

STUDIES ON THE POLLINATION ECOLOGY OF *TIPULARIA DISCOLOR* (ORCHIDACEAE)¹

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ABSTRACT

Tipularia discolor, a woodland orchid, flowers in mid-summer when reproductive activity is minimal within the herb synusia. *Tipularia* is insect-pollinated, and artificial crosses showed that seeds are produced after self-pollination, intra-inflorescence pollination, and outcrossing. The single nocturnal pollinator, *Pseudaletia unipuncta* (Noctuidae), located *Tipularia* populations within a day or two of anthesis. Pollinators were shown to be capable of utilizing portions of the inflorescences that contained the most nectar. After total nectar resources declined, pollinators were no longer active on the inflorescences, even though flowers and nectar were still available. The mode of pollinator activity seems to be closely related to floral morphology, although the moths are able, early in the flowering phenophase, to successfully obtain nectar without effecting any change in the reproductive status of flowers.

ASEXUAL REPRODUCTION is the predominant mode of propagation of perennial herbs in mesophytic temperate deciduous forests (Kawano, 1975; Struik, 1965; Whigham, 1974) and a large percentage of the net annual primary production is allocated to belowground storage structures (Abrahamson, 1979). Accordingly, most would be characterized as K-adapted species (Gadgil and Solbrig, 1972; Harper, 1977). In addition, most produce small numbers of large seeds and rely primarily on perennation by asexual reproduction (Kawano, 1970, 1975; Kawano and Nagai, 1975; Morgan, 1971; Stevenson, 1972; Whigham, 1974).

Tipularia discolor (Cranefly orchid), a species which is widespread throughout the eastern deciduous forest herbaceous flora (Braun, 1950), is of interest because it is one of the few woodland herbs that produces numerous seeds, a characteristic usually associated with species of early successional habitats, yet populations contain few seedlings and asexual reproduction is the primary mode of population maintenance (Whigham, unpublished). *Tipularia* exhibits other life cycle characteristics that are of ecological interest. *Tipularia* and *Aplectrum hyemale* (Auclair, 1972; Adams, 1970) are the only two orchids in eastern deciduous forests that produce leaves,

usually one per plant, that develop in the late summer, persist through the winter, and senesce in spring prior to the onset of flowering. Unlike *Aplectrum*, *Tipularia* flowers in mid-summer (July–August) when sexual reproduction within the herb synusia is minimal (Taylor, 1974). *Tipularia*, consequently, flowers when pollinator resources, pollen and nectar, are minimal. This paper reports results of studies of the reproductive ecology of *Tipularia*. Specific objectives were to determine: (1) the type of breeding system exhibited by *Tipularia*; (2) the relationship between flowering phenology and pollinator activity; and (3) the relationship between nectar production and pollinator activity.

The research was conducted at the Chesapeake Bay Center for Environmental Studies (CBCES), a 1,000-ha research facility of the Smithsonian Institution. The CBCES property is located on the watershed of the Rhode River (Correll, 1977), approximately 16 km south of Annapolis, Maryland. Upland vegetation consists of a series of mature and successional forests, as well as pasture and cultivated areas. The *Tipularia* research was performed in a mixed hardwood forest that has not been disturbed since the Civil War (1861–65).

Dominant overstory species include *Quercus alba*, *Fagus grandifolia*, *Carya glabra*, *Carya tomentosa*, *Quercus falcata*, *Liriodendron tulipifera*, and *Liquidambar styraciflua*. Understory vegetation consists of canopy transgressives and *Cornus florida*, *Carpinus caroliniana*, *Viburnum dentatum*, and *Lindera benzoin*. The herb synusia is diverse, with *Claytonia virginica*, *Dentaria laciniata*, *Podophyllum peltatum*, *Smilacina racemosa*, and

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Desmodium nudiflorum being abundant species. Small populations of *Tipularia* are distributed throughout the forest.

METHODS—Breeding experiments were conducted in 1977 and 1978. Inflorescences, which emerge in July, were located prior to anthesis and the following manipulations performed: (1) pollinia were removed from 151 flowers on inflorescences of seven plants (apomixis); (2) pollinia were also removed from 45 flowers on three inflorescences and remaining flowers (53) on the three inflorescences were used for a study of automatic selfing and were not manipulated. The three inflorescences were then covered with a nylon bag to exclude pollinators: (3) flowers on eight plants (133 flowers) were self-pollinated (autogamy) and inflorescences were covered with nylon bags; (4) flowers of eight plants (156 flowers) were outcrossed (xenogamy) and inflorescences covered with nylon bags; (5) intra-inflorescence crosses (geitonogamy) were made on five plants (78 flowers) and inflorescences were covered with nylon bags. Nylon bags were removed when floral parts started to senesce. In addition, all inflorescences within a control population of 100 (1977) and 118 (1978) plants were observed throughout the flowering period and the number of pollinated flowers counted on each inflorescence.

The activities of pollinators were studied during both flowering seasons. All plants in the marked populations were visited at 2 or 3 day intervals in 1977 and 1–3 day intervals in 1978. During each visit in 1977, we examined all flowers on each inflorescence and scored them according to the following scheme which was designed to permit us to determine when flowers were visited and the impact of the pollinators: (1) flower intact; (2) pollinia removed; (3) pollinia placed on flower but not on stigmatic surface; (4) pollinia placed on stigmatic surfaces.

Observations during the 1977 study indicated that pollinators might have removed nectar from some flowers without otherwise affecting them. In 1978, we repeated the study, but also measured the amount of nectar in the spur of each flower. Nectar was estimated from measurements of the length of the nectar column in the spur of each flower. The spurs of *Tipularia* flowers are translucent and, nearest the throat, the meniscus of the nectar column is clearly visible. Nectar column length (mm) was converted to μ l of nectar using a regression equation that was developed by measuring the length of the nectar column (X) in flowers and then extracting the nectar with 1- μ l micropi-

ettes. The linear regression equation was $Y = -.143 + .11475X$, where Y is nectar volume (μ l). The correlation coefficient for the equation was .93 ($n = 50$ flowers).

The study populations were observed daily in 1977, and it was apparent that the flowers were visited by nocturnal animals. In 1978, we attempted to collect pollinators by sampling for 5-hr periods, on four consecutive nights, during the peak in pollinator activity. The full moon conditions that prevailed during the four nights (August 13–16) enabled us to observe and collect pollinators that visited *Tipularia* inflorescences. Phenological aspects of the marked population were noted at monthly intervals between May, 1977 and October, 1978. Observations were increased to a weekly interval during the flowering phenophase.

RESULTS—Inflorescences began to emerge during the 1st wk of July in 1977 and the 2nd wk in 1978. Twenty-three percent of 100 marked plants flowered in 1977 and 8.9% of 118 in 1978. Only one marked plant flowered in both 1977 and 1978. This observation is consistent with other data (Whigham, unpublished) indicating that very few plants flower in consecutive years.

The species successfully set seed after self-pollination and both inter- and intra-inflorescence crosses. The highest success rates were for artificially selfed flowers, where $91.3 \pm 3.4\%$ (1 SE) of the flowers set seed. A slightly lower seed set success occurred in inter-inflorescence ($69.1 \pm 8.2\%$) and intra-inflorescence ($84.0 \pm 11.1\%$) crosses. The lower percentages reflect the abortion of developing fruits due to unknown causes in 1978. Flowers from all inter- and intra-inflorescence crosses made in 1978 began to develop fruits, but several aborted within two weeks. The low (23.9%) percentage of fruit development from flowers that had pollinia removed (no bag placed over inflorescence), indicates that only a small percentage of flowers on a given inflorescence are successfully pollinated. We anticipated that there would be no fruit development in flowers that were tested for apomixis by removing the pollinia and then covering the inflorescence with a nylon bag. No fruits developed from flowers tested for apomixis in 1977 but, unfortunately, in 1978 each inflorescence used to test for apomixis was also used to test for automatic selfing and thus had terminal flowers from which we did not remove the pollinia. Fruits developed from $15.9 \pm 5.0\%$ of the flowers used to test for apomixis and $12.8 \pm 8.9\%$ of those used to test for automatic selfing. After removal of the nylon bags, we noted, however,

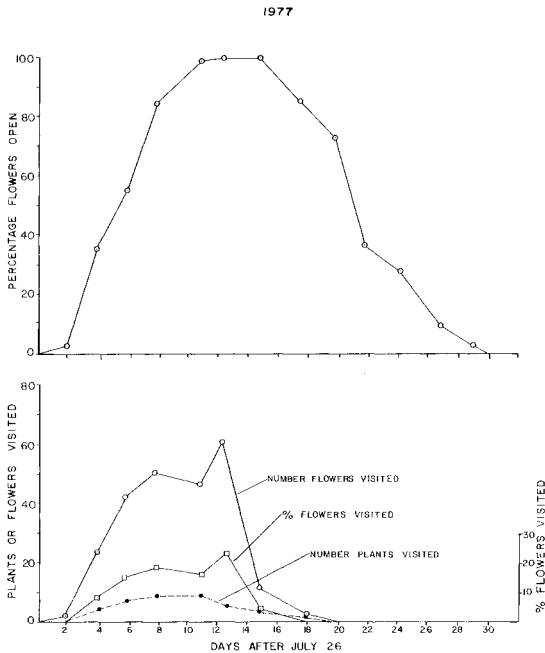


Fig. 1. Results of 1977 population census during the flowering phenophase. Pattern of flower availability is shown in upper graph. Temporal pattern of pollinator activity is shown in lower graph. Pollinator visitations were based on noting physical changes in the reproductive status of *Tipularia* flowers (see methods for explanation of procedure).

that pollinia had been removed from some flowers used to test for automatic selfing and had, somehow, been transferred to stigmatic surfaces of all flowers that developed into fruits. The data, therefore, suggest that *Tipularia* is not apomictic nor does it automatically self, because no fruits developed from any of the flowers that remained intact.

Anthesis began on July 26, 1977, and the last floral part had fallen by August 15. The flowering phenophase began later (August 4) in 1978, and lasted for 23 days, which was five days longer than in 1977 (Fig. 1, 2). In both years, all flowers had opened by the 12th day after initial anthesis (Fig. 1, 2). Plants were first visited by pollinators on day 4 after anthesis in 1977, and on the 5th day in 1978. Visitations continued for 13 (1977) and 15 (1978) days, with pollinator activities peaking on the 13th day after anthesis in 1977 and on the 11th day in 1978. Interestingly, pollinator visitations ceased while there were still open flowers. In 1977, open flowers were present for 15 days after the last definite pollinator visitation. In 1978, open flowers were present for only 3 days after the last recorded pollinator

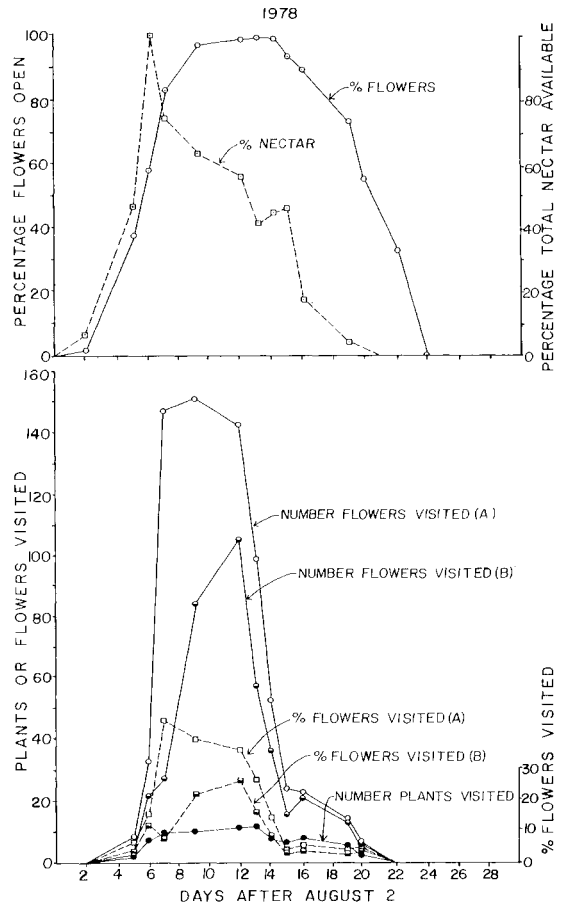


Fig. 2. Results of 1978 population census during the flowering phenophase. Patterns of flower and nectar availability are shown in upper graph. Pattern of pollinator activity is shown in lower graph (see methods for a description of procedure). Parts of the figure labeled B are visitations based on noting physical changes in reproductive status of *Tipularia* flowers. Parts labeled A are based on estimated total visits, which is the sum of B plus visitations during which nectar was removed, but no change in floral reproductive status was observed.

visitations. The maximum percentage of available flowers visited daily by pollinators was between 25% and 45% (Fig. 1, 2). Although each inflorescence was not visited daily, all were visited during the flowering phenophase.

Figure 2 shows the amount of nectar available during the flowering phenophase. Maximum nectar availability occurred very early and prior to anthesis of all flowers. Total available nectar then began to decline within the population once flowers began to be visited by pollinators and there were very few pollinator visitations after the large drop (30%) in nectar between days 15 and 16. The weighted mean

estimated amount of nectar production was $.98 \pm .08$ (1 SE) μl per flower and ranged from a high of $1.35 \pm .47$ μl per flower to a low of $.53 \pm .22$ μl per flower.

As mentioned in methods, it seemed that potential pollinators were capable of removing nectar without effecting any change in floral reproductive status. We attempted to clarify this question by comparing the number of flowers that showed any of the observed morphological changes, as well as had significant ($.23$ – $.25$ μl or more) changes in nectar volume (Method A, Fig. 2), with the number of flowers that only had observable morphological changes (Method B, Fig. 2). The two calculations differed, with the largest discrepancy found on the seventh day after anthesis. On that day, volumetric changes in nectar indicated that the reproductive status changed on only 18% of the 138 flowers that were visited. A large drop in total nectar also occurred on that day (Fig. 2). Differences between the methods (Fig. 2) of determining visitations then declined and averaged approximately 40% thereafter.

Three potential pollinator species, all of them moths, visited *Tipularia* inflorescences during each of the four observation periods. Two smaller geometrid moths (*Protoboarmia porcelaria* and *Xanthorhoe ferrugata*) were frequently seen, but only occasionally visited *Tipularia* inflorescences. When they did alight on a flower, they did not spend more than 2 or 3 sec and they flew away without visiting any other flowers. Examination of flowers after they had been visited, showed that the smaller moths did not effect any changes in the reproductive status of the flowers and, based on visual observations before and after visitations, they did not appear to remove nectar from spurs. Large noctuid moths (*Pseudaletia unipuncta*) were observed each night, and their flight pattern suggested they recognized *Tipularia* inflorescences. *Pseudaletia*, typically, was observed flying about 0.5–1 m above the ground. When individuals flew within approximately 3–5 m of an inflorescence, their pattern of flight suddenly altered, and they moved immediately to the inflorescence. *Pseudaletia* moths alighted on sepals and petals at the open throats of flowers. The heads then were inserted into the throat regions of flowers while nectar was removed. The moths' wings were continually in motion while flowers were visited, and they were able to move quickly from one flower to the next on a given plant. Each moth only visited a few flowers on an inflorescence before flying away. Visual observations of *Tipularia* inflorescences immediately

after the visitation showed, however, that nectar was removed from all flowers visited, while pollinia remained intact on some visited flowers.

DISCUSSION—Although orchid flowers are adapted for insect pollination (van der Pijl and Dodson, 1966; Vogel, 1978), Hagerup (1952) and Kirchner (1922) have shown that insect pollination of temperate orchids is not obligatory and that autogamy occurs in several species. *Tipularia* does not appear to be autogamous nor is outcrossing obligatory. Apomixis and automatic selfing in *Tipularia* can't be excluded, but we believe that it, at most, occurs only infrequently. Of the flowers that were being tested for apomixis and automatic selfing, fruits only developed on those that, for some unknown reason, had pollinia on the stigmatic surface.

Self-pollination resulted in successful seed production, but was only effected through the action of a pollinator. Seeds were also produced by intra-inflorescence crosses and outcrosses. The potential to produce seeds independent of the source of pollinia would be of selective advantage to *Tipularia* because there are so few pollinators present during the flowering phenophase (only one species observed during this study). Few other temperate orchids seem to exhibit this type of restricted pollinator-plant interaction. Stevenson (1972) found that *Goodyera pubescens*, a species with a flowering phenology similar to *Tipularia* was pollinated by *Bombus* during daytime and that the species was not autogamous, although he did not perform artificial crosses. Ackerman (1975) also has cited *Bombus* pollination of *Goodyera*. As noted (Hagerup, 1952; Ackerman and Mesler, 1979), some temperate zone orchids are capable of autogamy, but most appear to be insect pollinated. Nilsson (1978a) found that 28 species of lepidopterans, primarily Noctuidae, visited *Platanthera chlorantha* in Sweden. Moth-pollination of *Platanthera* has also been cited by Stoutamire (1944) and Smith and Snow (1976). *Epipactis palustris*, in Sweden (Nilsson, 1978b), was visited by a broad spectrum of insects. Of the species examined by Nilsson, the mode of pollination of *Platanthera* was somewhat similar to that observed in this study. *Platanthera* was also pollinated by moths and there was a specific relationship between size of moths' heads and morphology of the flower reproduction parts. Flowers of both *Platanthera* and *Tipularia* frequently are robbed of nectar (Fig. 2), seemingly without any effective change in the reproductive status of the flower. It seems like-

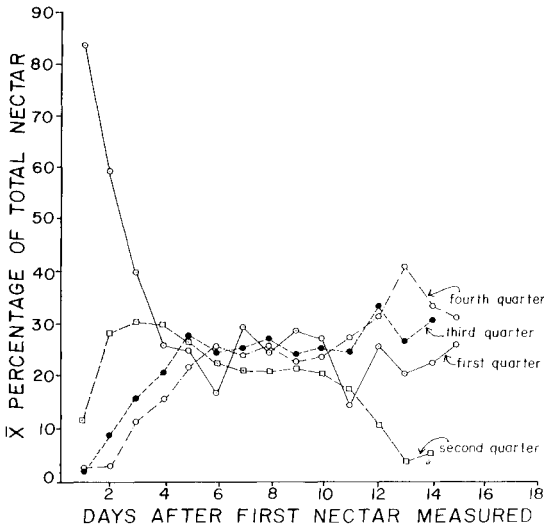


Fig. 3. Comparison of temporal availability of nectar in *Tipularia* inflorescences. Values represent mean daily percentages of nectar present in quarters of eleven inflorescences examined in 1978. Quarters represent, in ascending order beginning with the 1st quarter, sections of the inflorescence.

ly that a moth could remove nectar from full *Tipularia* spurs without effecting changes in the flower. When nectar volume is less, the moth, which has to probe deeper in the spur for nectar, touches the viscid disc of the pollinia with its eyes. When the moth's head moves further into the flower, the pollinia, now attached to the moth's eye, are pulled from the anther sac and moved to the region of the stigmatic surface, which is located immediately posterior to the anther sac. The stigmatic surface is very sticky and pollinia that contact it remain when the moth pulls away from the flower. Nilsson (1978a, 1978b) also found that deposition of pollinia in insect eyes is a common mode of pollen transfer in temperate orchids. As shown in Fig. 2, moths more frequently effect changes in the floral reproductive status after nectar begins to decline in the *Tipularia* population. Once pollinia were transferred to the stigmatic surface, almost all of those flowers developed fruits ($93.5 \pm 2.9\%$ in 1977 and $70.7 \pm 8.7\%$ in 1978).

Because of the interdependent nature of the moth and *Tipularia* breeding system, pollinator patterns at the population level are of interest. Fig. 1, 2 show that pollinators located *Tipularia* flowers within a day or two of the beginning of anthesis. Pollinator activity increased sharply and then declined, even though open flowers and nectar were available for a much longer period of time. Willson and Bertin (1979) have provided additional infor-

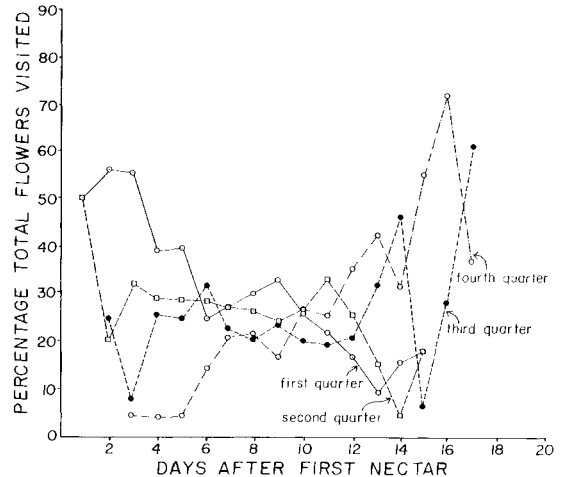


Fig. 4. Comparison of temporal patterns of pollinator visitations to portions of *Tipularia* inflorescences. All values represent mean daily percent of visitations to quarters of eleven inflorescences examined in 1978. Quarters represent, in ascending order beginning with 1st quarter, sections of the inflorescence. Visitations calculated as described for A in Fig. 2.

mation suggesting that *Pseudaletia unipuncta* exploits nectar for only a portion of the flowering period. They noted that *Pseudaletia* was a common pollinator of *Asclepias* for only 1 wk out of the total flowering period. *Pseudaletia* was seemingly able to respond to the total amounts of nectar available and, based on our technique for identifying visitations, ceased foraging for *Tipularia* nectar after nectar production declined below some minimum level. The production of nectar occurred acropetally on the inflorescence (Fig. 3) in synchrony with anthesis that also started at the base of the inflorescences. Initial pollinator visits were to the lower half of the inflorescences (Fig. 4), where almost all of the nectar was located (Fig. 3). After approximately 5 days, nectar was equally divided throughout the inflorescences (Fig. 3) and there was also indiscriminate use of the inflorescences by the pollinators (Fig. 4). After the 10th day, flowers on the upper half of the inflorescence contain most of the nectar, although the total amount was less (Fig. 2), and pollinators began selectively to utilize the upper halves of inflorescences (Fig. 4). The few visitations that occurred near the end of the flowering period (Fig. 1, 2) were restricted to the upper flowers (Fig. 4) where all of the nectar was found (Fig. 3). Pollinators therefore not only respond to overall levels of nectar availability within the flowering cycle of *Tipularia*, but also seem able selectively to utilize those specific areas of a given inflorescence where potential rewards are greatest.

LITERATURE CITED

- ABRAHAMSON, W. G. 1979. Patterns of resource allocation in wildflower populations of fields and woods. *Amer. J. Bot.* 66: 71–79.
- ACKERMAN, J. D. 1975. Reproductive biology of *Goodyera oblongifolia* (Orchidaceae). *Madroño* 23: 191–198.
- ACKERMAN, J. D., AND M. R. MESLER. 1979. Pollination biology of *Listera cordata* (Orchidaceae). *Amer. J. Bot.* 66: 820–824.
- ADAMS, M. S. 1970. Adaptations of *Aplectrum hyemale* to the environment: Effects of preconditioning on net photosynthesis. *Bull. Torrey Bot. Club* 97: 219–224.
- AUCLAIR, A. N. 1972. Comparative ecology of the orchids *Aplectrum hyemale* and *Orchis spectabilis*. *Bull. Torrey Bot. Club* 99: 1–10.
- BRAUN, E. L. 1950. Deciduous forests of eastern North America. Hafner Publ. Co., New York.
- CORRELL, D. L. 1977. An overview of the Rhode River watershed program. In: D. L. Correll [ed.], Watershed research in Eastern North America, CBCES, Smithsonian Institution, Edgewater, Maryland, p. 100–120.
- GADGIL, M., AND O. T. SOLBRIG. 1972. The concept of r and K selection: Evidence from wildflowers and some theoretical considerations. *Amer. Nat.* 106: 14–31.
- HAGERUP, O. 1952. Bud autogamy in some northern orchids. *Phytomorphology* 2: 51–60.
- HARPER, J. L. 1977. Population biology of plants. Academic Press, New York.
- KAWANO, S. 1970. Species problems viewed from productive and reproductive biology. I. Ecological life histories of some representative members associated with temperate deciduous forests in Japan—A preliminary discussion. *J. Coll. Lib. Arts, Toyama Univ.* 3: 181–213.
- . 1975. The productive and reproductive biology of flowering plants. II. The concept of life history strategy in plants. *J. Coll. Lib. Arts, Toyama Univ.* 8: 51–86.
- , AND Y. NAGAI. 1975. The productive and reproductive biology of flowering plants. I. Life history strategies of three wild *Allium* species in Japan. *Bot. Mag. (Tokyo)* 88: 281–318.
- KIRCHNER, O. 1922. Über selbstbestäubung bei den Orchideen. *Flora* 115: 103–129.
- MORGAN, M. D. 1971. Life history and energy relationships of *Hydrophyllum appendiculatum*. *Ecol. Monogr.* 41: 329–349.
- NILSSON, L. A. 1978a. Pollination ecology and adaptation in *Platanthera chlorantha* (Orchidaceae). *Bot. Notiser* 131: 35–51.
- . 1978b. Pollination ecology of *Epipactis palustris* (Orchidaceae). *Bot. Notiser* 131: 355–368.
- SMITH, G. R., AND G. E. SNOW. 1976. Pollination ecology of *Platanthera (Habenaria) ciliaris* and *P. blephariglottis* (Orchidaceae). *Bot. Gaz.* 137: 133–140.
- STEVENSON, J. C. 1972. Evolutionary strategies and ecology of *Goodyera* and *Spiranthes* species (Orchidaceae). Ph.D. Thesis, Univ. North Carolina, Chapel Hill.
- STRIJK, G. J. 1965. Growth patterns of some native annual and perennial herbs in southern Wisconsin. *Ecology* 46: 401–420.
- STOUTAMIRE, W. P. 1974. Relationships of the purple-fringer orchids *Platanthera psycodes* and *P. grandiflora*. *Brittonia* 26: 42–58.
- TAYLOR, F. G., JR. 1974. Phenodynamics of production in a mesic deciduous forest. In: H. Lieth [ed.], Phenology and seasonality modeling, p. 237–254. Springer-Verlag, N.Y.
- VAN DER PIJL, L., AND C. H. DODSON. 1966. Orchid flowers: their pollination and evolution. Coral Gables, Florida.
- VOGEL, S. 1978. IV. Floral ecology. Report on the years 1974 (73) to 1978. In: H. Ellenberg, K. Esser, H. Merxmüller, E. Schnepf, and H. Ziegler [eds.], *Progress in botany*, p. 453–481. Springer-Verlag, New York.
- WHIGHAM, D. F. 1974. An ecological life history study of *Uvularia perfoliata* L. *Amer. Midl. Nat.* 91: 353–359.
- WILSON, M. F., AND R. I. BERTIN. 1979. Flower-visitors, nectar production, and inflorescence size of *Asclepias syriaca*. *Can. J. Bot.* 57: 1380–1388.