<table>
<thead>
<tr>
<th><strong>Title</strong></th>
<th>Grafting of Vegetable Crops</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Author(s)</strong></td>
<td>ODA, Masayuki</td>
</tr>
<tr>
<td><strong>Editor(s)</strong></td>
<td></td>
</tr>
<tr>
<td><strong>Citation</strong></td>
<td>大阪府立大学大学院農学生命科学研究科卒業研究</td>
</tr>
<tr>
<td><strong>Issue Date</strong></td>
<td>2002-03-31</td>
</tr>
<tr>
<td><strong>URL</strong></td>
<td><a href="http://repository.osakafu-u.ac.jp/dspace/">http://repository.osakafu-u.ac.jp/dspace/</a></td>
</tr>
<tr>
<td><strong>Rights</strong></td>
<td></td>
</tr>
</tbody>
</table>
Grafting of Vegetable Crops

Masayuki ODA

(Laboratory of Plant Propagation, Graduate School of Agriculture and Biological Sciences
Osaka Prefecture University, Sakai, Osaka 599-8531, Japan)

Introduction

Grafting is essential for fruit production, and as a result, a great deal of information is available on the practice and physiology of grafting for fruit trees. Knowledge of fruit tree grafting can be gathered from papers, reviews (Andrews and Marquez, 1993; Rogers and Beakhane, 1957) and books (Rom and Carlson, 1987), but there is very little written on vegetable grafting.

The origin of grafting can be traced back to ancient times. The technique has been very popular for the propagation of fruit and nut trees. The Chinese knew about grafting since at least 1560 BC. In ancient Greece, Aristotle (384-322 BC) and Theophrastus (371-287 BC) discussed grafting in their writings with considerable expertise (Hartmann et al., 1997).

Early in the 20th century, grafting was applied to vegetables through studies on graft hybridisation, vernalization and photoperiodic responses in Solanaceae, Compositae, and other herbaceous plants (Oda, 1990).

Grafting was introduced into vegetable production to control soil-borne diseases and nematodes. In contrast, its use in fruit tree production was to propagate heterogeneous plants. Grafting was first used in vegetable production in Japan and Korea in the late 1920s, when watermelons (Citrullus lanatus) were grafted onto bottle gourd (Lagenaria siceraria) rootstocks (Lee, 1994). Scarlet eggplant (Solanum integrifolium) rootstocks were introduced into the production of eggplants (Solanum melongena) in the 1950s.

Grafting was later applied to the production of cucumber (Cucumis sativus) and tomato (Lycopersicum esculentum) from around 1960 and 1970, respectively (Oda, 1995, 1999). In 1990, grafted plants in Japan and Korea accounted for 59% and 81% respectively of the total area of fruit producing vegetables (i.e., watermelon, cucumber, melon (Cucumis melo), tomato and eggplant) in those countries (Lee, 1994).

Since the early 1990s, researchers in European countries have been eager in the use of vegetable grafting for least input sustainable agriculture. Information on vegetable grafting is sparse, however, so the aim of this review is to gather information that is available for easy reference.

I have concentrated mainly on papers written in English, so that readers can easily refer to the original papers. As a result, discussion on graft hybridisation papers written in Russian before 1960, and those on rootstock breeding and the practice of vegetable grafting written in Japanese, Korean and Chinese are limited. I believe that there are many books on vegetable grafting published in Japan and other Asian countries that could be useful for those involved in low input sustainable production of fruit-bearing vegetables. As those books are written in Asian languages, however, they will need to be translated before they can be of use to workers using vegetable grafting techniques in other countries.

General Effects of Grafting

Growth and yields

It is well known that rootstocks affect the growth and yield of scion plants. In vegetable crops, grafting is often introduced to give the crops vigour. Cucumber plants grafted onto pumpkin plants grown in sand culture were larger in dry mass than cucumber autografts (Shimada and Montani, 1977). Tomato plants grafted onto disease-resistant rootstocks 'K', 'KV', 'KVF', and 'KN' were more vigorous and
gave higher yields than ungrafted plants (White, 1963).

Grafting sometimes depresses the growth and yield of scion plants depending on the species or varieties of rootstocks used. Yields of tomato plants grafted onto Datura tatula were far below those of ungrafted tomato plants (Kramer, 1957) Solanum sodomaeum and S auriculatum rootstocks restricted the growth and reduced the fruit size of tomato scions. However, plants grafted onto S laciniatum withstood water-saturated soil conditions better than ungrafted tomato plants in the same field, and continued to grow for almost 2 months longer than ungrafted plants (Shackleton, 1965).

The growth of tomato plants grafted onto eggplants was restricted and the fruit yield was 1/5 of tomato autografts (Abdelhaffiz et al., 1975). The incidence of overgrowth and unmarketable fruit brought about by planting tomato plugs were controlled by grafting on some Solanum rootstocks (Oda et al., 2000).

Physiological disorders

Grafting can often cause physiological disorders and other undesirable characteristics in the ‘Prince’ melon, grafting onto squash (Cucurbita spp) rootstocks can cause physiological disorders such as green stripe, green spots and yellow mottle on fruit skin. It can also cause abnormal fermentation of fruit. Melon plants grafted onto ‘Shuntosa No 1’ (C maxima x C moschata) rootstock grew vigorously and produced higher yields than those of melon autografts, but there was a reduction in soluble solids in the fruit. A high level of green stripe and abnormal fermentation developed by the rootstock, but not much yellow mottle. When ‘Kongo’ rootstock (C moschata x C moschata) was used, the fruit grew large and contained a lot of soluble solids, but fermented abnormally. On ‘Shirakikuza’ (C moschata) rootstock, the scion plants did not grow so vigorously and a lot of yellow mottle developed, but there was little green stripe or green spot (Matsuda and Honda, 1981).

Sudden wilt was induced when watermelons were grafted onto bottle gourd (Lagenaria siceraria) and inoculated with an extremely low level of Fusarium oxysporum f sp lagenariae. The same level of inoculum failed to induce any marked symptoms when inoculated into bottle gourd seedlings or even into the stem or roots of rootstock. The inoculation of even a narrow part of the root system of the grafted plants was enough to bring about sudden wilt (Kuwata et al., 1981).

A fungus was isolated from rotted roots in the bottle gourd rootstocks of collapsed watermelon plants. The fungus was identified as Monosporascus cannonballus, which is known to be a soil-borne pathogen of melon. The pathogenicity of two isolates was positive on bottle gourd rootstock of watermelon plants as well as on melon plants under greenhouse conditions (Uenatsu et al., 1992).

Viral wilt

Wilt in grafted plants is sometimes attributed to viral diseases. Wilt was observed in cucumber plants grafted onto squash rootstocks. In the wilted plants, mosaic symptoms appeared on the upper leaves but not on the stem and roots. Ungrafted cucumber plants inoculated with leaf samples showing wilt symptoms in the field did not show any wilt symptoms. When ungrafted cucumber plants were inoculated with an isolated zucchini yellow mosaic virus (ZYMV), cucumber mosaic virus (CMV) and watermelon mosaic virus-2 (WMV-2) and their combinations, wilt symptoms were not observed in the ungrafted cucumber plants. In the grafted plants, however, inoculation with ZYMV and a combination of CMV and WMV-2 caused slight wilt in some plants. It was also found that mixed infection with CMV and ZYMV, or CMV, ZYMV and WMV-2 caused severe wilt on the grafted cucumber plants whereas single infection with ZYMV or mixed infection with CMV and WMV-2 caused only slight wilt. Inoculation with CMV or WMV-2, and with a combination of ZYMV and WMV-2 did not cause wilt symptoms (Iwasaki and Inaba, 1988).

The effect of 16 different cucurbit rootstocks on the incidence of viral wilt in grafted cucumber plants was studied (Iwasaki and Inaba, 1990). One leaf from cucumber plants
grafted onto various rootstocks was mechanically inoculated with a mixture of CMV and ZYMV, and the severity of viral wilt was classified as none (-), slight wilt (+), severe wilt (++), and plant death (+++). The plants grafted onto C. lanatus, C. melo 'reticulatus,' C. ficifolia, C. maxima, C. maxima x C. moschata and C. pepo showed severe wilt, with a high frequency of ++ and +++ scores. The plants grafted on Luffa cylindrica and Scolymus angulatus showed severe wilt. There was a low frequency of ++ and +++ scores on the 14th day after inoculation, and a high frequency on the 21st day after inoculation. The plants grafted onto Benincasa hispida, Cucumis melo var. acdulcus, Lagenaria siceraria var. hispida and L. siceraria var. microcarpa showed a low frequency of severe wilt on the 14th and 21st days after inoculation. Wilt was not observed in the plants grafted on C. melo var. conomon, C. sativus or L. siceraria var. gourda.

Flower formation

When gynomonoecious melons were grafted onto gynoecious melons, andromonoecious melons and pumpkin seedlings, the andromonoecious rootstock induced the formation of staminate flowers on the scion, and the pumpkin had a similar but smaller effect (Mockaitis and Kivilaak, 1964).

Flowering is generally promoted by grafting in cactus and sweet potatoes. In tomato, however, attempts to improve flowering by grafting were not successful (Coggins and Lesley, 1968).

Fruit quality

Rootstocks affect the quality of fruit born on scions. For example, it is well known that grafting onto some varieties of Cucurbita spp rootstocks degrades the fruit shape and taste of watermelon and melon. The fruit quality of tomato was slightly lowered by grafting, when compared with that from ungrafted plants in sterilised soil (Harnett, 1974).

Grafting tomato plants onto Solanum integrifolium increased the sugar contents of tomato fruit (Oda et al., 1996). Another study also showed that the quality of sugars and organic acids in tomato fruit were not affected by grafting onto three Solanum rootstocks (S. sisymbriifolium, S. torvum and S. toxicityramum) that are resistant to soil-born diseases (Matsuzoe et al., 1996).

A few types of Cucurbita spp rootstocks give rise to cucumber fruit known as 'bloomless.' When cucumber scions were grafted onto bloomless rootstock 'Unryu No.1,' bloom on the cucumber fruit disappeared. The main component of the bloom was silica, and the silica disappeared from leaves by grafting cucumber plants on bloomless rootstocks (Yamamoto et al., 1989).

Induced resistance to diseases and insect pests

Resistance to shoot diseases can be induced by grafting. Graft chimeras were generated using Lycopersicon pennellii and L. esculentum to determine the contribution of the three meristem layers (L1, L2 and L3) to trichome density, sugar ester production and aphid resistance. Sugar esters, in the form of tracylglucoses, have been implicated in aphid resistance in L. pennellii. One chimera possessed the epidermal layer (L1) of L. pennellii and the internal tissues (L2 and L3) of the aphid-susceptible L. esculentum. The second chimera had both the L1 and L2 of L. pennellii and the L3 of L. esculentum. Type IV trichome densities did not differ significantly among the chimeras and L. pennellii. Both chimeras accumulated sugar esters with similar sugar and fatty acid composition as L. pennellii. The concentration of epicuticular sugar ester on the chimeras was also comparable with that of L. pennellii. It was found that both chimeras are as resistant to aphids as L. pennellii. Resistance could also be reduced on the three types of plants by removal of the type IV trichomes. The amount and type of sugar esters produced were features determined by the genotype of the epiderms. These epidermal features were sufficient to account for the aphid resistance observed in L. pennellii (Goffreda et al., 1990).

The lowest number of eggplant shoots and L. orbonalis-infested fruit was recorded from plants grafted onto wild Solanum plants At
the flowering and early fruiting stages the incidence of whitefly (Bemisia tabaci) was low in grafted tomato plants. This may be why there was less infestation by tomato yellow leaf curl geminivirus, since this insect is the only vector of these diseases (Alam et al., 1994).

The induced resistance to parasites requires further study.

**Stress tolerances**

Grafting has been applied to vegetable production to increase stress tolerance. Some examples are the increased cold tolerance of cucumber plants grafted on Cucurbita ficifolia and increased drought tolerance of watermelon plants grafted on ‘Shuntosa’ (Cucurbita maxima x C. moschata).

Cucurbitaceae (Cucurbita spp., Lagenaria siceraria, cucumber, watermelon and melon) have also been tested as rootstocks to provide salt tolerance for cucumber scions. The root growth of Cucurbita spp. was less inhibited by NaCl concentrations of 0, 1,000 and 10,000 mg L⁻¹ than that of L. siceraria. When Cucurbita spp., L. siceraria, Benocasa hispida and cucumber plants were treated with NaCl in their nutrient solution, Cucurbita spp. and L. siceraria showed inhibition of top growth at 2,000 and 4,000 mg L⁻¹ NaCl, respectively, but B. hispida was less tolerant than the others. The concentration of sodium ions in the leaves of treated plants was 4.5% of dry weight in cucumber and watermelon, but 0.1% in Cucurbita spp. and L. siceraria. For cucumber plants grafted onto these rootstocks, the sodium ion concentration in leaves was 0.1%. This was much lower than the concentration of 3 to 5% found in the rootstocks (Matsubara, 1989).

Flooding reduced leaf photosynthetic rate, stomatal conductance, transpiration, soluble protein and activity of ribulose-1,5-bisphosphate carboxylase/oxygenase (Ribisco) in bitter melon (Momordica charantia) grafted onto Luffa cylindrica when compared with unflooded controls, but these reductions were less marked than in ungrafted seedlings (Liao and Lin, 1996).

**Responses to low root temperature**

**Growth** The growth responses to soil temperature of plants grafted onto various rootstocks were compared in cucumber, watermelon, eggplant and tomato (Okumura et al., 1986). The growth of cucumber plants grafted onto figleaf gourd (Cucurbita ficifolia) was suppressed at a high soil temperature but not at a low one, while ‘Shuntosa No 1’ (Cucurbita maxima x C. moschata) showed stable growth at various soil temperatures.

Watermelon plants grafted onto ‘Shuntosa No 1’ had the best resistance to low soil temperatures, and plants grafted onto bottle gourd (Lagenaria siceraria ‘Renshi’ and ‘Sakigake’) and pumpkin (Cucurbita moschata ‘No 8’) were second best. Watermelons grafted onto the same species (Citrullus lanatus) and onto wax gourd (Benocasa hispida) were inferior to the plants on the other rootstocks. Their growth was much better at higher soil temperatures up to 20°C, and the optimal soil temperature for their growth seemed to be 20-23°C.

Eggplants grafted onto ‘Tabyo VF’ (S. integrifolium x S. melongena) grew best at soil temperatures of 18 and 21°C, with the plants grafted on scarlet eggplant (S. integrifolium) and fox face (S. mammosum) coming second. The growth of ‘Torrum bijor’ (S. torvum) rootstock at these temperatures was slightly inferior to the other rootstocks, but this rootstock was the best for resistance to low soil temperatures (12 and 15°C).

In tomato plants, ‘LS-89 (Hawaii 7998)’ (L. esculentum) rootstock promoted growth at higher soil temperatures of 22 and 35°C, but suppressed it at lower soil temperatures. The growth of plants grafted onto ‘KNVF’ (L. esculentum x L. hirsutum) was not only excellent at lower soil temperatures of 10 and 13°C, but was also comparatively good at higher soil temperatures.

**Physiology** The growth of ‘Suyo’ and ‘Kurumeococha H’ cucumber plants was severely suppressed at root temperatures of 12 and 14°C, while figleaf gourd showed a high ability to tolerate these temperatures. The growth of ‘Suyo’ at low root temperatures was significantly improved when grafted onto figleaf
gourd. Highly significant correlations were observed between the degree of growth suppression and decrease in the nutrient concentrations of leaves, particularly in N, P and K (Tachibana, 1982).

The total lipid and lipid-phosphorus concentrations in roots increased at lower root temperatures. These were higher in figleaf gourd, which was more tolerant to low root temperatures than cucumber plant. Linolenate concentration continued to increase in figleaf gourd as the temperature became lower, until it made up 57% of total fatty acids at 12°C. In contrast, it increased only slightly at 15°C in cucumber cultivars (Tachibana, 1986).

The cytokinin concentrations in root xylem exudate of cucumber cultivars decreased sharply at lower root temperatures, whereas the concentrations of that from figleaf gourd were relatively constant at 14-23°C, and increased to a strikingly high level at 11°C. Cytokinin concentrations in roots were highest at 23°C in ‘Suyo’ cucumber, 17°C in ‘Kurumeo-chai H’ cucumber and 12°C in figleaf gourd. The most abundant cytokinin in the roots of figleaf gourd and probably of cucumber cultivars was zeatin riboside, and the composition apparently changed little with root temperature (Tachibana, 1988a).

The in vitro nitrate reductase (NR) activity from roots of both cucumber and figleaf gourd grown at a root temperature of 20°C was greatly reduced by low temperatures in the enzyme assay medium. In contrast, NR activity from roots of plants grown at root temperatures of 13°C or 20°C did not differ with the incubation temperature. The activity of root NR was very low compared to that of leaf NR, particularly in figleaf gourd. The absorption of nitrogen 15 nitrate at 13°C root temperature was significantly lower than that at 20°C, especially in cucumber. In both species, however, the assimilation and translocation of absorbed nitrogen 15 nitrate were little affected by root temperature. Roots of both species accumulated very slight amounts of reduced nitrogen 15. Most of the nitrogen 15 translocated to leaves was assumed to be in the form of nitrate. It was suggested that nitrate assimilation takes place predominantly in leaves in both cucumber and figleaf gourd, and that the nitrate assimilating capacity is not affected by low root temperature and therefore not responsible for the differential root-chilling tolerance of these plant species (Tachibana, 1988b).

The respiration of figleaf gourd roots was less susceptible to low temperatures than that of cucumber roots, and this difference was mainly caused by cytochrome respiration. Experiments with 2,4-dinitrophenol (DNP) indicated that the oxidative phosphorylation rate at low temperature was higher in figleaf gourd than in cucumber root. It was suggested that the response of cytochrome root respiration to low temperatures, coupled with oxidative phosphorylation, can account for the difference in root-chilling tolerance between cucumber and figleaf gourd (Tachibana, 1989).

Translocation of Matters between Scion and Rootstock

Minerals

Grafting affects the absorption and distribution of minerals in plants. Tomato plants were grafted onto an unspecified Solanum species, and the Solanum species was grafted onto tomato plants. All plants were then grown for 15 days in water culture containing radioactive phosphorus (P-32). Results showed an uneven distribution of phosphorus. The concentration in the tomato plants was generally lower than the Solanum species, and higher in the upper parts of the stem. In Solanum plants, the concentration was greater in the region of the graft union. When the radioactive phosphorus was injected into the scion or rootstock, it was translocated in either direction, but the distribution was again uneven (Glushenko and Drobbich, 1952).

By using auto- and hetero-grafts of tomato and Datura sp., it was confirmed that Datura roots had a greater ability than tomato roots to absorb phosphorus when the external phosphorus concentration was extremely low (Otsuka, 1968). In Cucumis melo grafted onto Cucurbita maxima x C moschata, organic nitrogen and fruit yield increased when compared with an ungrafted control, while the control had a higher concentration.
of nitrate, lower nitrate reductase activity and a greater concentration of total free amino acids and soluble proteins (Ruz and Romero, 1999) Nitrate, phosphate, calcium and magnesium absorbed from nutrient solution were higher in ungrafted cucumber plants than in the plants grafted onto figleaf gourd In ungrafted cucumber plants, phosphate absorption decreased markedly at a low air temperature of 10°C, when compared with grafted plants Oxygen consumption in roots was 1.5 times higher at 12°C in grafted cucumber plants than in ungrafted plants, while it was almost the same at air temperatures of 16°C and 24°C (Masuda and Gomi, 1984).

In soil culture, strong and weak symptoms of magnesium deficiency appeared on the leaves of cucumber plants grafted onto ‘Shintosa’ (C. maxima x C. moschata) and figleaf gourd, respectively. No symptoms were apparent, however, when the plants were grown in nutrient solution culture. The magnesium concentration in sap exudate was highest in ungrafted cucumber plants and lowest in the plants grafted on ‘Shintosa’ (Ikeda et al., 1986).

Grafting two melon cultivars onto three varieties of Cucurbita maxima showed that the use of different rootstock genotypes causes little change in the leaf content of macronutrients, particularly nitrogen and sodium. Nevertheless, the root genotype did determine the yield. There was a strong relationship between the variations in foliar concentrations of nitrogen and sodium and yield differences in grafted plants (Ruz et al., 1997).

In three hydroponically grown tomato cultivars including a rootstock cultivar resistant to bacterial wilt, the pathogen population in stems decreased with increased calcium concentration in the hydroponic solution (Yamazaki and Hoshina, 1995). Although calcium uptake by the shoot was increased by grafting onto a highly resistant cultivar, the development of the disease was not influenced by the difference in calcium uptake (Yamazaki et al., 2000).

An iron-inefficient tomato mutant ‘T3238fe’ was identified by growing tomato plants in solution cultures containing different Fe-

HEDTA concentrations. The cause of iron inefficiency in ‘T3238fe’ roots was identified by approach grafting ‘T3238Fe’ (iron-efficient) and ‘T3238fe’ strains. ‘T3238Fe’ tomatoes absorb more iron than ‘T3238fe’ and respond favourably to iron stress by releasing hydrogen ions from roots. This speeds up the reduction of Fe³⁺ to Fe²⁺, thus increasing the citrate concentration in the root. ‘T3238fe’ showed very little response to iron stress but it was unable to absorb and transport adequate iron to support growth (Brown et al., 1971).

Two cultivars/lines of dry bean (Phaseolus vulgaris), one resistant and one susceptible to iron deficiency chlorosis, were grown with their reciprocal grafts in a soil known to induce iron deficiency chlorosis. When susceptible scions were grafted onto resistant rootstocks, the leaves of the grafted plants were greener than the ungrafted susceptible plants. It was shown that rootstocks of dry beans appear to control chlorosis resistance, presumably due to root uptake or translocation of iron (Zaier et al., 1987).

Tomato ‘T3238’ and ‘Rutgers’ were grown in solution cultures containing different concentrations of boron. ‘Rutgers’ was about 15 times more efficient than ‘T3238’ in utilizing the boron in the growth medium, and translocated more boron to the top leaves. When plants developed symptoms of boron deficiency, there was no evidence of boron redistribution between tissues. Reciprocal grafts of ‘T3238’ and ‘Rutgers’ showed that boron transport is controlled by the roots (Brown and Jones, 1971).

Photosynthates

When carbon 14 was applied to muskmelon grafted onto figleaf gourd, little was translocated downward to the rootstock. This was apparent whether rootstock leaves were present or not (De Stigter, 1961).

In tomato plants, carbon 14 photosynthesize in the scion was not translocated to rootstock until 3 days after autografting. Hardly any of the carbon 14 photosynthesize assimilated by the leaves on the rootstock before grafting was translocated to the scion. Root activity, as shown by α-naphthylamine oxidising activity,
decreased rapidly after grafting, although its activity in plants with rootstock leaves was higher than in those without them. When the rootstock plants were grown under the shade before grafting, the rate of graft take and early vegetative growth of the scions was depressed. This depression, however, could be alleviated by keeping leaves on the rootstock. Early vegetative growth increased with an increasing number of leaves remaining on the rootstock, but the photosynthetic rate of scion leaves decreased (Yoshoka et al., 1981).

The translocation and distribution of photosynthetic assimilates from rootstock and scion leaves on eggplants were determined by exposing them to carbon 14 radio-labelled carbon dioxide 28 days after grafting. The growth of the scion was enhanced by increasing the number of leaves on the rootstock from 0 to 3. The percentage of rootstock carbon 14 assimilates transported from the rootstock leaves to the scion ranged from 40-45%, about 50% of the assimilates in the rootstock came from the scion leaves. The photosynthetic assimilates from both scion and rootstock leaves were translocated through the graft union, the amount was dependent on the degree of vascular connection established (Shishido et al., 1995).

The rate of assimilation between scion and rootstock has been compared between the compatible intrafamilial graft of tomato on potato and the less compatible interfamilial graft of bean (Vicia faba) on sunflower (Helianthus annuus). Transportation across the graft interface in the compatible heteroplastic and homoplastic combinations started 5-7 days after grafting. In the less compatible bean/sunflower grafts, labelled material from the scion could not be detected in the rootstock until 10 days after grafting. The maximum rate of transport from one partner to the other was significantly lower in bean/sunflower grafts than in the other systems. The first sieve tubes traversing the graft interface were found in tomato/potato grafts at the same time as the appearance of assimilates in the rootstock. The increasing numbers of sieve tubes in the graft unions correlated with an increasing rate of transported assimilates, indicating that the route of translocation was via the phloem (Rachow-Brandt and Kollmann, 1992a).

Carbon 14 translocation experiments, measurements of enzyme activity and microautoradiography showed that assimilates were partially unloaded on their way from scion to rootstock in graft unions of tomato/potato, bean/sunflower and autografts of tomato, bean and sunflower. Whereas sucrose was the main labelled assimilate in the internodes of all graft partners, in the graft unions most labelled assimilates were the monosaccharides glucose and fructose. Acid invertase was more active in graft unions than in scion or rootstock internodes. Efflux from the apoplast of stem tissues was higher in graft unions than in internodes. Labelled assimilates were fixed in young differentiating cells of the graft union (Rachow-Brandt and Kollmann, 1992b).

In Nicandra physalodes, there was only a small import of carbon 14-labelled sucrose into the rootstock 1-3 days after grafting. This was likely due to symplasmic or apoplasmic diffusion across the callus of the graft union. The translocation rate increased with the onset of phloem regeneration about 5-7 days after grafting. The interfamilial graft bean/Helianthus tuberosus was incompatible, as shown by a complete loss of vascular regeneration between rootstock and scion, and consequently by an extremely low rate of carbon 14-translocation into the rootstock (Wang and Kollmann, 1996).

Phloem transport was studied in cultured explant-grafts of tomato plants after application of carbon 14-sucrose and carboxyfluorescein (CF) to the scion. Sucrose translocation across the graft interface started 4 days after grafting and increased later. The observed translocation appeared to occur via wound phloem, since at this time the first complete wound-phloem bridge (shown as files of aniline-blue-positive sieve plates) traversed the graft interface. Seven days after grafting, sucrose transport across the graft interface returned to normal again. Carbon 14 profiles revealed accumulation of labelled carbon in sink tissues, where the basal callus of the rootstock, and temporarily the graft union.
itself, represent the main sinks for labelled sucrose. Moreover, there is little doubt that effective phloem translocation across the graft interface, visualised with CF, comes about by wound-phloem bridges reconnecting the cut vascular bundles of scion and rootstock (Schonung and Kollmann, 1995)

**Phloem proteins**

Phloem exudate proteins collected from *Cucumis sativus* grafted onto *Cucurbita ficifolia* or *Cucurbita maxima* were separated by SDS-PAGE. Phloem exudate from *Cucumis* scions contained least four novel protein bands that did not appear in *Cucumis* control plants, but corresponded in molecular weight exactly to complements of *Cucurbita* protein patterns, including the *Cucurbita* PP1 (filament-forming phloem protein) and PP2 (phloem lectin) subunits (Tiedemann and Carstens-Behrens, 1994).

When cucumber scions were grafted onto *Cucurbita* rootstocks, at least nine additional proteins appeared on sodium dodecyl sulfate-polyacrylamide electrophoresis gels of scion exudate, 9-11 days after grafting. These proteins corresponded exactly to those of the respective *Cucurbita* sp rootstock, including PP1 and PP2. The appearance of additional proteins was correlated with the establishment of phloem bridges across the graft union. In a few combinations of scion and rootstock species, some graft partners failed to show an exchange, but some behaved as donors for additional proteins and others could be donors or acceptors. Cucumber plants were consistently identified as acceptors, regardless of scion or rootstock (Golecki et al., 1998).

**Alkaloids**

When tobacco scions were grown on tomato rootstocks, no appreciable accumulation of nicotine occurred in the tobacco leaves or stems. In the reversed combination, however, nicotine was found in small quantities in the tomato stem and fruit, and large quantities of the alkaloid accumulated in the tomato leaves (Dawason, 1942).

No nicotine appeared in the scions of mahorka (*Nicotiana rustica*) grafted on tomato, potato, scented tobacco (*N. affinis*) or nightshade (*Solanum nigrum*). Tomato and potato on mahorka rootstock did not produce nicotine, but in scented tobacco grafted on mahorka, nicotine only appeared when the leaf area of rootstock was larger than that of the scion. It was suggested that in the scented tobacco scions on mahorka roots, nicotine is converted into some other unidentified alkaloid (Poda-Cikalenko, 1960).

*Solanum aculeatissimum* contains the steroid saponins aculeatiside A and aculeatiside B at high levels in the root, whereas they are lacking in all tomato organs. When *S. aculeatissimum* was grafted onto tomato, a small amount of steroid saponin was found in the leaves and the stem of *S. aculeatissimum* but not in the roots of the tomato. However, when tomato was grafted onto *S. aculeatissimum*, steroid saponin appeared only in the root of *S. aculeatissimum*. It has been suggested that steroid saponins are synthesised mainly in the roots of *S. aculeatissimum* (Ikenaga et al., 1990).

**Floral stimuli**

The translocation of floral stimuli has been studied using grafted plants. A qualitative short-day plant *Sicos angulatus* was induced to flower under non-inductive long-day conditions, not only by grafting onto plants of the same species that had been stimulated to flower, but also by intergeneric grafting onto the day-neutral plant *Cucumis sativus* or the quantitative short-day plant *Luffa cylindrica*. When a sufficient number of leaves on Sycos donors were exposed to long- or short-day stimuli, a far greater number of pistillate flowers formed on cucumber receptors grafted onto short-day donors than long day ones. It was suggested that the sex expression of flowering in Cucurbitaceae is influenced by the availability of floral stimuli (Takahashi et al., 1982).

In *Perilla*, a floral stimulus moved from the donor leaf with an area of only 1cm² was sufficient to induce flowering in the receptor plant, although a leaf area of more than 5cm² was needed to induce a maximum response. When N⁰-Benzylammonopurine (BA) was applied...
to the donor leaf of the stimulated plant before grafting it onto the non-stimulated receptor plant, the expression of the floral stimulus was inhibited. It was suggested that BA interferes with the movement of the floral stimulus from the donor leaf to the receptor plant (Suge, 1984).

Grafting experiments provided evidence that endogenous gibberellin did not function directly in the differentiation of floral primordia, but that it plays an important role in the development of floral organs once they have been differentiated. The grafting of 3-4 cm developing inflorescences onto vegetative rootstock induced flowering in 21.4% of cases (Suge, 1992).

By using grafting techniques, it has been suggested that the role of GA transport in early flower development in tomato is negligible, and GAs required for development have to be synthesized in the flower bud itself (Heuvel et al., 2000). Grafting experiments have also shown that some day-neutral *Cucurbita* plants produce a factor that inhibits flower formation in cucumber plants (Sato, 1996).

**Viruses**

When tomato plants were grafted onto *Lycium barbarum*, mosaic virus (TMV) was transmitted from a diseased *L. barbarum* to the tomato (Lumberk, 1951). Resistance to the four known Ohio pathogenic strains of TMV controlled through dominant alleles was transferred from *L. peruvianum* to *L. esculentum* (Alexander et al., 1963).

Alexander (1971) showed that there were three genes responsible for resistance to TMV, *Tm*, *Tm-2* and *Tm-2*'. The *Tm* gene controls viral multiplication, but does not stop the virus from spreading in the plants at a low density, while the *Tm-2* gene induces necrosis at the infected parts, preventing the spread of the virus in the plant. The *Tm-2* gene shows characteristics of both *Tm* and *Tm-2*', depending on the strain of TMV.

When plants with necrotic resistance to TMV were grafted onto susceptible plants or plants resistant to TMV through inhibition of virus multiplication, the grafted plants died of necrosis (Pilowski, 1971). In practice, the TMV-resistant genotypes of scion and rootstock varieties should match (Yamakawa, 1982). Recently, Japanese researchers have been studying the damage of grafted sweet pepper plants from the viewpoint of TMV-resistant genotypes.

**Graft Compatibility and Other Matters**

**Graft formation**

*Cucumis sativus* was grafted onto *Cucurbita ficifolia*, and the formation of the graft union was monitored. Firstly, an isolation layer and a parenchymatous callus formed at the graft interface. Symplastic contact between the cells, especially the sieve elements of rootstock and scion, was then observed. Symplastic phloem contact was demonstrated indirectly by serial sectioning of the graft union in the heterograft. Phloem development in the graft unions resulted in different numbers of connecting sieve tubes in each individual graft, but the average number of sieve tube connections in *Cucumis/Cucurbita* was much lower than in *Cucumis/Cucumis* (Tiedemann, 1989).

The development of the union of autografted pea (*Pisum sativum*) roots was observed under a microscope. Cell divisions were induced close to the wound in both partners in 1 to 2 days. The cytoplasm content increased in some cells of the pith, cambium, pericycle and cortex, and these cells proliferated to form both disorganised callus within the vascular cylinder and organised nodules of cells in the cortex of each partner. There was more proliferation proximal to the graft. The resulting cells invaded the graft gap, completely filling it and effecting union by the 7th day after grafting (day 7). The first wound-repair vascular tissue differentiated on day 4 in both partners. Wound-repair xylem bridged the union on day 7, phloem on day 8, and cambium by day 12. The necrotic layer (cells killed by cutting), initially thick, was disrupted by day 4 and disappeared, as did its phenolic staining properties, by the time of cohesion. During the development of the union, the cortex and pith proliferated most, the pericycle less and the endodermis and cambium least.
thus disproving the dogma that the cambium is the sole proliferative layer in graft formation (Stoddard and McCully, 1979).

Cellular events leading to graft formation in tomato autografts were studied morphologically using scanning and transmission electron microscopy. Initial adhesion in the pith of grafts was followed by the confrontation of new cells generated from the peripheral tissue of the rootstock and scion. Pectinaceous beads on the surfaces of these cells established a mechanical union between the cell surfaces, forming the functional equivalent of a middle lamella. Plasmodesmata formed de novo at the point of contact between opposing cells. These linked the cell membranes, forming a potential pathway for high specificity communication. Subsequently, wound vessels differentiated within the ‘callus’ at the graft union, and were connected into the vascular system of rootstock and scion by wound vessels differentiating from vascular and cortical parenchyma (Jeffree and Yeoman, 1983).

The formation of cytoplasmic cell connections was studied in vitro using Vicia faba grafted onto sunflower. Extremely thin wall parts with a striking endoplasmic reticulum (ER)-plasmalemma contact were observed in apical portions of protruding callus cells and in the contact zone between opposing cells. During subsequent thickening of the modified wall parts, cytoplasmic strands enclosing constricted ER cisternae were entrapped within the newly deposited wall materials. These cytoplasmic strands represented half plasmodesmata which, in the case of fusion with corresponding structures of adjoining cells across the loosened wall matrix, formed continuous cell connections. Golgi vesicles secreting wall material were involved in the process of forming half and continuous plasmodesmata, and followed the same mechanism of plasmodesmata development as described for isolated protoplasts in cell cultures (Kollmann and Glockmann, 1991).

Graft incompatibility

Beyrers (1974) reviewed the grafting of market solanaceous crops and showed that there is graft compatibility for tomato, eggplant, and other solanaceous plants Capsicum can only be grafted onto Capsicum. Andrews and Marquez (1993) reviewed in detail studies on the graft incompatibility of horticultural crops, mainly fruit trees.

The histophysiology of grafting was observed using compatible ('Doux' and 'Landes') and incompatible ('Yoloowonder' and 'Florida') pepper plants (Capsicum annuum) grafted onto tomato, with tomato autografts as a control. In compatible combinations and in controls, graft-bridging cambia were established some 10 days after grafting. A continuous vascular ring then formed progressively. In most incompatible combinations, no such vascular connection was established, or only a very weak one was differentiated. In incompatible grafts, both partners commonly formed well-developed wound periderms at the graft line. Wound periderms were decidedly less developed and normally appeared outside the vascular zone. Two days after grafting, slight peroxidase activity occurred in both partners in the outermost cells abutting the cut line. This activity progressively faded out from various zones of the graft line in compatible combinations, while the activity increased and persisted for several weeks in incompatible combinations. These enzyme activities seemed to be associated with the deposition of lignins, since the phloroglucinol-HCl test was clearly positive in older specimens precisely at the location where peroxidase was revealed. A number of characteristics readily distinguished compatible from incompatible grafts. In the latter, the vascular junction was absent, and accumulation of lignin and polyphenols at the level of the contact layer was always found (Deloire and Hebart, 1982).

In cultures of explant-grafts between compatible and incompatible combinations of three Solanaceous species (Lycopersicon esculentum, Nicandra physalodes and Datura stramonium), incompatible explant-grafts attained greater mechanical strength than corresponding whole-plant grafts. This characteristic of their development was a suitable criterion for judging the compatibility of explant-grafts. Explant grafting in culture might offer an ideal system for experimental
analysis of the cellular basis of incompatibility in internode grafting (Parkinson et al., 1987).

Monitoring graft take

Graft take has been judged through the observations of experienced farmers and researchers, but the mass production of grafted seedlings requires more objective methods for the determination of graft take.

One way of measuring graft take is through mechanical properties. By measuring tensile strength it was shown that the development of the graft continues steadily after the second day of grafting. It was also found that graft formation is the most rapid when the different stem tissues match up in the rootstock and the scion, and that the tensile strength of the graft is markedly reduced when the turgidity of the tissues is depressed (Roberts and Brown, 1961).

Breaking weight is another measure of the mechanical strength of graft unions. When this measure was used, it was shown that the strength of tomato graft unions increased in 2 phases over 7 days. The first phase lasted for about 4 days and was accompanied by active cell division and a large increase in the number of tracheidal elements. During the second phase, which lasted about 3 days, the tracheides continued to differentiate but with no increase in cell number. The change in breaking weight during the first phase was proportional to the amount of polysaccharides in the junction. The final restoration of vascular continuity taking place toward the end of the first phase and during the second was accompanied by a large increase in the number of tracheidal elements within the graft union at the time (Lindsay et al., 1974).

The measurement of electrical resistance across the graft interface of a tomato autograft was employed as a simple mean of detecting the success of graft union. The electrical resistance for the first 2-3 days increased rapidly in step with the formation and thickening of the isolation layer. In the next 3-8 days electrical resistance decreased steadily, as the isolation layer ruptured and disappeared during callus proliferation and interdigitation. Afterwards, resistance began to drop to the level of the intact stem, which seemed to indicate that sympatric connection and vascular unification had been completed in Amaranthus tricolor/Lycopersicon esculentum heterografts, the resistance increased steadily with the establishment of an isolation layer, which remained unruptured (Yang et al., 1992).

A thermal camera with an image processor has been used to evaluate the quality of graft take. When grafting is successful, water moves smoothly from the root to the leaves of the scion, reducing leaf temperature due to transpiration. The results showed that the leaf temperature of successfully grafted plants is 2-3°C less than that of badly grafted ones, which means the quality of graft-take may be estimated (Torn et al., 1992).

A displacement transducer was applied to tomato plants grafted by the tongue-approach method, to assess the functional hydraulic connection between rootstock and scion. The thickness of the scion and rootstock leaves was measured under conditions of repeated water stress for the scion roots. The change in leaf thickness corresponded with histological observations. The major hydraulic connection within the graft union of tomato became functional over a period of about 48h from the fifth day after grafting. This was consistent with the appearance of wound-xylem bridges at this time (Turquoise and Malone, 1996).

Electrical wave transmission from the scion to the rootstock across the grafting interface was related to histological changes during the development of the graft union (Lu and Yang, 1996).

Vessels at the joint of grafted plants were clearly distinguished with a 3-D-image-acquisition-machine vision system constructed with a microtome, a microscope, a CCD colour camera and an image processor (Nishura et al., 1999).

Plant hormones

It has been shown that natural gibberellin produced in the stem tip of pea plants does not reach the rootstock across the graft union, at least until after tissue union has occurred (Lockhart, 1957).

Two halves of an explanted internode of
**Lycopersicon esculentum**, *Datura stramonium* and *Nicandra physalodes* were grafted in sterile culture. The application of 0.2 to 2.0 mg L\(^{-1}\) IAA to the apical end of the internode was absolutely necessary for the successful formation of grafting. The addition of kinetin (0.2 mg L\(^{-1}\)) to the culture medium stimulated graft development but gibberellic acid (GA\(_3\); at 0.5 mg L\(^{-1}\)) was inhibitory (Parkinson and Yeoman, 1982).

The influence of IBA and 6-BA on graft formation in autografts of cucumber plants was determined using a similar technique. Growth regulators controlled the formation of graft unions by influencing the number of vascular bridges forming between the rootstock and scion. The best results were achieved when IBA (1.2 mg L\(^{-1}\)) was added to the scion media and 6-BA (0.3 mg L\(^{-1}\)) was added to the scion and rootstock media (Lu et al., 1996).

Cytokinin concentration in 'Keumdongee' oriental melon was not significantly influenced by the rootstock, although the highest concentration was observed in 'Keumdongee' grafted onto 'Chantozwa' rootstock (Kim et al., 1999).

**Other factors**

When cucumber plants were grafted onto vigorous cucumber varieties and grown in a closed hydroponic nutrient solution, there was an increase in later fruit yield (Asao et al., 1999).

It was shown that viruses in tomato and hot pepper could be readily transmitted from diseased plants to healthy plants mainly via root-tip grafting when the plants were grown in recirculating hydroponic nutrients. However, viruses were hardly transmitted at all when the hydroponic nutrient solution was regularly renewed (Park et al., 1999).

In grafted tomato plants, ascorbic acid in the scion leaves increased gradually over 2 days compared with control plants. It was suggested that the increase in ascorbic acid corresponded to curing or healing of the graft wound (Wadano et al., 1999).

**Rootstock Breeding and Grafting for Breeding**

**Resistance to soil-borne diseases**

The main purpose of vegetable grafting has been the control of soil-borne diseases and nematodes, so many resistant rootstock species and varieties have been searched for and bred.

**Solanaceous plants**

'Moneymaker' tomatoes grafted onto Dutch rootstock 'K' tomatoes showed much faster growth than ungrafted 'Moneymaker' plants, and total yields were higher when they were grown in a soil infected with root rot fungus (Derbyshire and Green, 1961). The use of *L. hirsutum* var. *glabratum* × *L. esculentum* F\(^1\) hybrid rootstocks resulted in a reduction of *Fusarium* infection (Harrison and Burgess, 1962), and gave yields 3 to 4 times greater than those of ungrafted plants, which showed magnesium deficiency in the scions (Smith and Proctor, 1965).

Soil sterilisation adequately controlled corky root (*Pyrenochaeta lycopersici*) infection in rootstock 'K' (Spender and Wechold, 1964). *Lycopersicum hirsutum* is resistant to corky root and *Dyodymella lycopersici* but lacks vigour, so *L. hirsutum* was crossed with tomato and the hybrid rootstock became vigorous (Bravenboer, 1962). Rootstock of University of Hawaii selection line 5808-2 was resistant to bacterial wilt and produced higher yields of commercial grade tomatoes compared with non-grafted tomato plants grown on non-infected soil (Obrero, 1969).

American tomato ‘OTB-2’ rootstock was resistant to bacterial (*Pseudomonas* sp.) and *Fusarium* wilt (Okuda et al., 1972). The interspecific hybrids between *L. esculentum* × *L. hirsutum* is commonly named ‘KNVF,’ which means resistance to corky root (K), root knot nematodes (N), *Verticillium* wilt (V) and *Fusarium* wilt (F). This variety gave a 600% increase in yield compared to the ungrafted control (Gndrat et al., 1977), and was resistant to *Fusarium* wilt J3 (*F. oxysorum* f. sp. *lycopersici* Race J3) and tomato brown root rot (*Pyrenochaeta lycopersici*) (Kunyasu and Yamakawa, 1983).

*Solanum* rootstocks were tentatively used.
for tomato grafting to introduce their stronger resistances to bacterial wilt and nematodes than those of the tomato plants. When tomato ‘Pusa Ruby’ was grafted onto bacterial-wilt resistant eggplant ‘Dingra’s Multiple Purple,’ higher yield was obtained than that of tomato plants grafted onto the same species (Tikoo et al., 1979).

Monografted ‘Momonato’ tomato plants were highly susceptible to all strains (1 to 5) of Pseudomonas solanacearum, whereas monografted S. toscarum were completely resistant to all five strains ‘Momonato’ tomato plants grafted onto S. toscarum were not infected by any of the P. Solanacearum strains. Monografted S. sspymbiosfericum were resistant to strain 3, whereas those of S. torvum were resistant to strains 1, 2 and 5. Tomato ‘Momonato’/S. torvum became susceptible to strain 5 S. sspymbiosfericum and S. torvum were susceptible to the other strains, but they became more susceptible when combined with ‘Momonato’ tomato scions (Matsuzoe et al., 1993b). Fruit yields of tomato plants grafted onto S. sspymbiosfericum and S. toscarum were about 100 and 80% respectively of monografted tomato plants, while those of S. torvum was in between these two figures (Matsuzoe et al., 1993a).

In eggplant, tomato rootstock ‘KNVF’ onto eggplant (Solanum spp.) increased the yield of high quality fruit and harvesting was not retarded (Gindrat et al., 1976). S. torvum and S. sanatwongsei have been used for breeding rootstocks resistant to bacterial wilt. ‘Dataro’ is highly resistant to bacterial and Fusarium wilt and has a high germination rate necessary for mechanized grafting. This variety was bred from S. melongena (Monna et al., 1997).

In green pepper (Capsicum annum), superior growth and yield from the scion was obtained by using interspecific hybrid C. annum ‘Murakaki’ x ‘No. 3341’ (C. chinense introduced from Bolivia) as a rootstock (Yazawa et al., 1980).

Cucurbitaceous plants Melon plants grafted onto Benacasa cerfera gave good results in trials of Verticillium and Fusarium-resistant rootstocks (Slobbe, 1965), and complete resistance to Fusarium oxysporum f. melo-

Resistance to nematodes Damage by nematodes (Meloidogynae spp.) in soil can be controlled through grafting onto resistant rootstocks. Resistance to root knot nematodes or Verticillium wilt was also introduced into the rootstock by crossing L. hirsutum with a tomato variety resistant to these parasites (Bravenboer, 1962).

Among the wild relatives, interspecific Solanum hybrids and amphidiploids of eggplants, immunity or high resistance was observed in S. khasianum, S. torvum and S. toscarum. Small swellings were formed in S. sspymbiosfericum, but nematode maturation/egg production did not occur. The susceptible factor of the grafted scion was not transmitted across the graft union to the rootstock. Eggplants (S. integrifolium), their hybrids and amphidiploid, and S. undulatum failed to show resistance against the root-knot nematode. Solanum mammosum and S. surattense were highly susceptible to M. incognita (Ali et al., 1992).

Tomato ‘Kyoryokubego’ were grafted onto themselves, tomato ‘LS-89 (Hawaii 7998),’ S. sspymbiosfericum, S. torvum and S. toscarum. Tomato plants were highly susceptible to M. incognita, whereas S. toscarum, and S. torvum were resistant. Small nodules were observed on the roots of S. sspymbiosfericum, but nematode maturation and egg production were not observed. The high resistance of these Solanum species to M. incognita.
persisted when they were used as rootstocks for 'Kyoryokubejii' (Matsuoe et al., 1993b)

A tomato line susceptible to root-knot nematodes was also grafted onto Solanum indicum, S. sisymbriifolium, S. torvum and 2 amphidiploids of crosses S. integrifolium x S. melongena ‘Dingaraj Multiple Purple (DMP)’ and ‘Uttara’ Thirty days after inoculating the second stage larvae of Meloidogyne incognita, a maximum of 95 galls/g root were formed on non-grafted tomato plants, 21 galls/g root in plants grafted on the amphidiploid of the cross S. integrifolium x ‘DMP’ and 0·3 galls/g root in plants grafted on the 4 other rootstocks (Mian et al., 1995)

Rootstocks resistant to root-knot nematode were selected from wild Cucumis species, C. melulfurus and C. angurra for muskmelon, cucumber and watermelon plants C. melulfurus was considered to be suitable as a rootstock for melon and cucumber plants (Igarashi et al., 1987)

Seed production

Grafting has been used for seed production. When breeding tomatoes resistant to corky root, repeated reciprocal grafting between tomato and Lycopersicon glandulosum did not increase interspecific fertility (Sztelyn, 1959)

Biennial seedlings were grafted either onto annuals (e.g. cabbage onto rape or mustard), or onto seed plants of the same species (e.g. beet and carrot), and seed was obtained during the first year (Kruuzlin and Svediska, 1959) Selected cauliflower curds were divided into portions, and were grafted onto rootstocks of winter cauliflower raised in pots in a greenhouse. Flowering shoots were produced 4-8 weeks after grafting and up to 2 g of seed was obtained from each curd portion (Watts and George, 1963)

Flower numbers in a sweet potato cultivar were increased for breeding purposes by grafting onto Ipomoea carnea ssp. fistulosa (Mart ex Choisy) (Lardizabal and Thompson, 1988) Grafting 4 sweet potato cultivars onto Ipomoea carnea ssp. fistulosa increased flower numbers, percentage capsule set and number of seeds in all cultivars, whereas the responses to growth regulator application were different among cultivars (Lardizabal and Thompson, 1990)

Chimeras

Chimeras have been produced to breed new varieties. Reciprocal grafts were made in Solanaceae species. When well established, the grafts were cut back to the point of union and adventitious buds arising from the callus were allowed to develop. Two periclinal chimeras were obtained from a graft of tomato on nightshade and the reciprocal graft (Misro, 1954) Chimera plants were obtained by co-culturing nightshade and tomato plants. Two periclinal chimeric shoots were also obtained from grafts between tomato and nightshade (Lindsay et al., 1995)

Graft hybrids

Vegetative hybridisation had been studied mainly in the USSR. Before the 1960s Yagishita and Hirata’s group continued the study on the heredity of graft-induced characteristics (Yagishita and Hirata, 1986, 1987, Yagishita et al., 1990) After analysis of soybean storage proteins with electrophoresis techniques, they suggested that genetic changes were transmitted to the progeny in an unstable manner (Hirata and Yagishita, 1986). Through a study of the stability of phenotypic changes and characteristics of the graft-induced variants in pepper, they considered that some of the characteristics of the rootstock were introduced into the progeny through selfed seeds from the scion, and the novel characteristics appeared as a result of graft induction (Taller et al., 1998)

A RAPD marker was found in rootstock cultivars of the pepper and in graft-induced variants, but was absent in the scion cultivars. It was concluded that new characteristics in pepper which are induced by grafting are stable, new traits, and can be used as a novel genetic source in breeding (Taller et al., 1999)

Other uses

Grafting has been applied to check the level of virus infection in strawberry. A modified leaf-petirole grafting technique was an efficient
way of indexing the various virus components found in strawberries (Miller, 1958). The resistance of vigorously growing lateral shoots to cucumovirus (CMV) was confirmed by a combination of grafting and inoculation methods (Yazawa et al., 1996).

Tuber sink potential in sweet potato was studied by using the grafts of several cultivars differing in sink potential. The tuber formation largely depended on the characteristics of the stock cultivar. The effects on tuber thickening were observed for both the scion and rootstock at the stage after the tuber formation. It was suggested that the tuber becomes the dominant sink for assimilate and the source activity was quantitatively influenced by tuber sink activity after the 10g D W tuber stage (Nakatani et al., 1988).

**New Grafting Methods**

**Conventional grafting**

Grafting methods to suit each vegetable have been studied. Cleft grafting was the most efficient and successful for eggplants (Honma, 1977). In tomato plants, the sowing date for production of rootstock and scion plants should be the same. Wedge-grafted seedlings should be illuminated with mercury vapour or white fluorescent lamps suspended 80 and 90 cm, respectively, above the plants (Pirog, 1982).

Inarching and cleft grafting was recommended for the grafting of melons (Trentini and Maoi, 1989). Cleft and tongue-approach grafting methods are popular for solanaceous and cucurbitaceous plants, respectively (Oda, 1995). The conventional grafting methods are explained with illustrations (Oda, 1999).

Recently, double-stem grafting methods have been developed using tomato plugs to reduce the number of seedlings for planting. First, a scion cut at the first internode is grafted onto a rootstock cut at the first internode to produce an ordinary graft. Next, the shoot cut from the rootstock is cut to the soil to produce a secondary rootstock, and punched scion with only two cotyledons is grown to generate two lateral shoots. Finally, the secondary scion with two lateral shoots is grafted onto the secondary rootstock 2 weeks after the first grafting. The number of grafted plants and harvestable shoots made with unit seeds can be increased with this technique (Kawai et al., 1996).

**Micrografting**

Micropropagation has been studied for the rapid propagation of regenerated plantlets. Although adventitious buds differentiated on the hypocotyl-derived callus in eggplants did not develop into shoots, they grew into a shoot by micrografting. Vigorous roots developed at the same time. Tomato hypocotyls were also successfully used as rootstock sources for this micrografting, but buds grafted onto watermelon or melon hypocotyls failed to develop into shoots (Yamamoto and Matsumoto, 1994).

Pea plants (*Pisum sativum*) regenerated from macerated vegetative apices and immature embryos rooted with low frequency (about 10%). However, 80 to 85% of pea shoots survived to maturity and produced seed when they were grafted onto rootstocks of the same cultivar (Natali and Cavallini, 1989).

Micrografting has been applied to the physiological study of grafting. Explants derived from the internodes of tomato, *Nicandra physalodes* and *Datura stramonium* were grafted in vitro, in order to elucidate the chemical basis of cell to cell contact and its possible role in differentiation. The factors involved in determining whether a graft will or will not be successful were present as normal constituents of the ungrafted internodal tissue, where they are associated with the cell wall and are released as a result of cellular contact between the rootstock and scion. These factors could not be transferred by the use of an inert interstock. Internode pectins from one species reduced the ability of homografts of other species to form vascular connections between rootstock and scion (Jeffree et al., 1987).

The function of phloem connection in regenerating *in vitro* grafts was investigated by giving carbon 14 labelled sucrose to the scion. The resulting carbon 14 profiles showed that sucrose translocation across the graft interface started 4 days after grafting.
increased later. The translocation appeared to occur via wound phloem, since at this time the first complete wound-phloem bridge traversed the graft interface. In 7-day-old autografts, sucrose transport across the graft interface returned to normal again. Carbon 14 profiles revealed accumulation of label in sink tissues, where the basal callus of the rootstock, and temporarily the graft union itself, represent the main sinks for labelled sucrose (Schoning and Kollmann, 1995).

Apical segments ranging from about 200µm down to 50µm in depth were successfully grafted back to their parent plants, and the apices developed completely normally. Complete shoots could be regenerated from an apical segment less than one-thousandth of a cubic millimetre in volume, containing about 600 cells, which was smaller than any graft previously recorded at the time of publication (Gulline and Walker, 1957).

**Tools**

Tying materials have been used for vegetables in the same way as they have for fruit trees. In vegetables, grafting clips have also been used in cleft grafting and slant-cut grafting (Oda, 1999). A hand-held grafting device constructed with changeable stainless-steel, single-edge razor blades, allows the operator to simultaneously cut a uniform wedge in the stems of *Phaseolus vulgaris* and its receptacle (White, 1979). Agricultural companies have now developed and released a great many materials and tools for grafting.

**Robots**

Basic factors in grafting cucurbitaceous vegetables such as variations of seedling shape, location for cutting and gripping plants, cutting methods, fixing materials and tools etc have been reviewed, with the aim of developing a grafting robot “Cutting-off Cotyledon Grafting, (CCG),” cutting the seedlings at the joint of the cotyledons, and cutting the seedlings at the joint of the cotyledons and the hypocotyl at an angle of 10° for scion and 30° for rootstock, have all been suggested as suitable methods for mechanisation (Suzuki et al., 1995).

The first grafting robot using the CCG method was manufactured in 1987 and the second in 1989. It took 3 seconds to make a grafted plant, and the survival rate was 95% (Onoda et al., 1992). It was judged that the demonstration model robot was practical and the technology was transferred to an agricultural machinery manufacturing company and then to the marketplace (Kobayashi et al., 1996).

Multiple grafting robots for plugs have been in place since 1987. A grafting plate was designed to grasp tomato plugs. This grafting plate consists of a hollowed plate (HP) and a drive plate (DP), which can be used for both scions and rootstocks. Plants are held in V-shaped hollows on the HP with spongy rubber mounted on the inner side of the DP. The axes of the scions and rootstocks firmly held. The HP and DP are fixed with holders. The axes are cut parallel to the lower and upper sides of the grafting plates for the scions and rootstocks, respectively. The grafting plate holding the scions is placed on that holding the rootstocks (Oda et al., 1994a).

Another mechanised method involves fixing the scion and rootstock with an adhesive (alkyl 2-cyanoacrylate) and a hardener (a polyalkylene glycol derivative). Adhesive grafting has been successful in eggplants, cucumber plants and grapevines (Morita, 1988). Chinese cabbages were also grafted onto turnip at the two-leaf stage using the adhesive and hardener (Oda and Nakajima, 1992).

A grafting robot using the above-mentioned techniques has been constructed and commercially released. The growth and yield of tomato (Oda et al., 1995) and eggplant (Oda et al., 1997) grafted by the robot were as good as those grafted by conventional manual grafting.

The grafting robot was applicable to solanaceous but not to cucurbitaceous plants. Robotic grafting requires a horizontal cut, in which the scion and rootstock are transversely cut at the hypocotyl level. The effects of this type of grafting on the survival rate and growth of cucumber plants grafted on *Cucurbita* spp was therefore studied.

Cucumber plants that were horizontally grafted at the hypocotyl had a low survival
rate, compared with conventional grafting. The low survival rate of horizontal-cut grafts at the hypocotyl was attributed to the loss of the cotyledons from the rootstock and a smaller number of vascular bundles coming into contact with the cut surfaces of the rootstock and scion. The survival rate was higher when the angle formed by the expanding direction of the cotyledons of the scion and rootstock was 90°, as opposed to 0°. This was associated with an increase in the number of vascular bundles coming into contact with the cut surfaces of the scion and rootstock (Oda et al., 1994b). When the difference in the diameter of the hypocotyls between the scion and rootstock was minimised, the survival rate and growth of cucumber scions cut horizontally and grafted onto *Cucurbita* spp. at hypocotyl level was enhanced (Oda et al., 1993). Finally, the low survival of cucumber plants grafted by horizontal-cut grafting is attributed to low pressure on the spliced cut surfaces and loss of cotyledons on the rootstock (Oda et al., 2000). The length of fig leaf gourd (*Cucurbita ficifolia*) hypocotyls, a rootstock for cucumber, was controlled for robotic grafting through seed treatment with uniconazole and spraying of gibberellic acid (Oda, 1994).

Kurata (1994) introduced grafting robots developed by Japan Tobacco Inc (JT), Techno Grafting Research Inc. (TGR), Bio-oriented Technology Research Advancement Institution (BRAIN) and Osaka Prefecture University (OPU). The BRAIN robot was manufactured in 1987, and the second one in 1989. It took about 3 seconds to graft a cucumber plant with a 98% survival after acclimatisation (Onoda et al., 1992). The OPU robot used a plug-in method, in which a pencil-tip shaped scion stem is plugged into the conical hole of the rootstock stem. The OPU robot could be used for tomato, eggplant and watermelon (Honami et al., 1992).

**New combinations for robotic grafting**

Various combinations of cruciferous plants were tried out to determine their suitability for robot grafting. The following scion/rootstock graft combinations were successful: cabbage/kale, kohlrabi/kale and cabbage/kohlrabi for *Brassica oleracea* combinations, Chinese cabbage/turmeric for *B. campestris* inter-varietal grafting, Chinese cabbage/kale, Chinese cabbage/cabbage, Chinese cabbage/Takana (*B. juncea*) and Takana/turmeric for inter-specific grafting among *B. oleracea*, *B. campestris* and *B. juncea*, and Chinese cabbage (*B. campestris*). Japanese radish (*Raphanus sativus*) and Japanese radish/cabbage (*B. oleracea*) for inter-generic grafting (Oda et al., 1992).

**Acclimatisation**

Various grafting methods have been applied to fruit bearing vegetables, but for grafted plants to survive, the acclimatisation conditions are very important. There are two types of acclimation: 'healing' of the cut surface and 'hardening' for the harsh conditions in the field (Oda, 1999). It is possible to acclimatise by enclosing the grafted plants in a plastic bag until graft union is complete (Denna, 1962).

For farmers, an acclimatisation tunnel constructed with agricultural materials is an effective way to achieve a high survival rate. High humidity and weak light at an intensity a little higher than the light compensation point prevented wilting of grafted tomato plants scions. These conditions were favourable for healing of the cut surfaces of grafts. When films reducing thermal radiation were put onto an acclimatisation tunnel, the temperature rise of the leaves was depressed so that the materials extended the favourable range of light intensity for heating of grafted plants (Nobuoka et al., 1996). Healing of the graft union is hastened by air movement at relatively high light intensity and absolutely high humidity, which allows the plants to grow without wilting (Nobuoka et al., 1997).

**References**

Abdelhaffez, A T, Harssema, H, and Verkerk, K., 1975 Effects of air temperature, soil temperature and soil moisture on growth and development of tomato itself and grafted on its own and eggplant rootstock *Sci Hortic.*, 3, 63-73.

Alam, M. Z., Ali, M., Akanda, A M., Choudhury, D A M., Haque, N M M., Hossain, M M.,
and Ogawa K. 1994. Trafting technology an integrated pest management for eggplant and tomato Bull Inst Tropic Agric Kyushu Univ., 17, 85-91

Alexander, L J 1963. Transfer of a dominant type of resistant to the four known Ohio pathogenic strains of tobacco mosaic virus (TMV) from Lycopersicon peruvianum to L. esculentum Phytopathol., 53, 869 (Abstr.)

Alexander, L J 1971. Host-pathogen dynamics of tobacco mosaic virus on tomato Phytopathol., 61, 611-617


Bravenboer, L. 1962. Control of soil-borne diseases in tomatoes by grafting on resistant rootstocks Proc 16th Int Hort Congr., Brussels, 1, 98


Coggins, C W and Lesley, J W. 1968. Attempts to improve flower bud retention and development in tomatoes with grafts, nutrition and growth regulators HortScience, 3, 237-238

Dawson, R F 1942. Accumulation of nicotine in recprocal grafts of tomato and tobacco Amer J Bot., 29, 66


Derbyshire, D M and Green, J A 1961. Tomato grafting solves the root rot problem Grower, 56, 911

Gindrat, D., Ducrot, V., and Caccia, R 1976. The control of Verticillum wilt of eggplant by grafting onto resistant tomatoes Revue Suisse de Viticulture Arboriculture Horticulture, 8(2), 71-76 (in French)


Harmett, R F 1974. Resurgence of interest in grafting techniques on heated tomato crops Grower, 82, 861-862


Heuvel, K J P T., Heijmen, P H F., Barendse,


Jeffree, C. E. and Yeoman, M. M. 1983 Development of intercellular connections between opposing cells in a graft union New Phytol., 93, 491-509

Jeffree, C. E., Yeoman, M. M., Parkinson, M., and Holden, M. A. 1987 The chemical basis of cell to cell contact and its possible role in differentiation Monograph, Britsh Plant Growth Regulator Group, 16, 73-86


Kramer, M. 1957. Physiological aspects of grafting Solanaceous plants Biologico, 23, 73-76. (in Spanish)


Lardizabalin, R. D. and Thompson, P. G. 1988 Hydroponic culture, grafting, and growth
regulators to increase flowering in sweet potato HortScience, 23, 993-995
Lardizabal, R D and Thompson, P G 1990 Growth regulators combined with grafting increase flower number and seed production in sweet potato HortScience, 25, 79-81
Limerick, J 1951 Experiments on grafting non-woody plants on woody plants 1 Grafting tomatoes and pepper on Lycium barbarum Sborn Ces Akad Zemed, 24, 335-340 (in French)
Lindsay, D W., Yeomann, M M., and Brown, R 1974 An analysis of the development of the graft union in Lycopersicon esculentum. Ann Bot., 38, 639-646
Lockhart, J A 1975 Studies on the organ of production of the natural gibberellin factor in higher plants Plant Physiol., 32, 204-207
Miller, P W 1958 Comparative efficiency of excised leaf-petiole grafts and stolon grafts for transmitting certain strawberry viruses Plant Dis Reprt., 42, 1043-1047
Morta, S 1988 The use of new binding agent in grafting of various fruits and vegetables Agriculture and Horticulture, 63, 1190-1196 (in Japanese)


Obrero, F P. 1969. Grafting tomatoes to control tomato bacterial wilt. Hawaii Fm Sci, 18(3), 1-4


Suge, H. 1984. Nature of floral stimulus in Perilla as studied by grafting. I. Method of evaluation and the movement of floral stimulus as affected by N6-Ben-
Grafting of Vegetable Crops

Zylamnonpurine Japan J Crop Sci., 53, 423-429
Szeyn, K 1959. Trials to overcome the incompatibility of crosses between Lycopersicium esculentum and Lycopersicium glandulosum by repeated grafting Euphytica, 8, 145-150
Tachibana, S 1982 Comparison of effects of root temperature on the growth and mineral nutrition of cucumber cultivars and figleaf gourd J Japan Soc Hor Sci, 51, 299-308
Tachibana, S 1988b The influence of root temperature on nitrate assimilation by cucumber and figleaf gourd J Japan Soc Hort Sci., 57, 449-447
Tachibana, S 1989 Respiratory Response of detached roots to lower temperature in cucumber and figleaf gourd grown at 20C root temperature J Japan Soc Hort Sci., 58, 333-337
Taller, J., Yagihata, N., and Hirata, Y 1999 Graft-induced variants as a source of novel characteristics in the breeding of pepper (Capsicum annuum L.) Euphytica, 108, 73-78
Tikoo, S.K., Mathar, P.J., and Kushan, R 1979 Successful graft culture of tomato in bacterial wilt sick soil Current Science, 48, 259-260
Watts, L E. and George, R A T 1963 Vegetative propagation of autumn cauliflower. Euphytica, 12, 341-345
Whute, M C 1979 A hand-held grafting device for making uniform wedge cuts Agron J., 71, 141-144
Whute, R A J 1963 Grafted glasshouse tomatoes give heavier crops NZ J Agric, 106, 247-248
Yagishita, N and Hirota, Y 1986 Genetic nature of bushy plant type in the variant strain induced by grafting in Capsicum annuum L Euphytica, 35, 17-23
Yagishita, N and Hirota, Y 1987 Graft-induced changes in fruit shape in Capsicum annuum L J Genetic analysis by crossing Euphytica, 36, 809-814
Yagishita, N, Hirota, Y, Mizukami, H, Ohashi, H, and Yamashita, K 1990 Genetic nature of low capsaicin content in the variant strains induced by grafting in Capsicum annuum L Euphytica, 46, 249-252
Yamazaki, H and Hoshina T 1995 Calcium nutrition affects resistance of tomato seedlings to bacterial wilt HortScience, 30, 91-93
Yazawa, S, Uemachi, T, Higashide, T, and Watanabe, H 1996. CMV resistance developed in vigorous-growing lateral shoots from virus infected plants of Capsicum annuum L Scientia Hort, 65, 295-304
Zuilstra, S, Groot, S P C, and Jansen, J 1994 Genotypic variation of rootstocks for growth and production in cucumber; possibilities for improving the root system by plant breeding Sci Hortic, 56, 185-196

(Received Oct 15, 2001, Accepted Jan 28, 2002)