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Effects of constant and alternating temperatures on breaking dormancy of Peach (*Prunus persica* L.) seeds

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فعـول درجـات الحـرارة القـارة و المتغيـرة على إيـزال غفـوة بـذور الخـوخ (Prunus persica L)

قمنا في هذه الدراسة بتقييم مفعول درجات الحرارة خلال التطبق على إيزال غفوة بدور أنواع الخوخ المختلفة من حيث حاجاتها للبرودة. تحسن الإنبات في 20 درجة حرارية بطريقة تصاعدية مع طول مدة التطبق. كما استجابت بعض أنواع البذور، ذات الحاجة المنخفضة للبرودة، لفرج حرارية أوسع. و قارنا كذلك مفعول تناوب دراجات حرارية متوسطة و مرتفعة مع درجات حرارة منخفضة و قارة. تناوب 10 درجة حسن الإنبات في حين كان مفعول 15 درجة و ما فوق متغيرا حسب أنواع البذور. و هكذا خفضت 15 درجة إنبات البذور ذات الحاجيات المرتفعة من البرودة فقط. أما 20 و 25 درجة فقد كان مفعولها سلبيا على الإنبات عند جميع أنواع البذور. تبين من خلال تجاربنا أن درجات الحرارة المتوسطة كان لديها مفعول إيجابي على الإنبات حين تخضع لها البذور في المراحل الأخيرة من التطبق. و قد تلعب هذه الدرجات الحرارية دورا هاما في توفير حاجيات البرودة في المناطق التي تمتاز بفصل شتاء دافيء.

الكلمات المفتاحية : بذور الخوخ - إنبات - التطبق - درجة الحرارة - البرودة.

Effets des températures constantes et alternées sur la levée de dormance des graines de pêcher (*Prunus persica* L.)

Les effets de températures constantes de stratification ont été évalués sur la levée de dormance des populations de graines de pêcher ayant des besoins en froid différents. La germination a été améliorée progressivement avec la durée de stratification. Certains types à faible besoins en froid ont répondu positivement à une large gamme de températures. Par ailleurs, les effets d'alternance de températures modérées et élevées ont été comparés à ceux de températures basses continues. La température de dix degrés a amélioré la germination, alors que la réponse à des températures supérieures a varié avec le type de graines. En effet, la germination a été inhibée par 20 et 25°C pour toutes les graines, tandis que la température de 15°C a eu un effet négatif seulement sur les graines àbesoins en froid élevés. Les températures élevées avaient un effet positif sur la stratification surtout quand elles étaient appliquées aux derniers stades de la stratification. Les températures modérées pourraient jouer un rôle important dans la satisfaction des besoins en froid dans les régions caractérisées par un hiver doux.

Mots clés : Pêcher- *Prunus persica*- Graine- Besoins en froid- Température- Germination- Inhibition

Effects of constant and alternating temperatures on breaking dormancy of Peach (*Prunus persica* L.) seeds

The effects of constant stratification temperatures on breaking peach seed dormancy were evaluated using seed populations with various chilling requirements. Germination at 20°C increased steadily with stratification time. Some low chilling populations exhibited a broader temperature response curve, and a higher optimum temperature for stratification. The effects of alternating moderate and high temperatures were evaluated in comparison with continuous low temperatures. Intermittent exposure to 10°C promoted subsequent germination. Response to intermittent exposure to temperatures of 15 to 25°C varied with seed type. Germination was consistently inhibited in high chill seeds by temperatures of 15°C or higher, whereas germination of low chill seeds was either promoted or unaffected. Temperatures of 20 and 25°C consistently inhibited germination, regardless of seed source. Further experiments with high chill seeds demonstrated that exposure to high temperature was most effective in the late stages of chilling. Such temperatures may play an important role in chilling accumulation in areas with mild winters.

Key Words: Peach- *Prunus persica*- Seed- Chilling requirement- Temperature- Germination- Inhibition

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INTRODUCTION

Peach seeds, like those of other temperate fruit trees, are dormant at maturity. Removal of such dormancy is usually accomplished by a moist cold treatment. The effective temperature range for removal of dormancy in Rosaceous seeds and buds lies between 0° and 10°, with an optimum between 2° and 7°C (Stokes, 1965 ; Seeley & Damavandy, 1985). However, the optimum temperature does not appear to be constant throughout the chilling period but rather changes with time. Saure (1985) speculated that as seeds or buds accumulate chilling the optimum is shifted to a higher temperature. Any temperature within the effective range can remove dormancy if allowed to act long enough. As a consequence, chilling requirement is sometimes expressed as the number of weighted chilling hours (Erez & Lavee, 1971). Exposure to high temperature normally has a negative effect on chilling accumulation. However, moderate temperatures can enhance chilling accumulation in bud of peach (Erez *et al.* ,1979), nectarine (Gilreath & Buchanan, 1981a), low chilling blueberry cv (Gilreath & Buchanan, 1981b), and sour cherry (Felker & Robitaille, 1985).

The objectives of this study were to determine: (a) the temperature response curves of several populations of peach seeds that differ in their chilling requirements, (b) whether the effect of temperature changes as stratification proceeds, (c) whether the promotive effect of alternating chilling with moderate temperatures observed in peach buds (Erez & Lavee, 1971) also occurs with seeds, and (d) the inhibitory effect of high temperatures in relation to cycle length and stage of stratification.

MATERIALS AND METHODS

Seeds of 5 different types were used: "Siberian C" (SibC) and "Lovell" both of which are used as rootstocks, a Chilean seedling, a Brazilian seedling (Pessigo), and a selection from Florida, "FL 9-4". These seed types have 800, 1000, 450, 450, and 150 hr chilling requirements, respectively. The seeds were obtained from the following sources: "Siberian C"-Hilltop Nurseries, Hartford, Michigan; "Lovell"- a commercial orchard in California; "Chilean"- Chile; "Pessigo"- Brazil; and "FL9-4"- Dr Wayne Sherman, University of Florida, Gainesville, FL. Dry peach pits were held at 5°C until used. Seeds were removed from the pits and soaked in a fungicide solution (0.03% "Captan" = N-[(trichloromethyl) thio]-4-cyclohexene-1,2-dicarboximide) for 24 h, then placed in petri dishes containing 2 layers of filter paper moistened with Captan solution. Four dishes (10 seeds per dish) were used per treatment. After stratification, for 2 to 8 weeks (336 to 1344 h), germination capacity was evaluated by holding seeds for 10 days at 20°C in the dark. The final germination (%) represents total germination during stratification plus 10 days at 20°C.

To determine if the effect of temperature alternation depends upon the chilling accumulated or stage of stratification, the chilling period was divided into 3 equal subperiods (I, II, III). The seeds were held at continuous 5, or at 5/10, 5/13, 5/15, or 5/20°C (16 h at 5°C and 8 h at higher temperature). Temperatures were alternated either throughout the stratification period, or only during one or two of the 3 subperiods. Within experiments, all seeds were exposed to 5°C for the same total number of hours.

For alternating temperature treatments, seeds were held in styrofoam boxes placed in a 5°C room. The temperatures within the boxes were raised for specified periods of time each day using a time-clock to illuminate an incandescent light bulb. A thermostat prevented overheating and a fan mixed the air inside the box to equalize temperature. Controls were kept at constant temperatures (5 for alternating and 20°C for continuous temperature treatments). Data were analyzed factorially (temperature x time of stratification), after arcsin transformation of percentage germination for each cultivar.

RESULTS AND DISCUSSION

1. Comparison of responses of 4 seed population to a range of constant temperatures

Germination of "Sib C" seeds increased steadily with time at both 0 and 5°C, and 0 was as effective as 5°C (Table 1). Temperatures of 15 and 20 were totally ineffective in stimulating germination, whereas 10°C resulted in about 20% germination after 1344 h. In "Chilean" seeds, 0 was more effective than 5°C at 336 and 672 h. Whereas other temperatures were ineffective. However, 0, 5, and 10°C were equally effective after 1008 hr, while 15 and 20°C were ineffective regardless of the stratification time. Although holding "Pessigo" seeds for 336 h at 0, 5, or 10 increased germination in comparison with seeds held at 15 and 20°C, response was essentially saturated within 672 h.

Some increase in germination was apparent following 672 h at 15°C. Five degrees was generally more effective than 0 or 10°C. In "FL9-4" seeds held at 5°C, little response was obtained by extending chilling beyond 336 h; however, at 0°C, 672 h of chilling was much more effective than 336 h. Response to 10 paralleled that of "Pessigo" seeds to 15, whereas neither 15 nor 20°C were effective.

Table 1. Effect of stratification temperature and duration on germination (%) (during stratification plus 10 days at 20°C) of peach seeds with varying chilling requirements.

Stratification temperature (°C)	Seed source				
	Time (h)	SibC	Chilean	Pessigo	FL 9-4
	336	3f	50b	43bcde	30b
0	672	38de	95a	50bcde	88a
	1008	68bc	85a	53bcde	93a
	1344	85ab	85a	53bcde	93a
	336	0f	28c	53abcd	90a
5	672	20e	65b	65ab	93a
	1008	50cd	90a	68a	93a
	1344	90a	90a	68a	93a
	336	3f	3d	38cdef	10c
10	672	0f	8d	60abc	18bc
	1008	0f	88a	55abcd	30b
	1344	23e	88a	55abcd	30b
	336	3f	3d	13g	10c
15	672	3f	10d	28efg	5c
	1008	8f	5d	35defg	10c
	1344	5f	5d	35defg	10c
	336	5f	0d	18fg	5c
20	672	5f	5d	18fg	5c
	1008	5f	13cd	18fg	5c
	1344	5f	13cd	18fg	5c

Values are for germination after holding seeds at 20°C for 10 days. Mean separation by Duncan's Multiple Range Test, 5% level.

2. Effects of constant 5°C vs. 5°C alternating with higher temperatures in a diurnal cycle

To evaluate the effects of continuously alternating temperatures in a diurnal cycle, four peach seed populations ("SibC", "Lovell", "Chilean" and "FL9-4") were exposed to diurnal cycles (16 h at 5 and 8 h at 10, 13, 15, 20, or 25°C). In "SibC" no difference was evident in percentage germination between treatments up to 672 chilling hour (Table 2). However, the 5/10 regime significantly promoted germination, once 896 h at 5°C were accumulated, in comparison with 1008 h at continuous 5°C, but differences thereafter were non-significant. Alternating 5°C with 15, 20 or 25 significantly reduced chilling accumulation and germination percentage remained very low even after the seeds held at these temperature regimes had accumulated 1344 h at 5°C. Similar results were obtained with "Lovell" (Table 2) except that 15°C only partially negated chilling accumulation.

Table 2. Effect of stratification temperature, (5°continuous vs5° for 16 h alternated with higher temperatures for 8 h) and duration on germination (%) of peach seeds with high chilling requirements.

Stratification at 5°C (h)	Temperature (°C)				
	5	5/10	5/15	5/20	5/25
Siberian C					
448	-	12fg	10fg	23cdef	18cdefg
672	7g	15defg	10fg	8g	18cdefg
896	85a	-	16cdefg	15defg	13efg
1008	65b	-	-	-	-
1120	-	98a	20cdefg	30c	18cdefg
1344	95a	98a	27cde	28cd	30c
168097a	-	-	-	-	
2016	98a	-	-	-	-
Lovell					
448	-	0f	2f	0f	0f
672	0f	3f	3f	3f	0f
896	-	47c	10ef	7ef	3f
100813ef					
1120	-	63b	12ef	17e	10ef
134470b	70b	33d	7ef	7ef	
168092a	82a	50c	30d	20de	
201695a	-	-	-	-	
2520	92a	-	-	-	-

Values are for germination after holding seeds at 20°C for 10 days. Mean separation within seed lots among stratification treatments, DMRT 5%. LSD=10 for SibC and 13 for Lovell at the 5% level.

On the first sampling date "Chilean" seeds held continuously at 5°C germinated best. However, after 672 h at 5°C, 5/10, 5/13, and 5/15°C all promoted, while 5/20°C inhibited germination relative to constant 5°C. Both promotive and inhibitory effects disappeared when the cycle was repeated until the seeds had accumulated 1008 h at 5°C (Table 3).

Table 3. Effect of stratification temperature, (5° continuous vs 5° for 16 h alternated with a higher temperature for 8 h) and duration on germination (%) of peach seeds with low chilling requirements

Stratification at 5°C (h)	Temperature (°C) 5	5/10	5/13	5/15	5/20
Chilean					
336	35c	2d	5d	0d	10d
672	60b	87a	85a	87a	45c
1008	90a	97a	90a	97a	95a
FL 94					
168	55de	62cd	55de	40e	20f
336	77bc	87ab	87ab	82ab	52de
672	92ab	95ab	97a	97a	95ab

Values are for germination after holding seeds at 20°C for 10 days. LSD= 10 for Chilean and 15 for FL 9 4. Mean separation within seed lots among stratification treatments, DMRT 5%.

With "FL9-4" seeds moderate temperatures (10, 13, and 15°C) alternating with 5°C were no more effective than continuous 5°C regardless of the chilling period. In contrast 5/20°C caused partial negation when seeds were stratified for short periods, but not after 672 h at 5°C (Table 3).

In a second experiment with "Siberian C", all treatments were continued until 1008 h at 5°C were accumulated (Table 4). Alternating temperature of 5/10°C again promoted germination while 5/15 and 5/20 inhibited it; 5/13°C reduced germination, but not significantly.

Table 4. Effect of stratification temperature, (5° continuous vs 5° for 16 h alternated with a higher temperature for 8 h for a total time of 1008 hr at 5°) on germination (%) of Siberian C peach seeds

Temperature (°C) 5	5/10	5/13	5/15	5/20
65b	82a	55b	17c	10c

Values are for germination after hloding seeds at 20°C for 10 days. LSD=26. Mean separation among stratification treatments, DMRT 5%.

3. Effects of alternating temperatures at various times during the chilling period

• Diurnal temperature alternation during only one of 3 subperiods

Temperatures of 13°, 15°, and 20° all inhibited germination significantly when alternated with 5°C during the first period (I) of stratification. However, they had no significant effect in subsequent two stages (Table 5). A promotive effect of 10° was not evident, probably because the control germinated well after 1344 h at 5°C; promotion might have been observed with a shorter chilling period.

Table 5. Effect of stratification temperature, (5° continuous vs 5° for 16 h alternated with a higher temperature for 8 h for a total time of 1344 h at 5°) on germination (%) of Siberian C peach seeds

Temperature (°C)	Subperiod I	II	III
5/10	80ab	77ab	85a
5/13	60bc	80ab	77ab
5/15	55c	75ab	82a
5/20	35d	70abc	75ab
Control (constant 5)	85a		

Values are for germination after holding seeds at 20°C for 10 days. LSD=30. Mean separation among stratification treatments, DMRT 5%.

• Diurnal alternation during 2 of the 3 subperiods

In contrast with the previous experiment, 5/13° and 5/15° regimes did not inhibit germination significantly (Table 6). However, in the first experiment seeds were chilled for a longer time (1344 vs 1176 h). Only the 5/20°C temperature regime significantly negated germination when applied during the first and second or first and third subperiods. None of the temperatures significantly inhibited germination in the later stages.

Table 6. Effect of stratification temperature, (5° continuous vs 5° for 16 h alternated with a higher temperature for 8 h for a total time of 1176 h at 5°C) on germination (%) of Siberian C peach seeds

Temperature (°C)	Subperiod I+II	I+III	II+III
5/10	78ab	88a	83ab
5/13	68ab	73ab	83ab
5/15	65ab	65ab	85ab
5/20	25c	25c	60b
Control (continuous 5)	78ab		

Values are for germination after holding seeds at 20°C for 10 days. LSD=29. Mean separation among stratification treatments, DMRT 5%.

In our experiments, maximum germination at 20°C was reached after exposure to 0, 5, and 10°C for 1008 h for low chilling peach cv ("Chilean", "Pessigo", and "FL 9-4") and 1344 h for "Sib C". "Chilean" and "Pessigo" exhibited a broader temperature response curve than did "SibC" or "FL9-4".

This supports the view that seeds and buds of low chilling cultivars may have a wider range of effective temperatures that break dormancy. Fifteen degrees by itself had little chilling effect. The efficiency of 10°C varied with stratification time and experiment as well as seed source.

Both promotive and inhibitory effects of temperature alternation have been reported. Spiers and Draper (1974) found no inhibitory effect of 18°C (10 h) in alternation with 7°C (14 h) on chilling accumulation in rabbiteye blueberry "Tifblue", but 20°C (7 h a day, 5 days a week) partially reduced the temperature effect.

The effect of insufficient chilling was more pronounced on flower than vegetative buds. Temperatures of 7/15°C for 14/10 h, or continuous 15, were as effective as constant 7°C on low chilling cv of blueberry but were ineffective in breaking rest of a high chilling cv (Gilreath & Buchanan 1981b). However, when 30°C was applied in a diurnal cycle (10 h) with 7°C, a partial negation of bud break resulted (Gilreath & Buchanan 1981b).

Fifteen degrees was not effective in breaking rest of "Montmorency" sour cherry flower buds, but a 5/15°C (16/8 h) cycle was more effective than constant 5°C (Felker & Robitaille, 1985).

Erez *et al.* (1979) exposed potted peach trees to diurnal temperature cycles (16 h at 4°C, 8 h at higher temperatures). Twenty degrees completely negated chilling accumulation, whereas temperatures below 18°C were not inhibitory. Intermittent exposure to 10 to 15°C enhanced chilling efficiency (response per hour of exposure to 4°C) relative to constant 4°C.

In one experiment 6/17°C (16/8 h) inhibited chilling accumulation in "Redhaven" buds while 6/15°C was without effect (Erez & Couvillonn 1987). However, in another experiment when 15°C was alternated with 6°C bud break was enhanced. Eighteen degrees was neutral and 21°C completely inhibited the effect of 6°C.

Ten degrees (8 h) alternating with 5°C (16 h) during stratification promoted germination at 20° of seeds with high chilling requirements. However, a temperature of 15°C or higher inhibited it. Similar observations were reported by Aduib & Seeley (1985). They found that 5/10°C promoted while 5/15°C inhibited the germination of "Halford" peach seeds when these temperatures were applied in a diurnal cycle of 16 h at 5°C and 8 hr at 10 or 15°C. Thus although 15°C hastens peach bud break, it inhibits the breaking of dormancy in peach seeds with high chilling requirements. This suggests that seeds may have a lower temperature threshold for enhancing the chilling effect on 16/8 h cycle.

In contrast 5/15°C cycles did not inhibit subsequent germination at 20°C of peach seeds with low chilling requirements. In fact both 13° and 15° promoted germination of "Chilean" peach seeds held for 672 h at 5°C.

When the total chilling period was divided into 3 equal subperiods and cycling was applied in different stages, 4/15°C (16/8 h respectively) it enhanced peach flower bud opening only when applied in the last stages. However, cycling was ineffective in promoting leaf bud break in this experiment; in fact, it inhibited leaf bud opening when applied during the first subperiod of the chilling period (Erez & Couvillon,1987).

When we applied similar treatments to peach seeds, temperatures of 13, 15, and 20°C inhibited germination significantly when alternated with 5°C during the first subperiod of stratification, but not during either of the last 2 subperiods. Temperature regimes of 5/13 and 5/15°C did not inhibit subsequent germination when applied during any two of the subperiods, and 20°C inhibited germination only when applied during the first and second or first and third subperiods. None of the temperature regimes significantly inhibited germination when applied throughout the second and third subperiods.

It is difficult to explain the inhibitory effect, in early stages, of a temperature regime such as 5/13° or 5/15°C. The same effect was apparent in peach buds (Erez & Couvillon, 1987).

CONCLUSION

The evaluation of the effects of constant and alternating stratification temperatures on peach seed populations with varying chilling requiremetns showed that:

- The effective temperature range for breaking dormancy is wider for types with low chilling.
- Moderate temperature (13-15°C) inhibited subsequent germination when altenated with 5°C in the first subperiod of stratification but not during either of the two last subperiods;
- Ten degrees (8 hours a day) in alternation with 5°C throughout the stratification period promoted chilling accumulation, while 15°C or higher inhibited it for seeds with high chilling requirements.
- Temperatures of 13 and 15°C, in alternation with 5°C, promoted chilling accumulation in low chilling peach seeds.

Moderate temperatures could play a key role in removing dormancy in areas with mild climates. Seeds with low chilling,in addition to the low number of hours they require, exhibit a broader temperature response curve.

The fact that the inhibitory effect of moderate temperatures on chilling accumulation varied with the time of application suggests that either the chilling unit changes with time, or that there are two distinct but overlapping reactions with two different optimum temperatures.

Therefore, the inhibitory effect of a high temperature in early stages could be compensated for by its promotive effect as chilling accumulates.

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