New Phytol. (1999), **143**, 339–349

Climatic determinants of budburst seasonality in four temperate-zone tree species

ISABELLE CHUINE* AND PIERRE COUR

Paléoenvironnements & Palynologie, UMR CNRS 5554, Institut des Sciences de l'Evolution de Montpellier, Université Montpellier II, France

Received 1 October 1998; accepted 8 April 1999

SUMMARY

Several physiological processes controlling tree phenology remain poorly understood and in particular bud dormancy. Many studies have emphasised the action of chilling temperatures in breaking dormancy. However, the effect of the preceding summer temperatures has rarely been investigated although there is some evidence that they may be involved in the settlement and intensity of dormancy as well as cold acclimation. In this paper, thermal time to budburst in relation to the duration of chilling outdoors, preceding summer temperatures and forcing temperatures was studied by outdoors experiments in seedlings of *Platanus acerifolia*, *Vitis vinifera*, *Quercus pubescens* and *Castanea sativa*. Results showed that temperatures of the preceding summer had no significant effect on the timing of budburst, *P. acerifolia* and *Q. pubescens* showed a very weak response to the duration of chilling, and the phenological characteristics of each species were found to be adapted to the climate conditions of its own geographical area. The phenological model used in this study explained 82–100 % of the variance of the data without taking into account summer temperatures. Thus, although summer temperatures may be well involved in the intensity of dormancy and cold hardiness, they do not significantly affect budburst and therefore may not need to be considered in phenological models for predicting budburst.

Key words: phenology, transfer experiments, bud dormancy, phenological models, budburst, climate.

INTRODUCTION

Since investigations by Reaumur (1735), it has been known that the phenology of temperate and boreal-zone tree species is influenced by meteorological conditions. Since then, extensive experimental and simulation work on tree phenology has provided much information on the relationship between bud growth and meteorological conditions (Robertson, 1968; Sarvas, 1974; Cannell & Smith, 1983; Kobayashi & Fuchigami, 1983; Murray *et al.*, 1989; Hänninen, 1991, 1995, 1996; Hänninen *et al.*, 1993; Kramer, 1994b). Research on tree phenology has known a revival in the last 10 yr due to climate warming. An important component of this recent work has involved obtaining accurate predictions of tree phenology (Hänninen, 1990; Hunter & Lechowicz, 1992; Kramer, 1994b; Chuine *et al.*, 1998, 1999). Phenological models are currently needed to allow reconstruction and prediction of the primary productivity of ecosystems (Lieth, 1971; Kramer & Mohren, 1996; Kramer *et al.*, 1996), evaluate the risks of frost damage in a warming climate (Cannell & Smith, 1986; Murray *et al.*, 1989, 1994; Prentice *et al.*, 1991; Hänninen *et al.*, 1993; Kramer, 1994a, 1995; Hänninen 1996), and develop accurate decision support tools for fruit growers and reforestation programmes.

Although accuracy of budburst models has been greatly improved in recent work (Kramer, 1994b; Chuine *et al.*, 1998, 1999), several physiological processes which control tree phenology remain poorly understood, such as how bud dormancy might be broken by the action of chilling temperatures (Perry, 1971) and how important is that trait for the timing of budburst. Bud dormancy is a physiological state usually defined as a period of reduced growth rate with few or no cell divisions and in which chilling temperatures are required for growth and development to recommence (Perry, 1971). However, the definition of bud dormancy is

*Author for correspondence : Department of Forest Sciences, University of British Columbia, 3041–2424 Main Mall, Vancouver, BC V6T 1Z4, Canada (tel + 1 604 822 6020; fax + 1 604 822 9102; e-mail ichuine@interchange.ubc.ca).

still controversial because it is a gradually changing state: the transitions are gradual from the state of imperceptible growth to the state of true winter rest, then to the state of quiescence (when the plant is capable of renewed vegetative growth). Many experiments have shown that bud dormancy can be naturally broken by chilling temperatures below 10°C during autumn and early winter (Perry, 1971; Hänninen & Backman, 1994; Sarvas, 1974), but the effect of preceding periods on dormancy and budburst has rarely been studied (Mauget, 1977; Champagnat, 1983). In particular, as dormancy takes place at the end of the summer, temperatures of the summer preceding budburst might be involved in the intensity of dormancy and consequently in the timing of budburst. Differences in the response of phenology to temperatures or in cold hardiness, which is closely related to dormancy (Campbell & Sorensen, 1973; Burr *et al.*, 1989; Qamaruddin *et al.*, 1993), between coastal and inland tree populations have already been reported (Sorensen, 1973; Rehfeldt, 1989; Aitken & Adams, 1997; White *et al.*, 1997). The correlation existing between summer and winter temperatures over a continental gradient raises the possibility that summer temperatures may be a proximal factor affecting the timing of budburst and cold hardiness of trees. If so, warm summers inland may lead to hardier and more dormant buds in prevision of more severe frosts in autumn, winter and spring, and in contrast to the mild summers on the coast which might lead to less dormant and less hardy buds in prevision of less severe frosts. Some studies suggest that this may be the case in conifers: cold hardiness has been shown to be positively correlated to summer temperatures (Balduman *et al.*, 1999) and coastal populations usually have fewer chilling requirements (suggesting that they are less dormant) than inland populations (Campbell, 1974; Sorensen, 1983). Consequently, budburst models which do not consider summer temperatures for predicting the timing of budburst, might be improved if the relationship between summer temperatures and budburst can be established.

In the present investigation, the effects of summer temperatures and autumn/winter chilling temperatures on budburst of four European species: *Platanus acerifolia*, *Castanea sativa*, *Quercus pubescens* and *Vitis vinifera*, have been studied experimentally over 2 yr in natural climatic conditions. A modelling analysis using recently developed phenological models was used to determine the date of transition between dormancy and quiescence, and therefore the precise amount of chilling and forcing temperatures received outdoors. The following questions were addressed in this study. Do summer temperatures have a significant effect on budburst of the following spring? Should they be taken into account in phenological models? What are the precise climatic determinants of budburst for the species studied?

MATERIALS AND METHODS

The experiment was conducted outdoors at two locations in southern France: the Centre National de la Recherche Scientifique experimental gardens on the outskirts of Montpellier (43.3°N, 3.6°E, 20 m above sea level) at 10 km from the Mediterranean and at the Office National des Forêts nursery in Roquedols near Meyrueis (44.4°N, 3.2°E, 650 m above sea level), on the north face of the Mont Aigoual, *c.* 100 km from the Mediterranean. Minimum and maximum temperatures averaged over the 2 yr of the experiment were 0.6°C, 8.6°C (Jan.) and 9.2°C, 25.5°C (July) at Roquedols and 4.4°C, 12.0°C (Jan.) and 17.3°C, 30.1°C (July) at Montpellier.

In September 1996, 2-yr-old seedlings of four species, *Platanus acerifolia* Willd., *Quercus pubescens* Willd., *Castanea sativa* Mill. and *Vitis vinifera* L. were obtained from nurseries in southern France. All seedlings of each species were grown from a single open pollinated parent, except those of *V. vinifera* which were grown from cuttings of clone 237 R 110. These four species are widespread in the region of Montpellier (southern France), as natural (*Q. pubescens*, *C. sativa*) or cultivated (*V. vinifera*) populations or ornamental trees (*P. acerifolia*), on plains (*Q. pubescens*, *V. vinifera*, *P. acerifolia*) or at altitude (*C. sativa*). In August 1996, the 2-yr-old seedlings of each species were repotted into 3-l plastic containers containing vegetable mould and fertilized peat (excepted *V. vinifera*, 0.5-l container containing marl with a high proportion of carbonate fine soil with fertilized peat). Trees were grouped by species in an open area of *c.* 20 m² at both stations (Montpellier and Roquedols) and were irrigated as required. For recording and water supply facilities seedlings were grouped by species. Air temperature was registered bi-hourly with a temperature recorder (type NG 5484, Jules Richard, Paris, France) in a meteorological house 10 cm above ground level.

Experiment I

Seedlings of each species were divided into three groups of seven individuals, except *P. acerifolia* for which 14 seedlings were divided in two groups. Group I1 remained in Montpellier, whereas groups I2 and I3 were moved to Roquedols, the high elevation site, on 30 September 1996. Groups I2 and I3 were kept 4 and 5 months, respectively, at Roquedols before being returned to Montpellier (Fig. 1). Phenological stage was recorded once a week from the beginning of the experiment to the first stage and then three times a week at each site according to the following scale: 1, slightly swollen; 2, swollen; 3, leaves visible in the bud; 4, leaves completely unfolded. A stage was considered reached on the Julian date when 50% of the buds of each seedling fulfilled the criterion.

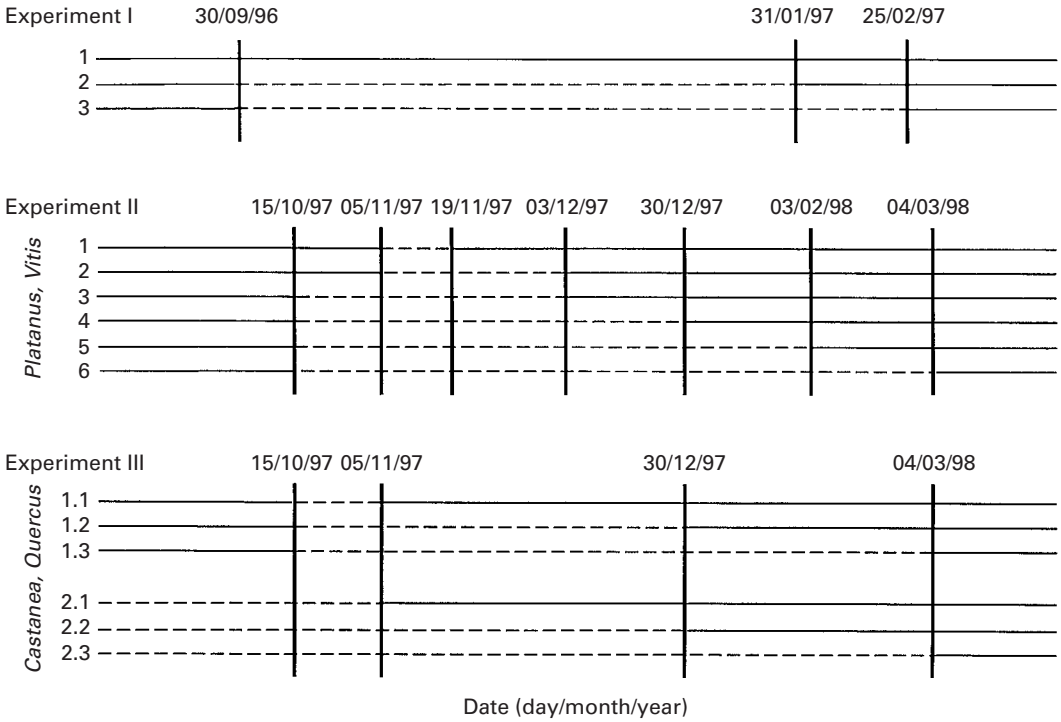

Fig. 1. Experimental design of Experiments I, II and III. Solid lines, time spent in Montpellier; broken lines time spent in Roquedols. Minimum and maximum temperatures for January and July: 4.4–12.0°C and 17.3–30.1°C, respectively, (Montpellier) and 0.6–8.6°C and 9.2–25.5°C, respectively (Roquedols). See the Materials and Methods section for details of the experimental groups.

Experiment II

Three-yr-old seedlings of *P. acerifolia* and *V. vinifera* were divided into six groups of seven seedlings. *Platanus* seedlings were first repotted into 15-l containers whereas *Vitis* seedlings were kept in their 3-l pots. All groups were kept in Montpellier until 15 October 1997, when they were all moved to Roquedols for 2 wk (group II1), 4 wk (groups II2), 7 wk (group II3), 10 wk (group II4), 15 wk (group II5) and 20 wk (group II6) (see Fig. 1). Budburst date was recorded as in Experiment I.

Experiment III

Three-yr-old seedlings of *C. sativa* and *Q. pubescens* were divided into six groups of seven individuals. During the summer 1997, groups III1.1, III1.2, III1.3 were kept in Montpellier, whereas groups III2.1, III2.2, III2.3 were kept in Roquedols (Fig.1). On 15 October 1997, seedlings were moved to Roquedols. After 2 wk (groups III1.1 and III2.1), 11 wk (groups III1.2 and III2.2) or 26 wk (groups III1.3 and III2.3) they were moved back to Montpellier where budburst date was recorded as in Experiment I (Fig. 1).

Data analysis

A priori *analyses of variance*. ANOVA was performed on the budburst dates of each species recorded in Experiments I, II and III to test the significance of the among-groups differences.

Modelling analysis. The Spring–Warming model (Cannell & Smith, 1983), SeqSar and Par1Sar models (Chuine *et al.*, 1999) were used in the present analysis because they have been shown to be the most effective models among the different ones proposed in the literature. The Spring–Warming model assumes that budburst occurs when a critical sum of degree-days (more generally named forcing units, F^*) above a certain threshold (Tb), cumulated from a fixed date (t_1) is attained (Table 1). The SeqSar and Par1Sar models assume that chilling, which also accelerates bud development from quiescence to budburst, breaks dormancy. In these models, the critical state of forcing (F^*) required for budburst is determined by the state of chilling (C^*) according to a negative exponential model ($F^* = a\,e^{-bC^*}$, where a and b are >0). F^* and C^* are sums of chilling and forcing units, respectively, which are both particular functions of the temperature. In the Par1Sar model, when a given date is reached, forcing temperatures become fully active and chilling temperatures remain active and accelerates bud development (Table 1). By contrast, forcing and chilling temperatures act sequentially in the SeqSar model, chilling temperatures are not active after the break of dormancy.

Internal validity. The daily mean temperature computed as the average of the daily maximum and

Table 1. *Description of the Spring–Warming, SeqSar and Par1Sar models*

y	Date of flowering
x_t	Daily mean temperature (°C)
$R_f(x_t)$	Forcing rate function
$R_c(x_t)$	Chilling rate function
S_f	State of forcing
S_c	State of chilling
C^*	Critical value of state of chilling for the transition from rest to quiescence
F^*	Critical value of state of forcing for the transition from quiescence to flowering
t_0	Date of onset of rest
t_1	Date of onset of quiescence
T_b	Base temperature
T_o	Optimal temperature of the rate of chilling
a, b	Constants ($a>0$, $b<0$)

Spring–Warming model

$$y \text{ such as } S_f(y) = \sum_{to}^{y} R_f(x_t) = F^*$$

$$R_f(x_t) = \begin{cases} 0 & x_t \leqslant Tb \\ x_t - Tb & x_t > Tb \end{cases}$$

For both the SeqSar and Par1Sar models

$$y \text{ such as } S_f(y) = \sum_{to}^{y} R_f(x_t) = F^*$$

$$F^* = a\, e^{bSc(y)}$$

$$R_f(x_t) = \begin{cases} 0 & \\ \dfrac{28.4}{1+e^{-0.185(x_t-18.4)}} & x_t \leqslant 0 \\ & x_t > 0 \end{cases}$$

$$R_f(x_t) = \begin{cases} 0 & x_t \leqslant -3.4 \text{ or } x_t \geqslant 10.4 \\ \dfrac{x_t+3.4}{T_o+3.4} & -3.4 < x_t \leqslant T_o \\ \dfrac{x_t-10.4}{T_o-10.4} & T_o < x_t < 10.4 \end{cases}$$

SeqSar model

$$S_c(t) = \sum_{to}^{tl} R_c(x_t)$$

$$S_f(t) = \sum_{tl}^{t} R_f(x_t)$$

Par1Sar model

$$S_c(t) = \sum_{to}^{t} R_c(x_t)$$

$$S_f(t) = \sum_{tl}^{t} R_f(x_t)$$

minimum temperatures was used for the modelling analysis. Each parameter of the three models were first fitted using the budburst dates obtained in Experiment II (*Platanus* and *Vitis*) and Experiment III (*Castanea* and *Quercus*), with a simulated annealing method described in Chuine *et al*. (1998). An F-test was used to assess the goodness of fit.

Model selection. A cross-validation method was used to assess the accuracy of prediction of the models as follows. We computed the percentages of variance explained by the models ($R^2 = (SS_{tot} - SS_{res})/SS_{tot}$, where SS_{tot} is the total sum of squares, and SS_{res} is the residual sum of squares) when used to predict the dates of budburst of Experiment I (i.e. R^2 values were computed on external data). The best model was selected according to the highest external R^2 value (models have similar degrees of freedom) .

Estimation. To obtain more reliable estimates, each parameter of the best model identified was fitted a second time to the budburst dates recorded in Experiments I and II (*Platanus* and *Vitis*), and Experiments I and III (*Castanea* and *Quercus*). Those estimates were used to determine how many chilling units were accumulated from the 1 October to the date of the onset of quiescence (t_1) and how many forcing units were accumulated from t_1 to the date of budburst for each group. This modelling analysis allowed us to compare precisely the different treatments for the data analysis (i.e. the amount of chilling units and forcing units achieved for the different groups).

RESULTS

Experiment I

ANOVA (Table 2) showed that the dates of budburst recorded in Experiment I were not significantly different among treatments (length of the period spent in altitude during dormancy) for *Platanus* ($P = 0.36$), almost significantly different for *Quercus* ($P = 0.054$) and significantly different for *Castanea* ($P < 10^{-4}$) and *Vitis* ($P < 10^{-6}$). Dates of budburst of *Platanus* were very similar (2.1 days of difference on average) between group I1, kept in Montpellier all the year and group I2, kept for 4 months at altitude (Table 3, Fig. 2). Budburst occurred first in group I1 for *Vitis*, whereas for *Castanea* and *Quercus* it occurred first in group I2 (Table 3, Fig. 2). *Vitis*

Table 2. *ANOVA and corrected design ANOVA on the dates of budburst recorded in Experiment I*

Species	Groups	R^2 (%)	F	P
Platanus		8.3	$F_{1,10} = 0.9$	0.36
Vitis	I1, I2, I3	75.7	$F_{2,19} = 29.6$	$<10^{-6}$
	I1, I2	88.4	$F_{1,13} = 98.3$	$<10^{-6}$
	I2, I3	50.8	$F_{1,12} = 12.4$	0.004
Castanea	I1, I2, I3	63.0	$F_{2,18} = 15.3$	$<10^{-4}$
	I1, I2	51.7	$F_{1,12} = 12.9$	0.004
	I2, I3	69.4	$F_{1,12} = 27.2$	$<10^{-6}$
Quercus	I1, I2, I3	30.6	$F_{2,16} = 3.5$	0.054
	I1, I2	6.4	$F_{1,12} = 0.8$	0.383
	I2, I3	67.2	$F_{1,10} = 20.5$	0.001

Groups I1 and I2 experienced the same forcing conditions, but different chilling conditions whereas groups I2 and I3 experienced the same chilling conditions, but different forcing conditions. R^2 = percentage of variance explained.

Table 3. *Mean Julian dates of budhurst (stage 4) of each group for the three experiments (I, II, III) (first column) and number of days to budburst after transfer to Montpellier (second column)*

Experiment		*Platanus*		*Vitis*		Experiment		*Quercus*		*Castanea*	
I	1	78.5	78.5	71.9	71.9	I	1	80.6	80.6	75.6	75.6
	2	75.5	44.5	77.1	46.1		2	77.9	46.9	70.9	39.9
	3			84.7	28.7		3	85.8	29.8	77.4	21.4
II	1	88.3	130.3	84.2	126.2	III	1.1	96.7	138.7	101.3	143.3
	2	84.7	112.7	79.4	107.4		1.2	98.0	97.7	91.9	102.3
	3	86.7	114.7	79.0	107.0		1.3	105.8	42.8	101.4	38.4
	4	88.0	89.0	83.1	84.1		2.1	84.0	140	90.4	146.4
	5	90.0	56.0	86.0	52.0		2.2	84.8	85.0	83.8	91.4
	6	107.0	44.4	101.7	38.7		2.3	99.0	36.0	93.0	30.0

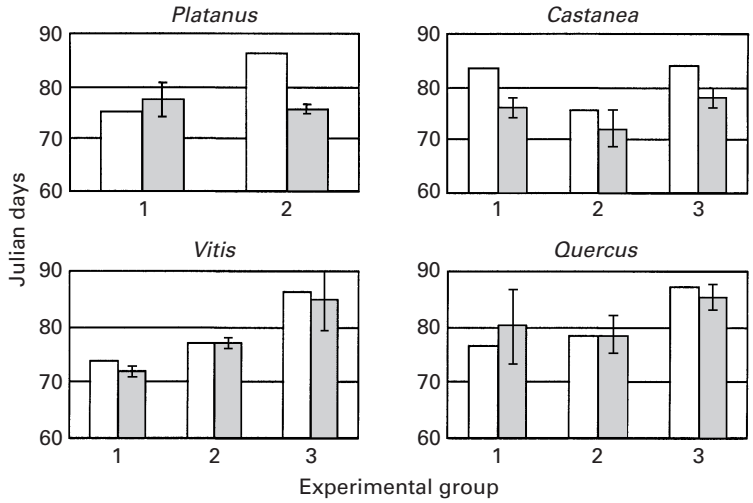

Fig. 2. Observed (filled bars) Julian dates of budburst for each species during Experiment I with standard errors and predicted (open bars) Julian dates of budburst according to the SeqSar model fitted on the dates of budburst recorded in Experiment II (*Platanus*, *Vitis*) or III (*Castanea*, *Quercus*). See the Materials and Methods section for details of the experimental groups.

showed the highest variance in budburst time among groups and the lowest within-groups variance, which is consistent with the fact that *Vitis* is a clone (Fig. 2).

Experiment II

ANOVA showed that the average dates of budburst of *Platanus* and *Vitis* were significantly different among treatments ($P<10^{-6}$, Table 4). As in Experiment I, *Vitis* showed the greatest variance among groups and the lowest within-group variance (Fig. 3).

Experiment III

ANOVA showed that the dates of budburst of *Castanea* and *Quercus* were significantly different between groups III1(.1, .2, .3) and III2 (.1, .2, .3) ($P<10^{-4}$ and $<10^{-5}$ for *Castanea* and *Quercus*, respectively) as well as among groups III1.1, III1.2 and III1.3 or III2.1, III2.2 and III2.3 ($P = 0.004$ and $P<10^{-4}$ for *Castanea* and *Quercus*, respectively).

Table 4. *ANOVA and corrected design ANOVA on the dates of budburst recorded in Experiment II*

Species	Groups	R^2 (%)	F	P
Platanus	II1–II6	87.8	$F_{5,32} = 46.1$	$<10^{-6}$
	II1–II4	22.2	$F_{3,23} = 2.2$	0.12
	II4–II6	93.1	$F_{2,14} = 94.1$	$<10^{-6}$
Vitis	II1–II6	94.8	$F_{5,33} = 121.2$	$<10^{-6}$
	II1–II4	71.3	$F_{3,21} = 17.4$	$<10^{-5}$
	II4–II6	94.1	$F_{2,18} = 142.7$	$<10^{-6}$

Groups II4–II6 have experienced the same chilling conditions, but different forcing accumulation rates, whereas groups II1–II4 have experienced the same forcing accumulation rate, but different chilling conditions.

If both factors (summer temperatures and chilling plus forcing temperatures) had a significant effect on budburst, their interaction was not significant for these two species (Table 5).

Seedlings of *Quercus* and *Castanea* kept at altitude during the summer (groups III2.1, III2.2, III2.3) broke bud 10.6 d and 8.9 d earlier, respectively, than

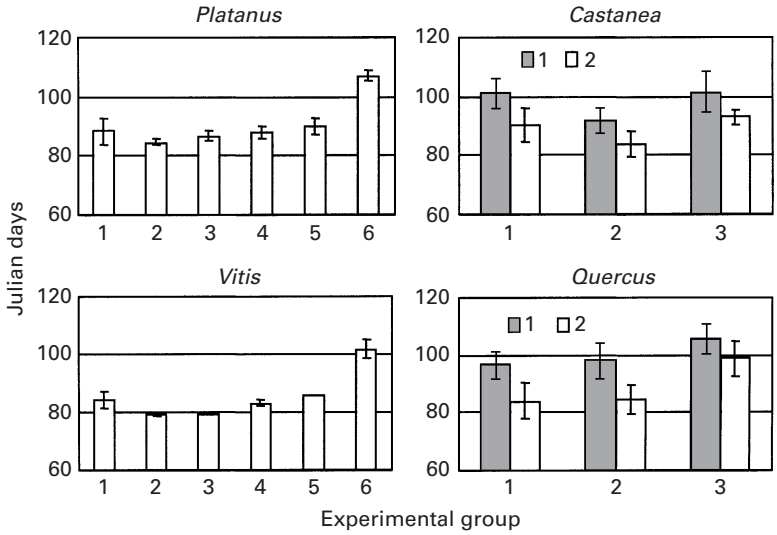

Fig. 3. Observed Julian dates of budburst for each species during Experiment II (*Platanus*, *Vitis*) and III (*Castanea*, *Quercus*) with standard errors. Filled bars, group III1 (.1, .2 .3); open bars, group III2 (.1, .2, .3). See the Materials and Methods section for details of the experimental groups.

Table 5. *ANOVA on the dates of budburst recorded in Experiment III*

Species	Factor	R^2 (%)	F	P
Castanea	(Chilling + forcing) temperatures	23.5	$F_{2,24} = 7.0$	0.004
	Summer temperatures	35.6	$F_{1,24} = 21.1$	0.0001
	Interaction	0.2	$F_{2,24} = 0.1$	0.927
Quercus	(Chilling + forcing) temperatures	31.4	$F_{2,30} = 16.1$	$<10^{-4}$
	Summer temperatures	36.1	$F_{1,30} = 37.0$	$<10^{-5}$
	Interaction	3.6	$F_{2,30} = 1.7$	0.204

those kept in Montpellier (groups III1.1, III1.2, III1.3) (Table 3). For both species, it seems that lower temperatures during the summer allowed earlier budburst irrespective of the temperature conditions of the autumn, winter and spring. The delay between groups III1 (.1, .2, .3) and groups III2 (.1, .2, .3) is on average greater than the delay between groups III1.1, III1.2, III1.3 or III2.1, III2.2, III2.3 induced by chilling temperatures, indicating that summer temperatures have a stronger effect than chilling and forcing temperatures on budburst.

For *Quercus*, the shorter the period spent at altitude, the earlier the date of budburst (budburst occurred first in groups III1.1 and III2.1), whereas for *Castanea*, the group kept 2 months at altitude budburst earlier than the group kept 3 wk and 26 wk at altitude (Fig. 3). *Castanea* thus requires more chilling temperatures in autumn than *Quercus*.

Modelling analysis

Internal validity. Percentages of variance explained by the SeqSar model were on average the highest (82–100 %), although the goodness of fit was only significant for *Quercus* and *Castanea* (Table 6). The Spring–Warming and Par1Sar models explained less variance than the SeqSar model and goodness of fit were not significant for any species. The Spring–Warming model was globally the least effective

Table 6. *Percentages of variance explained by the Spring–Warming (SW), SeqSar and Par1Sar models for each species*

Species	Model SW	SeqSar	Par1Sar
(a) ($n = 6$)			
Platanus	0.89 ns	0.83 ns	0.81 ns
Vitis	0.63 ns	0.82 ns	0.78 ns
Quercus	0.45 ns	0.96 ns	0.69 ns
Castanea	0.21 ns	1.00**	0.69 ns
(b) ($n = 8$ or 9)			
Platanus	0.77*	0.88*	0.79 ns
Vitis	0.74*	0.88*	0.83*
Quercus	0.73*	0.96***	0.69 ns
Castanea	0.80*	0.95**	0.80 ns

(a), models were fitted with the dates of budburst of Experiment II or III, (b), models were fitted with the dates of budburst of all experiments. ***, $P<0.001$; **, $P<0.01$; *, $P<0.05$; ns, not significant.

Table 7. *Amount of chilling units (C*) and forcing units (F*) accumulated by each group during each Experiment (I, II, III) according to the SeqSar model estimates*

		Platanus		*Vitis*				*Quercus*		*Castanea*	
Experiment		*C**	*F**	*C**	*F**	Experiment		*C**	*F**	*C**	*F**
I	1	51	472	25	475	I	1	25	610	56	410
	2	65	419	38	419		2	38	477	71	325
	3			38	419		3	38	477	71	325
II	1	43	504	22	492	III	1.1	20	678	49	460
	2	45	497	25	477		1.2	27	594	61	378
	3	50	473	29	458		1.3	27	594	64	363
	4	48	482	30	455		2.1	29	563	58	396
	5	48	482	30	455		2.2	34	516	68	338
	6	48	482	30	455		2.3	34	516	71	324

model (R^2 between 21 % and 89 %), although it performed almost as well as the SeqSar and Par1Sar model for *Platanus*. This result supports the hypothesis that *Platanus* is only weakly influenced by chilling temperatures.

Model selection. Accuracy of predictions of the SeqSar estimates, assessed with the external R^2 values, varies greatly among species (Fig. 2). It is very high for *Vitis* (R^2 = 0.92), much lower for *Quercus* (R^2 = 0.31) and zero for *Platanus* and *Castanea* despite the small differences in days between predictions and observations. The large differences in R^2 values obtained among species are probably due to the small amount of data available to estimate the goodness of prediction (n = 2 or 3) and to the small amount of data available to fit the model (n = 6).

Estimation. Estimates of the SeqSar model parameters (best model among the three tested), fitted on all dates of budburst recorded for each species, were used to calculate both chilling units from the 1 October to the date of the onset of quiescence (t_1) and forcing units from t_1 to the date of budburst for each group (Table 7). This analysis revealed two important results.

First, it showed that some groups experienced the same amount of chilling units. This was the case for groups I2 and I3 of *Vitis*, *Quercus* and *Castanea*, groups II4, II5 and II6 of *Vitis* and *Platanus*, and groups III1.2, III1.3 and III2.2, III2.3 of *Quercus*. This is because the onset of quiescence (t_1) occurred before groups I2, II4, III1.2 and III2.2 returned to Montpellier. As a result, all these groups, that returned after t_1 to Montpellier, shared the same amount of chilling units (which are not accumulated further after t_1). The amount of forcing units required for budburst (*F**), as a function of the chilling units received (*C**), was also the same for groups I2 and I3, groups II4, II5 and II6, groups III1.2 and III1.3, and groups III2.2 and III2.3 of *Quercus*. Thus, the differences observed among these groups within each experiment were only due to the faster accumulation of forcing units for those groups which returned earlier to Montpellier: *F** was reached earlier for group I2 than group I3, for group II4 than groups II5, II6 and for groups III1.2, III2.2 than groups III1.3, III2.3.

Second, the analysis showed that groups of *Quercus* and *Castanea* kept at altitude during summer 1997, cumulated more chilling units than the groups kept in Montpellier. During 12–15 October 1997, an unusually cool event occurred in southern France and the daily mean temperature dropped to 12–18°C at Montpellier and 4–9°C at Roquedols. As a consequence, seedlings were already in chilling conditions at the beginning of October at Roquedols. The number of forcing units needed for budburst was then less for groups III2.1, III2.2, III2.3 which budburst earlier than groups III1.1, III1.2, III1.3, respectively.

Corrected design analysis of variance

Since groups I1 and I2 and groups II1–4 returned to Montpellier before the date of quiescence (t_1), forcing took place for those groups in the same conditions in Montpellier (same accumulation rate of forcing units). Thus, differences between groups I1 and I2, and among groups II1–4 were only due to different accumulated chilling units (*C**). By contrast, groups I2, I3, as well as groups II4–6, experienced the same amount of chilling units but different conditions of forcing because they returned after t_1 to Montpellier (different rates of accumulation of forcing units). Thus, although experiments were undertaken in natural conditions, effects of chilling and forcing temperatures could be tested independently, as for controlled experiments. ANOVA on the dates of budburst of groups I1, I2 and I2, I3, and groups II1–4 and II4–6, were performed to assess the effects of chilling or forcing temperatures, all else being equal.

The ANOVA performed on the dates of budburst recorded in Experiment I showed that differences between groups I1 and I2 were not significant for *Quercus*, but highly significant for *Vitis* and *Castanea* (Table 2) whereas those in Experiment II showed that differences among groups II1–4 were not significant for *Platanus*, but highly significant for *Vitis* (Table 4).

DISCUSSION

Modelling analysis

During phenological experiments, delays between phenological stages are easily recorded, but the actual moment when bud dormancy breaks down and the period when chilling and forcing temperatures have an active effect on bud growth can only be determined by modelling. The identification of the actual date of quiescence (t_1) is especially important for outdoor experiments since, in contrast to glasshouse experiments, it is not possible to deduce it from the date of transfer from controlled chilling conditions to controlled forcing conditions. The different phenological models developed to predict dates of budburst of trees (Murray *et al*., 1989; Hänninen, 1990; Hunter & Lechowicz, 1992; Kramer, 1994b; Chuine *et al*., 1999) usually only use daily mean temperatures and not photoperiod. Since the photoperiod was very similar in both experimental sites, we were able to use those models to determine the date of quiescence for each group depending on the thermal treatment as well as the chilling and forcing temperatures reached for each group during the experiments. This modelling analysis allowed us to interpret the results of the different experiments more precisely by comparing quantitatively the different thermal treatments that have been applied. Phenological models can thus be used to analyse precisely and quantitatively the thermal conditions which affected phenology from the settlement of dormancy to budburst in natural populations.

Effects of summer temperature on budburst

The modelling analysis performed with the SeqSar model, which explained 82–100 % of the variance between groups, showed that chilling conditions experienced by groups III1.1, III2.1 or groups III1.2, III2.2 or groups III1.3, III2.3 were different. This was because chilling units can be accumulated very early according to the SeqSar model (possible accumulation from 1 September 1997 and possible range of chilling temperatures: −5°C to 20°C). From 1 September to 15 October 1997, chilling units had already been accumulated in both localities because of an unusual cool event, and the difference in the amount of chilling between altitude and lowland was sufficient to allow earlier budburst in groups III1 (.1, .2, .3) than in groups III2 (.1, .2, .3). The SeqSar model, which does not take into account the preceding summer temperatures, explains 82–100 % of the among-groups differences with the differences among the amounts of chilling units experienced only. Thus, according to this model, if chilling conditions had been exactly the same for groups III1 (.1, .2, .3) and III2 (.1, .2, .3), both would have budburst at the same time, whatever the thermal conditions of the preceding summer.

Our interpretation based on the SeqSar model might be, however, an artefact due to the large range of the potential chilling temperatures considered (−5°C; 20°C). To verify that it was not the case, the data was adjusted to the same model in which the chilling temperature range was constrained between −3.4°C and 10.4°C, as used in other studies (Hänninen, 1990, 1991, 1995; Hänninen *et al*., 1993; Kramer, 1994b) based on the results of Sarvas (1974). This additional constraint lead to a decrease of 14 % of the percentages of variance explained on average. Considering, in addition, that previously fitted optimal chilling temperatures (T_o) of each species were inside the range −3.4°C to 10.4°C, except for *Vitis*, the use of the SeqSar model with unrestricted range of chilling temperatures is legitimate.

The amount of chilling required for budburst is known to be dependent on genotype and the meteorological conditions of the preceding summer (Perry, 1971). These factors have been poorly studied compared with the action of winter and spring temperatures on the break of dormancy and the onset of growth. Experiments on *Juglans regia* have shown that the intensity of bud dormancy (number of days required for budburst when exposed continuously at 20°C) declines with altitude (Mauget, 1977), suggesting that mild summers can induce higher intensities of dormancy. Similar observations have also been made on *Corylus avellana* (Champagnat, 1983) and on different conifers species by comparing coastal and inland populations (Sorensen, 1973; Rehfeldt, 1989; Aitken & Adams, 1997; White *et al*., 1997). Other experiments have shown that higher intensities of dormancy do not necessarily induce larger chilling requirements, and thus, longer time to break dormancy. Strongly dormant buds may thus renew growth earlier than less dormant buds in the same thermal conditions (Mauget, 1977, 1980; Champagnat, 1983). The studies on *J. regia* (Mauget, 1977, 1980) strongly suggest that, dormancy intensity does not influence the timing of budburst if dormancy is totally broken by the end of December, whatever the thermal conditions experienced in the preceding summer and autumn. The accuracy of the SeqSar model apparently corroborates these experiments, showing that temperatures of the

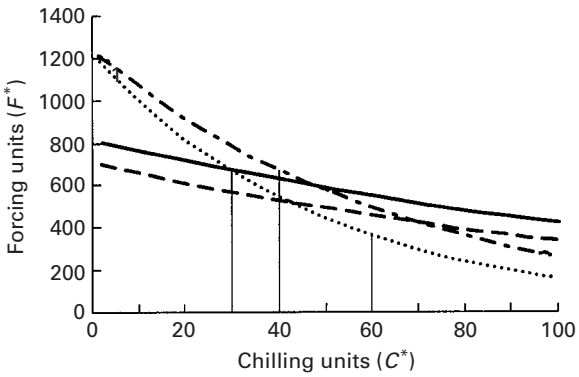

Fig. 4. Relationship between chilling units and forcing units for *Platanus acerifolia* (solid line), *Vitis vinifera* (dashed line), *Quercus pubescens* (dotted-dashed line) and *Castanea sativa* (dotted line) according to the SeqSar model estimates (see *Data analysis* in the Materials and Methods section). Vertical lines represent the conditions in Montpellier (*Platanus* and *Vitis* both experience, on average, 30 chilling units, whereas *Vitis* experiences 40, and *Castanea* 60, chilling units).

preceding summer do not influence the date of budburst although they could influence the intensity of bud dormancy. The performance of phenological models might therefore not be improved significantly by integrating events before the settlement of dormancy.

Effects of chilling temperatures on budburst

Our results show that chilling temperatures accelerate the renewal of growth once dormancy is broken; the more chilling units are cumulated, the less forcing units are subsequently needed for budburst and this corroborates various experimental results obtained for different tree species in controlled or semi-controlled conditions (Nienstaedt, 1966; Farmer, 1968; Nelson & Lavender, 1979; Mauget, 1983; Heide, 1993; Hänninen & Backman, 1994; Myking & Heide, 1995). However, marginal gains in forcing units decrease with chilling accumulation and tend to zero for very long chilling periods, as it occurs naturally in temperate and boreal climates. For that reason, one can wonder if, in natural conditions where chilling requirements are usually largely fulfilled (C^* on the asymptotic part of the curve, Fig. 4), the relationship between the length of the chilling period and the length of the forcing period required for budburst is important in predicting budburst timing. Our results show that this relationship might be important in Mediterranean natural conditions. Mediterranean winters are mild enough for average C^* to be far from the asymptotic part of the negative exponential curve: differences in C^* values lead to substantial differences in F^* values (Fig. 4).

However, the negative exponential relationship may be an artefact of budburst models for several reasons. First, the optimum chilling temperature and the size of the active chilling temperature range are still unknown with certainty and may differ from one species to another. Second, the period when chilling and forcing temperatures are active (or what is considered to be chilling and forcing temperatures) is also unknown. Hence, the negative relationship observed may be only due to a confusion in the occurrence of chilling and forcing temperatures as neither the chilling and forcing temperature ranges nor when they are active on bud growth, is known. Thus, at the end of the development, a decreasing amount of forcing temperatures is usually observed with an increasing amount of chilling temperatures, but a part of the chilling temperatures recorded may be inactive and a part of forcing temperatures may not have been taken into account, so that the relationship observed is artefactual.

Table 8. *Estimates of the SeqSar model fitted with the dates of all experiments* (*see Table 1*)

Species	T_o	a	b	t_1
Platanus	7.0	722.3	−0.0083	16/12
Vitis	−5.0	611.3	−0.0099	10/12
Quercus	1.7	1008.5	−0.0198	26/11
Castanea	1.9	987.4	−0.0156	11/1

According to the SeqSar model, the date of onset of quiescence (t_1) is very similar and very early for *Vitis*, *Platanus* and *Quercus* (10, 16 and 26 November, respectively). Dormancy is thus broken very early for these populations of southern France. This result is in agreement with experimental studies on *P. acerifolia* showing that in December, buds are able to renew growth because their dormancy is totally broken (Ricaud *et al.*, 1995).

Adapted responses to environment

According to the estimates of the SeqSar model (Table 8), two groups of species can be identified based on the relationships between chilling and forcing units (Fig. 4); *Vitis* and *Platanus*, and *Castanea* and *Quercus*. However, the average amount of chilling cumulated in southern France leads to similar amounts of forcing units needed for budburst in every species except *Castanea*, which requires much lower forcing temperatures and which had the curve with the steepest slope (Fig. 4). These phenological characteristics agree with the natural habitat conditions of each species. Seedlings of *Platanus*, *Quercus* and *Vitis* used in this study originated from southern France where the mean annual temperature is among the highest in France and where forcing temperatures are not a limiting factor. By contrast, *Castanea* predominantly grows at altitudes between 500 and 700 m in southern France (approximately the altitude of the experimental station at Roquedols) where the mean annual

temperature is 4°C lower than in the lowland, and where forcing temperature can be a limiting factor for budburst, flowering and fruit maturation. The low forcing temperature requirement of this species in southern France may be an adaptation to the sub-highland climate conditions. Another trait of *Castanea*, that corresponds to its climatic niche, is the longer period of accumulation of chilling units ($t_1 = 11$ January rather than 26 November to 16 December for the other species, Table 8) and the steep slope of the negative exponential (Fig. 4). Both ensure that a lower amount of forcing units will be necessary for budburst (not only more chilling units can be accumulated, but supplementary accumulated chilling units lead to substantial decreases in F^*).

Summer temperatures had no effect on the date of budburst, which was principally determined by the thermal conditions from quiescence to budburst and secondarily by chilling temperatures preceding this period. This result agrees with the few experiments, which have investigated the thermal conditions preceding the settlement of the dormancy (Mauget, 1977, 1980). Even if bud burst models do not simulate the exact biological process involved, this study shows that they can be helpful in analysing experimental data on phenology, in particular in natural conditions. Moreover, our results show that budburst may be weakly influenced by chilling temperatures in species such as *P acerifolia* or *Q. pubescens*. Two facts may explain this small influence of chilling temperatures on these Mediterranean species. First, they do not need high chilling requirements that would delay budburst because they experience little frost damage in their native geographical area. Second, high chilling requirements would be hardly fulfilled since autumn temperatures are rarely, on average, lower than 10°C around the Mediterranean; this is usually considered the upper limit of effective chilling temperatures. A very weak and non significant effect of chilling temperatures might also be found in other Mediterranean species, which seem to have adapted to very mild autumn and winter thermal conditions. However, the adaptive value of this trait corresponding to very mild autumn and winter conditions remains to be demonstrated.

ACKNOWLEDGEMENTS

The authors are very grateful to T. Lenormand for constructive criticisms on the manuscript, and to J. D. Thompson and G. A. O'Neill for the linguistic help. They are also grateful to D. D. Rousseau for financial support, and to S. Delavelle and J. L. Caporgneau for technical support, for having recorded the temperatures in Roquedols and for permission to conduct this experiment at the ONF of Roquedols. They are also very grateful to B. Delay, Director of the CEFE, CNRS Montpellier for allowing the experiment in the CEFE and to C. Collin for his help and technical support during the experiment. They would also like to thank G. Cambon, D. Duzer, J. Ferrier, R. Gros, A. Mignot, D. D. Rousseau and D. Vernier who participated in the transplantation of the trees and to P. Pichet who collected a part of the phenological observations in Montpellier in 1997. Last, we are grateful to H. Hänninen and anonymous referees for constructive comments on an earlier draft. Support was provided by a Bourse de Docteur Ingenieur of the Centre National de la Recherche Scientifique to I. C. This an ISEM contribution 99–047.

REFERENCES

Aitken SN, Adams WT. 1997. Spring cold hardiness under strong genetic control in Oregon populations of *Pseudostuga menziessi* var. *menziessi*. *Canadian Journal of Forest Research* **27**: 1773–1780.

Balduman LM, Aitken SN, Harmon M, Adams WT. 1999. Genetic variation in cold hardiness of Douglas-fir in relation to parent tree environment. *Canadian Journal of Forest Research* **29**: 62–72.

Burr KE, Tinus RW, Wallner SJ, King RM. 1989. Relationships among cold hardiness, root growth potential and bud dormancy in three conifers. *Tree Physiology* **5**: 291–306.

Campbell RK. 1974. Use of phenology for examining provenance transfers in reforestation of Douglas-Fir. *Journal of Applied Ecology* **11**: 1069–1080.

Cannell MGR, Smith RI. 1983. Thermal time, chill days and prediction of budburst in *Picea sitchensis*. *Journal of Applied Ecology* **20**: 951–963.

Campbell RK, Sorensen F. 1973. Cold-acclimation in seedling Douglas-fir related to phenology and provenance. *Ecology* **54**: 1148–1151.

Cannell MGR, Smith RI. 1986. Climatic warming, spring budburst and frost damage on trees. *Journal of Applied Ecology* **23**: 177–191.

Champagnat P. 1983. Quelques réflexions sur la dormance des bourgeons des végétaux ligneux. *Physiologie Végétale* **21**: 607–618.

Chuine I, Cour P, Rousseau DD. 1998. Fitting models predicting dates of flowering of temperate-zone trees using simulated annealing. *Plant, Cell and Environment* **21**: 455–466.

Chuine I, Cour P, Rousseau DD. 1999. Selecting models to predict the timing of flowering of temperate trees: implications for tree phenology modelling. *Plant, Cell and Environment* **22**: 1–13.

Farmer RE. 1968. Sweetgum dormancy release: effects of chilling, photoperiod, and genotype. *Physiologia Plantarum* **21**: 1241–1248.

Hänninen H. 1990. Modelling bud dormancy release in trees from cool and temperate regions. *Acta Forestalia Fennica* **213**: 1–47.

Hänninen H. 1991. Does climatic warming increase the risk of frost damage in northern trees? *Plant, Cell and Environment* **14**: 449–454.

Hänninen H. 1995. Effects of climatic change on trees from cool and temperate regions: an ecophysiological approach to modelling of budburst phenology. *Canadian Journal of Botany* **73**: 183–199.

Hänninen H. 1996. Effects of climatic warming on northern trees: testing the frost damage hypothesis with meteorological data from provenance transfer experiments. *Scandinavian Journal of Forestry Research* **11**: 17–25.

Hänninen H, Backman R. 1994. Rest break in Norway spruce seedlings: test of a dynamic temperature response hypothesis. *Canadian Journal of Forest Research* **24**: 558–563.

Hänninen H, Kellomäki S, Laitinen K, Pajari B, Repo T. 1993. Effect of increased winter temperature on the onset of height growth of Scots pine: a field test of a phenological model. *Silva Fennica* **27**: 251–257.

Heide OM. 1993. Daylength and thermal time responses of budburst during dormancy release in some northern deciduous trees. *Physiologia Plantarum* **88**: 531–540.

Hunter AF, Lechowicz MJ. 1992. Predicting the timing of budburst in temperate trees. *Journal of Applied Ecology* **29**: 597–604.

Kobayashi KD, Fuchigami LH. 1983. Modelling temperature effects in breaking rest in Red-osier Dogwood (*Cornus sericea* L.). *Annals of Botany* **52**: 205–215.

Kramer K. 1994a. A modelling analysis of the effects of climatic warming on the probability of spring frost damage to tree species in The Netherlands and Germany. *Plant, Cell and Environment* **17**: 367–377.

Kramer K. 1994b. Selecting a model to predict the onset of growth of *Fagus sylvatica*. *Journal of Applied Ecology* **31**: 172–181.

Kramer K. 1995. Phenotypic plasticity of the phenology of seven European tree species in relation to climatic warming. *Plant, Cell and Environment* **18**: 93–104.

Kramer K, Friend A, Leinonen I. 1996. Modelling comparison to evaluate the importance of phenology and spring frost damage for the effects of climate change on growth of mixed temperate-zone deciduous forests. *Climate Research* **7**: 31–41.

Kramer K, Mohren GMJ. 1996. Sensitivity of FORGRO to climatic change scenarios: a case study on *Betula pubescens*, *Fagus sylvatica* and *Quercus robur* in the Netherlands. *Climatic Change* **34**: 231–237.

Lieth H. 1971. The phenological viewpoint in productivity studies. In: UNESCO, eds. *Productivity of forest ecosystems*. Paris, France: UNESCO, 71–83.

Mauget J-C. 1977. Dormance des bourgeons végétatifs de Noyer (*Juglans regia* L.) cultivés sous différentes conditions climatiques. *Comptes Rendus de l'Académie des Sciences, Série D* **284**: 2351–2354.

Mauget J-C. 1980. Dormance et précocité de débourrement des bourgeons chez quelques cultivars de Noyer (*Juglans regia* L.). *Comptes Rendus de l'Académie des Sciences, Série D* **290**: 135–138.

Mauget J-C. 1983. Etude de la levée de dormance et du débourrement des bourgeons de noyer (*Juglans regia* L., cv. 'Franquette') soumis à des températures supérieures à 15°C au cours de leur période de repos apparent. *Agronomie* **3**: 745–750.

Murray MB, Cannell MGR, Smith RI. 1989. Date of budburst of fifteen tree species in Britain following climatic warming. *Journal of Applied Ecology* **26**: 693–700.

Murray MB, Smith RI, Leith ID, Fowler D, Lee HSJ, Friend AD, Jarvis PG. 1994. Effects of elevated CO_2, nutrition and climatic warming on bud phenology in Sitka (*Picea sitchensis*) and their impact on the risk of frost damage. *Tree Physiology* **14**: 691–706.

Myking T, Heide OM. 1995. Dormancy release and chilling requirements of buds of latitudinal ecotypes of *Betula pendula* and *B. pubescens*. *Tree Physiology* **15** 697–704.

Nelson EA, Lavender DP. 1979. The chilling requirement of western hemlock seedlings. *Forest Science* **25**: 485–490.

Nienstaedt H. 1966. Dormancy and dormancy release in white Spruce. *Forest Science* **12**: 374–384.

Perry TO. 1971. Dormancy of trees in winter. *Science* **171**: 29–36.

Prentice IC, Sykes MT, Cramer W. 1991. The possible dynamic response of northern forests to global warming. *Global Ecology and Biogeography Letters* **1**: 129–135.

Qamaruddin M, Dormling I, Ekkerg I, Eriksson G, Tillberg E. 1993. Abscissic acid content at defined levels of bud dormancy and frost tolerance in two contrasting populations of *Picea abies* grown in a phytotron. *Physiologia Plantarum* **87**: 203–210.

Reaumur. 1735. Observations du thermomètre, faites à Paris pendant l'année 1735, comparées avec celles qui ont été faites sous la ligne, à l'isle de France, à Alger et quelques unes de nos isles de l'Amérique. *Académie des Sciences de Paris* **545**.

Rehfeldt GE. 1989. Genetic variances and covariances in freezing tolerance of lodgepole pine during early winter acclimation. *Silvae Genetica* **38**: 133–137.

Ricaud S, Alaoui-Sossé B, Crabbé J, Barnola P. 1995. Dormance et croissance des bourgeons du platane hybride (*Platanus acerifolia*) en milieu urbain. *Canadian Journal of Botany* **73**: 130–140.

Robertson GW. 1968. A biometeorological time scale for a cereal crop involving day and night temperatures and photoperiod. *International Journal of Biometeorology* **12**: 191–223.

Sarvas R. 1974. Investigations on the annual cycle of development of forest trees. Autumn dormancy and winter dormancy. *Communicationes Instituti Forestalis Fenniae* **84**: 1–101.

Sorensen FC. 1973. Frequency of seedlings from natural self-fertilization in coastal Douglas-fir. *Silvae Genetica* **22**: 20–24.

Sorensen FC. 1983. Geographic variation in seedling Douglas-fir (*Pseudostuga menziessi*) from the western Siskiyou Mountains of Oregon. *Ecology* **64**: 696–702.

White MA, Thornton PE, Running SW. 1997. A continental phenology model for monitoring vegetation responses to interannual climatic variability. *Global Biogeochemistry Cycles* **11**: 217–234.