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Characteristics of galls formed by *Lipara pullitarsis* Doskočil & Chvála, 1971 (Diptera, Chloropidae) on common reed (*Phragmites australis* (Cav.) Trin. Ex Steud, 1841)

ABSTRACT

We studied galls formed by *Lipara pullitarsis* in the apical part of common reed stems, paying particular attention to the number and length of internodes that formed the basal part of each gall. *L. similis* galls were used only as a reference for the study of *L. pullitarsis* galls, as they were characterised by a uniform structural pattern and a shape similar to some galls produced by *L. pullitarsis*.

L. pullitarsis galls vary in shape. The species is found in conspicuous galls that are narrow at the base and have a wider apical part. It can also be found inside rod-shaped galls similar to those formed by *L. similis*. The shape of an *L. pullitarsis* gall is determined by the number and length of internodes that form its basal part, with the length of internodes III, IV and V being of the greatest significance.

Keywords: Diptera, *Lipara pullitarsis*, gall, *Phragmites australis*

INTRODUCTION

Four species of flies of the genus *Lipara* occur in Poland, namely *L. lucens* (Meigen, 1830), *L. similis* (Schiner, 1854), *L. rufitarsis* (Loew, 1858) and *L. pullitarsis* (Doskočil, Chvála, 1971). All four are monophagous, their host plant being the common reed (*Phragmites australis* Cav.) Trin. ex Steud). The feeding larvae distort the apical part of reed stems, forming so-called galls.

Individual species form galls of a unique shape (1, 8). *L. lucens* galls are shaped as a conspicuous cigar. Stems affected by *L. similis* are very difficult to distinguish from healthy reed stems that have not developed their inflorescences. Galls formed by *L. rufitarsis* and *L. pullitarsis* are very similar in external appearance, being narrow at the base and widened in the apical part. However, their internal structures are very different, with the larval chamber positioned below the growing point in the former and above it in the latter case (1).

Long-term observations of galls formed by *Lipara* flies have shown that *L. pullitarsis* forms not only galls of the type widely associated with this species (1, 8), but also ones that are rod-shaped and very similar in their external appearance to *L. similis* galls (2).

The life cycle of *L. pullitarsis*, and of all *Lipara* species, is closely related to the development of the host plant. *L. pullitarsis* females lay eggs on young reed shoots in May. The first-instar larva enters the inside of the stem to mult there and transform into the second- and third-instar larvae. In March of the following year, early pupae begin to appear in the decayed stem. In May, adults leave the galls and after copulation females begin to search for young reed shoots to place their eggs (3).

This paper describes the deformities (galls) produced by flies of the species *L. pullitarsis*. A detailed analysis of their internal structure will allow to identify those structural elements which determine the overall shape of the gall.

MATERIALS AND METHODS

We studied *Phr. australis* stems that bore signs of the presence of *Lipara* flies in their apical parts. The stems were sampled by sight in the Lublin administrative province in Poland between April and August 2014. Samples were taken in *Phragmition* reed beds growing over peat formations in the locality of Ciesacin and around the banks of Lake Moszne and in a wet meadow near Zemborzyce Reservoir. The reed stems collected in the field were photographed in the laboratory and sectioned with a stereoscopic microscope. The larvae obtained in this manner were subsequently determined using a specialised key for pre-imaginal forms (1). Stems with *L. pullitarsis* and *L. similis* larvae were subjected to a detailed laboratory examination which involved describing the external appearance of the deformity and its basal part, counting internodes within the basal part and measuring the length of each internode and stem diameter at the base just above the ground. The first internode with an abnormal length ratio when compared to the length of internodes in healthy stems was recognised as the lowest internode comprised within the basal part of a gall.

In order to identify the structural elements of the basal part that had a considerable influence on the overall shape of *L. pullitarsis* galls, we compared the findings of a detailed examination of the structure of galls formed by *L. pullitarsis* and *L. similis,* because some galls produced by larvae of the former species were quite similar to *L. similis* galls, which have a uniform structural design.

We collected a total of 532 fly-invaded reed stems. 319 colonised by *Lipara* flies of which measurements were carried out in 40 stems affected by *L. pullitarsis* and 20 affected by *L. similis*. Larvae were preserved in 75% ethyl alcohol with glycerol.

RESULTS

Lipara flies colonised 59.96% of all reed stems collected by us.The greatest number among these, 185, were colonised by *L. similis* and 134 were affected by *L. pullitarsis*, respectively accounting for 57.99% and 42.01% of all stems colonised by flies of the genus *Lipara* (Tab. 1).

Tab. 1. Statistics regarding common reed stems analysed in the study

Locality	Date	Number of stems analysed		Number of stems colonised by *Lipara* flies		Stems colonised by *Lipara* flies as a percentage of all stems analysed	Stems colonised by *Lipara* flies							
							L. pullitarsis		Stems colonised by *L. pullitarsis* as a percentage of stems colonised by *Lipara* flies	*L. similis*		Stems colonised by *L. similis* as a percentage of stems colonised by *Lipara* flies		
Ciesacin	12.04.2014	143	221	57	131	59.28 %	52	67	51.15 %	5	64	48.85 %		
	25.08.2014	78		74			15			59				
Lake Moszne	06.05.2014	190		106		55.79 %	47		44.34 %	59		55.66 %		
Zemborzyce Reservoir	30.07.2014	31	121	20	82	67.8 %	0	20	24.39 %	20	62	75.61 %		
	18.08.2014	90		62			20			42				
TOTAL		532		319		59.96 %	134		42.01 %	185		57.99 %		

GALL STRUCTURE

In a healthy reed stem, the growing point is surrounded by leaf sheaths growing out of nodes at the base of each internode (Fig. 1. A, B). A gall is formed by shortened internodes at the apex of a stem (basal part) and by leaves growing out of nodes, with the leaves responsible for the overall shape of the deformity (Fig. 1. C, D)

The overall external appearance of stems colonised by *L. similis* did not deviate from literature descriptions. On the other hand, the stems colonised by *L. pullitarsis* larvae did not demonstrate one structural pattern and varied considerably in shape. Some of them could be identified on the basis of relevant literature as they were wider in the apical part and narrow at the base (Photo1a), while others (Photo 1b) deviated substantially from that pattern and resembled the external appearance of galls produced by *L. similis* (Photo 1c). Thus, some representatives of *L. pullitarsis* and all of *L. similis* produced galls without an evident deformity in the apical part of the reed stem.

The basal parts of all *L. pullitarsis* galls had a cigar-like external appearance that sometimes was more or less deformed. The upper edge of the internodes forming the basal part of a gall often followed a slightly wavy line. The walls of the internodes were hard and thick. The lowest internodes had a narrower lower part (Photo 2a, 2b). All internodes had conspicuous longitudinal ribbing on their surface. The length of the internodes forming the basal part varied, with the longest ones found at the very bottom of the basal part and internodal length growing progressively smaller towards the growing point.

BASAL PART OF APICALLY WIDER GALLS OF *L. PULLITARSIS*

The basal part of typical *L. pullitarsis* galls was made up of 5–7 shortened internodes, with 90% of such galls comprising 6–7 internodes in their basal part. The overall length of such galls was between 37 and 112 mm (Tab. 2). The walls of the lowest-lying internodes in their basal part were more or less straight, demonstrating no evident abnormalities (Photo 2a). The diameter of those stems ranged from 3.2 to 5.3 mm (mean 4.29 mm) (Tab. 2). The length of the two topmost internodes ranged between 1 and 4 mm (Tab. 3). For the third internode in galls of this type, the mean length was 3.42 mm and the median was 3 (Tab. 3). A substantial increase in and diversification of internodal length could be seen from the fourth internode onwards (mean 7.05, median 4, SD 7.65) (Tab. 3), with each following internode (V, VI, VII) being longer than those above it (Tab. 3). Length differences between internodes IV–VII were much greater than in the case of those lying above them (I, II, III). The number of internodes in the basal part of a gall was found to be inversely related to their length. For example, the fifth internode in galls regarded as characteristic of *L. pullitarsis* colonisation ranged widely in length from 4 to 38 mm (Tab. 3). In galls with the basal part made up of 7 internodes, the mean

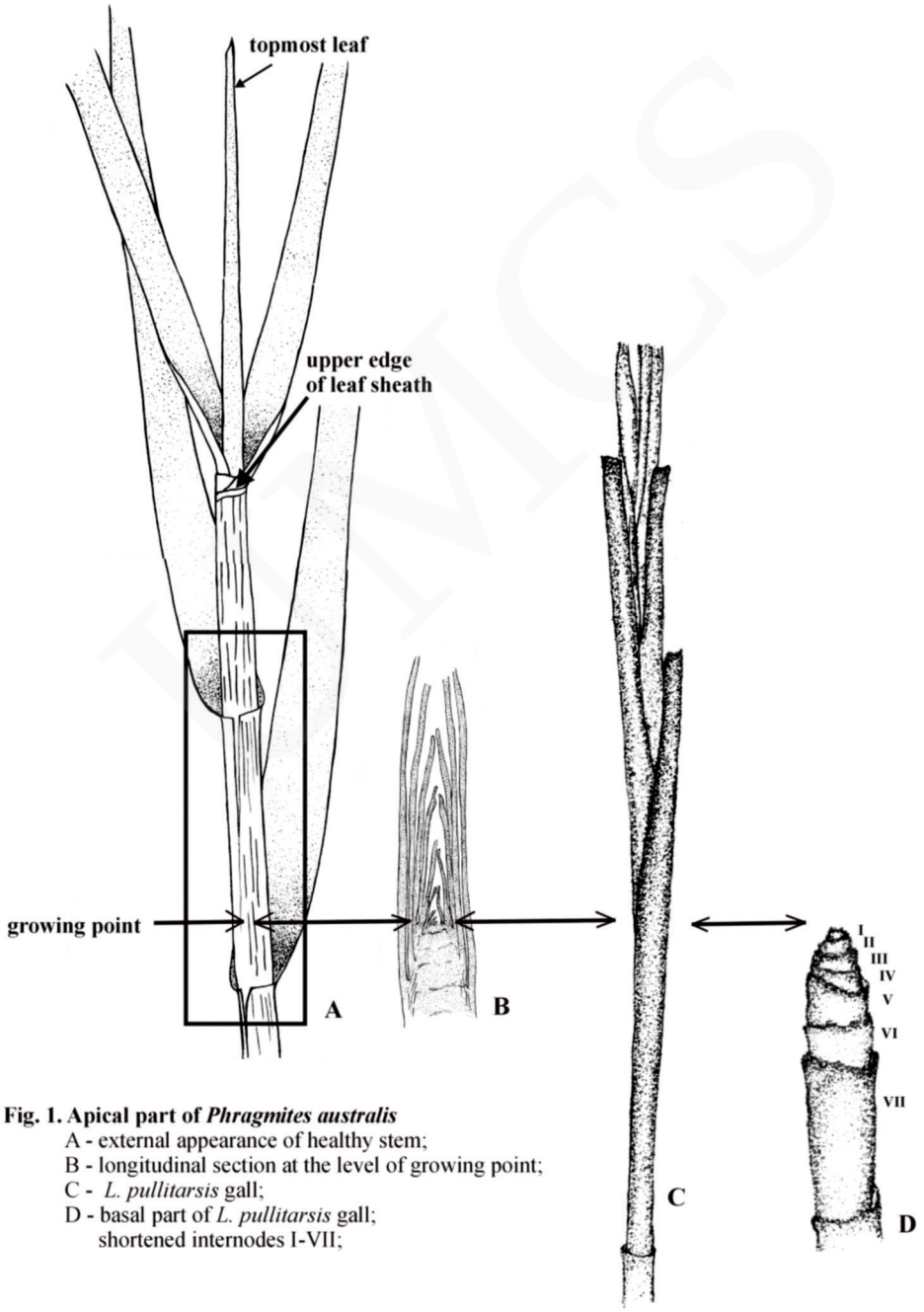
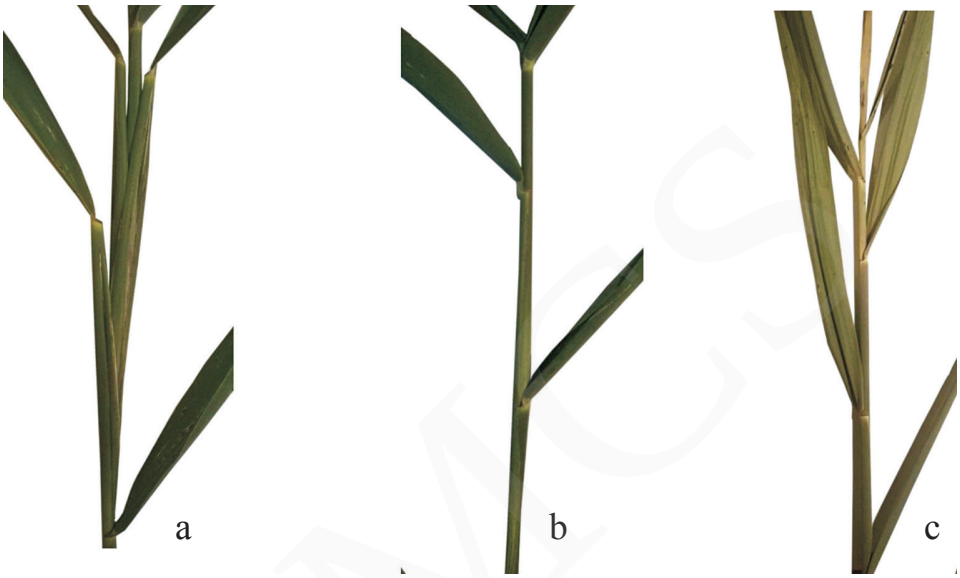


Fig. 1. Apical part of *Phragmites australis*
A - external appearance of healthy stem;
B - longitudinal section at the level of growing point;
C - *L. pullitarsis* gall;
D - basal part of *L. pullitarsis* gall;
shortened internodes I-VII;



Phot. 1. Galls produced by *Lipara* flies (external appearance): *L. pullitarsis* – (a–b), *L. similis* – c

Phot. 2. Basal part of galls with leaves removed: apically widened gall of *L. pullitarsis* – a, rod-shaped gall of *L. pullitarsis* – b; gall of *L. similis* – c

Tab. 2. Length of the basal part of galls and diameter of stems colonized by *L. pullitarsis* and *L. similis*

Gall	Number of internodes forming the basal part of gall														Stem diameter		Total stems analysed
	3			4			5			6			7				
	Number of stems	Length		Number of stems	Length		Number of stems	Length		Number of stems	Length		Number of stems	Length			
		Range	Mean		Range	Mean		Range	Mean		Range	Mean		Range	Mean		
Apically widened gall of *L. pullitarsis*	–	–		–	–	–	2	37–59	48	8	41–112	71.19	10	37–99	63	3.2– 5.3	4.29	20
Rod-shaped gall of *L. pullitarsis*	–	–		–	–	–	4	47–109	70.25	16	39–100	56	–	–	–	2.7–4.6	3.78	20
L. similis	2	16–26	21	11	32–55	43.6	6	33–68	52.92	1	52.5	52.5	–	–	–	3.3–5.1	4.22	20

Tab. 3. Internode length in apically widened galls of *L. pullitarsis*

Internode number	N	Length [mm]	Mean	Median	SD
		Range			
I	20	1–2	1.1	1	0.31
II	20	1–4	2	2	0.84
III	20	1–8	3.42	3	1.84
IV	20	2–28	7.05	4	7.65
V	20	4–38	14.55	11	12.44
VI	18	6–45	23	23	11.55
VII	10	20–40	31.9	30.5	9.18

length of internode V was 6.3 mm, increasing to 20.38 in galls comprising 6 shortened internodes, and to as much as 32.5 mm in those comprising five internodes in their basal part. Most galls contained an inflorescence inside.

BASAL PART OF ROD-SHAPED GALLS OF *L. PULLITARSIS*

The basal part of rod-shaped galls produced by *L. pullitarsis* was made up of 5 or 6 internodes, with 80% of such galls comprising 6 internodes. The overall length ranged from 39 to 109 mm (Tab. 2). The lowest-lying internode was considerably deformed, most commonly twisted, in the vast majority of the cases (Photo 2b). Stem diameter ranged from 2.7 to 4.6 mm (mean 3.78 mm) (Tab. 2.). The length of the two topmost internodes was in the range of 1–4 mm (Tab. 4). The mean length of the third internode in this type of galls was 4.1 mm and the median was 3.5 mm (Tab. 4). Much greater and varied lengths were seen from the 4th internode onwards (mean length 8.25, median 5.5, SD 7.11) (Tab. 4). Each following internode (V, VI) was longer than the one overlying it (Tab. 4). Length differences between internodes IV–VI were much more marked than those between the higher-lying internodes (I, II, III). The length of internodes IV–VI depended on the number of shortened internodes forming the basal part of a gall. For example, in galls with a basal part involving 6 internodes, the mean length of the 5th internode was 13.5 mm, compared to 40 mm in galls including 5 shortened internodes. Those stems had no inflorescences, except just a few, which, however, bore very small inflorescences.

BASAL PART OF *L. SIMILIS* GALLS

The basal part of galls produced by *L. similis* comprised 3 to 6 shortened internodes, with 85% of such galls involving 4–5 internodes. The overall length of a gall ranged from 16 to 68 mm (Tab. 2). All galls had the same structural design (Photo 2c). The stem diameter was 3.3-5.1 mm (mean 4.22) (Tab. 2.) The length

Tab. 4. Internode length in rod-shaped galls of *L. pullitarsis*

Internode number	N	Length [mm] Range	Mean	Median	SD
I	20	1–2	1.25	1	0.44
II	20	1–4	2.25	2	0.79
III	20	2–9	4.1	3.5	2.13
IV	20	4–33	8.25	5.5	7.11
V	20	6–61	18.8	12	13.99
VI	16	20–54	30.25	26	9.87

of the two topmost internodes ranged from 0.5 to 4 mm (Tab. 5). The mean length of the third internode was 4.92 mm, and the median was 4 (Tab. 5.). Much greater and varied lengths were seen from the 4th internode onwards (mean 23.56, median 26, SD 16.93) (Tab. 5.). Each following internode (V, VI) was longer than the overlying one (Tab. 5). Differences in the length of internodes IV–VI were much greater than in the higher-lying ones (I, II, III). The length of internodes IV–VI depended on the number of internodes forming the basal part of a gall. For example, in galls with the basal part made up of 5 internodes, the mean length of the 4th internode was 4.67 mm, while in galls with 4 shortened internodes, this index increased to 35.82 mm. Most of the stems in this group had no inflorescence.

COMPARISON OF DATA REGARDING THE BASAL PART OF GALLS PRODUCED BY *L. PULLITARSIS* AND *L. SIMILIS*

A detailed analysis of the structure of the basal part of *L. pullitarsis* and *L. similis* galls revealed significant differences in the number of shortened internodes and their length both between stems colonised by the same *Lipara* species and between the same internodes in both species.

Tab. 5. Internode length in galls of *L. similis*

Internode number	N	Length [mm] Range	Mean	Median	SD
I	20	0.5–2.5	1.1	1	0.45
II	20	1–4	1.9	2	0.84
III	20	1–21	4.92	4	4.81
IV	18	2–48	23.56	26	16.93
V	13	4–59	37.86	39	20.02
VI	19	44	–	–	–

The basal part in galls of *L. pullitarsis* comprised a much greater number of internodes (5–7) than the corresponding part of *L. similis* galls (3–6). Most stems colonised by the former species involved 6 or 7 shortened internodes, while *L. similis* most often produced galls by shortening 4 internodes, less often 5. Rod-shaped galls of *L. pullitarsis* most commonly involved 6 shortened internodes in their basal part.

Significant differences in the length of internodes forming the basal part of galls were seen with regard to the third internode and would grow steadily with each following (underlying) internode. Internode length was inversely related to the number of internodes forming the basal part of a gall. The mean lengths of internodes III, IV and V were lowest in typical galls of *L. pullitarsis* and highest in galls produced by *L. similis*. In rod-shaped galls of *L. pulitarsis*, these indices were always higher than the corresponding indices in typical galls of *L. pullitarsis* and lower than those measured in *L. similis* galls. Thus, they were closer to those measured in rod-shaped galls (produced by *L. similis*). These data show unequivocally that it is internodes III–V in the basal part of a gall that determine the gall's outer shape.

Each node in the basal part of a gall grows leaves that determine the overall shape of galls formed by *Lipara* flies. When the internodes are short, the leaf sheaths growing out of nodes at the base of these internodes deviate from the stem's main axis due to lack of space, resulting in galls of the shape commonly attributed to *L. pullitarsis*. In galls with the basal part formed by longer internodes, the leaf sheaths do not deviate, having enough space to grow in, and so they grow parallel to the stem, producing the rod-shaped appearance that is so much characteristic of *L. similis* galls.

DISCUSSION

It is generally believed that gall-forming insects actively change the shape of the host plant (8). Many authors of papers on flies colonising the common reed (1, 6, 7) are convinced that individual species of *Lipara* flies make galls of a shape characteristic of that particular species. This knowledge has been used to compile keys for determining common reed-dwelling insects (1, 4). Regrettably, the data contained in these publications are not very precise. The main focus is on typically shaped galls, documented in photographs, while the diversity of gall designs is ignored, which considerably hinders, or even completely precludes, the practical application of these keys. With regard to galls formed by *L. pullitarsis*, the difficulty is certainly related to the description of the gall given by Pokorny (6), which was too general and has been reproduced by other authors (1, 7). It has very often been stressed (1, 7) that the overall shape of an *L. pullitarsis* gall is similar to that of

galls produced by *L. rufitarsis*, another *Lipara* species, and the two may therefore be confused. Evidence for this statement is based on two different captions under the same photograph included in two different publications. The same gall (that is beyond doubt - it is strongly suggested by the overall shape of the deformity and the pattern of leaf blades in the apical part) was described as a typical *L. rufitarsis* gall in a collective paper by Chvála et al. (1) and as an *L. pullitarsis* gall in the paper by Pokorny (7). A gall of this species drawn by Nartshuk (5) is rod-shaped with a distinct widening in the middle part and so its external appearance differs from those of the galls in the photographs published in the papers by Pokorny (7) and Chvála et al. (1). The present paper adds to this diversity by giving evidence of gall forms similar to those produced by *L. similis*. These data show that the external appearance of galls formed by *L. pullitarsis* does not lend itself to sweeping statements, while also showing the formal diversity of galls that this species produces in the apical part of common reed stems.

Descriptions of the external appearance of galls are often accompanied by photographs and characterisation of the basal part of *L. pullitarsis* galls (1, 7, 8). Häfliger's (4) key ignores the external appearance of a gall, relying only on the presence of shortened internodes near the location of the larva within the stem as a determinant. Using this criterion, too, one cannot be certain which species actually dwells in a reed stem without sectioning it as, without leaves, the galls are very much similar to one another.

It is beyond doubt that the external shape of an *L. pullitarsis* gall is moulded by leaves growing out of nodes separating the internodes in the basal part. The leaves together with the shortened internodes comprise the segment of the plant deformed by *L. pullitarsis* (i.e. the gall). To date, no definite statements regarding this have been voiced. *L. pullitarsis* forms galls of diverse shapes. Considering this diversity, it should be borne in mind that attempts to determine the species of fly which has produced a particular gall on the basis of its external appearance or the structure of the basal part are generally doomed to fail. Determination will be most reliable when it is based on sectioning of the entire gall.

The formation of galls of different shapes is certainly associated with differences in the impact on the stem of different individuals of *L. pullitarsis*. Egg-laying by this species displays a similar lack of a uniform pattern. *L. pullitarsis* females, unlike females of other species of this fly genus, lay eggs on the entire surface of the plant (3), while the other species use specific locations (for example, *L. rufitarsis* lay their eggs only at the very tip of a leaf blade).

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