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EFFECTS OF HYBRID POPLAR (SALICACEAE) CLONE AND
PHENOLOGY ON GYPSY MOTH (LEPIDOPTERA: LYMANTRIIDAE)
PERFORMANCE IN WISCONSIN

James J. Kruse and Kenneth F. Raffa¹

ABSTRACT

Gypsy moth (*Lymantria dispar*) developmental interactions with two hybrid *Populus* species clones were studied in laboratory trials. Significant differences in larval performance were found between clones and within the same clone at different phenological states. No larvae were able to complete development on clone NM6. All gypsy moth larvae feeding on clone NC5271 survived when leaf flush was synchronized with gypsy moth eclosion in early May. However, neonates feeding on NC5271 foliage in July experienced increased mortality. Weights of surviving gypsy moth larvae feeding on NC5271 foliage in May versus July were not significantly different.

Hybrid poplars, *Populus* species, are currently being developed as rapidly grown short rotation trees for alternative sources of fiber and biomass (Hansen et al. 1983, Dickmann & Stuart 1983, Abrahamson et al. 1990). In previous evaluations in Wisconsin, 15 hybrid poplar clones were characterized for their primary growth rates, general mensurational properties, foliar chemistry, and susceptibility to several important endemic insect, fungal, and vertebrate pest species (Robison & Raffa 1994, 1996, 1997). This report considers the susceptibility of selected clones from this group on an impending invasive species that is currently becoming established in Wisconsin, and therefore must be considered in hybrid poplar plantings.

The gypsy moth, *Lymantria dispar* L., feeds on a wide range of host species throughout its native Eurasian and introduced North American distributions (Montgomery 1991). Although highly polyphagous, the gypsy moth prefers and performs particularly well on certain tree species, particularly oaks (*Quercus* spp.) and aspens (*Populus* spp.) (Barbosa & Capinera 1977, Hough & Pimentel 1978, Barbosa & Greenblatt 1979, Barbosa et al. 1983, Raupp et al. 1988, Gross et al. 1990, Sheppard & Friedman 1990, Hamilton & Lechowicz 1991, Stoyenoff et al. 1994 a, b). A variety of work has been done evaluating the suitability and acceptability of *Populus* to gypsy moth (Lindroth & Hemming 1990, Chilcote et al. 1992, Roth et al. 1994, Stoyenoff et al. 1994a, b, and c, Montgomery 1986, Roden & Surgeoner 1991).

The clonal nature of *Populus* species may have a major impact on gypsy moth growth and performance (Chilcote et al. 1992). Likewise, the performance of forest tent caterpillar (*Malacosoma disstria* Hbn.) varied widely

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among hybrid poplar (Robison & Raffa 1990, 1994, 1996, 1997) and native aspen (Lindroth & Bloomer 1991) clones. Hybrid poplar clonal variation can potentially be exploited to reduce pest impacts by using resistant native clones, selecting for enhanced resistance, and using deployment strategies that adversely affect pests (Dickmann and Stuart 1983). The purpose of this research was to test the growth and performance of gypsy moth larvae on two hybrid poplar clones that showed differential resistance to *M. disstria*. Phenological changes can partially explain differences in larval performance within and between tree species, and between years (Montgomery 1986, Meyer et al. 1987, Raupp et al. 1988, Gross et al. 1990, Sheppard & Friedman 1990, Chilcote et al. 1992, Hunter & Leckowicz 1992, Robison & Raffa 1997). Therefore, we conducted bioassays at two widely separated time periods to fully characterize clonal suitability.

MATERIALS AND METHODS

Poplar clones. Two hybrid poplar clones were chosen because of their differential suitability to an oligophagous Lepidopteran, the forest tent caterpillar. NM6 (*Populus nigra* X *P. maximowiczii*) is relatively resistant to *M. disstria* herbivory, while NC5271 (*P. nigra* 'Charkowiensis' X *P. nigra* 'Caudina') is highly susceptible (Robison & Raffa 1994).

Poplar clones were established in 1994 from frozen, 12 cm long, dormant hardwood cuttings taken from healthy trees growing outside at the University of Wisconsin-Madison West Madison Agricultural Research Station. Cuttings were planted in saturated Redi-Earth Peat-Lite® potting soil in 20 cm diameter plastic pots in the glasshouse. Trees were fertilized with 15 g per plant Osmocote® slow-release 17-6-12 plus micronutrients and flood irrigated regularly. Cuttings had taken root by fall, 1994, and overwintered outside. In April 1995, trees were transplanted in vermiculite/peat moss/Fafard® soil mixture (Carlin Sales, Milwaukee, WI). Glasshouse conditions were 16:8 L:D (moderately shaded with standard fluorescent lamp supplements), and fluctuated seasonally between 18–35 °C, and 25–100% RH.

Gypsy moths. Gypsy moth embryonated egg masses were obtained from culture NJ41 USDA-APHIS, Otis AFB, MA. Gypsy moths were reared in the University of Wisconsin-Madison Russell Laboratories quarantine facility. Upon removal from cold storage, gypsy moth egg masses were sterilized using a sodium hyperchlorite solution (2060 ml ddH₂O, 21 ml polyoxy-ethylene sorbitan monooleate, and 40 ml bleach), triple rinsed with ddH₂O and allowed to dry. Egg masses were placed in rearing containers and kept in a growth chamber at 16:8 L:D, 23–25°C and 100% RH. Upon eclosion, gypsy moth larvae were evenly distributed among mass rearing containers containing NC5271 or NM6 leaves, or synthetic diet (ICN Biomedicals, Aurora, OH). Each treatment received several hundred neonate larvae.

Effects of Clone. Leaf flush of glasshouse trees was timed to coincide with foliar development of *Populus* in the field. Gypsy moth egg masses were sterilized and incubated to synchronize eclosion and feeding at approximately seven days mean budbreak. Larvae were fed excised foliage daily or as required. Leaves were excised from trees in a destructive fashion so that no tree was used more than once for a feeding. Leaves were brought to the quarantine facility, disinfected with 10% Clorox® solution and rinsed with distilled water, before they were provided to larvae.

Larvae were allowed to feed for eight days. Sixty-five larvae feeding on NC5271 were randomly selected and weighed. Only 52 larvae feeding on NM6 remained alive at this point, so all living larvae on this clone were

weighed. After weighing, larvae were placed singly in 25 ml plastic rearing cups in a growth chamber at 16:8 L:D, 23–25°C, and 45–65% RH. Larvae were reared until pupation or death. Data included larval weight, survival, and development time. Due to high gypsy moth mortality on NM6, two successive instars were tested. Forty-five second and third instars were transferred from artificial diet to NM6 immediately after molting. Dead larvae were counted and removed until no larvae remained. Data were analyzed via one-way ANOVA (Abacus Concepts 1989), using weight measurements as a response and poplar clone as a treatment.

Effects of Phenology. Trees growing outside at the UW West Madison Agriculture Research Station were utilized in early July, during the time poplar in the field was actively growing. Gypsy moth egg masses were treated in the same manner as before. Leaves were excised from trees in a random fashion, and so that no tree was used more than once for a feeding. After eight days, no larvae feeding on NM6 remained alive, so only NC5271 data were collected and subsequently compared to the early season NC5271 data. All larvae alive on day eight were reared until death or to three weeks of age. Data were collected and analyzed as before.

RESULTS

Effects of Poplar Clone. Poplar clone NC5271 was a highly suitable host for gypsy moth. All gypsy moth larvae feeding on NC5271 in May survived through the third stadium (Fig. 1). At the time of weighing, most larvae feeding on NC5271 were mid to late-second instars. Of 20 larvae randomly selected for further rearing, 19 survived to pupation. Average pupal weight and development time for female gypsy moths were 1022 mg and 37.2 days, respectively. Pupal weights of gypsy moths that had been fed poplar clone NC5271 were greater than those of gypsy moths that had developed upon Red Oak (*Quercus rubra*) and Tamarack (*Larix laricina*) under identical conditions (Kruse, unpublished data).

Poplar clone NM6 was not a suitable host plant for gypsy moth development or survival (Fig. 1). Out of several hundred neonates assigned to the NM6 treatment in May, only 52 survived to the eighth day for weighing. Weights of larvae feeding on NM6 were significantly lower from the weights of larvae feeding on NC5271 (Table 1). By the tenth day after eclosion, only 13% of the 52 weighed gypsy moth larvae feeding on NM6 were still alive. All larvae feeding on NM6 were dead by day 14. All additional 45 gypsy moth second and third instars transferred to NM6 foliage from ICN diet were dead by the eighth day after transfer.

Effects of Phenology. NC5271 foliage in July was a poor host for neonate gypsy moth larvae. Eighty-nine percent of the gypsy moth larvae feeding on NC5271 foliage in July survived two weeks after eclosion. Between 14 and 18 days after eclosion, however, there was high mortality, with only 31% surviving through this interval. By the 21st day after eclosion, mortality leveled off, but only 25% of the original 65 larvae survived. Among survivors, however, larval weights eight days after eclosion between the July and May feeding dates were not significantly different (Table 1).

NM6 foliage in July was not a suitable host plant for gypsy moth development or survival. All larvae placed on NM6 foliage in July were dead by the fourth day of the assay (Fig. 1).

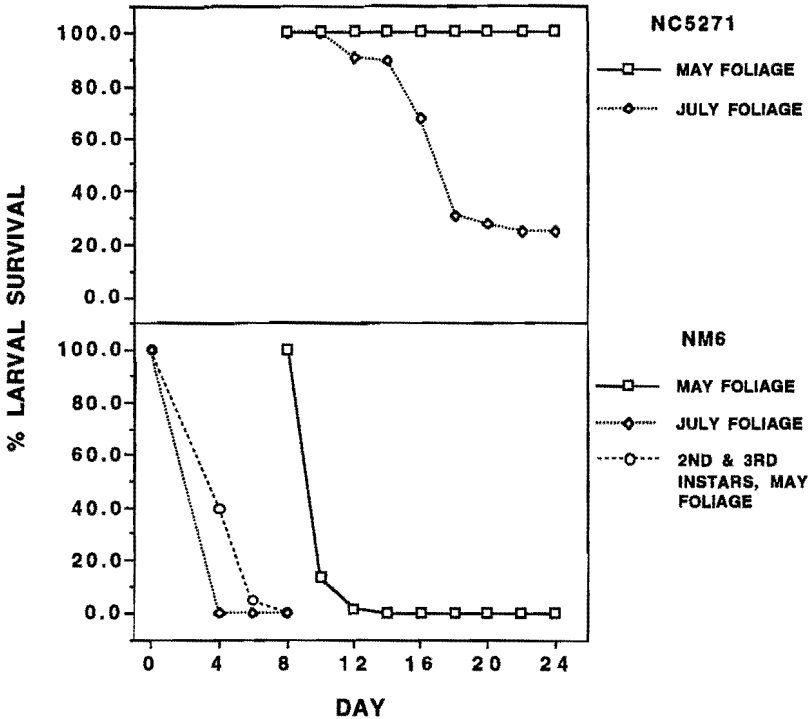


Figure 1. Effect of hybrid poplar clone and phenology on gypsy moth survival. Experiment began eight days after eclosion or the day of eclosion (day 0). Above: Neonates on NC5271 foliage in May (N=65), and in July (N=65); Below: Neonates on NM6 foliage in May (N=52), and in July (N=100), and second and third instars in May (N=45).

Table 1. Poplar clone and phenological effect on gypsy moth survival and weight. Means within a column followed by different letters are significantly different at alpha = 0.001, using Fishers Protected LSD technique.

Clone	Month	Days after Eclosion	Survival(%)	Mean Wt (mg)	SE	N
NC5271	May	8	100%	150 a	1.0	65
	July	8	24.62%	130 a	0.4	65
NM6	May	8	0	2 b	1.0	52
	July	0	0	—	—	100
	May	2-3 instars	0	—	—	45

DISCUSSION

Gypsy moth neonates placed on newly flushed NM6 foliage were unable to survive longer than two weeks. This is equivalent to the period Barbosa et al. (1983) found gypsy moth larvae survived on highly unsuitable host species. All larvae appeared to simply starve to death by day four on NM6 foliage in July.

NC5271 foliage is a highly suitable host for gypsy moth neonate larvae shortly after budbreak in May. However, neonate performance on NC5271 foliage in July demonstrated a phenological asynchrony. Over 75% of gypsy moth neonates feeding in July died on a food plant that was highly suitable at the time of spring leaf flush. Such phenological changes can occur over shorter periods of time than this study, as previously demonstrated by Raupp et al. (1988) on other tree species.

In previous studies, NM6 reduced both development and feeding by the forest tent caterpillar (Robison & Raffa 1994, 1996, 1997). In this study, behavior of gypsy moth in response to the poor quality NM6 host appeared to be related to dispersal. Large amounts of silk were observed in the NM6 containers, compared to the NC5271 containers, which may indicate that the former group of neonates were attempting to disperse. This finding is consistent with the view that gypsy moth initiate dispersal when confronted with unsuitable food (Leonard 1971, Capinera & Barbosa 1976, Lance and Barbosa 1981).

Larvae feeding on NC5271 foliage in July demonstrated high mortality, but the survivors were capable of attaining the size of the phenologically synchronized May predecessors. Likewise, Chilcote et al. (1992) found that gypsy moth can recover from initial losses in performance suffered on unsuitable foliage during early larval development.

The mechanism of resistance to gypsy moth in clone NM6 likely relates to phenolics, primarily phenolic glycosides. Resistance to Lepidoptera among native aspen, *Populus tremuloides*, is largely associated with high concentrations of compounds such as tremulacin, which cause reduced feeding and development in gypsy moth (Lindroth et al. 1987, Lindroth & Hemming 1990). Likewise, hybrid poplar clone NM6 foliage is nearly twice as high as in total phenolics than is clone NC5271 (Ramachandran et al. 1993), and application of foliar extracts from NM6 containing phenolic glycosides can reduce forest tent caterpillar feeding when applied to otherwise susceptible clones such as NC5271 (Robison & Raffa 1997).

These results suggest that NM6 is a good candidate for deployment in areas with threatening or repeated gypsy moth defoliation. Further studies are needed to further characterize the effects of NM6 on biological control agents, and its responses to multiple biotic and abiotic factors under field conditions.

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