Netherlands Journal of Agricultural Science 49 (2001) 53-66

Development of micropropagated potato plants over three phases of growth as affected by temperature in different phases

M. TADESSE[1,2], W.J.M. LOMMEN[1,*] AND P.C. STRUIK[1]

[1] Crop and Weed Ecology Group, Department of Plant Sciences, Wageningen University, Haarweg 333, NL-6709 RZ Wageningen, The Netherlands
[2] University of Asmara, P. O. Box 1220, Asmara, Eritrea
* Corresponding author (fax: +31-317-485572; e-mail: willemien.lommen@cwe.dpw.wau.nl)

Received 16 October 2000; accepted 10 August 2001

Abstract

To assess (after)effects of temperature on plant development, *in vitro* potato plantlets produced at 17 or 23 °C (normalisation phase, 3 weeks) were planted into soil in growth chambers at 18/12 or 26/20 °C (transplant production phase, 2 weeks), and transplanted to glasshouses at 18/12 or 26/20 °C (tuber production phase, 6 weeks). The latter phase commonly takes place in the field. Transition from one phase to another, especially from *in vitro* to *in vivo* conditions, greatly increased leaf growth and to a smaller extent stem growth. Within a phase, higher temperature stimulated vegetative growth, but delayed tuber formation and reduced tuber yields, harvest index (HI) and tuber dry matter concentration. Temperature during tuber production was more important for high yield than temperatures during earlier phases. Normalisation and transplant production temperatures nevertheless showed after-effects in later phases. Lower normalisation temperatures advanced plant development: they increased vegetative growth in subsequent phases and finally increased fresh tuber yield and HI. This could have yield advantages at locations where field production seasons are short. Lower transplant production temperatures reduced vegetative growth in the next phase, but enhanced early tuber production. Finally they increased tuber dry weight and HI when tuber production temperatures were high. This may increase yield at locations where field conditions delay tuber formation.

Keywords: acclimatisation, after-effects, harvest index, *in vitro*, normalisation, pre-treatments, *Solanum tuberosum* L., transplant, transplant shock, tuber production, temperature, tuber number.

Introduction

The conventional way of propagating potato (*Solanum tuberosum* L.) involves the repeated multiplication of potato seed tubers by clonal selection. The multiplication rate is only 12–20 per year (Beukema & van der Zaag, 1990) in areas where one crop

is grown per year. Potato is susceptible to many viral, bacterial and fungal diseases and the seed stock gradually degenerates with increasing numbers of field multiplication (Haverkort *et al.*, 1991). Micropropagation techniques have widely been introduced in potato seed production systems during the past few decades to alleviate problems associated with the conventional seed production system (Jones, 1988; Struik & Wiersema, 1999). These techniques have been developed because large numbers of disease-free plants can be produced within a short period of time all year round (Jones, 1988). The fastest production system constitutes four phases: *in vitro* multiplication (through nodal cuttings), normalisation (where single-node cuttings develop into rooted *in vitro* plantlets), transplant production (acclimatisation) and tuber production.

A possible advantage of this system is that plantlets can be manipulated in the normalisation and transplant production phases with different treatments (e.g. temperature sequences) to influence plant growth and development (Hussey & Stacey, 1981; Marinus, 1985; Charles *et al.*, 1992; Tadesse *et al.*, 2000, 2001a, b), for example towards a higher multiplication rate. In the last phase of the seed production system high tuber numbers and yields are required. There still is no clear view on how *in vitro* produced potato plantlets develop through all these phases and on how pretreatments affect further growth. In this paper we will focus on temperature effects during different phases on development of *in vitro* propagated potato plants.

Growth and development of potatoes are profoundly affected by high soil and air temperatures (Ewing, 1981). Potato is well adapted to a mean temperature of 17 °C. Haulm growth is enhanced by an increase in temperature until an optimum of 20–25 °C (Ingram & McCloud, 1984). High temperature delays stolon and tuber initiation (Gregory, 1956; Slater, 1963) and onset of early tuber growth, the optimum being 15–19 °C (Van Dam *et al.*, 1996). High temperature also delays and reduces partitioning of dry matter to the tubers resulting in low harvest indices (Struik & Ewing, 1995). Low temperature, therefore, may increase tuber yield. Low temperature, on the other hand, restricts haulm growth and promotes the accumulation of dry matter in the tubers (Menzel, 1985). A delay in tuber formation, however, may stimulate final yield when the growing season is sufficiently long to profit from the increased duration of ground cover (Struik & Ewing, 1995). At higher temperature, primary stolons are able to form branches and can have numerous potential tuber sites (Struik *et al.*, 1989). Higher temperature thus may increase tuber number.

Temperature in the seed production system mentioned above not only exerts effects through its direct influence on tuber formation but also through its effects on leaf area growth. Leaf area of *in vitro* propagated potato plantlets increased logistically through time in all phases of the production system (Tadesse *et al.*, 2001b). A boost in leaf area growth and a rapid increase in leaf number occurred when *in vitro* plantlets were transferred to soil (Tadesse *et al.*, 2001b). Higher temperatures did not significantly increase leaf area during normalisation and increased leaf area during transplant production (Tadesse *et al.*, 2001b). Growing potato plantlets at higher temperature during transplant production also promoted leaf area of plantlets during tuber production in the field (Tadesse *et al.*, 2001a). Higher leaf area may increase yield directly by enabling the plants to intercept more solar radiation. The effect of

pre-culture conditions on dry matter partitioning during later phases may also be important. A relatively high dry matter partitioning to tubers in the early phase of field growth may limit haulm growth in transplant crops from early cultivars, leading to low accumulated intercepted radiation and hence low yields (Lommen, 1999).

These general effects of temperature and pre-culture conditions may be relevant to seed production systems starting from *in vitro* plantlets and may determine the efficiency of the system. Information on the pattern of dry matter production of *in vitro* produced plantlets and the effect of pre-treatments on this pattern can help to understand yield formation processes within transplant crops. Temperature may also influence tuber number and average tuber weight.

This study, therefore, assesses dry matter production, tuber number, tuber weight, tuber fresh yield and harvest index of *in vitro* propagated potato plantlets over three phases of growth as influenced by temperature in various phases.

Materials and methods

Details on the experiment described in this paper have already been provided by Tadesse *et al.* (2001b). Here we briefly summarise the main aspects and provide further details relevant to this paper.

Potato culture and treatments

Potato (*Solanum tuberosum* L., cv. Gloria) plantlets were propagated *in vitro* by single-node cuttings. The plantlets were cultured on a standard medium containing MS salts (Murashige & Skoog, 1962) with vitamins, 25 g l^{-1} sucrose, 8 g l^{-1} agar and 0.0133 g l^{-1} alar-64% (daminozide). Viable nodes were cut from plantlets discarding tops and cultured in 25×150 mm culture tubes on a 10 ml medium (one per tube) at 17 or 23 °C and a photophase of 16 h for 21 days in the 'normalisation' phase.

Rooted *in vitro* plantlets were then planted in transplanting trays with small cells filled with potting soil at 75 plants m^{-2} and grown in chambers with day/night temperatures of 18/12 (optimal for tuber growth) or 26/20 °C (optimal for haulm growth), a photophase of 14 h and a RH of 80% for 14 days in the 'transplant production' phase.

At the end of the transplant production phase, plants were transplanted in 5-litre pots filled with potting soil to two glasshouses at an initially density of 16.0 plants m^{-2} subsequently declining, by repetitive sampling, to 12.8, 9.6 and 6.4 plants m^{-2}. The pots were placed in two glasshouses at a day/night temperature of 18/12 or 26/20 °C, a photophase of 16 h and a relative humidity of 80% for 42 days in the 'tuber production' phase.

Some plants were grown for an extra week in the normalisation and transplant production phases.

Experimental design

The experiment was carried out in a split-split plot design in 16 blocks where the tuber production temperature (TB) was randomised within the transplant production

temperature (TP) and the latter was randomised within the normalisation temperature (N). Harvests were carried out at 21 and 28 days after cutting (DAC) in the normalisation phase, at 7, 14 and 21 days after planting (DAP) in the transplant production phase, and at 7, 14, 28 and 42 days after transplanting (DAT) in the tuber production phase (Figure 1). The plants harvested at 28 DAC in the normalisation phase and at 21 DAP in the transplant production phase were grown one extra week in their respective phases compared to other plants, in order to follow up their pattern of undisturbed growth.

Measurements and statistical methods

Dry matter increase through time was analysed by destructively harvesting plants, separating the different plant parts and drying them in an oven at 105°C for 16 h. Leaf dry weight, above and below ground stem dry weight, tuber dry weight and total plant dry weight were measured. Stolons were included in the stem fraction. Roots were not harvested.

Tuber numbers (tubers with a diameter ≥ 0.5 mm) were counted. Tuber fresh yield was determined and average weight per tuber calculated. Harvest index (HI, the proportion of tubers in the total dry matter excluding roots) was calculated. Tuber dry matter concentration was calculated as the percentage of tuber dry weight in the tuber fresh weight.

Data were subjected to analysis of variance (ANOVA) using Genstat 5 release 3.22 (1995) and differences between treatments were analysed by LSD tests at $P < 0.05$.

Results

Normalisation phase

Effects of temperature. At the end of the normalisation phase (21 DAC), leaf and total dry weights were not significantly different between the two temperatures, but stem dry weight was significantly higher at higher than at lower temperature (Table 1). Leaf, stem and total dry weights were not significantly affected by temperature when the normalisation phase was extended one week (28 DAC, Table 1).

Transplant production phase

Effects of planting. One week after planting to soil (Table 1, 7 DAP) leaf and total dry weights were much higher than those of plants of the same age that were left an extra week under normalisation conditions (Table 1, 28 DAC). Also stem dry weights were higher.

After-effects of normalisation temperature. Plants pre-grown at the low normalisation temperature had higher leaf and total dry weight at 7 DAP in the transplant production phase than plants grown at the high temperature in the previous phase (Table 1). For stem dry weight, no after-effects of normalisation conditions were observed.

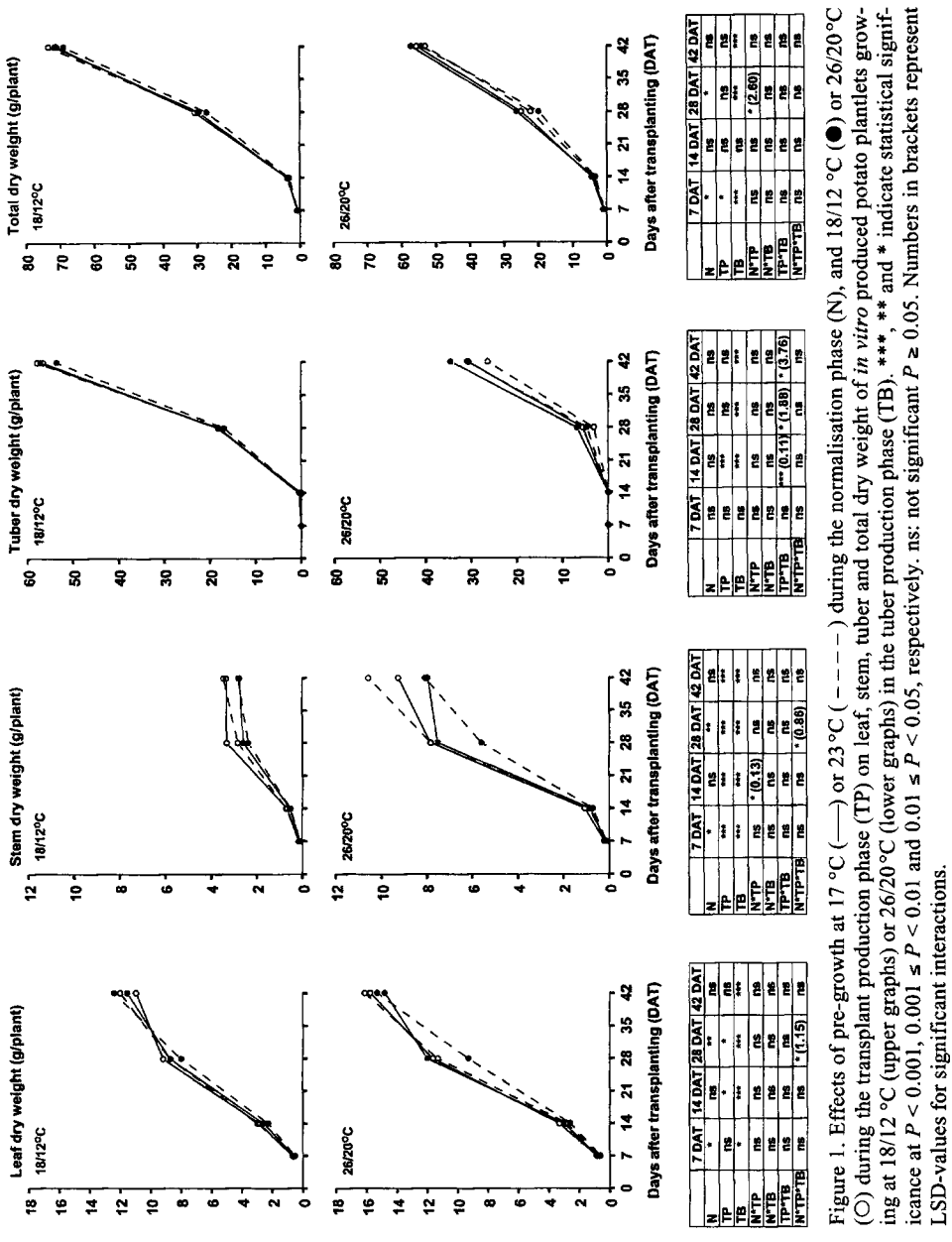


Figure 1. Effects of pre-growth at 17 °C (——) or 23 °C (– – – –) during the normalisation phase (N), and 18/12 °C (●) or 26/20 °C (○) during the transplant production phase (TP) on leaf, stem, tuber and total dry weight of *in vitro* produced potato plantlets growing at 18/12 °C (upper graphs) or 26/20 °C (lower graphs) in the tuber production phase (TB). ***, ** and * indicate statistical significance at $P < 0.001$, $0.001 \leq P < 0.01$ and $0.01 \leq P < 0.05$, respectively. ns: not significant $P \geq 0.05$. Numbers in brackets represent LSD-values for significant interactions.

Effects of temperature during transplant production. Higher temperature during transplant production resulted in significantly higher leaf, stem and total dry weights at the end of the phase (14 DAP, Table 2). Stolons appeared at the end of the transplant production phase (not shown). Tubers were not formed during this period.

Table 1. Leaf, stem and total dry weights (mg/plant) at the end of the normalisation phase (21 DAC), after 1 extra week under normalisation conditions (28 DAC), and 1 week after planting to the transplant production phase (7 DAP after 21 DAC) of *in vitro* propagated potato plantlets grown at two temperatures during the normalisation (N) and transplant production (TP) phase. DAC = days after cutting, DAP = days after planting.

Temperature (°C) during		Leaf	Stem	Total
N	TP			
End of normalisation phase (21 DAC)				
17	–	2.8	1.1	3.9
23	–	3.1	1.6	4.8
Significance [a]				
N		ns	**	ns
One extra week under normalisation conditions (28 DAC)				
17	–	4.5	1.9	6.4
23	–	4.3	2.3	6.6
Significance [a]				
N		ns	ns	ns
One week after planting (7 DAP after 21 DAC)				
17	18/12	19.4	2.9	22.3
23	18/12	14.7	2.6	17.3
17	26/20	24.5	4.2	28.6
23	26/20	19.5	3.8	23.2
Significance [a]				
N		*	ns	*
TP		ns	**	*

[a] *** $P < 0.001$, ** $0.001 \leq P < 0.01$, * $0.01 \leq P < 0.05$, ns: not significant $P \geq 0.05$; interactions were not significant.

When the transplant production period was extended one week, leaf dry weight was not different at the two transplant production temperatures (21 DAP), but stem and total dry weight were significantly higher at higher transplant production temperature (Table 2). Stem dry weight increased tremendously in this last week (Table 2).

Tuber production phase

Effects of transplanting. One week after transplanting to tuber production conditions, leaf dry weights of transplants (Table 2, 7 DAT) were higher than those of plants of the same age that were left an extra week under transplant production conditions (Table 2, 21 DAP).

After-effects of normalisation temperature. Plantlets produced at lower temperature during normalisation had higher leaf dry weight at 7 and 28 DAT in the tuber production phase than those produced at higher temperature during normalisation (Table 2, Figure 1). At 28 DAT this effect was only significant for plants grown at low transplant production temperature, followed by high temperature during tuber production. Plants pre-grown at lower normalisation temperature had a significantly

Table 2. Leaf, stem and total dry weights (mg/plant) at the end of the transplant production phase (14 DAP), after 1 extra week under transplant production conditions (21 DAP) and 1 week after transplanting to the tuber production phase (7 DAT after 14 DAP) of *in vitro* propagated potato plantlets grown at two temperatures during the normalisation (N), transplant production (TP), and tuber production (TB) phases. DAP = days after planting, DAT = days after transplanting.

Temperature (°C) during			Leaf	Stem	Total[b]
N	TP	TB			
End of transplant production phase (14 DAP)					
17	18/12	–	115	8	123
23	18/12	–	104	8	112
17	26/20	–	172	22	194
23	26/20	–	142	18	160
Significance[a]					
N			ns	ns	ns
TP			**	***	***
One extra week under transplant production conditions (21 DAP)					
17	18/12	–	345	133	504
23	18/12	–	343	128	489
17	26/20	–	433	189	657
23	26/20	–	365	168	539
Significance[a]					
N			ns	ns	ns
TP			ns	**	*
One week after transplanting (7 DAT after 14 DAP)					
17	18/12	18/12	611	113	740
23	18/12	18/12	539	98	651
17	26/20	18/12	692	164	863
23	26/20	18/12	614	125	751
17	18/12	26/20	831	168	1021
23	18/12	26/20	627	130	769
17	26/20	26/20	856	218	1075
23	26/20	26/20	754	198	956
Significancea					
N			*	*	*
TP			ns	***	*
TB			*	***	***

[a] *** $P < 0.001$, ** $0.001 \leq P < 0.01$, * $0.01 \leq P < 0.05$, ns: not significant $P \geq 0.05$; interactions were not significant.

[b] includes also tubers at 21 DAP and 7 DAT.

higher stem dry weight at 7 DAT in the tuber production phase (Table 2). This positive effect also showed at 14 DAT for plants growing at higher tuber production temperature, and at 28 DAT for plants growing at high tuber production temperature after low temperature during transplant production (Figure 1). There was no significant after-effect of normalisation temperature on tuber dry weight in the tuber production phase, but tuber dry weights tended to be higher for plants grown at a low temperature during normalisation (Figure 1). Tuber fresh weight was significantly

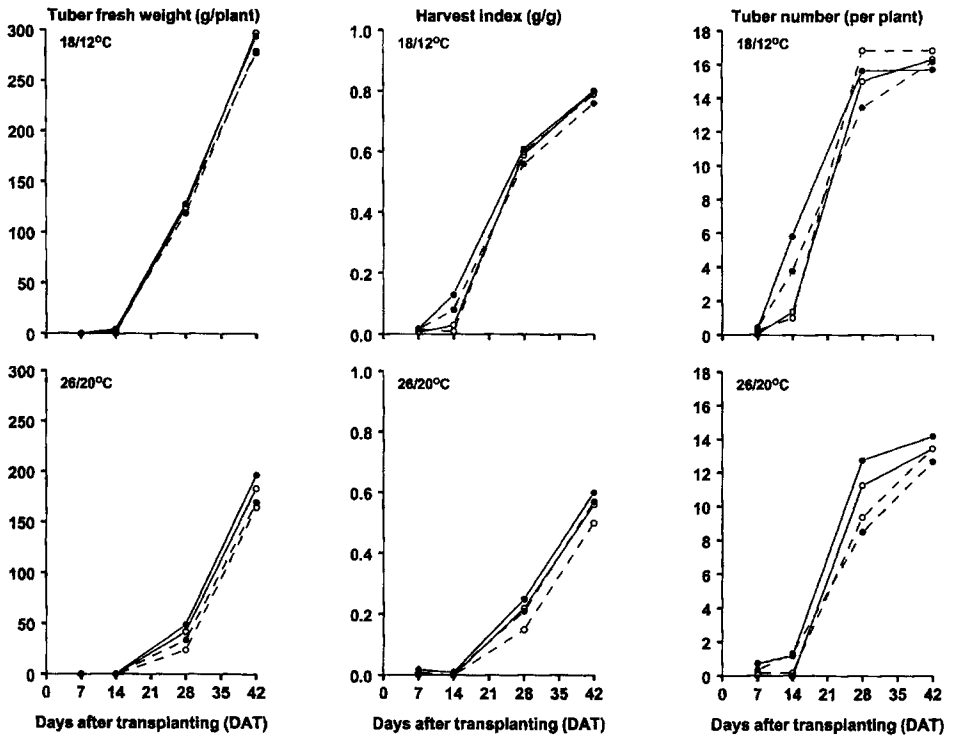
higher for plants produced at lower normalisation temperature, for plants growing at high temperature in the tuber production phase at 28 DAT and for all plants at the end of the tuber production phase (Figure 2). Plantlets pre-grown at lower normalisation temperature had a significantly higher total plant dry weight at both temperatures in the tuber production phase at 7 DAT (Table 2) and for plants grown at a high tuber production temperature at 28 DAT (Figure 1). Plants pre-grown at lower normalisation temperature also had a higher HI at the end of the tuber production period (Figure 2).

A lower normalisation temperature resulted in significantly more tubers at 28 DAT, but only at high temperature during tuber production (Figure 2). At the end of the tuber production period, there were no significant after-effects of the normalisation temperature on the average weight of the tubers produced or their dry matter concentration.

After-effects of transplant production temperature. Plants pre-grown at high transplant production temperature had significantly higher leaf dry weight at 14 and 28 DAT in the tuber production phase than those pre-grown at low temperature (Figure 1). At 28 DAT the effect was only significant for plants grown at the highest normalisation and tuber production temperatures. Stem dry weight in the tuber production phase was usually also significantly higher for plants pre-grown at higher transplant production temperature (Table 2, Figure 1). A higher transplant production temperature, however, resulted in significantly lower tuber dry weight for plants grown at the low tuber production temperature at 14 DAT, and for plants grown at the high tuber production temperature at 42 DAT (Figure 1). Plants pre-grown at high transplant production temperature also had significantly lower tuber fresh weights at 14 DAT (Figure 2), but only at low tuber production temperature. A higher transplant production temperature resulted in higher total plant dry weight at 7 DAT but this effect disappeared soon in the tuber production phase (Figure 1). Plants grown at higher transplant production temperature had a lower HI than those grown at lower transplant production temperature (Figure 2), but this effect was only significant at lower tuber production temperature at 14 DAT and at higher tuber production temperature at 28 and 42 DAT.

Plants pre-grown at high transplant production temperature had fewer tubers early in the tuber production phase than those pre-grown at low temperature. At 14 DAT the difference was only significant for plants growing at low temperature during tuber production (Figure 2). At the end of the tuber production period, there were no significant after-effects of the transplant production temperature on the average weight of the tubers produced or their dry matter concentration (Table 3).

Effects of temperature during tuber production. High tuber production temperature enhanced leaf and stem dry weights throughout the tuber production phase, compared to low temperature during this phase (Figure 1, Table 2). Tuber dry and fresh weights were significantly lower at higher tuber production temperature (Figures 1 and 2) except at 7 DAT, when tubers were barely present, and for fresh weight at 14 DAT for plants that received a high transplant production temperature and were least



	7 DAT	14 DAT	28 DAT	42 DAT
N	ns	ns	ns	**
TP	ns	***	ns	ns
TB	ns	***	***	***
N*TP	ns	ns	ns	ns
N*TB	ns	ns	* (13.5)	ns
TP*TB	ns	** (1.05)	ns	ns
N*TP*TB	ns	ns	ns	ns

	7 DAT	14 DAT	28 DAT	42 DAT
N	ns	ns	ns	*
TP	ns	***	ns	ns
TB	ns	***	***	***
N*TP	ns	ns	ns	ns
N*TB	ns	ns	ns	ns
TP*TB	ns	*** (0.03)	ns	* (0.04)
N*TP*TB	ns	ns	ns	ns

	7 DAT	14 DAT	28 DAT	42 DAT
N	ns	ns	*	ns
TP	*	**	ns	ns
TB	ns	***	***	***
N*TP	ns	ns	* (1.90)	ns
N*TB	ns	ns	ns	ns
TP*TB	ns	** (1.06)	ns	ns
N*TP*TB	ns	ns	ns	ns

Figure 2. Effects of pre-growth at 17 °C (——) or 23 °C (- - - -) during the normalisation phase (N), and 18/12 °C (●) or 26/20 °C (○) during the transplant production phase (TP) on tuber fresh weight, harvest index and tuber number of *in vitro* produced potato plantlets growing at 18/12 °C (upper graphs) or 26/20 °C (lower graphs) in the tuber production phase (TB). For significances, see Figure 1.

advanced in tuber formation. Total plant dry weight was higher at the higher tuber production temperature at the beginning of the phase (7 DAT, Figure 1, Table 2), but this effect was reversed later (Figure 1). After 7 DAT, HI was lower at higher tuber production temperature (Figure 2).

Tuber number was lower at higher tuber production temperature from 14 DAT onwards (Figure 2). At the end of the tuber production period, tubers produced at high temperature were significantly lighter and had a lower dry matter concentration than those produced at low temperature (Table 3).

Table 3. Tuber size and tuber dry matter concentration at the end of the tuber production phase (42 DAT), of *in vitro* propagated potato plantlets grown at two normalisation (N) x two transplant production (TP) x two tuber production (TB) temperatures. Average values over two normalisation temperatures, because main effect and interactions with normalisation temperature were not significant.

Temperature (°C) during		Tuber size (g/tuber)	Tuber dry matter concentration (%)
TP	TB		
18/12	18/12	19.1	19.3
26/20	18/12	18.1	19.9
18/12	26/20	13.7	17.3
26/20	26/20	13.7	16.1
Significance[a]			
N		NS	NS
TP		NS	NS
TB		***	***
TP*TB		NS	*** (1.22)[b]

[a] *** $P < 0.001$, ** $0.001 \leq P < 0.01$, * $0.01 \leq P < 0.05$, NS: not significant $P \geq 0.05$; interactions not listed were not significant.

[b] number in brackets represents the LSD.

Discussion

Effects of transition

Transition of plants from one phase to another had a strong stimulating effect on leaf growth and a smaller on stem growth (Tables 1 and 2). Tadesse *et al.* (2001b) showed that leaf area increased logistically in all phases of growth suggesting that growth limitations occurred. By transition, the growth limitations at the end of the previous phases must have been overcome. Differences in temperature, light quality, air composition, relative humidity, a higher light intensity or a better water or nutrient availability must have stimulated growth. The stimulating effect of transition on leaf growth was larger when *in vitro* plants were planted to transplant production conditions than when transplants were transplanted to tuber production condition, as manifested by 4 fold (Table 1) versus 2 fold (Table 2) higher leaf dry weight of plants one week after transition compared to plants of the same age that were not (trans)planted. This is consistent with the boost in leaf area increase of *in vitro* plantlets after planting to soil (Tadesse *et al.*, 2001b). Also transplanting, however, stimulated leaf growth, despite the transplant shock reported for leaf area increment by Tadesse *et al.* (2001b). The transplanting effect must have been smaller than the planting effect because of this transplanting shock and because growth limitation in the relatively short transplant production phase was less severe than in the normalisation phase (Tadesse *et al.*, 2001b).

Main effects of temperature

Effects of temperature on *in vitro* propagated potato plants were similar to the effects on tuber-derived plants in the same temperature range and were also more or less

consistent throughout the phases. The positive effects of high temperature on leaf dry weight at the end of the transplant production phase and throughout the tuber production phase, and on stem dry weight in all phases (Table 1 and 2, Figure 1) agree with reports for tuber-derived plants (e.g. Ben Khedher & Ewing, 1985; Basu & Minhas, 1991).

High temperature reduced tuber dry and fresh weights (Figures 1 and 2). Van Dam *et al.* (1996) also showed that low temperature (15–19°C) is optimal for tuber initiation and initial growth. High temperature delays tuber initiation (Struik *et al.*, 1989b) and strongly reduces the proportion of total dry matter partitioned to tubers (Ewing, 1981; Ben Khedher & Ewing, 1985; Struik *et al.*, 1989a; Bennett *et al.*, 1991; Wheeler *et al.*, 1986). Associated with this, tuber fresh yield, average tuber weight, tuber dry matter concentration and HI were all lower at higher temperature (Figure 2, Table 3). This agrees with the results reported for tuber-derived potato plants by Borah & Milthorpe (1962), Bodlaender (1963), Midmore (1984) and Van Dam *et al.* (1996). The higher tuber dry matter concentration at lower than at higher temperature is associated with the positive effect of relatively low temperature on starch synthesis (Wolf *et al.*, 1991; Lafta & Lorenzen, 1995). Tuber number usually was also lower at higher temperature (Figure 2; cf. Van Dam *et al.*, 1996; Struik *et al.*, 1989b).

Total plant dry weight was higher at higher temperature at the beginning of the tuber production phase (7 DAT) but this effect was reversed later (Figure 1). At that time leaf area index was high and high temperature therefore did not result in better light interception. Maintenance respiration may also have been large in the leafy crop at higher temperature, and may have lowered total plant dry weight. Total biomass generally is also reduced at high temperature (Nagarajan & Bansal, 1990), because haulm growth (with considerable formation of structural tissue) requires more energy than starch production in tubers (Penning de Vries *et al.*, 1974). In addition, lack of strong tuber sink may have reduced total dry weight increase as illustrated by a lower rate of assimilate transport from the leaves (Basu & Minhas, 1991; Lorenzen & Ewing, 1992) and the subsequent reduction in photosynthesis at higher temperatures.

After-effects of temperature pre-treatments

After-effects of the normalisation temperature were found in the transplant production phase and even in the tuber production phase. A lower normalisation temperature resulted in higher leaf and total dry weights at 7 DAP in the transplant production phase (Table 1). This was also found for above-ground leaf area after planting in several cultivars (Tadesse *et al.*, 2000). The stimulating effects of lower normalisation temperature in the next phase are opposite to the effects or tendencies found during the normalisation period itself (Table 1, Tadesse *et al.*, 2001b). This suggests that temperature during *in vitro* growth affected the habitus of the plants, with higher temperatures leading to smaller upper leaves and consequently to lower above ground leaf areas and leaf weights in the next phase. A lower normalisation temperature also resulted in higher leaf, stem and total dry weights early in the tuber production phase (Figure 1). The higher dry weights could be the direct result of higher

weights during transplant production (Table 2). A lower normalisation temperature resulted in a significantly higher tuber fresh yield and HI at the end of the tuber production phase, although the difference in tuber dry weight was not significant. Preculturing plants at low normalisation temperature resulted in more tubers at higher tuber production temperature at 28 DAT, but fewer at the final harvest when plants were grown at low temperature during tuber production. In general, the main effect of lower normalisation temperature seemed to advance plant development.

Effects of the transplant production temperature were also carried over to the tuber production phase. Contrary to temperature during the normalisation period, plants pre-treated with lower transplant production temperature had lower leaf, stem and total dry weights during parts of or the whole tuber production phase (Figure 1). Lower transplant production temperature resulted in higher tuber dry and fresh weights, more tubers, higher tuber fresh yield and higher HI during the first weeks of the tuber production phase indicating that low temperature during the transplant production period favoured tuberisation and advanced tuber formation. At final harvest, a lower transplant production temperature only increased tuber yields, HI and dry matter concentration of tubers when temperature during tuber production was high. After-effects of transplant production temperature on tuber characteristics therefore were more or less similar to after-effects of normalisation temperature, but were clearest at an earlier stage. Low temperature stimulates tuber formation and partitioning of dry matter to tubers (Menzel, 1985; Van Dam *et al.*, 1996). Previous research showed that higher transplant production temperature could increase leaf area (Tadesse *et al.*, 2001b) and leaf dry weight (Tadesse *et al.*, 2001a), and resulted in plants with higher ground cover at the end of a short growing season (Tadesse *et al.*, 2001a) indicating that high temperature promotes or prolongs haulm growth. Associated with this, tuber initiation and formation were delayed.

Optimum temperature combinations

Temperature of the tuber production phase was more important for final yield than temperature during earlier phases, with the lower temperature giving higher yields than the higher temperature. Temperature, however, is difficult to vary when tuber production takes place in the field. Significant two- and three-way interactions were found throughout the experiment, and the expression of the effects induced during earlier phases sometimes depended on the temperature during tuber production. To optimise the production of potato seed tubers, an appropriate combination of temperature treatments must be selected, depending on the expected field conditions.

Low temperature during normalisation mainly seemed to advance crop development (enhanced vegetative growth during transplant production combined with early tuber production and higher tuber fresh yield, average tuber weight and HI at the end of the tuber production phase). This therefore may thus result in high yields being achieved earlier, which is especially important when the growing season for field production is short.

Low temperature during transplant production seemed to stimulate tuber formation and partitioning of dry matter to tubers (higher tuber weight early during transplant production), but on the other hand reduced vegetative development. Ultimately,

lower temperature during transplant production increased tuber dry weight, tuber dry matter concentration and HI only at the higher temperature during tuber production, at which vegetative development was more pronounced and partitioning of dry matter to tubers reduced. Producing transplants at lower temperature therefore is especially important when field conditions are likely to limit dry matter partitioning to tubers, as is the case under warm conditions.

High transplant production temperatures increased leaf and/or stem dry weights in the tuber production phase, but delayed tuber formation. This could have yield advantages under conditions where a higher radiation interception is limiting yield and partitioning of dry matter to tubers is still sufficient to ensure higher tuber yields, for instance when field conditions are strongly inductive to tuberisation, e.g. cool conditions at short daylengths.

This research was carried out with the very early cv. Gloria. This cultivar shows a very strong partitioning of dry matter to tubers under field conditions (Lommen, 1999). Compared to other cultivars, its performance seemed to be more difficult to manipulate by temperature than that of other cultivars (cf. Tadesse et al., 2000, 2001a). We therefore expect after-effects to show even more clearly in other cultivars.

References

Basu, P.S. & J.S. Minhas, 1991. Heat tolerance and assimilate transport in different potato genotypes. *Journal of Experimental Botany* 42: 861–866.

Ben Khedher, M. & E.E. Ewing, 1985. Growth analysis of eleven potato cultivars grown in the greenhouse under long photoperiods with and without heat stress. *American Potato Journal* 62: 537–554.

Bennett, S.M., T.W. Tibbitts & W. Cao, 1991. Diurnal temperature fluctuation effects on potatoes grown with 12h photoperiod. *American Potato Journal* 68: 81–86.

Beukema, H.P. & D.E. Van der Zaag, 1990. Introduction to potato production. Pudoc, Wageningen, 208 pp.

Bodlaender, K.B.A., 1963. Influence of temperature, radiation and photoperiod on development and yield. The growth of the potato. Proceedings of the 10th Easter School Agricultural Science, University of Nottingham, pp. 199–210.

Borah, M.N. & F.L. Milthorpe, 1962. Growth of the potato as influenced by temperature. *Indian Journal of Plant Physiology* 5: 53–72.

Burton, W.G., 1989. The Potato. John Wiley & Sons, Inc., New York, 742 pp.

Charles, G.L., L. Rossignol & M. Rossignol, 1992. Environmental effects on potato plants *in vitro*. *Journal of Plant Physiology* 139: 708–713.

Ewing, E.E., 1981. Heat stress and the tuberization stimulus. *American Potato Journal* 58: 31–49.

Gregory, L.E., 1956. Some factors for tuberization in the potato. *Annals of Botany* 43: 281–288.

Haverkort, A.J., M. Van de Waart & J. Marinus, 1991. Field performance of potato microtubers as propagation material. *Potato Research* 34: 353–364.

Hussey, G. & N.J. Stacey, 1981. *In vitro* propagation of potato (*Solanum tuberosum* L.). *Annals of Botany* 48: 787–796.

Ingram, K.T. & D.E. McCloud, 1984. Simulation of potato crop growth and development. *Crop Science* 24: 21–27.

Jones, E.D., 1988. A current assessment of *in vitro* culture and other rapid multiplication methods in North America and Europe. *American Potato Journal* 65: 209–220.

Lafta, A.M. & J.H. Lorenzen, 1995. Effect of high temperature on plant growth and carbohydrate metabolism in potato. *Plant Physiology* 109: 637–643.

Lommen, W.J.M., 1999. Causes for low tuber yields of transplants from *in vitro* potato plantlets of early cultivars after field planting. *Journal of Agricultural Science, Cambridge* 133: 275–284.

Lorenzen, J.H. & E.E. Ewing, 1992. Starch accumulation in potato (Solanum tuberosum) leaves during the first 18 days of photoperiod treatment. *Annals of Botany* 69: 481–486.

Marinus, J., 1985. *'In vitro' multiplication of potatoes: Description of methods and experience in The Netherlands*. CABO, Wageningen, 21 pp.

Midmore, D.J., 1984. Potato (*Solanum* spp.) in the hot tropics. I. Soil temperature effects on emergence, plant development and yield. *Field Crops Research* 8: 255–271.

Menzel, C.M., 1985. The control of storage organ formation in potato and other species. A review. Part 1. *Field Crops Abstracts* 38: 527–537.

Murashige, T. & F. Skoog, 1962. A revised medium for rapid growth and bioassays with tobacco tissue cultures. *Physiologia Plantarum* 15: 473–479.

Nagarajan, S. & K.C. Bansal, 1990. Growth and distribution of dry matter in a heat tolerant and a susceptible potato cultivar under normal and high temperature. *Journal of Agronomy and Crop Science* 165: 306–311.

Penning de Vries, F.W.T., A.H.M. Brunsting & H.H. Van Laar, 1974. Products, requirements and efficiency of biosynthesis: a quantitative approach. *Journal of Theoretical Biology* 45: 339–377.

Slater, J.W., 1963. Mechanisms of tuber initiation. In: J.D. Ivins & F.L. Milthorpe (Eds), The growth of the potato. Proceedings of the 10th Easter School of Agricultural Science, University of Nottingham, pp 114–120.

Struik, P.C., J. Geertsema & C.H.M.G. Custers, 1989a. Effects of shoot, root and stolon temperature on the development of the potato (*Solanum tuberosum* L.) plant. I. Development of the haulm. *Potato Research* 32: 133–141.

Struik, P.C., J. Geertsema & C.H.M.G. Custers, 1989b. Effects of shoot, root and stolon temperature on the development of the potato (*Solanum tuberosum* L.) plant. III. Development of tubers. *Potato Research* 32: 151–158.

Struik, P.C. & E.E. Ewing, 1995. Crop physiology of potato (*Solanum tuberosum*): responses to photoperiod and temperature relevant to crop modelling. In: A.J. Haverkort & D.K.L. MacKerron (Eds), Potato ecology and modelling of crops under conditions limiting growth. Kluwer Academic Publishers, Dordrecht, pp. 19–40.

Struik, P.C. & S.G. Wiersema, 1999. Seed Potato Technology. Wageningen Pers, Wageningen, 383 pp.

Tadesse, M., W.J.M. Lommen & P.C. Struik, 2000. Effects of *in vitro* treatments on leaf area growth of potato transplants during acclimatisation. *Plant Cell Tissue and Organ Culture* 61: 59–67.

Tadesse, M., W.J.M. Lommen & P.C. Struik, 2001a. Effects of temperature pre-treatment of transplants from *in vitro* produced potato plantlets on transplant growth and yield in the field. *Potato Research* 44: 173-185.

Tadesse, M., W.J.M. Lommen, P.E.L. Van der Putten & P.C. Struik, 2001b. Development of leaf area and leaf number of micropropagated potato plants. *Netherlands Journal of Agricultural Science*, 49:15-32.

Van Dam, J., P.L. Kooman & P.C. Struik, 1996. Effects of temperature and photoperiod on early growth and final number of tubers in potato (*Solanum tuberosum* L.). *Potato Research* 39: 51–62.

Wheeler, R.M., K.L. Steffen, T.W. Tibbittis & J.P. Palta, 1986. Utilisation of potatoes for life support systems. II. The effects of temperature under 24-h and 12-h photoperiods. *American Potato Journal* 63: 639–647.

Wolf, S., A. Marani & J. Rudich, 1991. Effects of temperature on carbohydrate metabolism in potato plants. *Journal of Experimental Botany* 42: 619–625.