Trichomes of Nama (Hydrophyllaceae) That Produce Insect-active Compunds

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ABSTRACT

Nama hispidum, N. lobbii, N. rothrockii, and *N. xylopodum* have two basic types of trichomes on the adaxial and abaxial surfaces: glandular and nonglandular. *Nama hispidum* and *N. xylopodum* have (1) short semierect or intermediate-length acicular trichomes that often recurve toward the leaf surface and (2) short-stalked capitate glands. The larger acicular trichomes have micropapillae. *Nama lobbii* has long filiform trichomes and sessile capitate glands. *Nama rothrockii* has erect, smooth subulate trichomes and long-stalked capitate glands. Morphological diversity of trichomes in *Nama* and their possible functional significance as a predator defense are discussed.

Key words: Hydrophyllaceae, *Nama hispidum, Nama lobbii, Nama rothrockii, Nama xylopodum,* trichomes, insect growth regulators, juvenile hormones, antihormones.

INTRODUCTION

Leaf surface structures such as hairs and nettles act as physical barriers to reduce damage caused by the attack of insects. The density of pubescence (vesture) is correlated with a lower incidence of insect predation in soybean, French bean, cotton and other crops (Stipanovic 1983). These structures may act as spears to impale or elevate eggs and nymphs from the leaf surface, thus facilitating desiccation and increasing exposure to parasites, predators, and pathogens (Juniper and Southwood 1986). Glandular trichomes with enlarged terminal heads may release protective compounds upon contact with herbivores and other animals (Kelsey et al. 1984). Plants thus protected by trichomes and chemicals effectively deter insect herbivores from attacking the leaf and stem surfaces (Lin et al. 1987; Duffey 1986; Khan et al. 1986; Pillemer and Tingey 1976).

Some plants in the genus *Nama* contain insect antijuvenile hormones and/or juvenile hormone mimics. Binder et al. (1991) report antijuvenile hormones from *N. hispidum* Gray and *N. lobbii* Gray, antijuvenile hormones and juvenile hormone activity from *N. sandwicense* Gray, juvenile hormone activity from *N. rothrockii* Gray, and neither antijuvenile hormone nor juvenile hormone activity from *N. demissum* Gray, *N. densum* Lemmon, *N. jamaicense* L., *N. stevensii* Hitchcock, and *N. xylopodum* (Woot. & Standl.) C.L. Hitchcock. Insect antijuvenile hormones occur in *Nama hispidum, N. lobbii,* and *N. sandwicense* and have been detected in the glandular trichomes of *N. hispidum* var. *revolutum* Jepson (Binder et al. 1991). These antijuvenile hormones, precocene II and precocene I (6,7-methoxy-2,2-dimethylchromene, 7-methoxy-2,2-dimethylchromene) affect the growth, development, and physiology of insects (Bowers 1985) and, therefore, may provide substantial protection against herbivorous insects. Presence of antijuvenile hormones in glandular trichomes of *N. hispidum* (Binder et al. 1991) indicates that a phytochemical defense in some species of *Nama* may occur in the leaf surface structures. This study examines the trichome types of four species of *Nama*: *N. hispidum* and *N. lobbii*, which contain insect antijuvenile hormones, *N. rothrockii*, with juvenile hormone mimics, and *N. xylopodum*, which lacks both types of compounds, to determine if there are differences in trichome type that correspond to the type of or lack of insect-active phytochemicals from the different Namas.

MATERIALS AND METHODS

Plant Material

Specimens in the genus *Nama* were collected in 1987 at the following localities: *Nama hispidum* var. *revolutum*, Tucson, Pima Co. (collected in March 1987), Arizona, USA; *N. lobbii*, Eldorado Co., California, USA (*ARIZ 269694*); *N. rothrockii*, Inyo Co., California, USA (*ARIZ 269693*); *N. xylopodum*, Eddy Co., New Mexico, USA (*ARIZ 271153*). Voucher specimens for these species were identified by the author and are deposited at the University of Arizona Herbarium.

Leaf Preparation for Scanning Electron Microscopy

Air-dried leaves were fixed in formaldehyde:glutaraldehyde (4:1) in 100 mM sodium phosphate buffer,
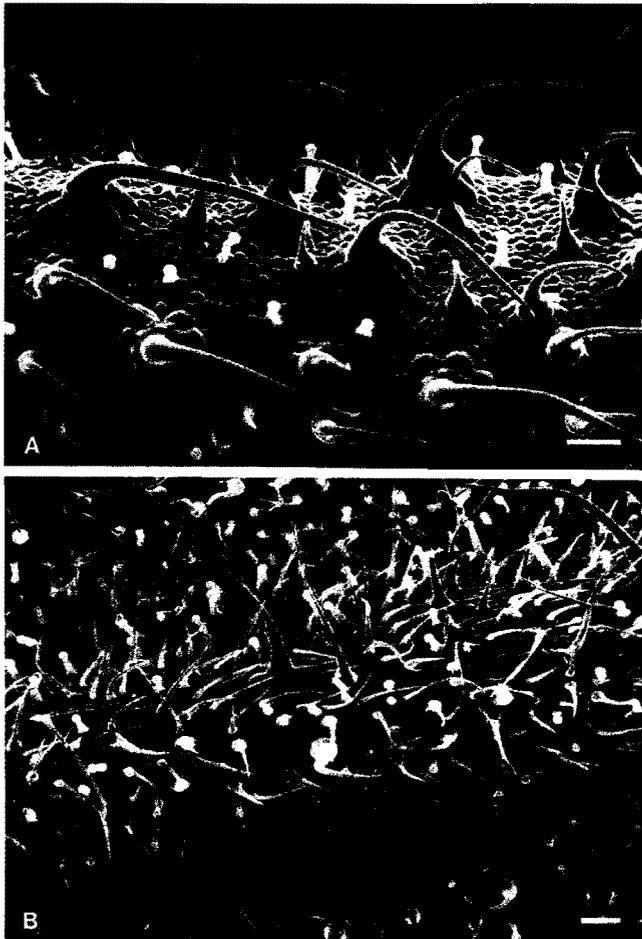
Fig. 1. Scanning electron micrographs of the adaxial (A) and abaxial (B) leaf surfaces of *Nama hispidum* var. *revolutum*. (Bars = 100 μm).
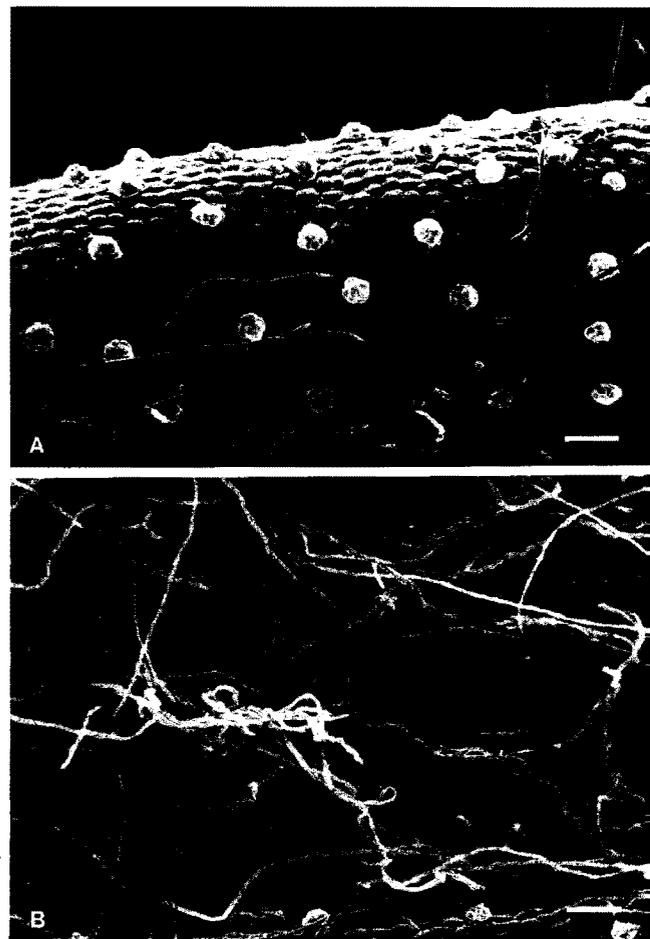

Fig. 2. Scanning electron micrographs of the adaxial (A) and abaxial (B) leaf surfaces of *Nama lobbii*. (Bars = 100 μm).

pH 7.2. Leaves were processed through an ethanol and Freon TF (Van Waters and Rogers, San Mateo, CA) dehydration series, critical point dried using liquid carbon dioxide, and sputter coated with gold (Postek et al. 1980). Scanning electron micrographs were taken of the adaxial and abaxial leaf surfaces on an International Scientific Instrument DS 130. Leaf surface structures are classified following the nomenclature of Theobald et al. (1979).

Measurements

Only the mean values and standard errors were cited in the results for all dimensions, which are based on ten measurements.

RESULTS

The adaxial and abaxial leaf surfaces of *N. hispidum* var. *revolutum* were densely covered with both long recurved and short, semierect, acicular, nonglandular trichomes and short-stalked, capitate, glandular trichomes. The larger acicular trichomes had micropa-

pillae. Recurved, acicular trichomes on the adaxial surface were 555 ± 40 μm long and were interspersed among shorter, semierect, acicular trichomes 185 ± 12 μm long. The distance between adaxial acicular trichomes was 345 ± 28 μm and 187 ± 23 μm, respectively, for long and short trichomes (Fig. 1A). Short-stalked capitate glands were 80 ± 5 μm long, and distances separating them on the adaxial surface were 211 ± 26 μm. Stomata on the adaxial surface were clearly visible. Long, recurved acicular trichomes on the abaxial surface were 537 ± 25 μm long and were interspersed among more numerous, semierect, acicular trichomes 178 ± 16 μm long (Fig. 1B). The distance between long and short, acicular trichomes was 760 ± 102 μm and 90 ± 10 μm, respectively. Short-stalked glands were 71 ± 5 μm long, and the distance between them was 113 ± 14 μm. Stomata on the abaxial surface were also clearly visible.

The adaxial and abaxial leaf surfaces of *N. lobbii* had filiform trichomes and sessile, multicellular, capitate glands. Filiform trichomes on the adaxial surface were widely distributed, 273 ± 45 μm apart and 420

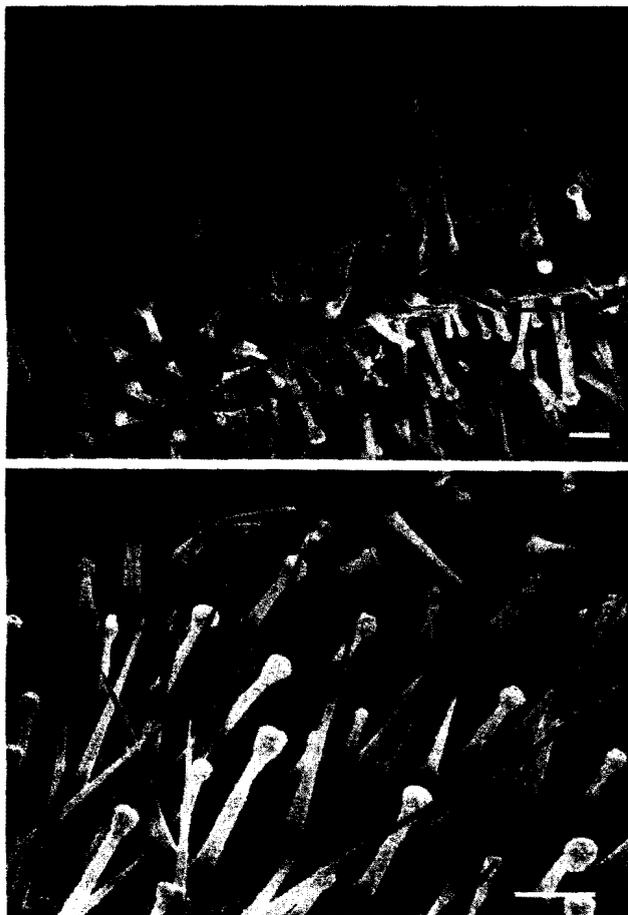Fig. 3. Scanning electron micrographs of the adaxial (A) and abaxial (B) leaf surfaces of *Nama rothrockii*. (Bars = 100 μm).

Fig. 4. Scanning electron micrographs of the adaxial (A) and abaxial (B) leaf surfaces of *Nama xylopodum*. (Bars = 100 μm).

± 36 μm long (Fig. 2A). Glands were uniformly distributed, 32 ± 2 μm high and 152 ± 11 μm apart. By contrast, the abaxial surface was nearly covered by the long, closely interwoven, filiform trichomes (Fig. 2B). Length and spacing among the filiform trichomes could not be determined. Glands on the abaxial surface were nearly completely obscured by the mat of filiform trichomes. Stomata were visible on the adaxial surface; on the abaxial surface stomata were mostly blocked from view by the long, tangled, nonglandular trichomes.

The adaxial and abaxial leaf surfaces of *N. rothrockii* had erect, smooth, subulate, nonglandular trichomes and multicellular, long-stalked, capitate glands (Fig. 3A, B). Erect subulate trichomes on the adaxial surface were in three size categories: 697 ± 14 μm, 368 ± 23 μm, and 133 ± 10 μm long. They were 697 ± 63 μm, 247 ± 16 μm, and 135 ± 11 μm apart, respectively. Uniformly distributed, long-stalked, capitate glands were 215 ± 35 μm long and were 382 ± 40 μm apart on the adaxial surface. Clusters of cells forming the enlarged terminal head of the glands on both surfaces were clearly visible. Stomata were visible on both leaf surfaces.
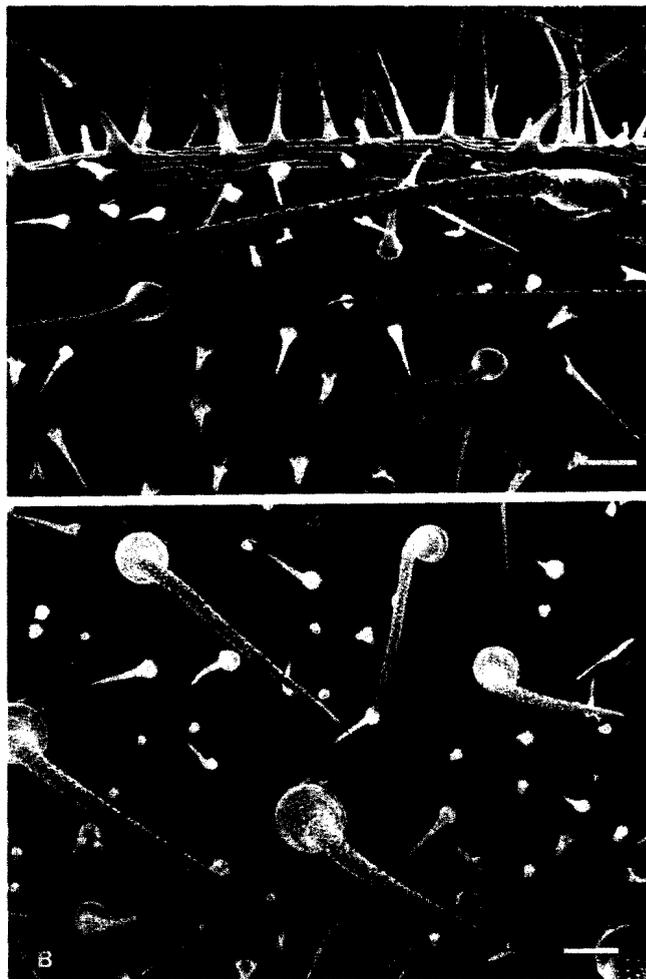
Erect trichomes on the abaxial surface were in two length categories: 445 ± 27 μm and 270 ± 24 μm long. Long trichomes were 380 ± 45 μm apart while short trichomes were 133 ± 16 μm apart. Long-stalked capitate glands were 126 ± 14 μm long and 145 ± 15 μm apart.

The adaxial and abaxial leaf surfaces of *N. xylopodum* had long acicular and short, semierect, nonglandular and short-stalked, capitate, glandular trichomes. Acicular trichomes on the adaxial surface were 515 ± 22 μm and 137 ± 12 μm long. They were 497 ± 48 μm and 237 ± 25 μm apart, respectively (Fig. 4A). Short-stalked glands on the adaxial surface were 62 ± 1 μm long. Recurved and semierect acicular trichomes on the abaxial surface were in two length categories: 515 ± 22 μm and 137 ± 12 μm long (Fig. 4B). They were 253 ± 22 μm and 158 ± 20 μm apart, respectively. Short-stalked glands on the abaxial surface were 42 ± 3 μm long and 106 ± 6 μm apart. Larger acicular trichomes had micropapillae. Stomata were visible on both leaf surfaces.

DISCUSSION

All leaves examined have nonglandular and glandular trichomes on the adaxial and abaxial leaf surfaces. The nonglandular trichome structure, however, is variable: those of *N. lobbii* are long, narrow and flexible and are tangled and interwoven into a dense sheet on the abaxial surface; those of *N. rothrockii* are cone-shaped, erect, and stiff; those of *N. hispidum* and *N. xylopodum* are semierect, intermediate-length, and recurve toward the leaf surface. All four species have multicellular glandular trichomes; those of *N. lobbii* are short and sessile; those of *N. rothrockii* are long and clavate; those of *N. xylopodum* and *N. hispidum* are narrow and capitate. Trichomes are known to help protect plants from insect herbivory (Kahn et al. 1986; Norris and Kogen 1980) and these leaf structures may have a similar contribution to the defense against herbivores in the genus *Nama*. Because of a lack of information about the insects associated with plants in *Nama*, defining a role for trichomes in the defense against insect predators must wait until further studies are completed.

Adaxial and abaxial surfaces in each species are morphologically different. Typically, the abaxial leaf surface is more densely populated with trichomes. The densely tangled abaxial trichomes of *N. lobbii* nearly cover the leaf surface and this feature may prevent or deter insect herbivore attack as it does in other plant genera (Norris and Kogan 1980). Similarly, the long nonglandular trichomes of *N. xylopodum* may protect the plant from attack as do similar structures associated with leaves of *Phaseolus vulgaris* L. (Pillemer and Tingey 1976). The well-protected abaxial surface may influence oviposition by certain types of insects such as moths, some of which are known to preferentially oviposit on the underside of leaves (Jackson et al. 1983, Navasero and Ramaswamy 1991).

Phytochemicals may account for some differences in leaf structures in *Nama*. *Nama hispidum* and *N. lobbii* produce antijuvenile hormones while *N. rothrockii* produces juvenile hormone mimics (Binder et al. 1991) and these phytochemicals may have an important role in defense against herbivores. Precocene II is recognized as a toxicant and mediator of insect behavior (Binder and Bowers 1991, 1992, 1993) and its presence in the trichomes of *N. hispidum* may assist the delivery of this compound at the appropriate time or to the best location for maximum protection from insects. For species of *Nama* examined in this study, however, there is no apparent relation between type of trichome and chemical defenses. *Nama hispidum* and *N. xylopodum* have similar trichomes, but *N. hispidum* and *N. lobbii* produce precocene. Moreover, *N. lobbii* has trichomes distinct in structure from both *N. hispidum* and *N. xylopodum*. *Nama rothrockii* produces

juvenile hormone mimics and has trichomes that are distinct from those of the above species. Trichome diversity in *Nama* may be species specific and not chemistry specific although further studies are needed on additional *Nama* species to delineate relationships of chemical biosynthesis of insect-active compounds and trichome types in *Nama*.

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