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## Seasonal decline in clutch size of the caddisfly *Glyptotendipes pallidus* (RETZIUS) (Trichoptera: Limnephilidae)<sup>1</sup>

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**Abstract:** At a lake in Central Sweden females of the caddisfly *Glyptotendipes pallidus* (RETZIUS) oviposited on aerial leaves of aquatic and riparian plants from mid July until early September. In captivity females oviposited on average 3.3 times (range: 1-6) with a mean clutch size of  $121 \pm 70$  eggs. There was, however, a seasonal decline in eggs produced per deposition. The longest oviposition period for a female in captivity lasted for 45 days. If a female will survive to lay her full complement of eggs (four to six clutches), this will render her a fecundity of more than 500-600 eggs depending on her condition and size.

**Key words:** Caddisfly, *Glyptotendipes pallidus*, oviposition, seasonal decline, clutch size.

### Introduction

In the end of 1980's and the beginning of 1990's when studying the egg laying of *Glyptotendipes pallidus* (RETZIUS) (GULLEFORS 1989) and *Nemotendipes punctatolineatus* (RETZIUS) (GULLEFORS 1994) I found that the numbers of eggs were smaller in the end of the season. Could it be the same females laying the larger egg masses in the beginning of the season as well as the smaller ones in the end of the season?

The figures in literature on the number of eggs laid by different caddisfly species (e. g. SILFVENIUS 1906, SILTALA 1907, HANNA 1961, HINTON 1981) seem to refer to as the total number of eggs produced by an individual. However, information on how many times the females oviposit is often lacking, therefore the figures are probably only parts of eggs laid than the total sum. Many limnephilids are thought to oviposit only once, for example *Limnephilus stigma* CURTIS, *L. rhombicus* L (NOVAK & SEHNAL 1963), *Limnephilus externus* HAGEN and *Anabolia bimaculata* (WALKER) (BERTÉ & PRITCHARD 1986). BERTÉ & PRITCHARD (1986) also believed that only a small minority of the most fecund females of *Nemotendipes hostilis* (HAGEN) oviposited a second time. However, ANDERSON (1974, 1977) found that the American limnephilid species *Pseudostenophylax edwardsi* (BANKS) and *Clistoronia magnifica* (BANKS) deposit more than one egg mass. The later egg masses were smaller and contained for *C. magnifica* also non viable eggs (ANDERSON 1977). The Japanese limnephilid species *Nothopsyche ruficollis* (ULMER) is able to oviposit more than one egg mass (NOZAKI & SHIMADA, 1997). In Sweden females of *Nemotendipes punctatolineatus* deposit 3-5 egg masses (GULLEFORS 1994).

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<sup>1</sup> This paper is dedicated to Prof. Dr. Hans Malicky on the occasion of his 75<sup>th</sup> birthday.

The egg-laying and egg masses of *Glyptotaelius pellucidus* were studied by CRICHTON (1987), OTTO (1987, 1993) and GULLEFORS (1989), but no information is given on how many times the females oviposit. They are, however, thought to oviposit more than once (GULLEFORS 1989). The number of eggs per mass (= clutch size) varied a lot and in Central Sweden I found some very small masses in the end of the egg laying period. This aroused the questions: how many times do the *G. pellucidus* females oviposit during the season?

### Material and methods

The main study area was the south shore of the lake Valasjön in Central Sweden (63°00'N/17°30'E). The study of egg-laying and egg masses was mainly carried out in 1988, 1989 and 1991. From egg masses taken in August 1990 larvae were reared to adults in the laboratory in the autumn 1990 and spring 1991. Adults emerged from December to January were transferred to small plastic cages (8 dm<sup>3</sup>). Some of the females were dissected for examination of ovarian maturation. A few females oviposited in the cages and their egg masses were used for rearing larvae to adults.



**Fig. 1:** Five egg masses of *Glyptotaelius pellucidus* on a leaf of grey alder (*Alnus incana*). The upper left egg mass was less than twelve hours old.

In the field egg masses laid by *Glyptotaelius pellucidus* were collected every tenth day in 1988 (21, 31 July, 10, 20 and 30 August) from the same area (aprox. 40 m<sup>2</sup>). All masses were laid on leaves of willow bushes (*Salix* spp.) and grey alder (*Alnus incana*) and a few on leaves of other plants. Most collected egg masses were laid during the same last night, but some of the masses were a few days old (Fig. 1). None was older than three days. Additional data were sampled from ponds and other lakes in Central and South Sweden in 1988-1991. Throughout the season 1989 (20 July - 4 September), in the same area as 1988, newly laid egg masses were looked for every day and if found, the first one or two masses observed were collected. The masses were

sampled in the morning, i.e. they were less than twelve hours old. The numbers of eggs in all collected masses were counted after being soaked in 5 % KOH for about an hour. The jelly was then dissolved and the eggs could be counted easily. The method is described by CRICHTON (1987). The sizes of five eggs from every collected mass in 1989 were measured with an ocular micrometer to nearest 0.01 mm.

In 1991 the egg-laying of ten females in captivity was studied. Two females and one male, which were reared from an egg mass laid in laboratory in February, hatched on 11 June and 13 June respectively. The three specimens were kept together for 34 days (13 June - 16 July), then the females were placed in separate cages (No 1 and 2). I saw copulations on 29 June and 30 June, respectively. The other eight females were captured at night (about 23.00-24.00 hrs); one on 21 July and seven females on 27 July. When captured the females were active and behaved as if they were going to lay eggs (cf. GULLEFORS 1989). Normally, the egg-laying period of *G. pellucidus* in this part of Sweden starts around 20 July and lasts until the first week in September in the studied area. The egg masses are fewer in the beginning of the

period (CRICHTON 1987 and GULLEFORS 1989), and the peak oviposition period for *G. pellucidus* in Central Sweden is from the last days of July to mid August (GULLEFORS 1989). The captured females were thus not suspected to have oviposited before they were captured. All ten females were kept separately in the plastic cages until they died late in the season. In the cages there were fresh twigs with leaves of *Salix* spp and a vial cap with honey water. During the day the cages were kept indoors at a temperature of about +18 °C and put outside during the nights (19.00-08.00 hrs) at a temperature of about +15 °C. To prevent desiccation and maintain humidity, the cages were covered during the day with moist pieces of cloth. The cages were examined carefully every morning to see if there had been any oviposition during the night. The captured females laid egg masses on the leaves and a few times on the roof and the walls of the cages. The dates of the ovipositions were recorded and the numbers of eggs counted. To test the viability, the eggs of the last deposition (11 September) were counted without being soaked in 5 % KOH solution, and the egg development was studied after that. Clutch size is given as mean and SD.

When the female was dead the right forewing was measured to nearest 0.1 mm,. Larvae were collected in the field on 28 October 1990, and 9 May 1991.

## Results

### Egg deposition in the field

The clutch size was  $125 \pm 59$  ( $n=68$ , range: 32-280) in 1988 and  $113 \pm 59$  ( $n = 28$ , range: 37-238) in 1989. The clutch size decreased during the season both in 1988 ( $r = 0.78$ ,  $p < 0.001$ ) and in 1989 ( $r = 0.91$ ,  $p < 0.0001$ , Fig. 2). The eggs from different egg masses of the whole sample 1989 ( $N = 140$ ) measured  $0.60 \text{ mm} \times 0.48 \text{ mm}$  ( $SD$ : 0.03 for both measurements). No correlation between egg size and clutch size was found and nor differed the egg size during the season.

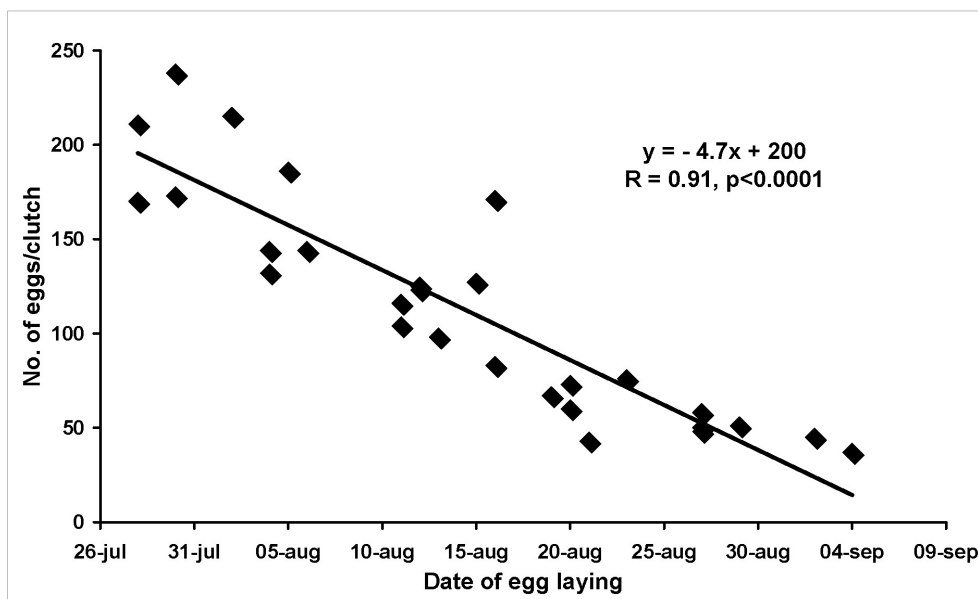


Fig. 2: Seasonal decline in clutch size of *Glyphotaelius pellucidus* in 1989.

## The adult females and egg deposition in captivity

The egg-laying period of the ten females in captivity lasted from 28 July to 11 September, i. e. 45 days. The clutch size decreased from the first laid masses to the last ones, with the exception of the sixth clutch of female no. 8 (Table 1).

After put into the cages three of the females laid their first eggs during the same night (27/28 July). The other ones deposited their first egg masses after 8 to 10 days (Table 1). The ten females laid together 33 clutches with an average of  $121 \pm 70$  eggs per mass (range 15-272). The time between two consecutive ovipositions was on average  $9.1 \pm 3.7$  days (range 6-21, median: 8). On two occasions six females laid eggs the same nights (4 and 10 August). It was raining both nights. Two other rainy nights (27 August and 5 September) four and three females, respectively, oviposited simultaneously. The two females emerged on 11 June in aquaria (No. 1 and 2) laid only one and two egg masses, respectively. They survived as adults for 58 and 84 days. With a supposed emergence on 11 June or earlier for all ten females, their adult lifetime was of  $81.8 \pm 15.5$  days (range 58-101). Eight of the females died within one to seven days after their last deposit. Two extremes lived for 21 and 22 days after their last oviposition. Embryos and later larvae developed within the last laid egg mass (11 September). The size of the forewing of the females was  $17.9 \pm 0.5$  mm.

**Tab. 1:** The egg laying of ten females of *Glyptotaelius pellucidus* in captivity in 1991.

	JULY			AUGUST				SEPTEMBER		
Female/Week										
No.	30	31	32	33	34	35	36	37	Sum	Died
1	-	-	191	-	-	-	-	-	191	7 Aug
2	-	230	126	-	-	-	-	-	356	2 Sep
3	-	220	142	-	-	79	40	-	481	7 Sep
4	-	250	170	-	98	72	38	-	628	6 Sep
5	-	-	272	-	-	136	38	-	446	9 Sep
6	209	148	-	-	-	-	-	-	357	8 Aug
7	-	185	131	-	-	-	-	-	316	17 Aug
8	185	-	114	94	-	66	15	31	505	18 Sep
9	157	-	120	66	-	50	-	-	393	29 Aug
10	-	128	98	-	60	33	-	-	319	19 Sep

### Female No.

- 1 Fore wing 17.5 mm. Emerged in aquaria 11 June, mated 30 June, drowned stuck to the egg mass on 7 Aug. The female had small undeveloped ovaries 2.5 mm, about 100 small undeveloped eggs (0.22 mm x 0.2 mm).
- 2 Fore wing 17.0 mm. Emerged in aquaria 11 June, mated 29 June with the same male as female no. 1. Contained four mature eggs when she died.
- 3 Fore wing 18.3 mm. Captured on 21 July. Contained 32 mature eggs when she died.
- 4 Fore wing 17.5 mm. Captured on 28 July. No eggs left when she died.
- 5 Fore wing 18.3 mm. Captured on 28 July. More than 30 mature eggs when she died.
- 6 Fore wing 18.9 mm. Captured on 28 July. 15 mature eggs.
- 7 Fore wing 18.1 mm. Captured on 28 July. About 120 small undeveloped eggs in ovaries together with spermatophore with sperms.
- 8 Fore wing 17.8 mm. Captured on 28 July. One single egg left.
- 9 Fore wing 17.5 mm. Captured on 28 July. 15 mature eggs left.
- 10 Fore wing 17.8 mm. Captured on 28 July. Five mature eggs and sperms in the spermatheca.

## Larvae

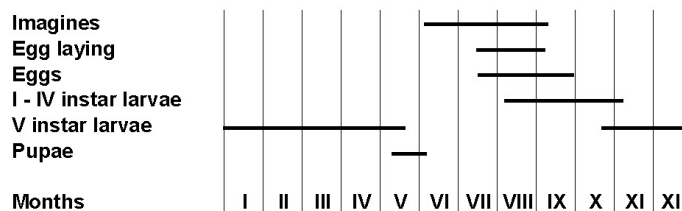
Three V instar larvae were collected in the lake Valasjön on 9 May 1991. Two of them pupated in aquaria on 11 May and the third on 14 May. They swam as pupae on 30 May (two males) and 1 June (a female) respectively. They all died without casting their pupal skins. Thus the pupation period was 19-20 days in 16 °C laboratory water and probably a few days more in the colder water in the field, suggests a hatching period in the beginning of June.

The head widths of three larvae collected on 28 October 1990 were 2.0, 1.8 and 1.5 mm respectively, indicating they were V instar larvae or for the last one maybe a IV instar larva (WALLACE, WALLACE & PHILIPSON 2003).

In egg masses brought to the laboratory, eye spots of the first developed embryos were seen after four days, and larvae hatched in the jelly within two weeks. The development rate in the same egg mass varied. Eggs, embryos and first instar larvae could be seen together in the same mass.

From egg masses brought to laboratory in August and September 1990 the first larvae pupated in the end of November. Adults emerged from mid December to late January. Females emerged with undeveloped ovaries, stage "A" according to the classification established by NOVAK & SEHNAL (1963). Some of the females laid eggs in the plastic cages. From an egg mass deposited in February adults emerged in the beginning of June.

The life cycle of *G. pellucidus* in Central Sweden is summarised in Fig. 3.



**Fig. 3:** Schematic presentation of the life cycle of *Glyptotaelius pellucidus*.

## Discussion

In southernmost Sweden *G. pellucidus* females have a quiescence period from the emergence in early June until the egg laying period starts (SVENSSON 1972), and that corresponds well for Central Sweden. The peak oviposition period in Central Sweden is from late July to mid August (GULLEFORS 1989), when some females oviposit for the first time and others lay their second or third clutch.

The number of eggs in the consecutive laid egg masses were on average two thirds of the previously laid mass (Table 1) and if the secondly laid egg mass has 185 eggs then the first one will have about 277 eggs. However, very few masses had over 250 eggs. The largest clutch in the studied area contained 280 eggs (1988) and in a neighbouring area (Rudtjärnen, 63°01'N/17°35'E), the largest egg mass contained 285 eggs. Also the female no. 8, with only 185 eggs in her first laboratory clutch, had probably not oviposited earlier, since she oviposited six times with the first laid mass already at the same night she was captured.

Although it seems as the females in this study had not started their egg laying before they were captured, though it might be possibly that the females nos 9 and 10, with a first clutch size in captivity of only 157 and 128 eggs, could have oviposited earlier.

The two females reared in laboratory from eggs to adults oviposited only one and two times, respectively. Their condition seemed not to be as good as the females captured in the field.

From the laid egg masses and the discover of further eggs in their ovaries after death, it appears as if a *G. pellucidus* female survives to lay her full complement of eggs (4 to 6 batches) then she will produce more than 500 and for some females more than 600 eggs. This estimate excludes the females nos. 1 and 2, and includes assumed earlier laid egg masses ( $\approx 250$  eggs) of females nos. 9 and 10, and it also includes the findings of ovary eggs in the dead females.

Adult caddisflies consume very little food and for the egg development all resources must be gained during the immature stages. Obviously the clutch size is reduced throughout the life of female insects which do not obtain much resources for eggs in the adult stage.

There is a limit to how many eggs can be squeezed inside the body of a female. CRICHTON (1987) reported 349 as the largest clutch number found in South England and that corresponds well with the largest egg masses (339 and 326 eggs) I have found in South Sweden (the shore of Insjön (59°20'N/18°22'E)). The sizes of the females depositing these large egg masses are unknown. However, no correlation could be found between body size and clutch size of the ten studied females in captivity.

*Glyphotaelius pellucidus* and *Nemotaulius punctatolineatus* are the only North West European limnephilids which oviposit on aerial leaves of aquatic and riparian plants. They are closely related and were for a long time considered as one genus (*Glyphotaelius*). Both species oviposit several times with decreasing clutch size (GULLEFORS 1994 and this study).

Considering the variation in clutch size for several studied limnephilids, e.g. *Limnephilus externus* (136-405), *Anabolia bimaculata* (84-211), and *Nemotaulius hostilis* (64-607) (BERTÉ & PRITCHARD 1986), it seems very likely that these species deposit several egg masses, more than the two ovipositions suggested for a minority of the most fecund *N. hostilis* females (BERTÉ & PRITCHARD 1986).

Clutch size variation within limnephilid species has been poorly studied. The found egg mass is often regarded containing all the eggs producible by a single female. Probably females of most limnephilid species, like *G. pellucidus* and *N. punctatolineatus*, oviposit several times and with a seasonal decline in clutch size and the total fecundity per female is therefore considerably higher than usually reported in literature.

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## Zusammenfassung

An einem zentralschwedischen See legen die Weibchen von *Glyptotendipes pallidus* ihre Eier außerhalb des Wassers an Blätter von Wasser- und Uferpflanzen von Mitte Juli bis Anfang September. In Gefangenschaft legten die Weibchen 1–6 (im Mittel 3,3) Eiballen bei einem Mittel von  $121 \pm 70$  Eiern pro Ballen. Mit der Jahreszeit nahm die Eizahl pro Gelege ab. Die längste Ablageperiode eines Weibchens in Gefangenschaft dauerte 45 Tage. Bei einer völligen Ablage aller Eier beträgt die Nachkommenschaft mehr als 500–600 Eier je nach der Kondition und Größe des Weibchens.

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