Growth, Opium Gum Yield, and Photoperiod Response of Five Opium Poppy Accessions

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Abstract. Flowering time, growth, and opium gum yield from five seed sources (T, L, B1, B2, B3) of opium poppy (*Papaver somniferum* L.) collected from different latitudes in three Southeast Asian countries were determined. Plants were grown in six growth chambers at a 11-, 12-, 13-, 14-, 15-, or 16-hour photoperiod with a 12-hour, 25/20 °C thermoperiod. Flower initiation was observed under a dissecting microscope (40×) to determine if time to floral initiation was identical for all accessions across a wide range of photoperiods. The main capsule was lanced for opium gum at 10, 13, and 16 days after flowering (DAF). Plants were harvested at 21 DAF for plant height, leaf area, and organ dry-weight determinations. In a 16-hour photoperiod, flower initiation was observed 10 days after emergence (DAE) for B1 vs. 8 DAE for the other four accessions. Flowering time was affected most by photoperiod in B1 and least in B2. Flowering times for B3, L, and T were similar across the range of photoperiods. B2, B3, and L had the highest gum yields per capsule; even though B1 had the greatest total plant biomass, it produced the lowest gum yield. There was no difference among accessions in the average ratio of gum : individual capsule volume. For the ratio of gum : capsule dry weight, only the difference between T and B1 was significant. Capsule size did affect these ratios slightly. T had a larger gum : volume ratio for larger capsules, and B3 had a smaller gum : dry-weight ratio for heavier capsules. Flowering time varied up to 40%, capsule dry weight up to 41%, and opium gum yield up to 71% for the five accessions across all treatments. No relationship was found between flowering time and the latitude where the seed sources were collected. Time to flower initiation could not be used to predict time to anthesis because floral development rates varied significantly among accessions and photoperiods. Capsule volume and dry weight were useful in estimating gum yield.

Southeast Asian countries, especially Burma and Laos, are major producers of the opium poppy from which the narcotic drug heroin is manufactured. Production figures are used to make decisions about foreign aid and the allocation of resources to the War on Drugs. The U.S. Dept. of Agriculture (USDA) Agricultural Research Service (ARS) is developing methods for improving estimates of opium yields, an essential component for production estimates.

A strong relationship between gum yield and capsule dry weight per unit land area was established during two field studies in Thailand (USDA–ARS, Systems Research Labo-

ratory, 1992). To determine how variable this relationship might be, we examined plants from seed sources collected over a wide geographical range in Burma, Laos, and Thailand, and compared the results with data from the field studies.

Since sampling for yields is necessarily limited and yet is a highly variable component of production, a simulation model that describes the effects of major environmental variables on yield is being formulated. Photoperiod is one of the most important variables that influence growth and flower development (Acock et al., 1996a; Gentner et al., 1975; Wang et al., 1997a, 1998) and thus can have a significant effect on opium yield. Opium poppy var. album DC is a long-day plant with a critical photoperiod near 16 h. In a 16-h pho-

toperiod and 25 °C day/20 °C night temperatures, album DC flowers in 32 d. To accurately predict flowering time one must know how variable photoperiod response is among cultivars.

The main objective of this study was to investigate variability in growth, flowering time, and opium gum yield among accessions grown at different latitudes in Southeast Asia. Specific questions to be answered were: Do accessions from various geographical locations respond differently to photoperiod? Can flowering time be predicted from time to flower initiation or from other more “observable” stages? Do accessions differ in opium gum yield? Is the opium gum yield from plants grown in controlled-environment chambers similar to that from field-grown plants? Does the ratio of opium gum yield : capsule volume or capsule dry weight differ among cultivars? Does this ratio depend on the size of individual capsules?

Materials and Methods

Seed source selection. Five seed sources (B1, B2, B3, L, and T) of opium poppy were selected from Burma, Laos, and Thailand (Table 1). These five sources represent common cultivars from the main growing regions in these countries, and were grown in widely varying latitudes.

Plant culture and photoperiod treatments. Seeds were sown in 3.75-L pots filled with a Jiffy Mix growing medium (Jiffy Products, Batavia, Ill.) consisting of 1 Canadian sphagnum peat : 1 vermiculite (by volume). The growing medium was amended with a slow-release fertilizer Osmocote (14.0N–6.1P–11.6K) (Scotts-Sierra Horticultural Products Co., Marysville, Ohio) at a rate of 3 g·L⁻¹. Dolomitic lime was added to adjust the pH of the medium to 6.5. The pots were placed in six reach-in controlled-environment chambers (Environmental Growth Chambers, Chagrin Falls, Ohio), with 30 pots in each chamber (six pots per accession). The six chambers were set at a photoperiod of either 11 (0800–1900 HR), 12 (0800–2000 HR), 13 (0800–2100 HR), 14 (0800–2200 HR), 15 (0800–2300 HR), or 16 (0800–2400 HR) h. Each chamber was provided with a combination of six high-pressure sodium and six metal halide lamps that were arranged alternately in three rows. Photosynthetic photon flux density inside the growth chambers was maintained at 1000 ± 100 μmol·m⁻²·s⁻¹ at the top of the plant canopy. Temperatures during the entire experimental period were controlled at 25 ± 1 °C during the

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Table 1. Seed source, accession code, plant identification number, and main flower characteristics of five opium poppy accessions.

Accession code	ID no.	Seed source	Latitude	Flower color	Petal edge
B1	01179	Burma	20°40′	White	Smooth
B2	01286	Burma	22°53′	White	Serrated
B3	01425	Burma	24°04′	White	Smooth
L	01556	Laos	21°41′	Purple	Smooth
T ^z	00064	Thailand	18°33′	White	Serrated

^zvar. album DC.

first 12 h (0800 to 2000 HR), and at 20 ± 1 °C for the remaining 12 h (2000–0800 HR) in the 24-h period in all six chambers. Peter's water-soluble fertilizer (20.0N–8.7P–16.6K) (Scotts Co.) was applied weekly in 250 mL of water at a concentration of 100 mg·L⁻¹ of N beginning 7 d after emergence (DAE). Plants were watered as needed during the experiment.

Plants were thinned to one per pot at 14 DAE. The dates on which seedlings emerged and the first flower opened were recorded for each plant. Apical meristems were observed daily on spare plants under a dissecting microscope beginning 5 DAE. A flower was considered to be initiated when a morphologically distinct flower structure was visible under a 40x dissecting microscope. Time to other floral development stages, including flower bud appearance (i.e., flower bud first visible after the leaves surrounding it were carefully separated) and hook stage (i.e., flower peduncle has a distinct hook and flower bud points downward) were also recorded.

Plants were harvested 21 d after flower (DAF). At harvest, plant heights, leaf areas, and numbers of main stem leaves for all six plants of each accession in each chamber were recorded. Each plant was separated into leaves, stems, main capsules, and branch capsules, and each organ was oven-dried at 72 °C and weighed.

Opium gum collection. The main capsule from each plant of a total of six plants per accession in each photoperiod was lanced three times at 10, 13, and 16 DAF. The total dry weight of gum for each main capsule is the sum of these three lancements. The technique of opium gum collection used has been reported previously (Acock et al., 1996b). A three-bladed knife, with blades 2 mm apart, was used to make longitudinal incisions from the base to the top of the capsule. Each incision was 1 mm deep into the capsule wall. At each lancing, two cuts were made, resulting in a total of six longitudinal incisions. The milky latex that exuded from the incisions dried to a dark brown gum overnight and was scraped off the capsule the following day onto preweighed aluminum foil. The gum was then dried in an oven at 72 °C for 72 h and weighed.

Capsule volume determination. Main capsule volumes were calculated to determine the relationship between opium gum yield and capsule size. Capsule volume (cm³) was calculated by assuming the capsule to be an ellipsoid:

$$\text{Capsule volume} = 4/3\pi a^2b$$

where $a = 0.5 \times \text{capsule diameter (cm)}$ and $b = 0.5 \times \text{capsule height (cm)}$. Diameter and height from each capsule were measured at 4, 7, 10, 13, 16, and 19 DAF.

The standard errors of the means ($n = 6$) for DAF and growth variables were calculated and presented for each accession. A scaled Weibull distribution function,

$$y = a \cdot \exp[-(x/b)^c] + d$$

(Abernethy, 1996), was chosen to describe the relationship between photoperiod and flowering time for all five accessions. Of the four

parameters of this equation, only parameters c and d have significant meaning for our study. Parameter c reflects sensitivity of flower development to photoperiod. A small c indicates less sensitivity. Parameter d is the minimum days to flower.

Results and Discussion

Floral initiation and development. Time to flower initiation in a 16-h photoperiod differed among accessions (Table 2). Flower initials were observed at ≈ 8 DAE for B2, B3, L, and T, and at 10 DAE for B1. Time to floral initiation for T coincided with the minimum number of inductive photoperiods required for rapid flowering in transfer experiments (Wang et al., 1997b). Plants exposed to a 16-h photoperiod for ≤ 7 d before transfer to a noninductive 9-h photoperiod flowered in >120 d, whereas those exposed for ≥ 8 d flowered in <48 d (Wang et al., 1997b). Eight days appeared to be critical for floral initiation and subsequent rapid development. The plant continued to initiate leaves on the main stem if it received a 16-h photoperiod for <8 d. Main stem leaf number at flowering was 14 for plants receiving a 16-h photoperiod for 8 d, but 45 for those receiving a 16-h photoperiod for 7 d (Wang et al., unpublished).

Development rates to other more observable stages also differed significantly among accessions (Table 2). For example, a) time to flower initiation was the same for B2, L and T, but time to anthesis was significantly later in B2 than in L and T, and b) the times from flower initiation to flower bud appearance and from flower bud appearance to hook stage were longer for B1, but that from hook stage to anthesis was shorter in B1 than in L. Therefore, flowering time for different accessions

cannot be predicted from the time to flower initiation or from times of other individual development stages or vice versa.

Growth. Growth variables differed among accessions for all photoperiods (Table 3). Capsule dry weight varied by 41%, and total plant dry weight by as much as 77% for the five accessions across all photoperiod treatments. There were no significant interactions between accession and photoperiod. Data for plants grown in the 15-h photoperiod are presented and are typical of all photoperiods (Table 3). B1 plants were the largest among the five accessions, probably because they had the longest vegetative period. L and T plants flowered earlier and were smaller than plants of the other three accessions (Table 3), but had more dry matter partitioned into capsules and less into stems and leaves (data not shown).

Photoperiod response. Time to flowering varied among the accessions from the five seed sources (Fig. 1). B1 flowered last in all six photoperiods. It flowered in 37 d in a 16-h photoperiod, and at a 25/20 °C thermoperiod, 6 d later than T, but flowered in 117 d in the 12-h photoperiod, 30 d later than T. B2 flowered first in the 11- and 12-h photoperiods. It flowered in 63 d in the 12-h photoperiod, 14 d earlier than T, but flowered in 35 d in the 16-h photoperiod, 4 d later than T. Time to flowering in 13- vs. 14-h photoperiods differed significantly for all five accessions. The change in development rate appeared to be most rapid in B1, intermediate in B3, L, and T, and slowest in B2 in response to photoperiod. The possible minimum days to flowering (i.e., parameter d) were 38, 35, 34, 33, and 30 for B1, B2, B3, L, and T, respectively. These minimum values could be obtained when photoperiod was ≥ 16 h at a thermoperiod of 25/20 °C.

There was no relationship between flower-

Table 2. Time to various floral development stages in five seed sources of opium poppy grown in a 16-h photoperiod and at a 12-h thermoperiod of 25/20 °C.

Developmental stage	Accession code				
	B1	B2	B3	L	T
<i>Time from emergence to various stages (d)</i>					
Flower initiation	10.4 a ^z	8.3 b	8.5 b	8.5 b	8.5 b
Flower bud appearance	27.8 a	25.3 a	26.0 a	22.8 b	22.0 b
Hook stage	35.6 a	32.7 ab	32.5 b	29.3 c	28.5 c
Flowering	38.8 a	37.3 a	36.5 ab	34.2 bc	32.2 c
<i>Duration between two stages (d)</i>					
Flower initiation to appearance	17.4 a	17.0 a	17.5 a	14.3 b	13.5 b
Flower bud appearance to hook	7.8 a	7.3 ab	6.5 b	6.5 b	6.5 b
Hook to flowering	3.2 b	4.7 a	4.0 ab	4.8 a	3.7 b

^zMean ($n = 6$) separation within rows by LSD, $P \leq 0.05$.

Table 3. Plant height and dry weight in five seed sources of opium poppy grown in a 15-h photoperiod and at a 12-h thermoperiod of 25/20 °C. Plant growth variables were measured at 21 d after flower opening.

Accession code	Plant ht (cm)	Dry wt (g/plant)					Total plant
		Leaf	Stem	Capsule			
				Main	Branch	Total	
B1	64.7 a ^c	9.8 a	8.4 a	1.75 a	7.23 a	9.0 a	27.1 a
B2	60.2 b	7.9 b	6.7 b	1.68 a	5.19 b	6.9 b	21.5 b
B3	49.8 c	6.5 bc	4.4 c	1.66 a	5.08 b	6.7 b	17.6 c
L	42.8 d	4.1 d	3.2 d	1.97 a	4.43 b	6.4 b	13.8 d
T	49.6 c	5.2 cd	5.1 c	0.82 b	4.15 b	5.0 c	15.3 cd

^zMean ($n = 6$) separation within rows by LSD, $P \leq 0.05$.

ing time and the latitude where the seeds were obtained. For example, B3 and T were collected from the extremes of the latitude range of this study (Table 1), but their flowering times were similar in all five photoperiods.

Capsule growth and opium gum yield. The capsule grew rapidly after flowering. Main capsules from all accessions reached maximum size 13 d after first open flower (data not shown). The second lancing date, the same date when the capsule reached maximum size, generally produced the greatest gum yield among the three lancements for all five accessions (Table 4). Opium gum dry weight per main capsule varied up to 71% among the five accessions. B2, B3, and L had the highest total gum yields, while B1 had the lowest. The gum dry weight : capsule dry-weight ratio [i.e., gum yield on a capsule dry-weight ($\text{mg}\cdot\text{g}^{-1}$) basis], was significantly different among accessions only when T was compared with B1.

Our growth chamber results on opium gum yield were similar to those obtained under field conditions in Thailand (USDA-ARS, Systems Research Laboratory, 1992). The comparable average opium gum yield was $38 \text{ mg}\cdot\text{g}^{-1}$ for three lancements in the Thailand study and $41 \text{ mg}\cdot\text{g}^{-1}$ averaged over the five accessions in this study.

Our results did not show a difference among accessions in gum yield on a capsule volume basis (average of $4.0 \text{ mg}\cdot\text{cm}^{-3}$ for all five accessions). This ratio is also similar to that reported for field-grown poppy in Thailand. Further studies are needed to determine whether this ratio is stable for all accessions and under all environmental conditions. At present, the average ratio of gum dry weight : capsule volume can be used to estimate opium gum yield in Southeast Asian poppy.

Effect of capsule size on the gum to volume and dry-weight ratios. No trend was observed in the ratio of gum : volume as a function of individual capsule volume for B1, B2, B3, or L. There was a slight ($r^2 = 0.12$) but significant tendency for this ratio to increase with larger capsules of T. Similarly, there was no trend observed in the ratio of gum : dry weight as a function of individual capsule dry weight for B1, B2, L, or T but a slight ($r^2 = 0.16$) but significant trend for the ratio of gum : capsule dry weight to decrease with heavier capsules of B3. Since the average opium gum yield for the average capsule weight and volume was similar for all accessions tested (Table 4), these tendencies do not appear to seriously affect the use of capsule dry weight and volume to estimate gum yields in Southeast Asia.

Conclusions

Plants from the five different seed sources collected from Burma, Laos, and Thailand differed in flowering response to photoperiod. Among the five accessions tested, B1 was a late-flowering accession and showed the most change in flowering time in response to photoperiod. B2 was an early-flowering accession least affected by photoperiod in terms of days to anthesis. Flowering time of all accessions

Fig. 1. Time from emergence to flower (d) as a function of photoperiod (h) at a 12-h thermoperiod of 25/20 °C in five seed sources of opium poppy. Vertical bars represent the standard error ($n = 6$) and are shown only where they are larger than the symbol size.

Table 4. Main capsule opium gum dry weight and the ratios of opium gum dry weight to capsule dry weight and to capsule volume in five seed sources of opium poppy. Capsules were lanced three times at 10 (first), 13 (second), and 16 (third) d after flowering.

Accession code	Gum dry wt ($\text{mg}/\text{capsule}$)				Ratio of gum : capsule dry wt ($\text{mg}\cdot\text{g}^{-1}$)	Ratio of gum dry wt : capsule vol ($\text{mg}\cdot\text{cm}^{-3}$)
	First lancing	Second lancing	Third lancing	Total		
B1	12.8 c ²	15.1 b	10.4 c	37.0 c	34.1 b	3.46 a
B2	14.4 bc	22.3 a	22.4 a	59.1 ab	38.8 ab	3.47 a
B3	23.5 a	23.1 a	19.5 ab	63.1 a	41.4 ab	3.57 a
L	20.4 ab	20.5 ab	17.6 ab	57.7 ab	39.8 ab	4.69 a
T	15.4 bc	20.4 ab	15.1 bc	49.7 bc	50.9 a	4.84 a

²Mean ($n = 6$) separation within rows by LSD, $P \leq 0.05$.

was less in a 14-h than in a 13-h photoperiod. No relationship between flowering time and latitude where seeds were collected was found.

Our results indicate that time to flower initiation cannot be used for predicting time to anthesis or vice versa, because floral development rate varied significantly among accessions for a given photoperiod. Plant growth and opium gum yield differed among accessions. Capsule dry weight varied up to 41%,

and opium gum dry weight as much as 71%, for five accessions across the six photoperiod treatments. B1 had a longer vegetative stage, but did not produce larger main capsules or higher gum yield.

Opium gum yield (on a capsule dry-weight or volume basis) in growth chambers in this study was similar to that reported for field-grown plants in Thailand. There was no difference among accessions in the ratio of gum dry

weight : individual capsule volume. The ratio of gum : capsule dry weight was highest in T. Measurements of capsule volume and dry weight to estimate the opium gum yields in Southeast Asia appear adequate.

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