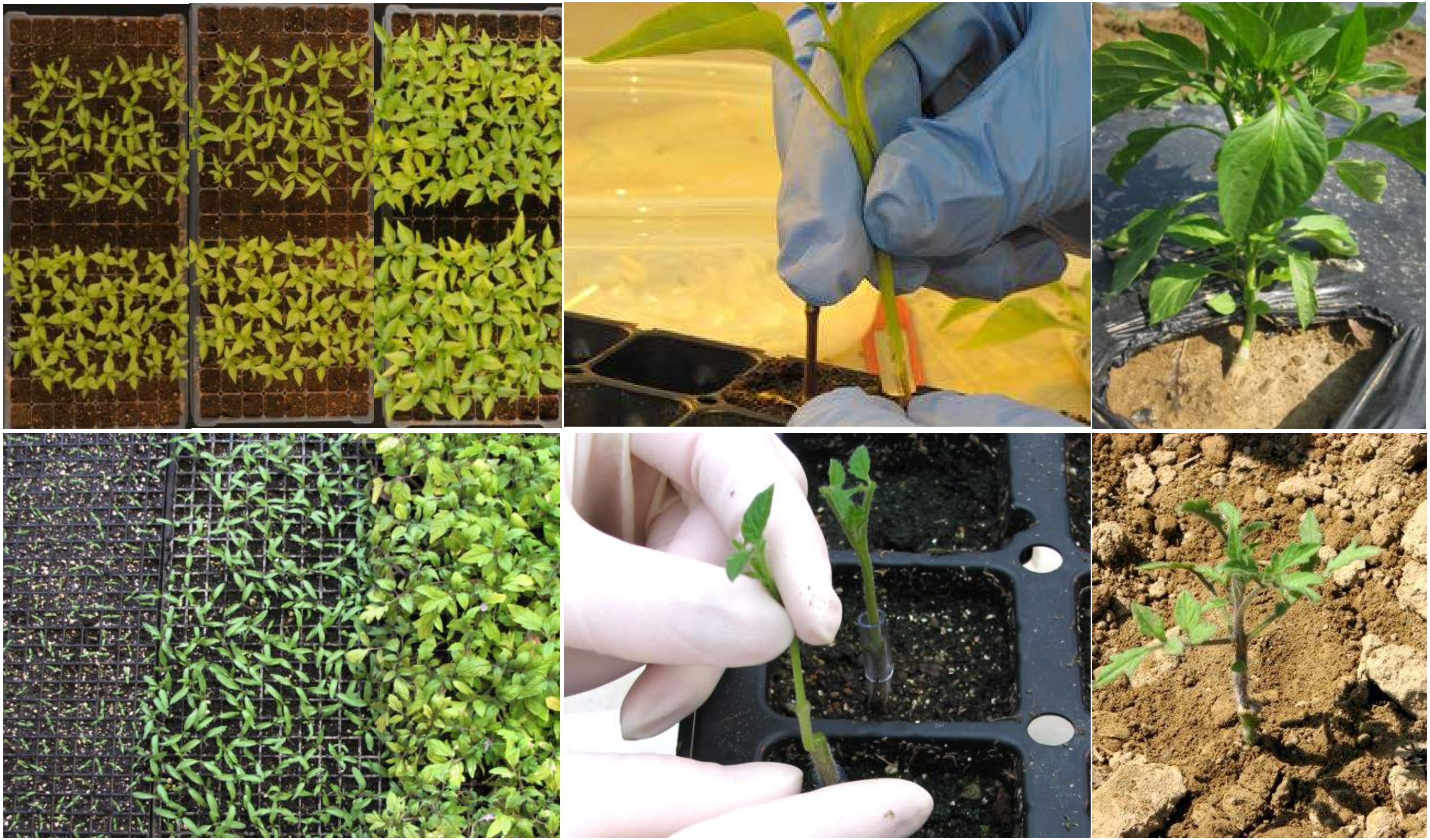
Grafted tomato (left) and pepper (right) plant ready for transplanting.

Setting plants in field can be done by hand or by machine.

A healthy grafted plant in the field. The graft union remains 2.5 cm (1 in.) above the soil line to prevent the scion from developing roots, which is unwanted when the scion is known to be susceptible to a disease known to be in the soil.

The above image depicts that some clips (especially tube) will be removed automatically after planting by normal stem growth.



For additional information, please visit the grafting information portal at http://www.vegetablegrafting.org.

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THE OHIO STATE UNIVERSITY EXTENSION

Extension FactSheet

Plant Pathology, 2021 Coffey Road, Columbus, Ohio 43210

Hot Water Treatment of Vegetable Seeds to Eradicate Bacterial Plant Pathogens in Organic Production Systems

Sally A. Miller
Melanie L. Lewis Ivey

One way plant pathogens are introduced into a crop is on infested seeds. Bacterial pathogens are particularly notorious for this method of spread. In general, the earlier a pathogen comes in contact with the crop, the greater the potential for a serious disease problem to develop. This is why it is very important to start with "clean" seeds (i.e. those that are free of pathogens). Bacterial pathogens on or within the seeds can be killed by treating the seeds with hot water.

When hot water-treating vegetable seeds it is critical to follow the instructions exactly, as seeds may be damaged by the treatment and/or the pathogen may not be completely eliminated. A few seed companies hot water-treat their seed prior to sale. Check all seed packages before applying the treatment to be certain that they have not hot water-treated the seeds. Seeds may be damaged if they are hot water-treated twice. In addition, old or poor quality seed can be injured by seed treatments. **Therefore, it is recommended that a small sample be treated and tested for germination prior to treating the entire seed lot (see method below).** The treatment should only be applied to raw seeds (without pelleting or films). Since seeds used in organic production systems are not treated with a synthetic fungicide to control fungal pathogens that cause damping-off, good cultural, biological and sanitation procedures are critical to prevent the introduction of these fungi. Such practices include:

1. Keeping the greenhouse CLEAN and not allowing seedlings, planting mix, or plants to come in contact with outside soil.
2. Only using well or city water to water seedlings and plants.
3. Using a pathogen-free planting mix. Mixes containing a high quality compost can be suppressive to *Pythium* and some other damping-off organisms.
4. Using an Organic Materials Review Institute (OMRI)-approved biopesticide if damping-off continues to be a problem.
5. Maintaining greenhouse environmental conditions that are optimal for seed germination but not for pathogen development. **Do not over-water seedlings, allow soil temperatures to become too cool, or overheat the greenhouse.**
6. If available, select disease-resistant varieties.

Hot Water Treatment

Properly used, hot water treatment kills most bacterial disease-causing organisms on or within seeds. This treatment is suggested for seeds of eggplant, pepper, tomato, carrot, spinach, lettuce, celery, cabbage, turnip, radish, and other crucifers. **Seeds of cucurbits (squash, gourds, pumpkins, watermelons, etc.) can be severely damaged by hot water and thus should NOT be treated.**

Instructions

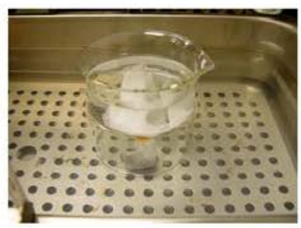
A. The following equipment and supplies are needed to hot water treat organic vegetable seeds:

- Water bath (preferably two: one for pre-warming and one for treatment; Sources: Fisher Scientific Co., Thomas Scientific, VWR Scientific)
- Thermometer
- Cotton cloth, cotton bags, or nylon bags
- Screen for seed drying

B. How to Hot Water-Treat Seeds

Step 1: Wrap seeds loosely in a woven cotton (such as cheesecloth) or nylon bag.

Step 2: Pre-warm seeds for 10 minutes in 100°F (37°C) water.



Step 3: Place pre-warmed seeds in a water bath that will constantly hold the water at the recommended temperature (see table that follows). **Length of treatment and temperature of water must be exactly as prescribed.** If water is too hot or treatment is too long, seeds may be damaged.

Type of seeds	Water temperature °F	Water temperature °C	Minutes
Brussels sprouts, eggplant, spinach, cabbage, tomato	122	50	25
Broccoli, cauliflower, carrot, collard, kale, kohlrabi, rutabaga, turnip	122	50	20
Mustard, cress, radish	122	50	15
Pepper	125	51	30
Lettuce, celery, celeriac	118	47	30

Step 4: After treatment, place bags in cold tap water for 5 minutes to stop heating action.

Step 5: Spread seeds in a single, uniform layer on screen to dry.

How to Test for Seed Germination After Hot Water Treatment

1. Mix seeds thoroughly in each seed lot and count out 100 seeds per seed lot.*
2. Treat 50 of the seeds exactly as described in the fact sheet.
3. After treated seeds have dried, plant the two groups of seeds separately in flats or pots containing planting mix according to standard practice. Label each group as "treated" or "untreated".
4. Allow the seeds to germinate and grow until the first true leaf appears (to allow for differences in germination rates to be observed).
5. Count seedlings in each group separately.
6. Determine the percent germination in each group:

$$\text{percent germination} = \frac{\text{number of seedlings emerged}}{\text{number of seeds planted}} \ (X\ 100)$$

7. Compare percent germination in each group: they should be within 5% of each other.

* If seed supply is limited, use a smaller number (at least 30) of seeds to test germination.

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THE OHIO STATE UNIVERSITY EXTENSION

Extension FactSheet

Plant Pathology, 2021 Coffey Road, Columbus, Ohio 43210

Hot Water and Chlorine Treatment of Vegetable Seeds to Eradicate Bacterial Plant Pathogens

Sally A. Miller

Melanie L. Lewis Ivey

One of the ways plant pathogens are introduced into a crop is on seeds. Bacterial pathogens are particularly notorious for this means of dissemination. In general, the earlier a pathogen comes in contact with the crop, the greater the potential for a serious disease problem to develop. This is why it is very important to start with "clean" seed. Clean seed can be obtained by applying one of the treatments described below to kill bacterial pathogens on and/or within the seed.

When treating vegetable seeds it is critical to follow the instructions exactly, as seeds may be damaged by the treatment and/or the pathogen may not be completely eliminated. In addition, old or poor quality seed can be injured by seed treatments. **Therefore, it is recommended that a small sample be treated and tested for germination (see method below) prior to treating the entire seed lot**. Treatments should be done on raw seed only, since the treatment will destroy any seed pelleting and will wash off any fungicide that may have been applied to the seed. If fungicide treated seeds are used, the fungicide washed off must be disposed of properly. After the treatment, seed may be treated with Thiram to prevent damping-off caused by various soilborne fungi.

Hot Water Treatment

Properly used, hot water treatment kills most bacterial disease-causing organisms on or within seed. This treatment is suggested for seeds of eggplant, pepper, tomato, carrot, spinach, lettuce, celery, cabbage, turnip, radish, and other crucifers. **Seeds of cucurbits (squash, gourds, pumpkins, watermelons, etc.) can be damaged by hot water and thus should not be treated.**

Instructions

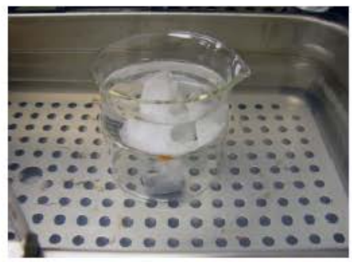
A. The following equipment and supplies are needed to hot water treat vegetable seed.

- Water bath (preferably two: one for pre-warming and one for treatment; Sources: Fisher Scientific Co., Thomas Scientific, VWR Scientific)
- Thermometer
- Cotton cloth, cotton bags, or nylon bags
- Screen for seed drying

B. How to Hot Water Treat Seed.

Step 1: Wrap seeds loosely in a woven cotton bag (such as cheesecloth) or nylon bag.


Step 2: Pre-warm seed for 10 minutes in 100°F (37°C) water.

Step 3: Place pre-warmed seed in a water bath that will constantly hold the water at the recommended temperature (see table that follows). **Length of treatment and temperature of water must be exactly as prescribed.**


Step 4: After treatment, place bags in cold tap water for 5 minutes to stop heating action.

Step 5: Spread seed in a single, uniform layer on screen to dry. Do not dry seed in area where fungicides, pesticides, or other chemicals are located.

Step 6: Dust seed with Thiram 75WP (1 tsp/1 lb seed) once the seed is completely dry.

Seed	Water temperature		Minutes
	°F	°C	
Brussels sprouts, eggplant, spinach, cabbage, tomato	122	50	25
Broccoli, cauliflower, carrot, collard, kale, kohlrabi, rutabaga, turnip	122	50	20
Mustard, cress, radish	122	50	15
Pepper	125	51	30
Lettuce, celery, celeriac	118	47	30

Chlorine Treatment

Chlorine treatment effectively removes bacterial pathogens on the seed surface. Unlike hot water treatment it does not eliminate pathogens within the seed. Chlorine treatment is recommended for both large- and small-seeded vegetables if the seeds have not been treated by any other method and the possibility of pathogens being carried inside the seeds is not a concern.

Instructions

A. The following equipment and supplies are needed to chlorine treat vegetable seed.

- Chlorox (5.25% hypochlorite)
- Surfactant (e.g., Activator 90 or Silwet)
- Glass beakers or jars
- Stirring stick
- Screen for seed drying

How to Test for Seed Germination After Hot Water or Chlorine Treatment

1. Mix seeds in each seed lot and count out 100 seeds per seed lot.
2. Treat 50 of the seeds exactly as described in the fact sheet.
3. After treated seeds have dried, plant the two groups of seeds separately in flats containing planting mix according to standard practice. Label each group as "treated" or "untreated."
4. Allow the seeds to germinate and grow until the first true leaf appears (to allow for differences in germination rates to be observed).
5. Count seedlings in each group separately.
6. Determine the % germination in each group:

$$\frac{\text{\# seedlings emerged}}{\text{\# seeds planted}} \times 100$$

7. Compare % germination in each group: they should be within 5% of each other.

Conversions:

8 oz = 1 cup

16 oz = 1 pint

32 oz = 1 quart

128 oz = 1 gallon

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