SUSCEPTIBILITY OF WILLOWS TO OVIPOSITION BY THE WILLOW SAWFLY, *NEMATUS OLIGOSPILUS*

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ABSTRACT

Different combinations of shoots of 30 *Salix* species, clones or identified hybrids, were exposed to *Nematus oligospilus* to determine which were preferred for oviposition. Single *N. oligospilus*, enclosed in gauze cages, chose from 9 - 12 terminal shoots in which to oviposit. There was wide variation in fecundity (49.3 ± 4.3 eggs/female; range 0-100). Sawflies oviposited in all tree-willows (subgenus *Salix*) screened - a selection dominated by clones of *S. matsudana* x *S. alba* hybrids, but wide variation in the number of eggs laid/shoot prevented a significant preference ranking. Shrub willows (subgenus *Caprisalix*) were, with one exception, not selected for oviposition.

Keywords: sawfly, willow, host preference, oviposition.

INTRODUCTION

Nematus oligospilus Förster (Hymenoptera: Tenthredinidae), a sawfly that feeds exclusively on willows (*Salix* spp.), was discovered in Auckland on 19 February 1997 (Berry 1997). Adult *N. oligospilus* disperse rapidly by flight, and by the autumn of 1997 the species was widespread throughout the Auckland isthmus (Baker *et al.* 1997), and had been reported in Rotorua (A. Wilkinson pers. comm.), and possibly Tauranga (C. Winks pers. comm.). In Auckland, the first record of adult *N. oligospilus* emerging from diapause was on 20 October 1997, and by April 1998 larvae were widespread in the Bay of Plenty (C. Stace pers. comm.) and Poverty Bay, where large numbers were found to be damaging several different willow types (B. Dobbie pers. comm.). *N. oligospilus* produces adults of both sexes in the Northern Hemisphere, but is apparently thelytokous where it is found in the Southern Hemisphere (Urban and Eardley 1995), including New Zealand.

Pontania proxima (Lepeletier) (Hym. Tenthredinidae), the only other sawfly found on willows in New Zealand, produces leaf-galls and is generally an insignificant pest. In contrast, *N. oligospilus* larvae are external leaf-feeders, with a demonstrated ability in other countries to defoliate and even kill willow trees. This new pest thus poses a particular threat to soil stabilisation and river bank erosion control programmes, which rely on relatively few species and cultivars of *Salix*. Most of these have been bred in New Zealand over the past 30 years. The genus *Nematus* contains both monophagous and polyphagous species (e.g. Roininen and Tahvanainen 1989) and so we initiated a research programme to identify any *Salix* phenotypes or genotypes in the willow collection at the HortResearch Aokautere Research Centre Nursery, Palmerston North, that were resistant or highly susceptible to *N. oligospilus* damage. Our intention was to identify any commercially planted species, clones or hybrids that might be especially at risk. Such data would help plant breeders to select suitable *Salix* genotypes for the future, and regional councils to manage existing soil erosion and river bank stabilisation programmes. Here we report on initial experiments carried out in 1997-1998 designed primarily to rapidly screen a large number of willows for oviposition acceptability.

MATERIALS AND METHODS

Experiment 1

Dormant cuttings of twenty eight willow species/clones or hybrids from Aokautere were rooted in pots at the Mount Albert Research Centre in early spring 1997, and shoots from primary buds used for oviposition experiments. Terminal shoots, all *ca.* 30cm long, were cut from selected willow hosts, and placed singly in 100mm x 25mm glass tubes filled with water, through a parafilm seal. Excess foliage was removed so that each shoot had 6 fully expanded leaves below the developing terminal leaves/bud. An array of tubes with shoots was supported in a square piece of 2.5cm thick polystyrene, in holes cut at 10cm centres.

The array of willow hosts, was placed within a 90cm x 90cm x 60cm nylon gauze covered cage. A single adult *N. oligospilus*, less than 24h old, was added through a velcro-lined door, and held at laboratory ambient temperature (*ca.* 21°C) beneath supplementary neon strip lighting on a 16h photoperiod. Cages were examined daily until the female died, when the willow shoots were examined individually and the eggs laid on each shoot were located and counted. The leaf aspect (upper or lower surface), leaf position (from oldest to youngest on the shoot), and shoot position within the array for each of the eggs laid were recorded.

Experimental larvae were reared on *S. fragilis*, which was neither one of the experimental willow species nor a parent of any of the hybrids screened. The first experiments used F₁ adults from the diapausing generation. Initially, the host range of *N. oligospilus* on *Salix* was unknown, and neither the cage size nor the daily supply of females was sufficient to test the 28 willow-types simultaneously. Consequently, the willows were divided into 3 groups, each of 9 or 12 broadly similar genotypes (when possible), and each group was screened for differences in acceptance for oviposition. Each female was given a choice of either, arranged in a 3 x 3 or 4 x 3 array within the cage. Each array was replicated 5 times, with fresh females and shoots on each occasion.

Experiment 2

To examine whether the most favourable or unfavourable hosts in an array had influenced oviposition on the remaining hosts, and in an attempt to 'benchmark' field records, we carried out a second series of experiments. Using the same procedures as above, we offered *N. oligospilus* females (a) 3x3 arrays of the 3 most 'susceptible' and (b) the 3 most 'resistant' willows from the first series, each array with 3 shoots of each of the 3 willows. There were insufficient shoots to re-test *S. pentandra* and *S. alba vitellina* was substituted in the array; (c) a mixture of the 2 most 'resistant' and 2 most 'susceptible' *S. matsudana* x *S. alba* hybrids from the first series, in a 4x3 array, again with 3 shoots of each willow. Two other willow species, crack willow (*S. fragilis*) and tortured willow (*S. matsudana* 'Tortuosa'), have been observed to be favourable hosts in Auckland, and so (d) nine *S. fragilis* and (e) nine *S. matsudana* 'Tortuosa', each in a 3x3 array were also offered to single sawflies. Data were analysed by comparison of means, with *t*-tests of significance.

RESULTS AND DISCUSSION

Lifespan

Females lived for 4.9±0.4 days (range 2-12) in the cages. The mean of *ca.* 5 days did not include the 0-24h pre-test adult period, nor did it take account of any response to unfavourable host plants, but is consistent with South African data (Urban and Eardley 1995).

Fecundity

Realised fecundity of those females which laid eggs (n=15 in first series, and n=11 in second series) was 49.3 (±4.3) eggs/female. The maximum realised fecundity was 100 eggs from one female, and the maximum oviposition rate was 21 eggs/day. Leaf position was a significant factor in oviposition, with more eggs laid on the oldest (or lowest) leaves and fewer on the youngest (or highest) leaves of the shoots (P<0.001). It was not determined whether this difference was due to physical

or chemical factors. Eggs were laid on both the upper and lower leaf surfaces, but three times as many eggs were laid on the upper surface ($P < 0.001$). These data are similar to those recorded in South African laboratory experiments (Urban and Eardley 1995).

TABLE 1: Mean number of eggs laid/plant on *Salix* shoots. Data from experiments 1 and 2 (e2) combined.

Register no. ¹	Species	Clone name	Willow type ²	Mean no. eggs laid/plant ($\pm$ se)
PN220 (e2)	<i>S. viminalis</i>	Gigantea	O	0.00
PN229 (e2)	<i>S. aegyptiaca</i>	Aegyptiaca	O	0.00
PN386 (e2)	<i>S. schwerini</i>	Kinuyanagi	O	0.00
PN215	<i>S. x reichardtii</i>	Pussy willow	S	0.4 $\pm$ 0.2
NZ1184 (e2)	<i>S. matsudana x alba</i>	Moutere	T	0.6 $\pm$ 0.4
NZ1001 (e2)	<i>S. matsudana x alba</i>	Cannock	T	4.4 $\pm$ 1.7
PN249	<i>S. purpurea</i>	Booth	O	0.7 $\pm$ 0.2
PN608	<i>S. purpurea</i>	Irette	O	5.1 $\pm$ 3.1
PN227	<i>S. matsudana</i>	Kew	T	1.9 $\pm$ 0.8
NZ1179	<i>S. matsudana x alba</i>	Makara	T	2.2 $\pm$ 1.0
PN694	<i>S. matsudana</i>	Tsinan	T	2.4 $\pm$ 0.7
PN695	<i>S. matsudana</i>	Shanghai	T	2.6 $\pm$ 1.5
NZ1002	<i>S. matsudana x alba</i>	Aokautere	T	3.6 $\pm$ 1.6
92-12-2	<i>S. matsudana x alba</i>	-	T	3.8 $\pm$ 1.6
NZ1149	<i>S. matsudana x alba</i>	Wairakei	T	4.0 $\pm$ 2.4
NZ1040 (e2)	<i>S. matsudana x alba</i>	Tangoio	T	4.6 $\pm$ 1.3
NZ1087	<i>S. purpurea</i>	Tortuosa	T	5.2 $\pm$ 3.8
PN693	<i>S. matsudana</i>	Pohangina	O	5.8 $\pm$ 2.1
NZ1317	<i>S. matsudana x alba</i>	Hathaway	T	5.9 $\pm$ 1.0
92-5-1	<i>S. matsudana x pentandra</i>	-	T	6.0 $\pm$ 1.9
PN218 (e2)	<i>S. fragilis</i>	-	T	6.5 $\pm$ 1.8
92-21-18	<i>S. matsudana x pentandra</i>	-	T	6.6 $\pm$ 1.2
NZ1003 (e2)	<i>S. matsudana x alba</i>	Te Awa	T	6.7 $\pm$ 1.0
NZ1130 (e2)	<i>S. matsudana x alba</i>	Hiwinui	T	7.5 $\pm$ 1.5
92-17-1 (e2)	<i>S. matsudana x alba vitellina</i>	-	T	7.8 $\pm$ 5.2
PN353	<i>S. alba vitellina</i>	Westhaven	T	2.6 $\pm$ 0.8
PN357 (e2)	<i>S. alba</i>	I 2-59	T	7.8 $\pm$ 3.4
PN731 (e2)	<i>S. nigra</i>	Black willow	T	0.7 $\pm$ 0.4
PN253	<i>S. pentandra</i>	Bay willow	T	9.0 $\pm$ 0.6
				4.5 $\pm$ 1.4
				9.0 $\pm$ 3.4
				10.8 $\pm$ 2.1
				6.9 $\pm$ 2.8
				14.0 $\pm$ 5.5
				10.5 $\pm$ 3.4
				14.7 $\pm$ 5.4

¹ Register number of the genotype at the Aokautere Plant Nursery, Palmerston North.

² O and S = *Salix* (*Caprisalix*); osiers and salallows, or shrub willows. T = *Salix* (*Salix*); tree willows.

Host preference

Oviposition selection was very variable between replicates, probably due to a combination of individual sawfly preference behaviour and variable shoot attractiveness. It proved difficult to maintain a continuous supply of equally healthy shoots from 30 different willows, and so, in some experiments, one or two shoots died or lost condition. These shoots were then either unavailable for oviposition or perhaps were relatively less attractive.

In Experiment 1, *N. oligospilus* showed no consistent preference for any host. Individual females often laid most of their eggs in one or two plants within an array, but a willow-type which received many eggs in one replicate frequently received no eggs in another. Because of the variation between replicates of an array, the oviposition data from the 3 different groups were amalgamated (Table 1). *S. viminalis* 'Gigantea', *S. aegyptiaca* and *S. schwerini* 'Kinuyanagi' were the only plants in which no sawfly laid eggs. Two sawflies laid one egg each in *S. x reichardtii* 'Pussy Galore'. These willows, together with *S. purpurea*, are all osier or sallow willows, in the subgenus *Salix* (*Caprisalix*), and were consistently (with the exception of *S. purpurea*) rejected for oviposition. *N. oligospilus* oviposited in shoots of all three clones of *S. purpurea*, with no significant difference in numbers laid per experiment ($P > 0.1$). All of the remaining hosts were tree willows in the subgenus *Salix* (*Salix*), and were acceptable for oviposition. Most eggs were laid in three species (*S. alba*, *S. nigra* and *S. pentandra*) although the numbers per plant were not directly comparable with others, and may have been lower if they had not been within the array which also included *S. viminalis* 'Gigantea', *S. aegyptiaca* and *S. schwerini* 'Kinuyanagi'. Oviposition on the large group of clonal selections from *S. matsudana* and the various *S. matsudana* x *S. alba* hybrids ranged from 0.6 - 7.8 eggs/female (Table 1).

In Experiment 2, *S. viminalis* 'Gigantea', *S. aegyptiaca* and *S. schwerini* 'Kinuyanagi' were again generally not selected for oviposition. Females usually died without ovipositing in these species, although a total of 8 eggs were laid in two *S. aegyptiaca*. Variation among eggs laid per host per replicate was even greater than in the first series, although *S. nigra* remained the most attractive host. Preference for the four *S. matsudana* x *S. alba* hybrids changed from the first series, and NZ1001 and NZ1184 were favoured more than NZ 1003 and NZ1130. The apparent inconsistencies between the data in the two series were adequately explained by a lack of control of the experimental variables, and showed that a more detailed statistical analysis of oviposition preference, especially among the tree willows, was not valid. Both *S. fragilis* and *S. matsudana* 'Tortuosa' were attractive hosts for ovipositing sawflies.

CONCLUSION

These experiments were not designed to provide a statistically valid ranking of resistance to willow sawfly oviposition among *Salix* genotypes. However, they do show that *N. oligospilus* exhibited clear aversion to ovipositing in osiers/sallows and shrub willows in *S. (Caprisalix)* (except *S. purpurea*), but general acceptance of the tree willows, *S. (Salix)*, including all of the hybrids and clones (the NZ/PN series) bred at Aokautere and selected for use in New Zealand. Sawflies oviposited freely in the experimental 'Tortured willow' (*S. matsudana* 'tortuosa') and 'Crack willow' (*S. fragilis*), as also observed in Auckland suburbs, and provided some confidence that the experimental procedures were appropriate. Thus most of the tree willows planted in hillside and river bank stabilisation programmes over the past 30 or so years may, to some degree, be vulnerable to *N. oligospilus* attack. *S. fragilis*, which are unwanted trees in some wetland conservation sites, are also vulnerable.

As larvae are responsible for defoliation, oviposition is not necessarily a good predictor of larval feeding preferences. However, we chose to examine oviposition first because we believe that the evolution of preferential selection of genotypes for oviposition is likely to precede larval host preference selection (e.g. Roininen and Tahvanainen 1989). We plan to evaluate larval feeding and development in a range of *Salix* hosts, and then to investigate oviposition and larval development together, using a smaller and more select group of *Salix* genotypes.

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