ISEA PAS

Acta zoologica cracoviensia, 68 (1) 2025
Kraków, 2025
http://www.isez.pan.krakow.pl/en/acta-zoologica.html

e-ISSN 2300-0163
https://doi.org/10.3409/azc.68.01

Variation in egg numbers of the meadow spittlebug *Philaenus spumarius* (L.) associated with particular dorsal colour phenotypic groups among New Zealand, Turkish and Welsh populations

Selcuk Yurtsever

Received: 6 March 2025 *Accepted 15 September 2025* *Available online: 18 November 2025* *Issue online 28 November 2025*

Original article

Yurtsever S. 2025. Variation in egg numbers of the meadow spittlebug *Philaenus spumarius* (L.) associated with particular dorsal colour phenotypic groups among New Zealand, Turkish and Welsh populations. *Acta zoologica cracoviensia* **68**: 1-12.

Abstract. Detailed egg number variations for the polymorphic meadow spittlebug *Philaenus spumarius* (L.) that were obtained from continuous mass breeding experiments in fully laboratory conditions are reported for the first time. Genetic crossing experiments with certain colour/pattern phenotypes were set up using unmated virgin insects. Three generations were reared during the three-year period of the experiments. A total of 21,213 eggs were produced through 247 pairings. The general median number of eggs per pairing was 76. Egg numbers for the insects from New Zealand, Türkiye and Wales (UK) were comparable. Although the maximum number of 378 eggs was produced by a *Typicus* phenotype from Türkiye, the medians showed no sign of statistical differences among the three populations. However, the median number of eggs among the F1, F2 and F3 generations were different. The median test applies to the Wales data regarding the combined egg numbers of Melanic, *Typicus* and *Trilineatus* groups of phenotypes. The Melanic group of females produced more eggs (81) than *Typicus* (42) and *Trilineatus* (72), although with no statistically significant differences. The methods used allow for the maintenance of *P. spumarius* as a stock for consecutive overlapping generations, which may facilitate the management of future ecological and evolutionary investigations in this spittlebug as a dependable laboratory model organism. The results also will contribute to the debates and implications for breeding studies of *P. spumarius* in the field of population ecology.

Key words: polymorphism, spittlebug, rearing, egg number, New Zealand, Türkiye, Wales.

Selcuk Yurtsever, Biology Department, Science Faculty, Trakya University, Türkiye.
✉ *E-mail: selcuk22yurt@gmail.com*

I. INTRODUCTION

Visual polymorphism in a diverse array of animals has been foundational for elucidating evolutionary processes (Poulton 1890; Ford 1955; Kettlewell 1973; Majerus 1998). Inherited colour polymorphisms in insects provide a raw material for deterministic (Brakefield & Willmer 1985; Cook & Saccheri 2013) and stochastic effects (Oxford 2005), in order to reveal how evolution acts to shape population variations. Elytral colour/pattern variations of several sympatric and allopatric *Philaenus* (Homoptera; Auchenorrhyncha) species in Europe and the Mediterranean area (Maryańska-Nadachowska et al. 2010; Seabra et al. 2010; Tanyeri et al. 2023) have provided good examples for evolutionary genetic studies. The meadow spittlebug *Philaenus spumarius* (Linnaeus, 1758) is a well-studied insect in terms of its heritable colour/pattern polymorphism (Yurtsever 2000a). Owing to its highly polyandrous behaviour (YURTSEVER 2001) and polyphagous nature on numerous plant species (HALKKA & HALKKA 1990; YURTSEVER 2000a; THOMPSON et al. 2023), *P. spumarius* abundantly occupies a wide variety of terrestrial habitats throughout the Holarctic region. It ranges from the Azores (BORGES et al. 2018) through the Urals (MAYOROVA & KOLOVA 2019) to the Far-East (SHIH & YANG 2002) and North America (WEAVER & KING 1954; HAMILTON 1982; THOMPSON et al. 2023). Introduced populations are present on both of the major islands in New Zealand (ARCHIBALD et al. 1979; THOMPSON 1984; SYRETT & SMITH 1998; HODGE & KEESING 2000). However, Hawaii is the only tropical area in which the species has been recorded (DAVIS & MITCHELL 1946). Those two populations possibly originated from Europe and the US, respectively. However, the wide geographic distribution of the species is strictly limited by chilling conditions in the cooler arctic and is limited in warmer regions due to the heat or cold tolerance of the delicate nymphs (HALKKA & HALKKA 1990; YURTSEVER 2000a).

The best detailed information on *P. spumarius*, dating from the early 1900s, can be found in the work of WEAVER and KING (1954). Their remarkable monograph gives a comprehensive account of the species' life cycle, ranging from the egg stage to polymorphic adults. Population genetic studies with advanced molecular applications have also made outstanding contributions to novel spittlebug investigations (MARYAŃSKA-NADACHOWSKA et al. 2010; SEABRA et al. 2010). By analysing significant numbers of published papers, *P. spumarius* studies can be divided into two different kinds of disciplines. The first deals with its ecology, including surveys of its noxious status as a plant pest. The second approach involves evolutionary biology, concerning the heritable colour/pattern polymorphism exhibited among several species in the genus *Philaenus*.

Since the nymphs and adults of *P. spumarius* feed on the xylem sap of plants, it has been cited as a serious plant pest by many authors. Although it does not always noticeably harm the host plants (WHITTAKER 1973; HAMILTON 1982; SYRETT & SMITH 1998), heavy densities of the nymphs may sometimes cause severe damage to economically valuable crops and cultivated plants (MUNDINGER 1946; WIEGERT 1964; MEYER & ROOT 1993). *P. spumarius* has also been viewed as a vector for some microbial plant diseases. For example, it has been stated in some earlier literature that *Xylella fastidiosa,* the causal agent of Pierce's disease, is transmitted by *P. spumarius* from diseased to healthy grape vines in the US. (DELONG & SEVERIN 1950). Recently, *P. spumarius* has been declared as a serious threat to susceptible olive trees in Italy and to almond trees with grape vines in Spain, as a carrier of the re-emerging pathogen *Xylella fastidiosa* (CORNARA et al. 2018). Accordingly, its vector status has led to great attention from agricultural research and the number of papers regarding its harmful impact have multiplied sharply during the last decade.

The inheritance of the elytral colour/pattern polymorphism of *P. spumarius* and the geographic variation of the phenotypes have been investigated in detail. As in remarkable examples of evolutionary biology (KETTLEWELL 1973; BRAKEFIELD & WILLMER 1985; OXFORD 2005; COOK & SACCHERI 2013), this polymorphism has provided good evidence about the modes of evolution acting on heritable traits in natural populations. The nomenclature of the colour/patterns morphs of *P. spumarius* has long been debated by many authors (YURTSEVER 2000a; TANYERI et al. 2023). Eleven common morphs are generally suggested as the principal phenotypes (STEWART & LEES 1996; YURTSEVER 2000a). The genetic basis for these phenotypes has been established by laboratory genetic crossing experiments (HALKKA et al. 1973; STEWART & LEES 1988; YURTSEVER 2000a; SILVA et al. 2015). It has been found that there is a single pigmentation locus with seven different alleles (Table 1) for expressing particular phenotypes.

HALKKA et al. (1973) and STEWART & LEES (1988) established a special nomenclature and three letter abbreviations for describing the common phenotypes, according to a complex genetic dominance hierarchy among them. These phenotypes are grouped into two main categories as melanics (dark coloured) and non-melanics (light coloured), according to the elytral colour designs (Fig. 1). For example; *typicus* (TYP) and *populi* (POP) are non-melanics and are produced by the same allele, called '*t*'. The *trilineatus* (TRI) phenotype is the third non-melanic produced by the allele '*T*'. The remaining phenotypes named in Table 1 are melanics. TRI is top and POP/TYP are bottom in the genetic dominance hierarchy (HALKKA & HALKKA 1990; STEAWART & LEES 1996; YURTSEVER 2000a). A variety of evolutionary forces have played a role on the occurrence and geographic variations of the phenotypes in natural populations.

The highest frequency of melanics have been reported from some Welsh populations (Stewart & Lees 1996), where strong natural selection was effective and resulted in industrial melanism. The existence of only a few alleles and the lack of many phenotypes in the remote islands of Portugal (Borges et al. 2018), Türkiye (Yurtsever 2018) and New Zealand (Lees 1993) are associated with random genetic drift events, such as founder effects.

Studies on the biology of *P. spumarius* in natural conditions date back to the early 1900s (refs. in Weaver & King 1954). Although some details about its life history may be found in much earlier papers (refs. in Weaver & King 1954; Hamilton 1982), the first oviposion information (Barber & Ellis 1922) involves a description of the eggs with figures of captured individuals in special rearing cages on *Setaria glauca* plants. Later, the life history of *P. spumarius* was investigated by rearing caged insects in the field (Mundinger 1946) and under greenhouse conditions (Ahmed & Davidson 1950). Mass breeding of *P. spumarius* for genetic crossing experiments in semi-natural conditions was conducted in some Finnish (Halkka et al. 1973) and Portuguese (Silva et al. 2015) populations. Recent ecological studies have examined whether the phenology of its life history shows variations in certain climatic conditions among some Spanish populations (Morente et al. 2021; Lago et al. 2023). Seok & Nam (2000) investigated insect-plant interactions using *P. spumarius* with *Gastropysa viridula* in greenhouse conditions at the campus of Lancaster University in the UK.

Table 1

Nomenclature of the Melanic, TRI and TYP group phenotypes with their abbreviations and the alleles for producing them in the meadow spittlebug *P. spumarius* (Halkka et al. 1973; Stewart & Lees 1988)

	Abbreviation	Allele
Non-melanic phenotypes		
Populi	POP	t
Typicus	TYP	t
Trilineatus	TRI	T
Melanic phenotypes		
Marginellus	MAR	M
Lateralis	LAT	L
Flavicollis	FLA-F	F
Gibbus	GIB	C
Leucocephalus	LCE	C
Quadrimaculatus	QUA	O
Albomaculatus	ALB	O
Leocophthalmus	LOP	O

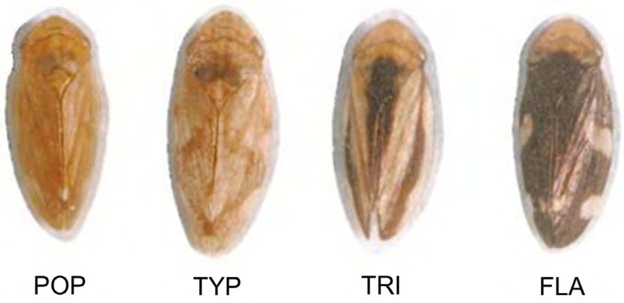

Fig. 1. Four of the common dorsal colour/pattern phenotypes for *P. spumarius*. The non-melanics, from left to right are: POP, TYP, TRI; a melanic phenotype: FLA. Abbreviations are defined in Table 1.

A study by Witsack (1973) is the first example of the rearing of *P. spumarius* in fully controlled laboratory conditions. Mass breeding of *P. spumarius* in the laboratory was successfully conducted by Lees and his co-workers in the late 1980s (Stewart & Lees 1988; West & Lees 1988), which involved many controlled genetic experiments. However, these studies principally focused on experimental work associated with the adult spittlebugs or that involved some stages of nymphal development. The rearing of several other cercopid species has been achieved in laboratories (Whittaker & Tribe 1998; Peck et al. 2004; Garcia et al. 2011; Chen & Liang 2012; Wang et al. 2021). To our knowledge, however, there is no published detailed information available on mass rearing with the egg quantification of the cercopid *P. spumarius* in fully controlled laboratory conditions. The current paper presents information gathered from mass laboratory breeding experiments involving three generations of the adult, nymph and egg numbers, from populations sourced from New Zealand, Türkiye and Wales. The data was derived from a PhD study (Yurtsever 1997), which was conducted for genetic polymorphism experiments in *P. spumarius*.

II. MATERIAL AND METHODS

Laboratory stock and breeding experiments

The breeding material for this study was obtained from field sites involving three different countries.

The spittlebugs were initially collected as nymphs, and these specimens were used as a stock for further experiments in the laboratory located at the University of Wales in Cardiff (Presently: Cardiff University) in the UK. The New Zealand stock was obtained from Lincoln (near Christchurch) in the east of the South Island (YURTSEVER 2000b; 2002a). The British stock, including polyandry data (YURTSEVER 2001), originated from Wales; and the Turkish spittlebugs were obtained from Kirklareli in Türkiye (YURTSEVER 2002b).

Philaenus spumarius is a univoltine and hemimetabolous insect; thus, it undergoes two separate egg diapauses (ovarian and overwintering diapauses) during a single life span, as in many other univoltine insects (WITSACK 1973; WEST & LEES 1988). After the egg stage, there are five nymphal instars (YURTSEVER 2000a) before it reaches the adult stage (Fig. 2). When the adults emerge from the final instar they are colourless, but in about one hour they are fully pigmented and begin to mate. Therefore, females and males must be separated at the fourth or fifth instar stage for the purpose of experiments. The segregation of the sexes at the last instar stage was not difficult to perform under a microscope.

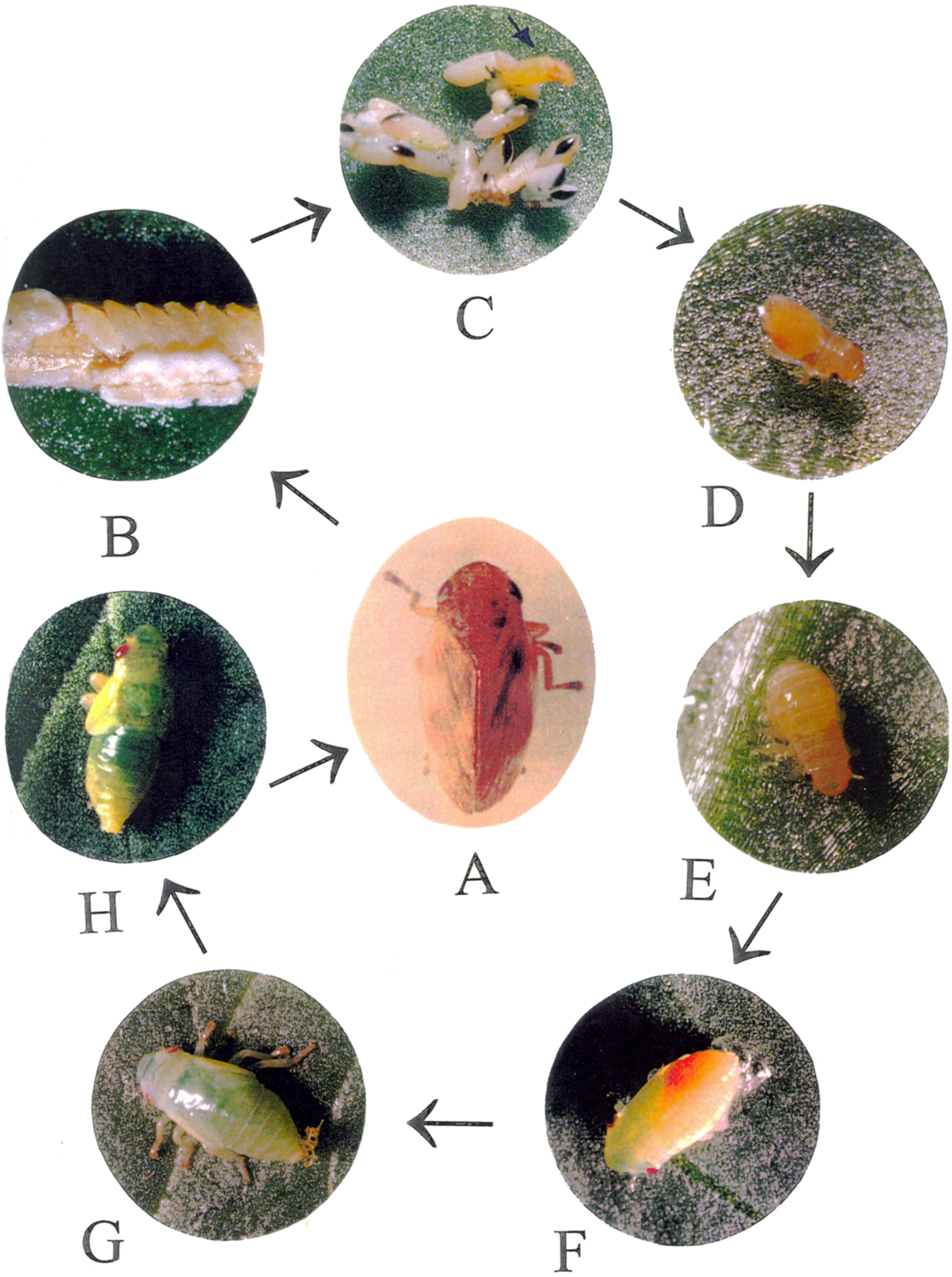

Fig. 2. Life cycle of *P. spumarius*. A – adult, B – newly-laid eggs, C – mature eggs and a nymph is (arrowed) just hatching out, D – first, E – second, F – third, G – fourth, H – fifth instar nymphs, respectively.

The sexing using a toothpick did not result in any harm to the delicate nymphs. Information on the sexing of spittlebug nymphs is only found in a few early papers (Kershaw & Muir 1922; George 1928), but those methods required dissection of the insect.

Certain photoperiod and temperature regimes were used, in order to shorten the life cycle in laboratory conditions (West & Lees 1988). In this study, all of the spittlebugs were reared on the dwarf broad bean (*Vicia faba* cv. 'The Sutton') (Fig. 3A) in breeding cages. Approximately ten spittlebugs were maintained in each cage. Cylindrical transparent breeding cages (12 x 38 cm) were made from a 0.16 mm acetate sheet and fine gauze (Fig. 3B). Two different thermostatic chambers (CT-a and CT-b) with 80% humidity were used (2 x 2 x 3 m) for rearing the insects. CT-a had an 18:6 L:D (light:dark) photoperiod at 17°C that was used to accelerate the egg hatching and nymphal development. CT-b had a 12:12 L:D photoperiod at 12°C, and served to terminate the ovarian diapause and thus, induced the females to lay eggs.

The females and males were sexed at the fifth instar stage and were kept in separate cages before the experimental treatments. Sexing of the nymphs was performed using a toothpick under a binocular stereomicroscope. The ventral side of the abdomen was gently pressed using the toothpick to see the outer parts of the genitalia (Fig. 4).

Fig. 3. Breeding of *P. spumarius*. A – broad bean (*Vicia faba*) plant and spittle masses produced by *P. spumarius* nymphs, B – breeding cages in the controlled room.

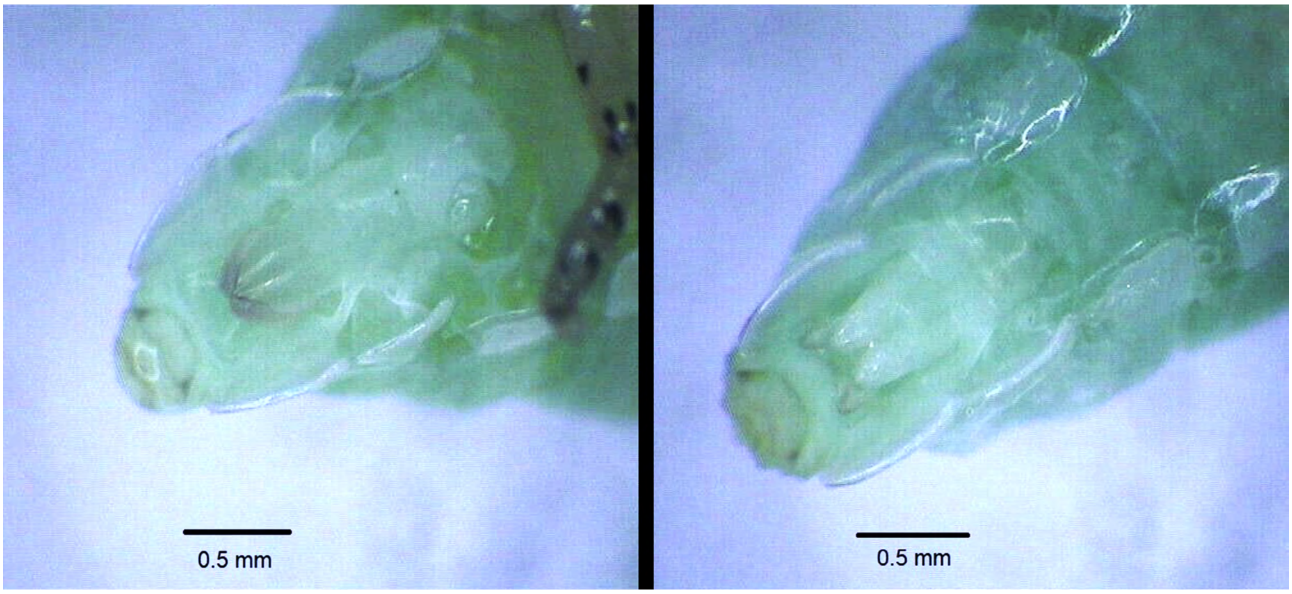

Fig. 4. Sexing for the fifth instar nymphs, female on the left and male on the right.

Each individual genetic cross was set up with unmated adult virgin pairs. Most of the pairs were readily mated in these cages; many pairs were even mated in the hand-made aspirator which was used for handling the adult insects. If the pairs were unwilling to mate, they were kept in small clip cages attached to the leaves of the broad bean plant until they mated, and were then released into their own breeding cage. In the polyandry experiments, each female insect mated with multiple males at appropriate intervals (see YURTSEVER 2001 for details). Clean wheat straw materials were placed on the soil in the breeding cages for the oviposition of the females. Gravid females also frequently preferred to lay eggs on the dead leaves of the broad bean plants.

The egg clusters (Fig. 2) were collected from the cages and were placed in Petri dishes with a Plaster of Paris base with water containing 'Nipagin' as an antibiotic, which protected the eggs against fungal and other deleterious infections. Eggs were kept in the fridge at 5°C for about 80 days. The petri dishes were inspected at four-day intervals to ensure that the Plaster of Paris remained moist. By exposing them to this chill period, the overwintering diapause was broken in nearly all the fertilised eggs. A black spot (lid) where young insects would leave the egg appeared on the tips of the fertilised eggs (Fig. 2C) showing they were ready for hatching. Unfertilised eggs had a bone-white colour with a small orange spot only. The fertilised eggs were removed from the fridge for post diapause development in breeding cages, where they were placed on the surface of moist soil in CT-a. The eggs synchronously hatched and tiny nymphs emerged in about four weeks, then crawled onto the broad bean plants to produce spittle until they became adults in about one month. In this way, the life cycle was generally reduced from one year to less than eight months. The insects were examined under a binocular stereo microscope at a x20 magnification.

Statistics

The non-parametric, Kruskal-Wallis, One-Way ANOVA test was used for comparing the group medians, where appropriate. Samples with a sample size n<3 were excluded from the analyses, to hold the statistical assumptions. When a Kruskal-Wallis test yielded a significant main effect, Dunn's pair-wise comparisons were applied amongst the independent groups to understand where the differences existed. The non-parametric Mann-Whitney-*U*-test was used for comparing the medians of the two groups. The IBM SPSS (Ver. 22; 2021) statistics software was used for the analyses.

III. RESULTS

Variation in egg numbers produced by the insects obtained from New Zealand, Turkiye and Wales

Details of the summary statistics for the eggs with the statistical results are presented in Tables 2-4.

A total of 21,213 eggs were counted from 247 pairings during three generations of the insects, resulting in four data sets (Table 2). The number of the eggs produced by the mated female insects showed a great deal of variation. The general median was 76 eggs (mean 85.88 and standard error 4.33). The number of eggs per female ranged from 3 to 378. The highest number of eggs was produced by a spittle bug from the Turkish stock. However, the highest median value for the eggs was found in polyandry data originating from the Welsh stock. The sample size was highest in the Welsh data, with 155 pairings, while being lowest in the Turkish and polyandry data. The New Zealand data with 47 pairings was mid-range. Interestingly, the sample sizes and mean numbers of eggs from the Turkish and polyandry stocks were very close. However, the median numbers were more variable, being highest in polyandry but lowest in the Welsh stock. Moreover, the median was the same for the New Zealand and Turkish data. Despite these variations, the median number of eggs showed no signs of statistically significant differences at the 0.05 level (Table 2).

The number of the eggs produced by the females in the three different generations of the three separate

Table 2

Summary statistics and Kruskal-Wallis test results of the egg numbers for *P. spumarius* by data sets (n = number of crosses)

Data set	n	Mean	Median	Min	Max
New Zealand	47	82.98	76	3	240
Polyandry	22	106.30	100	5	354
Türkiye	23	97.60	76	3	378
Wales	155	82.13	67	4	367

H = 1.08; df = 3; p = 0.782

data sets also showed interesting variations (Table 3). Since the polyandry experiments involved only one generation, this data was not included in the generation analyses. According to pair-wise comparisons, the median number of F2 progeny was lower than in F1 and F3 in all three data sets (Table 3). However, the three generations showed no statistically significant differences (p>0.05) in the median number of eggs regarding the data from Wales (Table 3). On the other hand, the statistical analyses revealed that the median egg numbers obtained in the F3 generation was significantly higher than the median numbers of eggs found in the F1 (p<0.05) and F2 (p<0.01) progeny in the New Zealand data. Nonetheless, there was no difference between F1 and F2 (p>0.05) in the medians. Moreover, the median of the eggs for the F1 progeny was significantly (p<0.01) higher than the median egg number found for the F2 progeny in the Turkish stock, but the data of the F3 generation was not possible to utilise for a statistical test, due to an inappropriate sampling size (n<3) of only 2.

Table 3

Summary statistics and Kruskal-Wallis test results of the egg numbers for *P. spumarius* by F1, F2 and F3 offspring in the New Zealand and Wales data sets. Mann-Whitney-*U* test results for the Turkish data (n = number of crosses)

	n	Mean	Median	Min	Max
New Zealand					
F1	12	77.30	67.50	15	236
F2	22	59.23	58.50	3	120
F3	13	128.50	128.00	7	240
H = 10.11; df = 2; p = 0.006; Median of F3, significantly higher than F1 (p = 0.02) and F2 (p = 0.001)					
Türkiye					
F1	12	138.00	96.50	9	378
F2	9	31.11	25.00	3	102
F3	2	155.00	155.00	24	285
Median of F1, significantly higher than F2. Mann-Whitney-U Test, W = 172.0, p = 0.005 (F3 is not included in the statistics, due to n<3)					
Wales					
F1	49	85.96	71	5	215
F2	64	77.86	64	4	216
F3	42	84.20	69	5	367
H = 0.51; df = 2; p = 0.773					

Variation in egg numbers produced by *P. spumarius* belonging to the Melanic, *Trilineatus* and *Typicus* phenotype groups

The maximum number of the eggs produced by particular phenotypic pairings (female x male) were as follows: Türkiye, 378 eggs (TYPxTYP); Wales, 367 eggs (MARxFLA); New Zealand, 240 eggs (FLAxFLA); and Polyandry (crosses in which females mated with more than one male), 354 eggs (TYPxLCE and xTRI).

Egg numbers produced by the females of particular phenotype groups (Melanic, *Trilineatus* and *Typicus*) were only possible to compare in the Welsh material (Table 4), because the data for the Welsh specimens was large enough to see differentiations among these groups. Nine groups of crosses show interesting variations in the egg numbers. On average, Melanic females produced higher number of eggs than the TRI and TYP females, with the TRI females being the least productive (Table 4), but the differences were not statistically significant (Table 4). Likewise, there were no significant differences (p>0.05) among the combined crosses in which females were Mel, TRI and TYP.

Hybrid crosses of *P. spumarius* between Wales (WA) x New Zealand (NZ) and between WA x Türkiye (TR) involving F1 and F2 generations were possible to examine, in order to determine if they were able to produce fertile eggs (Table 5). Both of the crosses produced fertile eggs and offspring. However, the median

Table 4

Summary statistics and Kruskal-Wallis test results of the egg numbers for *P. spumarius* by genetic crosses (Female x Male) between particular phenotypic groups in the Wales data set (n = number of crosses)

Cross	n	Mean	Median	Min	Max
Mel x Mel	31	95.00	76.0	5	367
Mel x TRI	5	123.40	116.0	63	202
Mel x TYP	17	99.90	107.0	5	207
TRI x Mel	7	67.90	42.0	15	216
TRI x TRI	12	72.40	39.5	8	180
TRI x TYP	11	70.50	65.0	7	200
TYP x Mel	24	82.58	81.5	15	172
TYP x TRI	9	79.10	72.0	12	154
TYP x TYP	32	87.20	64.5	9	311
H = 6.75; df = 8, p = 0.563					

Table 5

Summary statistics and Mann-Whitney-*U* test results of the eggs for *P. spumarius* from hybrid crosses among the New Zealand, Turkish and Welsh populations (n = number of crosses)

Data set	n	Mean	Median	Min	Max	Sum
Wales x New Zealand	17	64	65	1	200	1088
Wales x Türkiye	8	126.6	129	49	223	1013
W = 183.5; p = 0.031						

egg number obtained from the WA x NZ (median: 65) crosses was much lower than that found in the WA x TR (median: 129) crosses, and it was a statistically significant difference ($p<0.05$).

IV. DISCUSSION

The results demonstrate that *Philaenus spumarius* can be reared with a shorter life cycle in a laboratory, with manageable difficulties, provided that certain photoperiod, temperature and humidity requirements are met.

The phenology of the seasonal timing of the life cycle events for *P. spumarius* has variations, since local populations of this species are exposed to a broad range of climates in its wide distribution throughout the world. Therefore, shorter and longer life cycles in cooler and warmer climates, respectively, are predictable. In New Zealand (LEES 1993), the life cycle of the species is shifted by six months, to correspond to the seasonality of the Southern Hemisphere. WHITTAKER & TRIBE (1998) experimentally showed that raising the temperature has a significant impact on the earlier hatching of the eggs in some British *Neophilaenus lineatus* populations. Recently, THOMPSON & MOSCOVITCH (2022) found a decline of melanic forms of *P. spumarius*, based on their long-term study conducted in northern Minnesota. These two studies suggest that spittlebugs may prove to be a useful indicator of the accumulated climate changes in a broad range of geographical areas. Adult meadow spittlebugs can be found even in winter months (ZEYBEKOGLU et al. 2004) in the northern Turkish population. Although the impact of predictable global warming on this population cannot be excluded, this issue may be addressed more precisely in further studies, because the adults of some populations are able to survive until frost occurs in the late autumn (WEAVER & KING 1954). Interestingly, WITSACK (1973) in a study of some German *P. spumarius* stated that it would be possible to obtain two generations of this spittlebug in laboratory conditions. Strikingly, MORENTE et al. (2021), working on a local Spanish population, claimed that three generations per year under laboratory conditions was possible. However, these authors did not support this assertion with actual examples. Species have a characteristic set of genetically determined eco-physiological responses (TAUBER & TAUBER 1981). The diversity and the complexity of insect life cycles contribute to the difficulty in analysing the genetics of these cycle adaptations (GILL et al. 2017), which requires a substantial body of experimental data. Abundant species such as *P. spumarius* are exposed to very diverse climatic conditions, which may reflect in correspondingly significant variations in the seasonal cycles among geographic populations. Thus, further progress in understanding the evolution of seasonal cycles depends on advances being made in a number of important areas (TAUBER & TAUBER 1981; GILL et al. 2017) of genetic and ecological investigations.

The current study has demonstrated that it is possible to obtain overlapping generations in one year, by shortening the *P. spumarius* life cycle to less than eight months. However, obtaining multiple generations in one year in artificial conditions still remains a challenge. Natural populations of *P. spumarius* appear to be univoltine. However, there is one possible caveat to this. Hawaii has no low elevation and no cold climate, but substantial populations of *P. spumarius* have persisted there for decades. It appears that no one has ever studied their voltinism, which would make an interesting study. Perhaps it all has to do with the colder temperatures that can be found on higher levels of volcanos. The incidence of diapause may be quite variable from species to species, but also between different populations of the same species (TAUBER & TAUBER 1981; GILL et al. 2017). The results currently reported are similar to the those found in the studies of WITSACK (1973), WEST & LEES (1988) and STEWART & LEES (1988), who pioneered the method of artificially breaking the two obligatory diapauses to shorten the life cycle of *P. spumarius* in laboratory conditions.

A detailed description of *P. spumarius* eggs was given by BARBER & ELLIS (1922). MUNDINGER (1946) reported that the number of eggs per female varied between 18-50 in New York State in the month of September. According to Hamilton (1982), gravid females lay relatively few eggs, generally not exceeding 35 in number, but on occasion reaching up to 50, while some females may lay only about 10 eggs. Weaver & King (1954) counted the egg masses on different types of plant materials in northern Ohio. They mentioned that the average number of eggs produced by an individual female had never been clearly estimated, due to the difficulty of keeping adults alive in cages. As is noted in the current report, Weaver & King (1954) used wheat straw for the oviposition of the females and found that the type of oviposition site influenced both where the eggs masses are found and the total number of eggs that were laid. Interestingly, the presence of straw was apparently positively associated with an increase in the egg deposition. Hence, the use of different egg deposition materials for *P. spumarius* may be examined in future research, which may reveal interesting behaviours.

A detailed quantification of the egg numbers produced per female for *P. spumarius* is presented here for the first time. Although some egg quantities are given in previous *P. spumarius* studies, they have concerned incidental counts, not intentional egg production experiments. It is interesting to note that the number of eggs, for a single female Turkish specimen, reached 378. However, the median numbers of the eggs among the insects from the three populations did not show statistically significant differences. It seems that the insects adapted well to the photoperiod and temperature regimes with humidity in the breeding laboratories. It has been pointed out that repeated copulations of polyandrous females (Yurtsever 2001) may be associated with a high number of eggs. However, in the present work, the median egg counts of polyandrous females did not significantly differ from the median egg counts obtained from the females that mated with the same male. However, polyandry may have a positive impact on the genetic diversification of the offspring.

Hybrid crosses of *P. spumarius* between Wales (WA) x New Zealand (NZ) and between WA x Türkiye (TR) involving F1 and F2 generations produced fertile eggs and offspring. It is interesting to observe that the median egg numbers obtained from the WA x NZ crosses were much lower than those are found in WA x TR, which was possibly due to a lowered fitness and fecundity, and the sexual incompatibility of the hybrids; the offspring in these insects involved a mixed parentage from the different populations mentioned.

With regards to the egg counts produced by the females of different phenotypic categories, the results did not show any statistically significant differences. The median number of eggs produced by the Melanic, TRI and TYP group females was around 90. In a similar study, Silva et al. (2015) successfully reared Portuguese *P. spumarius* on *Carpobrotus edulis* plants, in semi-natural breeding conditions. They tested the productivity of TYP, TRI and MAR females in their breeding experiments. In contrast to the present study, they found that a higher number of eggs was laid by TRI females than those laid by the TYP and melanic MAR phenotypes. They found strong evidence that *Trilineatus* has an increased survival rate or a higher fertility than the other phenotypes. They also suggested that the TRI phenotype has an intrinsic capability against food shortages and the deleterious effects of temperature. Hence, temperature plays some role in the broad and complex selection regime, as a balancing factor in the polymorphism of *P. spumarius*. This has been demonstrated in some Scottish (Berry & Willmer 1986) and North American populations (Thompson & Moscovitch 2022). It will be worthwhile to test this hypothesis in the laboratory under controlled experimental conditions and, furthermore, to check the effects in natural populations in order to support the associations between particular genetic traits and evolutionary forces favouring a high fecundity of the TRI phenotype in particular climates. Yurtsever & Sal (2003) found that the proportion of TRI specimens did not change significantly in the summer and autumn seasons; thus, there was no association between the phenotype categories and the seasons, involving particular adaptive selective forces.

Even though the phenology of the life cycle in natural populations may show geographic variations, it is possible that every individual female has a certain egg production capacity. The diversity and sophistication of reproductive strategies in insects illustrate their adaptations to diverse ecological roles and their success in the evolutionary struggle for survival. Genetic, physiological and ecological factors have fundamental impacts in shaping insect reproductive strategies (Adams & Tariq 2024). One of these factors is the impact of natural enemies, ranging from spiders to birds (Yurtsever 2000a), that may negatively influence the reproductive potential of *P. spumarius* females in natural populations. Thus, it is worth noting that during the study presented here, only a few instances of natural enemies were observed in adult *P. spumarius* including the dipteran parasitoid *Verralia aucta* and some unknown species of mould, possibly *Entomophthora* sp. However, during our experiments the eggs were never seen exposed to any kind of predation or any notorious effects, possibly due to their 'styrofoam like' protection material.

The applicability of the breeding method presented here may be re-examined and may be improved in future studies. The current results emphasise the importance of *P. spumarius* as a dependable model organism for laboratory stock rearing. Despite the fact that this spittlebug is univoltine in natural populations, it is possible to produce eggs, nymphs and adults of a known lineage for experimental purposes year-round, which may facilitate management studies for this spittlebug as a plant pest. *P. spumarius* can be considered as a reliable model organism for future evolutionary studies to understand the interactions of different selective forces acting on the maintenance of polymorphism. Future research may also engage simulated novel laboratory experiments to investigate the particular fitness of phenotypes under different environmental conditions, such as aspects of the microclimate, resistant cultivars and parasite-predator interactions. Also, latest studies (Lovell-Read et al. 2022; Lago et al. 2023; Visnovsky et al. 2024) regarding the co-evolution among the host plants, pathogens and the pest status of *P. spumarius* involving mathematical modelling with interesting computer designs will be a stimulating area for evolutionary biologists. Above all, knowledge of the evolution of seasonal cycles provides a key to understanding how organisms exploit the environment, as well as how they interact (Seok & Nam 2000) and how they evolve (Tauber & Tauber 1981). Consequently, the findings presented here will potentially contribute to debates and the implications regarding breeding studies of *P. spumarius* in the fields of ecology and genetics.

V. ACKNOWLEDGEMENTS

I am most grateful to the anonymous referees for kindly help to improve this manuscript.

VI. CONFLICT OF INTEREST

The author declares no conflict of interest.

REFERENCES

Adams I., Tariq M. 2024. An overview of reproduction in Insects. *Asian Journal of Advances in Agricultural Research,* **24**: 133-147. https://doi.org/10.9734/ajaar/2024/v24i7529

Ahmed D.D., Davidson R.H. 1950. Life history of the meadow spittlebug in Ohio. *Journal of Economic Entomology,* **43**: 905-908. https://doi.org/10.1093/jee/43.6.905

Archibald R.D., Cox J.M., Deitz L.L. 1979. New records of plant pests in New Zealand. III. Six species of Homoptera. *New Zealand Journal of Agricultural Research*, **22**: 201-207. https://doi.org/10.1080/00288233.1979.10420862

Barber G.W., Ellis W.O. 1922. Eggs of three Cercopidae. *Psyche,* 29: 1-3. https://doi.org/10.1155/1922/71368

Berry A.J., Willmer P.G. 1986. Temperature and the colour polymorphism of *Philaenus spumarius* (Homoptera: Aphrophoridae). *Ecological Entomology,* **11**: 251-259. https://doi.org/10.1111/j.1365-2311.1986.tb00301.x

Borges P.A.V., Rodrigues A.S.B., Silva S.E., S.E., Seabra S.G., Paulo O.S., Quartau, J.A. 2018. New data on polymorphism of the meadow spittlebug *Philaenus spumarius* (L.) (Hemiptera: Aphrophoridae) from the island of São Miguel (Azores). *Zootaxa,* **43**: 144-150. https://doi.org/10.11646/zootaxa.4369.1.9

Brakefield P.M., Willmer P.G. 1985. The basis of thermal melanism in the ladybird *Adalia bipunctata*: Differences in reflectance and thermal properties between the morphs. *Heredity,* **54**: 9-14. https://doi.org/10.1038/hdy.1985.3

Chen X., Liang A-P. 2012. Laboratory Rearing of *Callitettix versicolor* (Hemiptera: Cicadomorpha: Cercopidae), With Descriptions of the Immature Stages. *Annals of the Entomological Society of America*, **105**: 664-670. https://doi.org/10.1603/AN12052

Cook L.M., Saccheri I.J. 2013. The peppered moth and industrial melanism: evolution of a natural selection case study. *Heredity,* **110**: 207-211. https://doi.org/10.1038/hdy.2012.92

Cornara D., Bosco D., Fereres A. 2018. *Philaenus spumarius*: When an old acquaintance becomes a new threat toEuropean agriculture. *Journal of Pest Science,* **91**: 957-972. https://doi.org/10.1007/s10340-018-0966-0

Davis C.J., Mitchell A.L. 1946. Host records of *Philaenus spumarius* (Linn.) at Kilauea, Hawaii National Park (Homoptera: Cercopidae). *Proceedings of the Hawaiian Entomological Society,* **12**: 515-516.

Delong D.M., Severin H.H.P. 1950. Spittle-insect vectors of Pierce's disease virus. I. Characters, distribution, and food plants. *Hilgardia,* **19**: 339-381. https://doi.org/10.3733/hilg.v19n11p339

Ford E.B. 1955. Polymorphism and taxonomy. *Heredity,* **9**: 255-264. https://doi.org/10.1038/hdy.1955.24

Garcia J.F., Simone S., Prado J.D., Vendramim J.D., Botelho P.S.M. 2011. Effect of sugarcane varieties on the development of *Mahanarva fimbriolata* (Hemiptera: Cercopidae). *Revista Colombiana de Entomologia,* **37**: 16-20. https://doi.org/10.25100/socolen.v37i1.9028

George C.J. 1928. The morphology and development of the genitalia and genital ducts of Homoptera and Zygoptera as shown in the life histories of *Philaenus* and agrion. *Journal of Cell Science*, **287**: 447-483. https://doi.org/10.1242/jcs.s2-72.287.447

Gill H.K., Goyal G., Chahil C. 2017. Insect Diapause: A Review. *Journal of Agricultural Science and Technology,* **7**: 454-473. https://doi.org/10.17265/2161-6256/2017.07.002

Halkka O., Halkka L. 1990. Population genetics of the polymorphic meadow spittlebug, *Philaenus spumarius* (L.). *Evolutionary Biology,* **24**: 149-191.

Halkka O., Halkka L., Raatikainen M. 1973. The genetic basis of balanced polymorphism in *Philaenus* (Homoptera). Hereditas, **74**: 69-80. https://doi.org/10.1111/j.1601-5223.1973.tb01105.x

Hamilton K.G.A. 1982. The Insects and Arachnids of Canada, Part 10. The Spittlebugs of Canada, Homoptera: Cercopidae. Ottawa-Ontario: Agriculture Canada, 102 pp.

Hodge S., Keesing V. 2000. Abundance and polymorph frequency of *Philaenus spumarius* (L.) (Hem., Homoptera, Auchenorrhyncha) on crops in the Selwyn District, Canterbury, New Zealand. Entomologists *Monthly Magazine,* **136**: 131-135.

Ibm Spss Statistics For Windows, (ver. 22). 2021. IBM Corp., Armonk, N.Y., USA). https://www.ibm.com/support/pages/downloading-ibm-spss-statistics-22

Kershaw J.C., Muir F. 1922. The Genitalia of the Auchenorhynchous Homoptera. *Annals of the Entomological Society of America,* **15**: 201-211. https://doi.org/10.1093/aesa/15.3.201

Kettlewell B. 1973. The Evolution of Melanism. Oxford: Clarendon Press. 423 pp.

Lago C., Giménez-Romero A., Morente M., Matías M.A, Moreno A., Fereres A. 2023. Degree-day-based model to predict egg hatching of *Philaenus spumarius* (Hemiptera: Aphrophoridae), the main vector of *Xylella fastidiosa* in Europe. *Environmental Entomology,* **52**: 350-359. https://doi.org/10.1093/ee/nvad013

Lees D.R. 1993. Novel New Zealand populations of the meadow spittlebug *Philaenus spumarius* (Cercopidae). *Proceedings of the 8th Auchenorrhyncha Congress, Delphi, Greece,* 95-97.

Lovell-Read F.A., Parnell S., Cunniffe N.J., Thompson R.N. 2022. Using 'sentinel' plants to improve early detection of invasive plant pathogens. *PLoS Computitional Biology,* **19**: e1010884. https://doi.org/10.1371/journal.pcbi.1010884

Majerus M.E.N. 1998. Melanism: Evolution in Action. Oxford University Press. 338 pp. https://doi.org/10.1093/oso/9780198549833.001.0001

Maryańska-Nadachowska A., Drosopoulos S., Lachowska D., Kajtoch L., Kuznetsova V.G. 2010. Molecular phylogeny of the Mediterranean species of *Philaenus* (Hemiptera: Auchenorrhyncha: Aphrophoridae) using mitochondrial and nuclear DNA sequences. *Systematic Entomology,* **35**: 318-328. https://doi.org/10.1111/j.1365-3113.2009.00510.x

Mayorova E.Y., Kolova U.V. 2019. Polymorphism of *Philaenus spumarius* Linnaeus, 1758 (Hemiptera: Auchenorrhyncha: Aphrophoridae) in the Altai Mountains. *Journal of Siberian Federal University,* **12**: 398-409. https://doi.org/10.17516/1997-1389-0050

Meyer G.A., Root R.B. 1993. Effects of herbivorous insects and soil fertility on reproduction of goldenrod. *Ecology,* **74**: 1117-1128. https://doi.org/10.2307/1940481

Morente M., Cornara D., Moreno A., Fereres A. 2021. Parapause breakage as a key step for the continuous indoor rearing of *Philaenus spumarius*. *Journal of Applied Entomology,* **145**: 1062-1067. https://doi.org/10.1111/jen.12931

Mundinger F.G. 1946. The control of spittle insects in strawberry plantings. *Journal of Economic Entomology,* **39**: 299-305. https://doi.org/10.1093/jee/39.3.299

Oxford G.S. 2005. Genetic drift within a protected polymorphism: Enigmatic variation in color-morph frequencies in the candy-stripe spider, *Enoplognatha ovata Evolution,* **59**: 2170-2184. https://doi.org/10.1554/05-046.1

Peck D.C., Morales A., Castro U. 2004. Alternative methods for rearing grass-feeding spittlebugs (Hemiptera: Cercopidae). *Neotropical Entomology,* **33**: 307-314. https://doi.org/10.1590/S1519-566X2004000300005

Poulton E.B. 1890. The Colours of Animals. Their meaning and use, especially considered in the case of insects. Bond Street, N,Y D. Appleton & Co. 336 pp. https://doi.org/10.5962/bhl.title.11353

Seabra S.G., Pina-Martins F., Marabuto E., Yurtsever S., Halkka O., Quartau J.A., Paulo O.S. 2010. Molecular phylogeny and DNA barcoding in the meadow-spittlebug *Philaenus spumarius* (Hemiptera, Cercopidae) and its related species. *Molecular Phylogenetics and Evolution*, **56**: 462-467. https://doi.org/10.1016/j.ympev.2010.03.023

Seok K.O., Nam S.H. 2000. Interactions between insect species feeding on *Rumex obtusifolius*: The effect of *Philaenus spumarius* feeding on the Ecology of *Gastrophys viridula*. *Korean Journal of Ecology,* **23**: 175-180.

Shih H-T., Yang J-T. 2002. Checklist of Aphrophoridae (Homoptera: Cercopoidea) from Taiwan. *Formosan Entomologist,* **22**: 193-214.

Silva E.S, Rodrigues A.S.B., Marabuto E., Yurtsever S., Borges P.A.V., Quartau JA., Paulo O.S., Seabra S.G. 2015. Differential survival and reproduction in colour forms of

Philaenus spumarius give new insights to the study of its balanced polymorphism. *Ecological Entomology,* **40**: 759-766. https://doi.org/10.1111/een.12252

Stewart A.J.A, Lees D.R. 1988. Genetic control of colour/pattern polymorphism in British populations of the spittlebug *Philaenus spumarius* (L.) (Homoptera: Aphrophoridae). *Biological Journal of the Linnean Society,* **34**: 57-79. https://doi.org/10.1111/j.1095-8312.1988.tb01948.x

Stewart A.J.A., Lees D.R. 1996. The colour/pattern polymorphism of *Philaenus spumarius* (L.) (Homoptera: Cercopidae) in England and Wales. *Philosophical Transactions of the Royal Society of London B*, **351**: 69-89. https://doi.org/10.1098/rstb.1996.0005

Syrett P., Smith L.A. 1998. The insect fauna of four weedy *Hieracium* (Asteraceae) species in New Zealand. *New Zealand Journal of Zoology,* **25**: 73-83. https://doi.org/10.1080/03014223.1998.9518138

Tanyeri R., Yurtsever S., Tezcan S. 2023. Parallel colour/pattern polymorphism in three sympatric spittlebug species (*Philaenus spumarius*, *P. signatus* and *Mesoptyelus impictifrons*) (Hemiptera: Aphrophoridae) in Mediterranean Türkiye. *Zoology in the Middle East,* **69**: 384-393. https://doi.org/10.1080/09397140.2023.2279359

Tauber C., Tauber M.J. 1981. Insect Seasonal Cycles: Genetics and Evolution. *Annual Review of Ecology and Systematics,* **12**: 281-308. https://doi.org/10.1146/annurev.es.12.110181.001433

Thompson V. 1984. Colour polymorphism in the introduced spittlebug *Philaenus spumarius* (Homoptera: Aphrophoridae) in New Zealand. *New Zealand Entomologist,* **8**: 86-88. https://doi.org/10.1080/00779962.1984.9722475

Thompson V., Harkin C., Stewart A.J.A. 2023. The most polyphagous insect herbivore? Host plant associations of the meadow spittlebug, *Philaenus spumarius* (L.). *PLoS One*. **18**: e0291734. https://doi.org/10.1371/journal.pone.0291734

Thompson V., Moscovitch R.M. 2022. Decline of melanic color forms in Northern Minnesota populations of *Philaenus spumarius* (L.) (Hemiptera: Aphrophoridae). *Entomologica Americana,* **128**: 1-10. https://doi.org/10.1664/NYES-D-22-00001

Visnovsky S.B., Kahn A.K., Nieto-Jacobo F. 2024. Multiple genotypes of a quarantine plant pathogen detected in New Zealand indigenous plants located in a botanical garden overseas. *Plant Pathology.* **74**: 1-10. https://doi.org/10.1111/ppa.14026

Wang Q., Bai L., Chen X., Liang A. 2021. Laboratory rearing of *Abidama liuensis* (Hemiptera: Cercopidae) and description of immature stages. *European Journal of Entomology,* **118**: 82-89. https://doi.org/10.14411/eje.2021.010

Weaver C.R., King D.R. 1954. Meadow spittlebug *Philaenus leucophtalmus* (L.). *Ohio Agricultural Experiment Station Research Bulletin,* **741**: 1-99.

West J., Lees D.R. 1988. Temperature and egg development in the spittlebug *Philaenus spumarius* (L.) (Homoptera: Aphrophoridae). *Entomologist,* **107**: 46-51.

Whittaker J.B. 1973. Density regulation in a population of *Philaenus spumarius* (L.) (Homoptera: Cercopidae) *Journal of Animal Ecology,* **42**: 163-172. https://doi.org/10.2307/3410

Whittaker J.B., Tribe N.P. 1998. Predicting numbers of an insect (*Neophilaenus lineatus* Homoptera in a changing climate. *Journal of Animal Ecology,* **67**: 987-991. https://doi.org/10.1046/j.1365-2656.1998.6760987.x

Wiegert R.G. 1964. Population energetics of meadow spittlebugs (*Philaenus spumarius* L.) as affected by migration and habitat. *Ecological Monographs,* **34**: 217-241. https://doi.org/10.2307/1948501

Witsack W. 1973. Experimental and ecological investigations on forms of dormancy in Homoptera-Cicadina (Auchenorrhyncha). 2. On ovarian parapause and obligatory embryonic diapause in *Philaenus spumarius* (L.) (Aphrophoridae). *Zoologische Jahrbücher Systematik,* **100**: 517-562.

Yurtsever S. 1997. Inheritance of colour/pattern variation in the meadow spittlebug *Philaenus spumarius*. *Unpublished Ph.D. thesis, University of Wales*, Cardiff. 271 pp.

Yurtsever S. 2018. Limited polymorphism in two spittlebugs, *Philaenus spumarius* (Linnaeus) and *P. signatus* Melichar (Hemiptera: Cercopidae) in island populations from Western Turkey. *Zoology in the Middle East,* **64**: 169-173. https://doi.org/10.1080/09397140.2018.1444568

Yurtsever S. 2000a. On the polymorphic meadow spittlebug, *Philaenus spumarius* (L.) (Homoptera: Cercopidae). *Turkish Journal of Zoology,* **24**: 447-460. https://journals.tubitak.gov.tr/zoology/vol24/iss4/13

Yurtsever S. 2000b. Inheritance of the two dorsal colour/pattern phenotypes in New Zealand populations of the polymorphic meadow spittlebug *Philaenus spumarius* (L.) (Homoptera: Cercopidae). *Journal of the Royal Society of New Zealand,* **30**: 411-418. https://doi.org/10.1080/03014223.2000.9517632

Yurtsever S. 2001. Multiple paternity in the meadow spittlebug *Philaenus spumarius* (Cercopidae). *Acta Zoologica Cracoviensia,* **44**: 173-184.

Yurtsever S. 2002a. Hybrid crosses of the meadow spittlebug *Philaenus spumarius* (L.) (Hom., Cercopidae) between the Turkish and Welsh populations. *Entomologist's Monthly Magazine,* **138**: 149-155.

Yurtsever S. 2002b. Hybrid crosses of the meadow spittlebug *Philaenus spumarius* (L.) (Homoptera: Cercopidae) between New Zealand and Welsh populations. *New Zealand Journal of Zoology,* **29**: 245-251. https://doi.org/10.1080/03014223.2002.9518308

Yurtsever S., Sal G. 2003. Population genetics of *Philaenus spumarius* (Homoptera, Cercopidae) on the Istranca Mts: Phenotypic diversity, allele frequency and sex ratio. *Biologia,* **58**: 929-934.

Zeybekoglu U., Yurtsever S., Turgut F. 2004. Polymorphism of *Philaenus spumarius* L. (Hemiptera: Cercopidae) in the Samsun (Mid-Black Sea Region) populations of Turkey. *Annales de la Société entomologique de France,* **40**: 277-283. https://doi.org/10.1080/00379271.2004.10697426