OBSERVATIONS ON GALL MIDGES OF WHITE AND RED CLOVER¹

Waarnemingen inzake de galmuggen van witte en rode klaver

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INTRODUCTION

From 1956 to 1958 an investigation was undertaken in order to find out whether gall midges might be responsible for the often insufficient development and growth of white clover in artificial pastures. In addition it was considered appropriate to start a study of the morphology, biology, distribution and economic importance of the gall midge species which are present on white and red clover in the Netherlands.

METHODS

The methods for this study included field observations and laboratory experiments. Samples of white and red clover flower heads were taken on various dates from various areas to provide material for experimental purposes. These flower heads were placed in lamp glasses, the bases of which were inserted in glass cylinders containing an inch or two of water (Plate 1A). The lamp glasses were covered with muslin sewn on to iron rings. Gall midge larvae, which normally pupate in soil, dropped into the water from which they could easily be extracted. These larvae were transferred to wide lamp-glasses containing fibre and standing in soil filled flower pots (Plate 1B). In these emergence pots pupation and subsequent emergence could take place.

Some of the adults obtained in this way were mounted for identification and some were used in breeding experiments. These breeding experiments were carried out in small wooden-framed cages, covered with nylon gauze. These cages were kept in an unheated greenhouse. The breeding experiments gave further details concerning the life history of the clover midges and their damage with its economic importance.

The figures were drawn by the author with the aid of a camera lucida from slides in his collection.

RESULTS

Gall midges of white clover

Gall midges or Cecidomyidae are Dipterous insects. They are small and delicately built. The species occurring on white clover in the Netherlands may be divided into the following groups: a. phytophagous species, b. predators and c. species whose life history is unknown.

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a. Phytophagous species

Dasyneura trifolii (F. Loew 1874)

The female midge lays her eggs in the young, unexpanded leaflets. Under the influence of the larvae, which at first are white, changing to orange at maturity, the leaflets remain folded along the midrib so that the upper surfaces are contiguous (Plate 2A). This gall becomes hard, but rather fleshy and inflated towards the midrib; the inflated part is yellowish-green or reddish. Usually only one of the leaflets is affected, sometimes two. The full-grown larvae, which are about 2 mm long, pupate in the gall in a white silken cocoon except for the overwintering larvae which pupate in the soil in the following spring. Just before emergence of the midge the pupa penetrates through the closed leaflets until it partially protrudes.

There are several generations a year. Hibernation takes place in the larval stage in the soil.

On the ventral surface of the prothorax of the matured legless larvae a characteristic organ typical for Cecidomyid larvae can be distinguished. It is the sternal spatula, anchor process or breastbone. This is a strongly sclerotised rod, deeply bilobed and free anteriorly, and with its posterior extremity rounded and sunk into a pocket formed by the body-wall (fig. 1b). The spatula is only present in the full-grown larva; the exact function of this organ is still uncertain

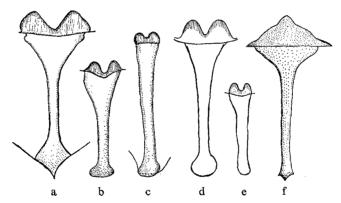


Fig. 1. Breastbones of / Borststaafjes van:

- a. Tricholaba trifolii (Rübs.) × 150.
- b. Dasyneura trifolii (F. Loew) × 150.
- c. Phaenobremia aphidivora (Rübs.) × 150.
- d. Clinodiplosis leguminicola Milne. (After MILNE / Naar Milne) × 180.
- e. Isodiplosis deutera Milne. (After MILNE / Naar Milne) × 180.
- f. Campylomyza ormerodi (Kieffer). (After Smith / Naar Smith) × 150.

The size of the midges is about 1.5 mm. The abdomen of the male ends in a tong-shaped hypopygium, which is characteristic for the genus *Dasyneura* (fig. 2). The wings are well developed and show a simple venation. The antennae have two basal segments and generally 12 or 13 flagellar segments. Each flagellar segment is single noded, with applied circumfila. The first two flagellar segments are fused.

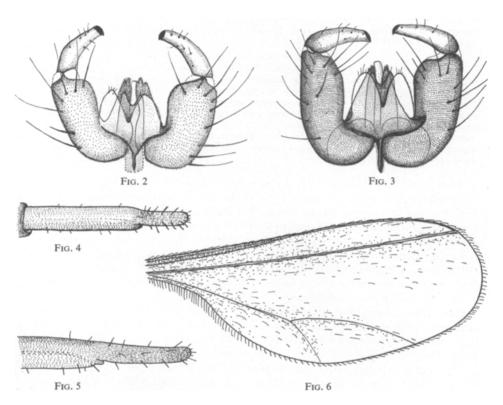
The abdomen of the female ends in an extensile ovipositor (fig. 4). The flagellar antennal segments are barrel-shaped and covered with setae.

BARNES (1946) records that this gall midge species occurs in Europe and in the U.S.A. He mentions as host plants: *Trifotium pratense* L., T. repens L., T.

medium L., T. fragiferum L. en T. arvense L. In the Netherlands Dasyneura trifolii has been sampled from T. repens, T. pratense and Medicago lupulina L.

According to Barnes (1946) the larvae of another gall midge, *Tricholaba trifolii* Rübs., live as inquilines in the galls of *D. trifolii*. The larvae of *Lestodiplosis trifolii* Barnes are predators and feed upon the larvae of *D. trifolii*. The latter two species will be discussed elsewhere in this paper.

Although *D. trifolii* occurs throughout the Netherlands, no damage of economic importance was observed during our investigations. No further particulars concerning this question could be obtained from the literature.



- Fig. 2. Male genitalia of *Dasyneura trifolii*. × 200. *Copulatie-orgaan van* Dasyneura trifolii (*mannetje*).
- Fig. 3. Male genitalia of *Dasyneura gentneri*. × 287. *Copulatie-orgaan van* Dasyneura gentneri (*mannetje*).
- Fig. 4. Ovipositor of *Dasyneura trifolii*. × 287. *Legbuis van* Dasyneura trifolii.
- Fig. 5. Ovipositor of *Dasyneura gentneri*. × 287. *Legbuis van* Dasyneura gentneri.
- Fig. 6. Wing of Dasyneura gentneri. \times 33. Vleugel van Dasyneura gentneri.

Dasyneura gentneri Pritchard (1953)

The females of this species oviposit on the open florets of white clover. The just hatched larvae enter the floret through the calyx and corolla to the ovary, where they destroy the developing seed. No distinct gall is caused, but sometimes the flower heads look distressed. After a fortnight the larvae are full-grown; they are pinkish-yellow in colour then. The anchor process differs from that of Dasyneura trifolii and D. leguminicola in having smaller lobes and a shorter and broader stem (Plate 2C and D). In general, the spatula of D. leguminicola is larger and more heavily sclerotised than that of D. gentneri. During the summer the larvae pupate in the flower heads. According to Milne (1960) D. gentneri may complete four generations annually. The larvae of the last generation hibernate in the soil, in which pupation also takes place in the next spring.

The adults of *D. gentneri* are smaller than those of *D. leguminicola* and more slender than those of *D. trifolii*. Another difference between both species is in the male genitalia. In *D. leguminicola* the basal clasp segment ends in a broad, lightly sclerotised and regular apodeme (fig. 21). In *D. gentneri* the basal clasp segments end in a slender, strongly sclerotised, somewhat irregular apodeme (fig. 3). In the female of *D. gentneri* the ovipositor is more broadly rounded apically than in *D. leguminicola* (figs. 5 and 22). The wings are well developed and show a simple venation (fig. 6). The antennae of the males of *D. gentneri* consist of two basal segments and generally 13 or 14 flagellar segments; the female antennae have 11 or 12 flagellar segments.

PRITCHARD (1953) found *D. gentneri* for the first time on Ladino clover (*Trifolium repens latum*) in the U.S.A. MILNE (1960) records this species from red clover (*T. pratense*), Alsike clover (*T. hybridum* L.) and white clover from England and Germany. MARKKULA (1959) reared *D. gentneri* from white clover in Finland. In the Netherlands we sampled this gall midge from white clover at Bennekom, Kapelle, Leur and Wageningen. According to MILNE (1960) it seems probable that the larvae of the Cecidomyid *Lestodiplosis trifolii* feed on *D. gentneri* in the flower heads. The same author records *Prosactogaster error* Fitch as a parasite of *D. gentneri*.

Field-observations and literature-studies showed that *D. gentneri* is of no economic importance for the Netherlands. Bishop (1954), however, records that this species infested nearly 100% of the flower heads of a seed crop in South-Oregon (U.S.A.) in 1951. The number of infested florets varied from 7 to 75% per flower head. In Central-Oregon it was observed in 1952, that 78 to 83% of the Ladino clover flower heads had been infested and this led to an important loss of yield.

b. Predators

Lestodiplosis pallidicornis Kieffer (1912a)

Specimens of the genus Lestodiplosis were frequently reared from white and red clover flower heads. Many species of this genus feed in their larval stage on the larvae of other gal midge species. The fact that the Lestodiplosis species are difficult to separate morphologically made an exact identification of our material rather troublesome. Kieffer (1912a) described the female of Lestodiplosis pallidicornis as a predator of Dasyneura flosculorum Kieffer on Trifolium medium. However, according to Barnes (1946), Metcalfe (1933) and

PRITCHARD (1953), D. flosculorum is synonymous with D. leguminicola, which lives on several Trifolium species but mainly on T. pratense (red clover).

In 1928 Barnes described the female of Lestodiplosis trifolii as a predator of Dasyneura trifolii. The latter species causes leaf-galls on Trifolium arvense, T. fragiferum, T. medium, T. pratense, T. repens and Medicago lupulina. MILNE (1960) reared Lestodiplosis trifolii from flower heads of white and red clover and described the male. The same author stated that the larvae fed on Dasyneura leguminicola and Tricholaba trifolii in observation tube experiments and that it seems probable that they also feed on D. leguminicola, D. gentneri and Tr. trifolii in the flower heads. On examination, our specimens, reared from the flower heads of white and red clover agreed with Lestodiplosis pallidicornis and L. trifolii in morphology and biology. As the predator as well as the prey prove to be polyphagous species, the author prefers to identify the Dutch Lestodiplosis species as Lestodiplosis pallidicornis Kieffer by reason of priority.

The larvae of *L. pallidicornis* are very mobile. When they meet a Cecidomyid larva, they pierce the skin of their victim with their mouth parts and suck the body fluid (Plate 2B). The mature larvae descend to the soil to pupate. According to MILNE (1960) pupation sometimes takes place in the flower heads; no cocoon is spun.

The larvae of *L. pallidicornis* have long, two-segmented antennae. The mouthparts are well developed and sclerotised. A peculiarity of this genus is that there is no sternal spatula. The full-grown larvae are 2.5 to 3 mm long; they vary in colour from pinkish orange to dark orange-red. Fig. 18c shows the anal segment of *L. trifolii*.

The adults are about 1 to 1.5 mm long. They are slender and delicately built. The male antennae consist of two basal segments and 12 flagellar segments. The first two flagellar segments are fused. The third flagellar segment is binodose. The basal node is globular and bears two whorls of regular circumfila and one whorl of setae; the distal node is more elongate and bears one whorl of circumfila and one whorl of strong setae (fig. 9). The female antennae also have two basal segments and 12 flagellar segments and the first two flagellar segments are fused. Each flagellar segment is cylindrical with a distinct long neck; it bears long stout setae and two whorls of applied circumfila joined laterally by a longitudinal series (fig. 10).

The wings are pubescent, with pigmented areas and a simple venation (fig. 7). The basal clasp segments of the male genitalia have a large triangular lobe on the inner side. The upper lamella is deeply emarginate with broadly rounded lobes; the lower lamella is as long as the upper lamella and broadly rounded apically (fig. 8).

The female abdomen ends in a short, lamelliform ovipositor, composed of two oval lamellae and a small ventral lobe. The lobes are set with setae and microtrichia (fig. 11).

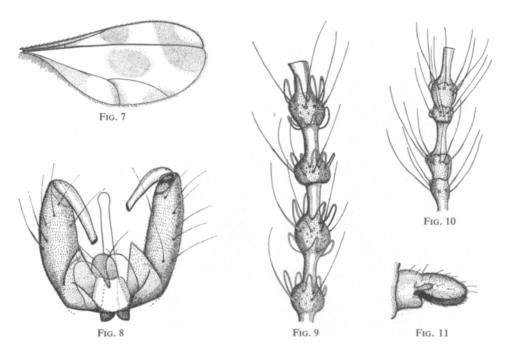
Little is known concerning the life history of L. pallidicornis. Only small numbers were bred from our clover samples and it does not seem likely that this species is of economic importance. Kieffer (1912a) reared the midges from Dasyneura leguminicola on Trifolium medium in France.

MARKKULA (1959) records it as a predator of *Dasyneura gentneri* on white clover in Finland. In the Netherlands *L. pallidicornis* was reared from samples of white and red clover, collected near Hemmen and Kapelle. Presumably the larvae fed on *D. gentneri*, *D. leguminicola*, *D. trifolii*, *Clinodiplosis* sp. and *Tricholaba trifolii*.

c. Species, whose life-history is unknown

Brachyneura squamigera (Winnertz 1853)

The Cecidomyid *Brachyneura squamigera* has not yet been found in the Netherlands, but it is recorded here for the sake of completeness.



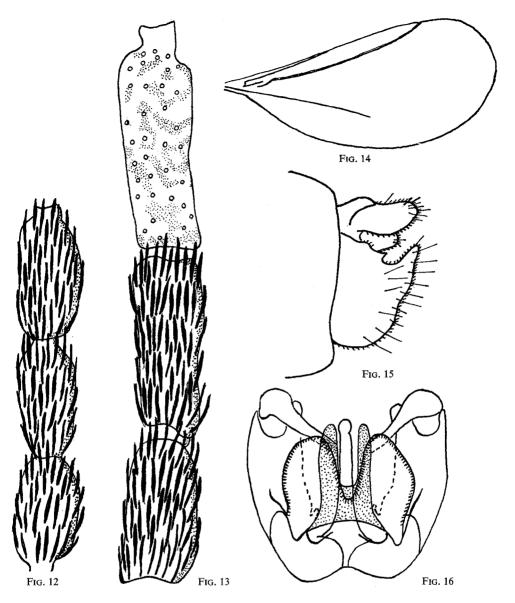
- Fig. 7. Wing of *Lestodiplosis pallidicornis*. × 33. *Vleugel van* Lestodiplosis pallidicornis,
- Fig. 8. Male genitalia of *Lestodiplosis pallidicornis*. × 200. *Copulatie-orgaan van* Lestodiplosis pallidicornis (mannetje).
- FIG. 9. Second and third flagellar segments of *Lestodiplosis pallidicornis* (male). × 215. Tweede en derde flagellumlid van Lestodiplosis pallidicornis (mannetje).
- Fig. 10. Second and third flagellar segments of Lestodiplosis pallidicornis (female). × 215. Tweede en derde flagellumlid van Lestodiplosis pallidicornis (wijfje).
- Fig. 11. Ovipositor of *Lestodiplosis pallidicornis*. × 215. *Legbuis van* Lestodiplosis pallidicornis.

WINNERTZ described this species in 1853. His description was amplified by EDWARDS (1937) and MILNE (1960). Adults were bred by BARNES in 1954 from samples of white clover from Oxfordshire, Shropshire and Cardiganshire in Great Britain. The antennae in both sexes have two basal segments and 10 flagellar segments. The flagellar segments are cylindrical with a short neck (figs. 12 and 13); they are covered with scales. Circumfila are present in the male, but they are difficult to observe. The wings show a simple venation (fig. 14). In the male genitalia the upper lamella is bilobed; the lower lamella is divided into two linear lobes (fig. 16). The ovipositor of the female is short and lamellate (fig. 15)

The life history of this species is still unknown. However, it does not cause damage of economic importance in white clover in England.

Gall midges of red clover

The gall midge species, occurring on red clover in the Netherlands, may be divided into: a. phytophagous species, b. inquilines, c. predators, d. species whose life-history is unknown.



- Fig. 12. Ventral view of first three flagellar segments of *Brachyneura squamigera* (female). After MILNE. × 360. Ventraal aanzicht van de drie eerste flagellumleden van Brachyneura squamigera (wijfje). Naar Milne.
- Fig. 13. Ventral view of first three flagellar segments of Brachyneura squamigera (male); scales removed from third segment to show insertions of circumfila (circles) and points of insertion of scales (dots). After MILNE. × 360.
 Ventraal aanzicht van de drie eerste flagellumleden van Brachyneura squamigera (mannetje); van het derde lid zijn de schubben weggelaten om de inplantingen van de zintuigharen (kringetjes) en de schubben (puntjes) te tonen. Naar Milne.
- Fig. 14. Wing of Brachneura squamigera, denuded of scales. After Milne. \times 60. Vleugel van Brachyneura squamigera, waarvan de schubben zijn weggelaten. Naar Milne.
- Fig. 15. Ovipositor of *Brachyneura squamigera* (female), lateral view. After MILNE. × 105. Lateraal aanzicht van legbuis van Brachyneura squamigera (wijfje). Naar Milne.
- Fig. 16. Male genitalia of Brachyneura squamigera. After Milne. × 360. Copulatie-orgaan van Brachyneura squamigera (mannetje). Naar Milne.

a. Phytophagous species

Dasyneura leguminicola (Lintner 1879) syn. trifolii Lintner (1879), flosculorum Kieffer (1890)

The species was described by LINTNER (1879) under the name *Cecidomyia leguminicola* from red clover in the U.S.A. The males are about 2 mm long and red-brown coloured. The females are bright-red and 2.5 to 3 mm long. The morphology of this gall midge agrees very closely with that of *Dasyneura trifolii* and *D. gentneri*, but as a rule the adults of *D. leguminicola* are larger. The male antennae have two basal segments and 14 or 15 flagellar segments (fig. 19); the females have 13 or 14 flagellar segments (fig. 20).

The male genitalia and the ovipositor of the female have already been discussed under *D. gentneri* (see p. 164). The wings are well developed and show a simple venation (fig. 23). The mature larvae are pink; the sternal spatula has also been discussed under *D. gentneri* (see p. 164). Figure 17a shows the anal segment of the mature larva.

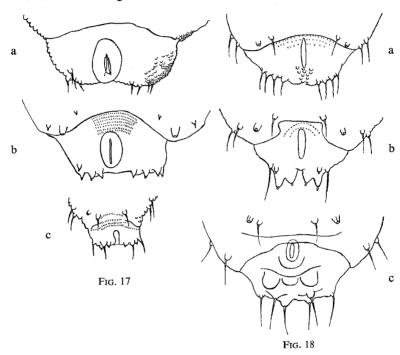


Fig. 17. Anal segments of larvae of / Achterlijfsegmenten van larven van:

- a. Dasyneura leguminicola (Lintner) × 80.
- b. Tricholaba trifolii Rübs. × 80.
- c. Isodiplosis deutera Milne. (After MILNE / naar Milne) \times 80.

Fig. 18. Anal segments of larvae of / Achterlijfsegmenten van larven van:

- a. Phaenobremia aphidivora (Rübs.) × 80.
- b. Clinodiplosis leguminicola Milne. (After MILNE / naar Milne) × 80.
- c. Lestodiplosis trifolii Barnes. × 80.

In general *D. leguminicola* starts emerging in the first half of May. The females always deposit their eggs on green heads of red clover (Plate 3B). The length of the egg stage lasts 3 to 4 days. After hatching, the young pale yel-

PLATE 1



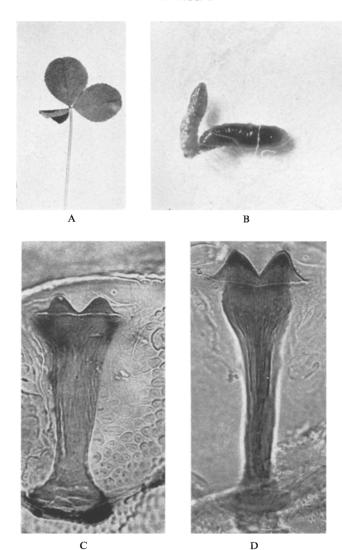


В

- A Lamp glass depot with water to collect mature gall midge larvae from infested clover flower heads.
 - Lampeglasdepot met water om volwassen galmuggen uit aangetaste klaverbloemen te verzamelen.
- B Lamp glass depot with peat, in which mature gall midge larvae can pupate.

 Lampeglasdepot met turfmolm om volwassen galmuglarven te laten verpoppen.

PLATE 2



- A Leaf galls on white clover, caused by the larvae of Dasyneura trifolii.

 Bladgallen op witte klaver, veroorzaakt door de larven van Dasyneura trifolii.
- B Larva of Lestodiplosis pallidicornis, sucking a larva of Dasyneura leguminicola. $8 \times .$ Larve van Lestodiplosis pallidicornis, die een larve van Dasyneura leguminicola leegzuigt.
- C Breastbone of *Dasyneura gentneri* larva. × 300. *Borststaafje van de larve van* Dasyneura gentneri.
- D Breastbone of *Dasyneura leguminicola-*larva. × 300. *Borststaafje van de larve van* Dasyneura leguminicola.

PLATE 3





В





D

- A Flower heads of red clover, infested by *Tricholaba trifolii*-larvae. Bloemhoofdjes van rode klaver, aangetast door de larven van Tricholaba trifolii.
- B Green head of red clover selected for oviposition by Dasyneura leguminicola.

 Groen bloemhoofdje van rode klaver, uitgezocht voor eiafzetting door Dasyneura leguminicola.
- C Normally developed flower head of red clover. Normaal ontwikkeld bloemhoofdje van rode klaver.
- D Flower head of red clover infested by Dasyneura leguminicola larvae.

 Bloemhoofdje van rode klaver, aangetast door de larven van Dasyneura leguminicola.

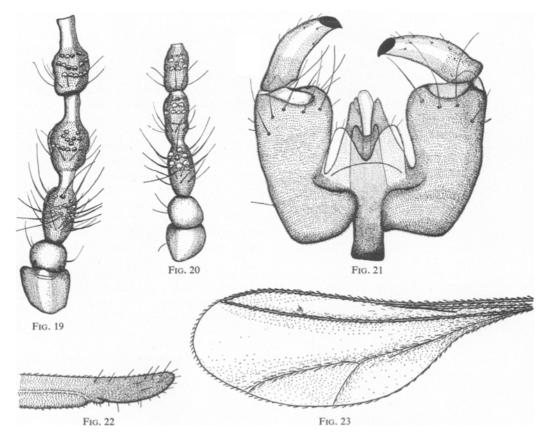


Fig. 19. First three flagellar segments of *Dasyneura leguminicola* (male). × 215. *Drie eerste flagellumleden van* Dasyneura leguminicola (mannetje).

- Fig. 20. First three flagellar segments of *Dasyneura leguminicola* (female). × 215. *Drie eerste flagellumleden van* Dasyneura leguminicola (wijfje).
- Fig. 21. Male genitalia of *Dasyneura leguminicola*. × 430. *Copulatie-orgaan van* Dasyneura leguminicola (*mannetje*).
- Fig. 22. Ovipositor of *Dasyneura leguminicola* (female). × 287. *Legbuis van* Dasyneura leguminicola (wijfje).
- Fig. 23. Wing of Dasyneura leguminicola. × 33. Vleugel van Dasyneura leguminicola.

lowish larva enters the undeveloped floret and starts feeding upon the ovary. The floret does not develop normally and remains closed. An infested flower head can easily be recognized by its irregular blossoming (Plate 3D); a head free from midge infestation blossoms uniformly (Plate 3C). Breeding experiments showed that the first symptoms of infestation can be noticed 15 to 18 days after oviposition.

The mature larvae migrate from the flower heads and pupate in the soil within a silken cocoon. The pupal stage lasts 11 to 16 days. Usually two com-

plete generations can develop annually. The larvae of the last generation hibernate in the soil and pupate in the following spring. Our observations showed that the larval stage of the first generation usually takes 16 to 33 days and that of the second generation 291 to 356 days. Wehrle (1929) states that a number of larvae of the first generation do not transform to adults until the following spring. Our investigations confirmed this phenomenon so that the larval stage of the first generation may sometimes last about a year.

D. leguminicola is recorded by Wehrle (1929) from Trifolium pratense and T. hybridum in the U.S.A. MILNE (1960) reared this species from red clover in England. He did not succeed in breeding it on white clover. Markkula (1959) reared D. leguminicola from red clover in Finland. Barnes (1946) records that this Cecidomyid also occurs in Canada and Norway and that it is reported under the name D. flosculorum from Central-Europe, Sweden and England.

In the Netherlands we collected *D. leguminicola* from red clover near Barneveld, Beerta, Bennekom, Breedebroek, Gronsveld, Heel, Hemmen, Herveld, Hoeven, Keer, Maasbracht, Mensingeweer, Oudesluis, Schore, Ulrum, Wageningen, Wessem and Wieringewerf. Near Wieringewerf it was also sampled from Alsike clover (*T. hybridum*).

Ir. M. J. F. KOOPMAN (Government Seed Testing Station) and Dr. B. K. BOOM (Institute of Horticultural Plant Breeding) kindly placed seeds of several Trifolium species at our disposal. Thus it was possible to undertake breeding experiments on Trifolium agrarium L., T. alexandrinum L., T. arvense L., T. campestre Schreb., T. diffusum Ehrh., T. fragiferum L., T. glomeratum L., T. hybridum L., T. incarnatum L., T. lappaceum L., T. parviflorum, T. pratense L., T. repens L. and T. squarrosum L. It appeared that oviposition took only place in the green flower heads of T. alexandrinum, T. fragiferum, T. glomeratum, T. hybridum and T. pratense L. The eggs hatched and the larvae developed normally to adults.

Field observations have shown that the life cycle of *D. leguminicola* fits well into the cultivation of red clover in the Netherlands. Commonly a seed crop is cut twice a year and the seed is gathered from the second cut. The crop is cut for the first time about mid June. At this time the larvae of the first gall midge generation are already full-grown and they have migrated from the flower heads to pupate in the soil. When the second flight period sets in, the seed crop is susceptible to gall midge infestation again. The second cut takes place when most full-grown larvae of the second gall midge generation have already left the flower heads.

Some of these larvae partly develop into midges in the same year. Some of the first emerged females can still oviposit in the flower heads before harvesting is finished, but the larvae have no opportunity to complete their development.

Several authors have reported injury of economic importance caused by *D. leguminicola* in Europe, the U.S.A. and Canada. LINTNER recorded in 1879 that serious depredation was caused upon clover-seed in Western New York. In Seneca Count some fields kept for seed proved not to be worth the cutting.

LOCHHEAD estimated in 1903 the average total value of clover seed produced in Ontario for one year at two million dollars. The loss caused by *D. leguminicola* was at least half a million or, on the average, a quarter of the crop. FLETCHER (1903) considered the Cecidomyid as the most destructive enemy of seed crops in Canada. Jarvis reported in 1906 that *D. leguminicola* every year

caused an immense depreciation in the yield of seed. He estimated that the loss in Ontario ranged in various localities, all the way from 25 to 75 per cent. MARKKULA (1959) studied this species in Finland from 1956 to 1958 and states that it is of economic importance in the production of red clover seed. Details concerning the life history and economic importance are further given in the papers of Wehrle (1929), Metcalfe (1933), Barnes (1946) and Milne (1960).

Up till now no information was available as to whether *D. leguminicola* is of economic importance for seed crops in the Netherlands. Four fields, kept for seed, were therefore examined in August 1958 by collecting three samples of 500 flower heads from each field. The samples were taken all over the fields by persons who were not acquainted with the subject and the type of damage. From each sample the number of infested flower heads was counted and percentaged. We did not count the third sample when no important differences between the first two samples had been observed. Table 1 shows the results.

Table 1. Percentage of infested flower heads on some seed crops of red clover in the Netherlands.

Percentage aangetaste bloemhoofdjes op enige zaadpercelen van rode klaver in Nederland

Locality Plaats	Date Datum	Soil Grondsoort	Sample nr. Monster no.	Percentage of flower heads attacked Percentage aangetaste bloemhoofdjes
Hemmen	15-8-1958	clay / klei	1	3.6
		- '	2	2.4
		ļ	3	2.4
Barneveld	18-8-1958	sand / zand	1	1.4
			2	1.4
		1.	3	not counted / niet geteld
Wessem	20-8-1958	clay / <i>klei</i>	1	0.8
			2	1.2
			3	not counted / niet geteld
Ulrum	22-8-1958	clay / klei	1	0.4
			2	0.2
			3	not counted / niet geteld

An examination of 100 normal blossoms and 100 heavily infested flower heads collected from a seed crop near Hemmen showed that actually the latter were not completely excluded from seed-gathering. The 100 normal flower heads contained 8790 well developed florets; this is an average of 88 per flower head. The 100 heavily infested ones contained 2805 normal florets; this is an average of 28 per flower head. This means that about 33 % of the florets in the infested flower heads did take a part in the process of seed formation and this fact reduces partially the very small seed loss as suggested in table 1.

As the damage caused by *Tricholaba trifolii* cannot be distinguished in the field from that of *D. leguminicola*, the seed loss is partially caused by the first species too. During our investigations it sometimes proved very difficult to collect the number of 100 flower heads, needed for one sample. Mr. H. EELDERINK (Nationale Coöperatieve Aan- en Verkoopvereniging voor de Landbouw "Centraal Bureau", G.A.), who is familiar with the problems concerning the breeding of clover seed in the Netherlands, informed us that he had never

observed a remarkable infestation by *D. leguminicola* on the seed crops. This is presumably due to the rather wide crop rotation in our country.

On account of the above mentioned investigations and facts we conclude that the gall midge *Dasyneura leguminicola* is of no economic importance in the production of red clover seed in the Netherlands.

Contarinia sp.

Barnes (1954) records that he found what were thought to be *Contarinia* larvae in one sample of red clover from Sussex, England. His diagnosis was based on the colour and the shape of the anal segment as well as the fact that they "jumped" on the laboratory bench. MILNE (1960) mounted and examined Barnes' material and confirmed that they were indeed *Contarinia* larvae. Figs. 24g and 25 show the sternal spatula and the anal segment of this larva.

Species of the genus *Contarinia* are known as phytophagous. As the *Contarinia* larvae were only found in one sample in England, it is still uncertain whether they are of economic importance. Up till now no *Contarinia* larvae have been collected from red clover flower heads in the Netherlands. However, they are only mentioned for the sake of completeness here.

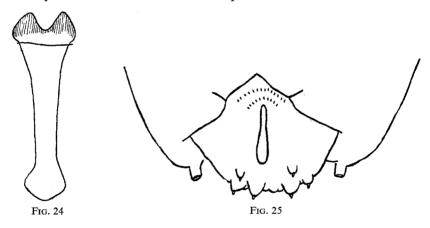


Fig. 24. Breastbone of Contarinia sp. After Milne. × 240. Borststaafje van Contarinia sp. Naar Milne.

Fig. 25. Anal segment of larva of Contarinia sp. After Milne. × 160. Laatste achterlijfsegment van larve van Contarinia sp. Naar Milne.

Tricholaba trifolii (Rübsaamen 1917)

Numerous Cecidomyids of the genus *Tricholaba* were reared from the flower heads of red clover. Their identification, however, was rather difficult. The type species, *T. trifolii*, was described by RÜBSAAMEN (1917) from leaf galls, caused by *Dasyneura trifolii* on white clover in Germany in 1917. At the same time he described the female of *T. similis*.

The yellow red larvae of the latter species live as inquilines in the galls, caused by *Dasyneura viciae* Kieffer. The host plant of *D. viciae* was not recorded in this description. The morphological differences between both species

are very small and RÜBSAAMEN thought that *T. similis* might be a form of *T. trifolii*. MILNE (1960) records that BARNES in 1929, 1930 and 1954 bred adults of the genus *Tricholaba* from red clover samples from various counties in England. The same author mounted a number of these specimens and bred new material from numerous other samples between 1955 and 1957. An examination of this material led to the conclusion that the British species differed in habitat and morphology from *T. trifolii* and *T. similis* and MILNE therefore described his species as new under the name *Tricholaba barnesi* Milne (1960). Another, undescribed species of the genus *Tricholaba* is represented in the BARNES collection; it was reared from the galls of *Dasyneura lupulinae* Kieffer on *Medicago lupulina*. MILNE separated the *Tricholaba* species under discussion as follows:

Species	Female	Male
T. trifolii	Node of 3rd flagellar segment $3\frac{1}{2}$ to 4 times longer than the stem	3rd flagellar segment $10\frac{1}{2}$ to 11 times the diameter of the proximal stems
T. barnesi	Node of 3rd flagellar segment 5 times longer than the stem	3rd flagellar segment 9 times the diameter of the proximal stems
T. sp.	No female examined	3rd flagellar segment 11 times the diameter of the proximal stems

An exact measurement, carried out on 50 males of our *Tricholaba*-material, showed us that one specimen agreed with *T. barnesi*, eleven specimens with *T. trifolii* and two specimens with the undescribed species from *Medicago lupulina*. The remaining 36 specimens showed all kinds of intermediate forms and did not fit in Milne's key for separation. Investigations of 20 females showed that 5 specimens agreed with *T. trifolii*; the characteristics of *T. barnesi* were not observed at all. In two specimens the node of the 3rd flagellar segment was 1.3 and 1.5 times longer than the stem respectively. This proportion varied in the 13 remaining specimens from 3.1 to 4.8, making an exact identification impossible.

It is clear that only a small number of our specimens could be identified and that most of them showed the characteristics of *Tricholaba trifolii*. As neither MILNE, nor ourselves have been able to examine the biological interrelationships of these *Tricholaba* species by further breeding experiments and testing of host plants, we consider it advisable for the present to identify our *Tricholaba* species from red clover flower heads as *Tricholaba trifolii*, because this name has priority.

The males are 2 to 2.5 mm long. The abdomen is red. The antennae consist of two basal segments and 12 flagellar segments. The first two flagellar segments are fused. The third flagellar segment is binodose (fig. 27). It has two whorls of hairs and three whorls of circumfila. The nodes and the stems are provided with microtrichia. The abdomen ends in a hypopygium, from which the upper and lower lamellae are deeply incised and bilobed. The basal and distal clasp segment are well developed and covered with small setae and microtrichia (fig. 26).

The females are 2.5 to 3 mm long. The abdomen is orange red. The antennae have two basal segments and 12 flagellar segments, of which the first two are fused. The flagellar segments are cylindrical; they end in a short stem with microtrichia (fig. 29). Each node bears two whorls of applied circumfila and two whorls of hairs. The abdomen ends in a lamelliform ovipositor

with two elongate, oval lamellae and a small ventral lobe; they are covered with setae and microtrichia (fig. 30). The wings of this species are elongate (fig. 28).

The feeding habits of *T. trifolii* are very similar to those of *Dasyneura leguminicola*. The females select the green flower heads of red clover for oviposition. The eggs are orange red. The larvae live at the expense of the developing young seeds. The infested flower heads show an irregular blossoming and the damage can hardly be distinguished in the field from that caused by *D. leguminicola* (Plate 3A). The full-grown larvae are about 3 mm long. Their colour is yellow ochre. The sternal spatula has a long narrow stem and is bilobed apically (fig. 1a). Figure 17b shows the anal segment of the larva. The larvae pupate in the soil within a cocoon. There are at least two generations annually.

Samples, taken from a seed field near Hemmen, showed that *D. leguminicola* and *T. trifolii* can occur at the same time in the same crop and that the latter species is more common than was thought. In 1956 we examined the larvae found in all samples collected from this field and 9.9 per cent were *T. trifolii*. This percentage increased to 51.4 in 1957 and to 98.1 in 1958. In 1959 a sample was collected from wild red clover along a ditch near this seed field. All the larvae belonged to *T. trifolii*.

In the Netherlands we collected *T. trifolii* from red clover seed crops near Barneveld, Hemmen, Keer and Wessem. It was sampled from wild red clover along road ditches near Heel, Hemmen and Nuth.

As the damage caused by *T. trifolii* cannot be distinguished from that of *D. leguminicola* in the field, we conclude that neither species is of economic importance in the production of red clover seed in the Netherlands.

b. Inquilines

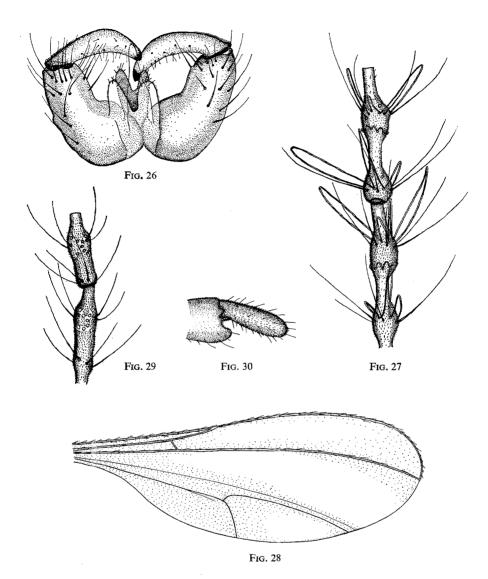
Clinodiplosis sp. and Clinodiplosis leguminicola Milne (1960)

The larvae of the genus *Clinodiplosis* commonly live as inquilines in plant galls caused by Cecidomyids and other insects. They may also be found in the autumn in deceasing parts of plants. Only one species is known as a gall maker. The mature larvae are 2 to 2.5 mm long and almost orange in colour.

The Clinodiplosis species are morphologically very similar and difficult to separate. One of the most useful characteristics is the upper lamella in the male genitalia. Two main types have been described in the literature. In Clinodiplosis cilicrus Kieffer (1889) the upper lamella is bilobed; each lobe is incised obliquely on the inner side (fig. 34). In C. coriscii Kieffer (1896) each lobe is divided into two smaller lobes apically (fig. 32).

Both main types were found in male midges bred from red clover samples collected near Hemmen, and we have identified them for the present as *Clinodiplosis* sp. Not only the main types, but all kinds of intermediate forms are described under several names in the literature, and all types may be found in material from the same origin, which suggests that many species are synonymous.

The midges are slender and delicately built. The males are 2 to 2.5 mm long. The antennae have two basal segments and 12 flagellar segments. The third flagellar segment is binodose (fig. 33). The basal node is subglobular and bears one whorl of regular circumfila and one whorl of hairs. The distal node is elongated and slightly constricted medially; it bears two whorls of circumfila and one whorl of hairs. The first two flagellar segments are fused. Figures



- Fig. 26. Male genitalia of *Tricholaba trifolii*. × 200. *Copulatie-orgaan van* Tricholaba trifolii (*mannetje*).
- Fig. 27. Second and third flagellar segments of $Tricholaba\ trifolii\ (male). imes 215.$ Tweede en derde flagellumlid van Tricholaba trifolii (mannetje).
- Fig. 28. Wing of *Tricholaba trifolii*. × 25. *Vleugel van* Tricholaba trifolii.
- Fig. 29. Second and third flagellar segments of *Tricholaba trifolii* (female). \times 215. Tweede en derde flagellumlid van Tricholaba trifolii (wijfje).
- Fig. 30. Ovipositor of *Tricholaba trifolii*. × 215. *Legbuis van* Tricholaba trifolii.

32 and 34 show the slender male genitalia with the variable upper lamella. The female antennae have two basal and 12 flagellar segments of which the first two are fused. The flagellar segments are cylindrical with a distinct stem and slightly constricted medially (fig. 36); they bear two whorls of applied circumfila and two whorls of hairs. The abdomen ends in a lamelliform ovipositor, covered with setae and microtrichia (fig. 37). The wings show a simple venation (fig. 35). Figure 1d shows the sternal spatula and figure 18b shows the anal segment of the larva.

The feeding habit of the larvae is still unknown. They neither disturb the normal development of their host, nor destroy the young seeds in the florets and therefore they are of no economic importance in the production of red clover seed in the Netherlands.

MILNE (1960) bred a *Clinodiplosis* species from red and white clover which did not agree with any of the described species or the undescribed species in the BARNES collection. Therefore he described it under the name *Clinodiplosis leguminicola* sp.n. The upper lamella in the male genitalia (dorsal plate) is deeply cleft, the lobes extending to smaller lobes apically on outer side only (fig. 43). *C. leguminicola* has not yet been found in the Netherlands and is recorded here for the sake of completeness.

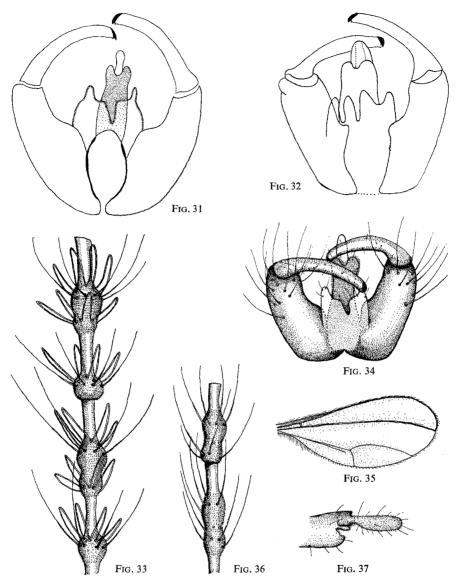
c. Predators

Phaenobremia aphidivora (Rübsaamen 1891)

This species was reared from red clover samples collected near Hemmen. The larvae prey upon aphids which live on the clover plants. However, the influence of this Cedicomyid on the aphid population in our seed crops is not yet exactly known. The eggs of P. aphidivora are only laid on plants, infested by aphids; they are elongated elliptical, 0.3 mm long and orange red. The larvae hatch after some days; they are very active and crawl about the plant till they meet an aphid. Then they transfix the skin of their victim with their mouth parts, preferring the joints between the legs, the antennae or the abdomen. After this, they start sucking the body fluid. When there is plenty of food and when the weather conditions permit the larvae may be full-grown within a week. Pupation takes place in the upper surface of the soil within a dark brown cocoon. The pupal stage lasts about two or three weeks. Several generations develop annually. It is very curious that the aphids seldom struggle when they are attacked by the gall midge larvae. This is probably due to a poison, excreted by the larvae when they transfix the aphid skin, as is supposed by Voukassovitch (1925). A strong preference was shown for the small immature aphids during the feeding period. They become completely shrivelled after a gall midge attack. The later instars show a more normal appearance, and they are seldom sucked completely. In both cases the sucked bodies can be found on the plant, hanging by their rostrum.

P. aphidivora is a very polyphagous species with a wide geographical distribution. In the Netherlands it is reported by Nijvelot (1954, 1955) as a predator in the field of Aphis gossypii Glover, A. urticata Gmelin, Brevicoryne brassicae (L.), Cavariella pastinacae (L.), Chaitophorus beuthani (Börner) and Hvalopterus pruni (Geoffr.).

Under experimental conditions it developed on Aphis fabae Scop., A. pomi Geer, Brevicoryne brassicae, Chaitophorus populeti (Panzer) and Pentatrichopus



- Fig. 31. Male genitalia of Clinodiplosis leguminicola. After MILNE. × 180. Copulatie-orgaan van Clinodiplosis leguminicola (mannetje). Naar Milne.
- Fig. 32. Male genitalia of *Clinodiplosis* sp. in the flower-heads of red clover. × 200.

 Copulatie-orgaan van Clinodiplosis sp. (mannetje) van de bloemhoofdjes van rode klaver.
- Fig. 33. Second and third flagellar segments of Clinodiplosis sp. (male). \times 215. Tweede en derde flagellumlid van Clinodiplosis sp. (mannetje).
- Fig. 34. Male genitalia of *Clinodiplosis* sp. in the flower-heads of red clover. × 200. *Copulatie-orgaan van Clinodiplosis sp. (mannetje) van de bloemhoofdjes van rode klaver.*
- Fig. 35. Wing of *Clinodiplosis* sp. \times 25. *Vleugel van* Clinodiplosis sp.
- Fig. 36. Second and third flagellar segments of Clinodiplosis sp. (female). \times 215. Tweede en derde flagellumlid van Clinodiplosis sp. (wijfje).
- Fig. 37. Ovipositor of *Clinodiplosis* sp. × 215. *Legbuis van Clinodiplosis sp.*

fragaefolii (Cock.). The same author records in 1957 that, in England, P. aphidivora has been reared from Acyrthosiphon pisum (Harris), Aphis pomi, A. fabae, A. gossypii Glover, Brachycaudus cardui (L.), B. helichrysi (Kltb.) Brevicoryne brassicae, Cryptomyzus ribis (L.), Hyalopterus pruni, Macrosiphoniella sanborni (Gil.), Myzus ornatus Laing and M. persicae (Sulz.). MILNE (1960) records that he succeeded in breeding P. aphidivora on Aphis fabae. Hyperomyzus lactucae (L.), Macrosiphum rosae (L.), Megoura viciae Buckt., Microlophium evansi (Theob.) and Myzus cerasi (F.) under experimental conditions. According to NIJVELDT (1957), midges were bred from larvae which fed on A. fabae, Myzus persicae and Periphyllus acericola (Wlk.). In the same paper it is reported that in Germany, where P. aphidivora was originally collected by RÜBSAAMEN (1891) from an unidentified aphid on Heracleum sphondvlium L., this Cecidomvid has also been reared from Brevicorvne brassicae. In Israël P. aphidivora has been collected from Aphis nerii Fonsc. and from Hyalopterus pruni (NIJVELDT, 1957). In the I.P.O. collections there are specimens of P. aphidivora sent by Dr. B. MAMAJEV (Russia) and Dr. P. STARY (Czechoslovakia). The Russian material was bred from Aphis farinosa Gmelin; the Czech specimens were collected in the field from colonies of Acyrthosiphon malvae (Mosley), A. fabae, A. idaei v. d. G., A. farinosa, A. intybi Koch, A. pomi, A. salviae Wlk., A. sambuci L., A. urticata, Brachycaudus sp., B. cardui, B. rumexicolens (Patch.), Dactynotus sp., D. cichorii (Koch.) Dysaphis sp., D. devecta (Wlk.), D. plantaginea (Pass.), Eriosoma patchae B. et Bl., Hayhurstia atriplicis (L.), Hyadaphis sp., Hyalopterus pruni, Hyperomyzus lactucae (L.), Lipaphis ervsimi (Kltb.), Macrosiphoniella sp., M. absinthii (L.), Metopeurum fuscoviride Stroyan, Microlopium evansi, Myzus sp., M. cerasi, Nasonovia nigra H.R.L., N. ribis-nigri (Mosley), Protaphis sp. and of unidentified aphids on Artemisia vulgaris L. and Anagallis coerulea (Schreb.) Vollm.

The mature larvae of *P. aphidivora* are 2.5 to 3 mm long. The colour is yellowish-orange. Figures 1c and 18a show the sternal spatula and the anal segment. The males are about 2 mm long. The abdomen is orange brown. The antennae have two basal segments and 12 flagellar segments; the first two flagellar segments are fused. The third flagellar segment is binodose. The basal node is globular and bears one irregular whorl of applied circumfila and one irregular whorl of hairs (fig. 39); the distal node is cylindrical and bears one irregular whorl of applied circumfila and one regular and one irregular whorl of hairs. The stems are without microtrichia. The male hypopygium is strongly haired (fig. 40). The lower lamella is cordate, the upper lamella is divided into two triangular lobes.

The females are slightly longer than the males and more pink in colour. The antennae consist of two basal segments and 12 flagellar segments, of which the first two are fused. The flagellar segments are cylindrical with a distinct neck (fig. 38). They bear two whorls of applied circumfila and two whorls of strong hairs with smaller hairs medially. The ovipositor is lamelliform and covered with hairs and microtrichia (fig. 41). The wings show the known

simple venation (fig. 42).

It is not exactly known on which aphid species the *Phaenobremia* larvae fed on the flower heads in the Netherlands. It is clear, however, that their feeding habits are of no economic importance in the production of red clover seed in our country.

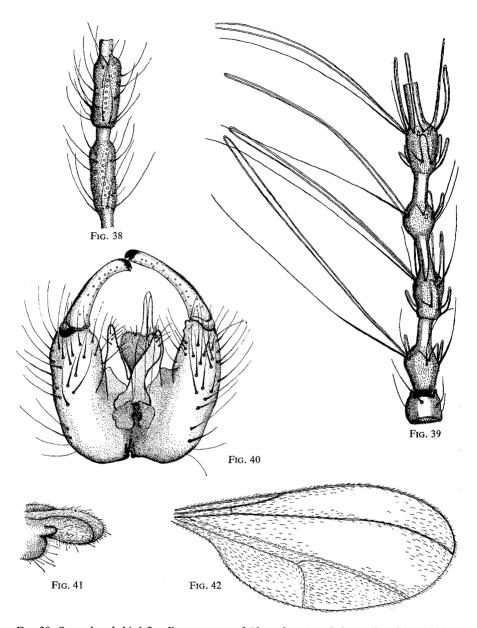


Fig. 38. Second and third flagellar segments of *Phaenobremia aphidivora* (female). \times 215. Tweede en derde flagellumlid van Phaenobremia aphidivora (wijfje).

- Fig. 39. Two first flagellar segments of *Phaenobremia aphidivora* (male). \times 215. Twee eerste flagellumleden van Phaenobremia aphidivora (mannetje).
- Fig. 40. Male genitalia of *Phaenobremia aphidivora*. × 200. Copulatie-orgaan van Phaenobremia aphidivora (mannetje).
- Fig. 41. Ovipositor of *Phaenobremia aphidivora*. × 215. *Legbuis van* Phaenobremia aphidivora.
- Fig. 42. Wing of *Phaenobremia aphidivora*. \times 25. *Vleugel van* Phaenobremia aphidivora.

Species whose life history is unknown

Ametrodiplosis sp. (Diplosis Loew 1888, Clinodiplosis Rübsaamen 1895, Contarinia Kieffer 1896, Ametrodiplosis Rübsaamen 1911, Löwodiplosis Kieffer 1912b, Cyrtodiplosis Kieffer 1912b, Ametrodiplosis Rübsaamen 1917)

Male specimens of the genus Ametrodiplosis were reared from red clover flower heads, collected near Hemmen. They are characterized by a conspicuous thickening at the base of the wing (fig. 45). The midges are 1.8 to 2 mm long and orange in colour. The antennae have two basal segments and 12 flagellar segments, from which the first two are fused. The third flagellar segment is binodose (fig. 43). The basal node is subglobular, bearing one whorl of circumfila and one whorl of hairs; the distal node is elongated, and also bears one whorl of circumfila and one whorl of hairs. The male genitalia are very similar to the genus Clinodiplosis (fig. 44). The wing venation is rather simple (fig. 45).

We did not succeed in breeding females. Fig. 46 shows the first two flagellar segments of a female of *Ametrodiplosis thalictricola* Rübs.

MAMAJEV published in 1961 a paper on the genus Ametrodiplosis. Some known species are discussed and some new species are described in this publication. The species A. viciae Mamajev lives in the unopened flowers of Vicia cracca L.; the larvae pupate in the soil. The larvae of A. auripes (F. Loew) live in subterranean, globular bud galls on Galium mollugo L.; they pupate in the galls. The larvae of A. medialis Mamajev live together with those of Dasyneura crataegi Winn. in terminal rosettes of leaves on Crataegus oxyacantha L.; pupation takes place in the soil. A. thalictricola Rübs. deforms the fruits of Thalictrum flavum L.; the mature larvae pupate in the soil. The species A. crassinerva lives in the flowers of Stachys silvatica L.

Some of our specimens were sent to Dr. Mamajev for examination; he observed a close similarity between this species and A. viciae. As no Ametro-diplosis species had been reared from red clover flower heads before, we identify our specimens for the present as Ametrodiplosis sp. As the life history of this species is still unknown it is impossible to say whether it is of any economic importance in the production of red clover seed in the Netherlands.

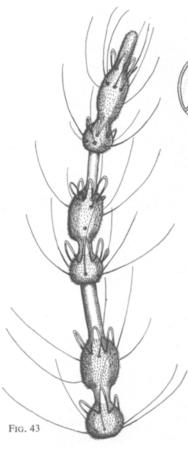
Campylomyza ormerodi (Kieffer, 1913)

Though this Cecidomyid is still unknown in the Dutch fauna, it is of interest to record it in this paper because many larvae have been found in diseased red clover plants in England. It was thought that *C. ormerodi* must be regarded as a primary agent. Barnes (1946), however, believes that this is still an open question because, according to Mac Dougall (1913), an eelworm, *Ditylenchus dipsaci* (Kühn), is almost invariably found in or about the diseased plants.

KIEFFER described this gall midge in 1913 under the name Amblyspatha ormerodi. It was later redescribed by EDWARDS (1938) as Campylomyza ormerodi.

Fig. 1f shows the sternal spatula of a mature larva. The male is about 2 mm long. The antennae have two basal segments and 12 flagellar segments. The flagellar segments have one node with a distinct long neck (fig. 49). The nodes are covered with long hairs and have some lamelliform sense organs on the distal part. The hypopygium (fig. 47) is firmly built and strongly haired. The upper and lower lamellae are broadly rounded.

The female is about 3.5 mm long. The antennae have two basal segments and 10 to 12 flagellar segments. Every flagellar segment has a short, compact node with a short neck (fig. 50). The



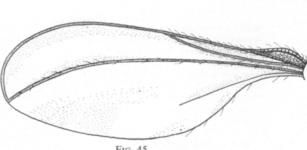
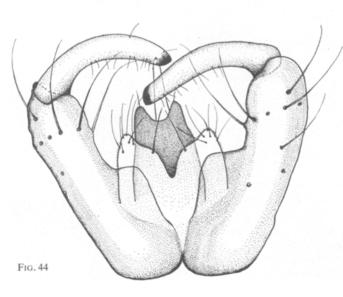
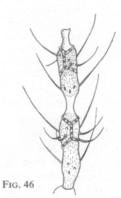


Fig. 45

- Fig. 43. Three distal flagellar segments of Ametro-diplosis sp. (male). × 215. Laatste drie flagellumleden van Ametrodiplosis sp. (mannetje).
- Fig. 44. Male genitalia of Ametrodiplosis sp. × Copulatie-orgaan van Ametrodiplosis sp. (mannetje).
- Fig. 45. Wing of *Ametrodiplosis* sp. \times 25. *Vleugel van* Ametrodiplosis *sp*.
- Fig. 46. First two flagellar segments of Ametrodiplosis thalictricola (female). After RÜB-SAAMEN. × 215. Twee eerste flagellumleden van Ametrodiplosis thalictricola (wijfje). Naar Rübsaamen.

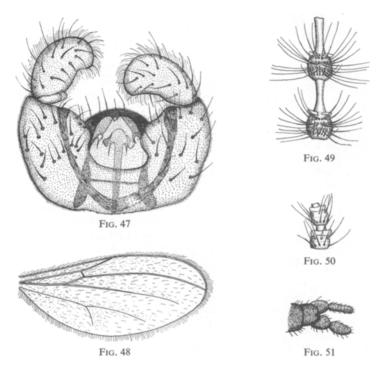




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nodes are provided with stout hairs and with a collar shaped sense organ distally. The abdomen ends in a lamelliform ovipositor (fig. 51); the lamellae are strongly haired. The wing venation is more complicated than in the other gall midge species recorded in this paper (fig. 48). It is characteristic of the subfamily of the Lestremiinae. According to BARNES (1946), C. ormerodi has only been found in England on red clover (Trifolium pratense) and on Kidney Vetch (Anthyllis vulneraria L.).

The figures are drawn by the author from specimens, presented by the late Dr. BARNES.



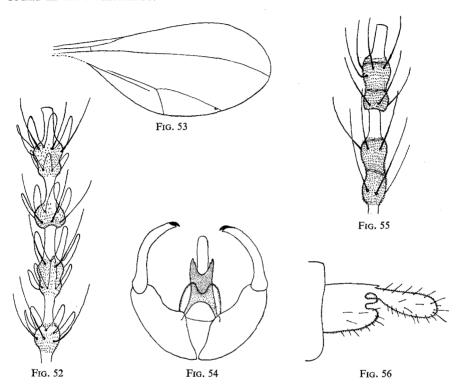
- Fig. 47. Male genitalia of *Campylomyza ormerodi*. × 215. *Copulatie-orgaan van* Campylomyza ormerodi (*mannetje*).
- Fig. 48. Wing of Campylomyza ormerodi. \times 25. Vleugel van Campylomyza ormerodi.
- Fig. 49. Second and third flagellar segments of Campylomyza ormerodi (male). × 215. Tweede en derde flagellumlid van Campylomyza ormerodi (mannetje).
- Fig. 50. Second and third flagellar segments of Campylomyza ormerodi (female). \times 215. Tweede en derde flagellumlid van Campylomyza ormerodi (wijfje).
- Fig. 51. Ovipositor of Campylomyza ormerodi. \times 215. Legbuis van Campylomyza ormerodi.

Giardomyia britannica Milne (1960)

The genus Giardomyia was described by Felt in 1907; he classed seven species in this genus with photopila as type. These species occur only in the U.S.A. Milne, however, reared a species of this genus from red and white clover in England and described it under the name Giardomyia britannica. Presumably only one generation develops annually. Figs. 53, 54 and 56 show the wing, the male genitalia and the lamelliform ovipositor respectively. Fig.

52 shows the second and third flagellar segments of the male and fig. 55 the second and third flagellar segments of the female.

Little is known concerning the biology of G. britannica. It is possible that the larvae may cause galls on the clover flower heads but it seems unlikely that the species is of economic importance. Up till now G. britannica has not been found in the Netherlands.



- Fig. 52. Second and third flagellar segments of Giardomyia britannica (male). After MILNE. × 780.

 Tweede en derde flagellumlid van Giardomyia britannica (mannetje). Naar Milne.
- Fig. 53. Wing of *Giardomyia britannica*. After Milne. × 30. *Vleugel van Giardomyia britannica*. *Naar Milne*.
- Fig. 54. Male genitalia of Giardomyia britannica. After Milne. × 180. Copulatie-orgaan van Giardomyia britannica (mannetje). Naar Milne.
- Fig. 55. Second and third flagellar segments of Giardomyia britannica (female). After Milne. × 180.

 Tweede en derde flagellumlid van Giardomyia britannica (wijfje). Naar Milne.
- Fig. 56. Ovipositor of Giardomyia britannica. After Milne. × 105. Legbuis van Giardomyia britannica. Naar Milne.

Isodiplosis deutera Milne (1960), f. n. sp.

Some midges belonging to the genus *Isodiplosis* were reared from samples of red clover collected near Hemmen. This genus was described by RÜBSAAMEN in 1912 with *I. involuta* as type species. The larvae of *I. involuta* were found

in Germany in leaf rolls, caused by the Curculionid Byctiscus betulae (L.) (Rhynchites betuleti F.) on Vitis vinifera L. The Isodiplosis larvae lived in the leaf rolls together with the larvae of the Byctiscus and of the gall midge Clinodiplosis rhynchitou Rübs. Presumably the larvae of I. involuta fed on decomposition products or fungus spores.

MILNE (1960) reared an *Isodiplosis* species from the flower heads of red clover and Alsike clover in England during 1955 and 1956. He distinguished this species from *I. involuta* on the feeding habits and described it under the name *Isodiplosis deutera*. His type material, however, is rather scarce, viz. 3 male cotypes, 2 female cotypes and 2 larval slides which are all deposited in the Barnes collection. Our material is also rather scarce (5 males) and this prevents a comprehensive comparative study concerning the morphology of both species. In view of the close similarity between the morphology and the feeding habits we identified our specimens as *Isodiplosis deutera* Milne. This is the first record for the fauna of the Netherlands.

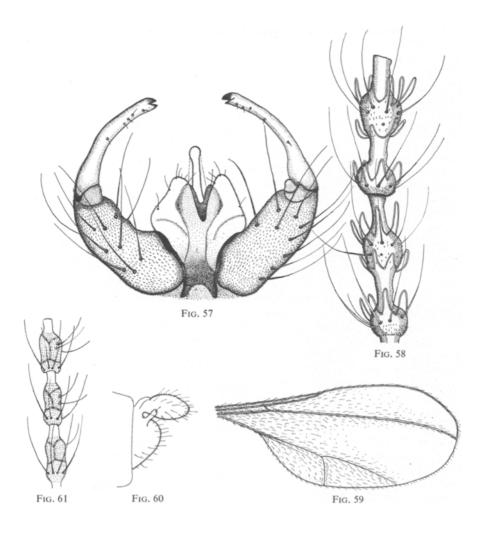
The mature larvae are 1 to 1.4 mm long. They are pale pinkish-orange in colour. Figures 1e and 17c show the sternal spatula and the anal segment. The males are about 1 mm long and pale orange in colour; the antennae have two basal segments and 12 flagellar segments. The first two flagellar segments are fused. The third flagellar segment is binodose (fig. 58). The basal node is subglobular and bears one whorl of circumfila and one whorl of regular hairs. The distal node is longer and slightly constricted medially; it bears two whorls of circumfila and one whorl of regular hairs. There are very few microtrichia on the dorsal surface of the basal node. The male genitalia are rather slenderly built (fig. 57). The upper and the lower lamellae are deeply incised; the lobes of the upper lamella are rather broad, the lobes of the lower lamella are slender and parallel.

The females are slightly longer than the males. They are very pale pinkish-orange in colour. The antennae are composed of two basal segments and 12 flagellar segments. The first two flagellar segments are fused. The third and the fourth are cylindrical with a distinct neck (fig. 61); they bear two whorls of applied circumfila joined laterally by a longitudinal series, a single basal whorl of hairs, and two or three tubercle-based hairs half way up the node on the ventral side. The ovipositor is short and lamelliform (fig. 60); it is composed of two oval lamellae and a smaller ventral lobe. The wings show a simple venation (fig. 59).

The feeding habit of *I. deutera* is not yet exactly known. It may also feed on decomposition products or fungus spores, since MILNE (1960) found numerous larvae in a sample of dead and decayed heads of Alsike clover collected in the field. He concluded that there are at least two generations, and possible more, in a year. It is unlikely that *I. deutera* is of any economic importance in the production of red clover seed in the Netherlands.

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- Fig. 57. Male genitalia of *Isodiplosis deutera*. × 430. *Copulatie-orgaan van* Isodiplosis deutera (mannetje).
- Fig. 58. Second and third flagellar segments of Isodiplosis deutera (male). \times 215. Tweede en derde flagellumlid van Isodiplosis deutera (mannetje).
- Fig. 59. Wing of *Isodiplosis deutera*. \times 25. *Vleugel van* Isodiplosis deutera.
- Fig. 60. Ovipositor of *Isodiplosis deutera*. After Milne. \times 105. *Legbuis van* Isodiplosis deutera. *Naar Milne*.
- Fig. 61. Third and fourth flagellar segments of *Isodiplosis deutera* (female). After Milne. × 180.

 Derde en vierde flagellumlid van Isodiplosis deutera (wijfje). Naar Milne.

economic importance of *D. leguminicola* and to Mr. F. Scheffel (I.P.O.) for taking the photographs, inserted in this paper. I am much indebted to Mr. D. Hille Ris Lambers (T.N.O., Bennekom) for the correct nomenclature of the aphids dealt with under *P. aphidivora*. Finally, special gratitude is due to Miss Margaret K. Arnold of Rothamsted Experimental Station, Harpenden, England, for her valuable advice in the use of the English language.

SAMENVATTING

In deze publikatie worden de resultaten vermeld van een onderzoek naar de galmugfauna van rode en witte klaver in Nederland. Laboratoriumproeven en veldwaarnemingen hebben uitgewezen, dat *Dasyneura gentneri* Pritchard, *D. leguminicola* (Lintner) en *Tricholaba trifolii* Rübs. zich voeden ten koste van het zich ontwikkelende klaverzaad. Hun optreden is echter niet van economische betekenis voor de teelt van klaverzaad in Nederland.

Dasyneura trifolii (F. Loew) verwekt bladgallen op witte klaver, maar door deze aantasting worden de planten niet in hun normale groei belemmerd. De larven van Lestodiplosis pallidicornis Kieffer en van Phaenobremia aphidivora (Rübs.) zijn roofvijanden; zij voeden zich respectievelijk met larven van allerlei andere galmugsoorten en met bladluizen, die op rode en witte klaver leven. De soorten Ametrodiplosis sp., Clinodiplosis sp. en Isodiplosis deutera Milne f.n.sp. leven in de bloemhoofdjes van rode klaver, maar hun juiste levenswijze is nog niet bekend. MILNE (1960) en BARNES (1946) vermelden van witte en rode klaver in Engeland nog de soorten Brachyneura squamigera (Winnertz), Campylomyza ormerodi (Kieffer), Clinodiplosis leguminicola Milne en Giardomyia britannica Milne. Deze soorten werden door ons in Nederland tijdens het onderzoek niet gevonden.

Deze publikatie vermeldt verder bijzonderheden inzake de levenswijze, de morfologie en de ontwikkelingscyclus van de behandelde galmugsoorten, terwijl ook aandacht is besteed aan hun economische betekenis voor de teelt van witte en rode klaver in Nederland

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