

# Aphid–*Buchnera*–Ant symbiosis; or why are aphids rare in the tropics and very rare further south?

Evgeny Perkovsky<sup>1\*</sup> and Piotr Wegierek<sup>2</sup>

<sup>1</sup>Schmalhausen Institute of Zoology, National Academy of Sciences of Ukraine, ul. Bogdana Khmel'nitskogo 15, Kiev 01601, Ukraine.  
Email: perkovsk@gmail.com

<sup>2</sup>Department of Zoology, University of Silesia, Bankowa 9, 40-007 Katowice, Poland.  
Email: piotr.wegierek@us.edu.pl

\*Corresponding author

**ABSTRACT:** At least since the Cretaceous Terrestrial Revolution, the geographical distribution of aphids, particularly in the Northern Hemisphere, has been strongly affected by the low thermal tolerance of their obligatory bacterial symbiont, *Buchnera aphidicola*, which was why the aphids switched to obligate parthenogenesis in low latitudes. Hormaphidids and greenideids penetrated into the tropics only after the Oligocene strengthening of climate seasonality, and specialisations of the tropical representatives of these families did not allow them to spread further south (in the case of cerataphidines), or only allowed in few cases (in the case of greenideids).

Aphids suffered from the Mesozoic–Cenozoic boundary extinction event much more strongly than other insects. The extinction was roughly coincidental with the establishment of the tight symbiosis of aphids with formicine and dolichoderine ants, which was accompanied by the flourishing of all three groups.

In the Cretaceous, all of the representatives of extant and subfamilies occupied positions that were subordinate to Armaniinae and Sphecomyrminae. Prior to large ant colonies evolving their efficient ant–aphid mutualism, the aphids remained unprotected before the growing ant predation. The origin of the aphid trophobiosis with large colonies of Formicinae and Dolichoderinae has resulted in the steep decline of aphids left beyond that ant–aphid symbiotic network. By at least the basal Eocene (unlike the Late Cretaceous), ant proportions in the entomofauna increased sharply, and evident dominants emerged. Even now, aphid milkers from small colonies (hundreds of specimens) never protect their symbionts, and homopteran-tending ants are more likely to be dominant, with large colonies of  $10^4$ – $10^5$  workers.

The mutualistic ant–aphid system failed to cross the tropical belt during the Cenozoic because of *Buchnera*'s low heat tolerance. As a result, the native southern temperate aphid fauna consists now of seven genera only, five of which are Late Cretaceous relicts. Some of them had relatives in Late Cretaceous amber of the Northern Hemisphere.

**KEY WORDS:** Aphidoidea, *Buchnera aphidicola*, coevolution, Formicidae, geographical and geological distribution

“After having studied aphids during half a century I have met riddles difficult to solve and questions difficult to answer. These questions are concerning the following fields: The choice of host plants, host alternation, variation in sizes of populations, morphology, geographical distribution, palaeontology and evolution” Ole E. Heie (2009)

The peculiar features of the geographical distribution of aphids in the Late Eocene formed the riddle that drew the senior author to this problem. One of the considerable differences in the composition between the Baltic and Rovno amber faunas of the same age is the fact that aphids are underrepresented in the Rovno fauna. They make up 4–6.5 % of all insects (excluding Entognatha) in the representative Baltic amber collections from Sambia (Sontag 2003; Perkovsky *et al.* 2007) and only 2.8 % in the representative Rovno amber collection of the Schmalhausen Institute, National Academy of Sciences of Ukraine; the source area of Late Eocene Baltic amber was Russo–Scandia (northern Subparatethys Sedimentation Province). The source area of Late Eocene Rovno amber is the Volyn region on the south coast of



Subparatethys (Ivanov *et al.* 2016, fig.1), so the climate of the Rovno amber forest (Ukraine) was warmer than that of the Baltic amber (Perkovsky *et al.* 2010).

These differences are especially notable in the representation of *Germaraphis* Heie, 1967, which is a dominant genus of late Eocene amber aphids. These aphids make up 5.7 % of all the insects (excluding Entognatha) in the representative collection of Baltic amber from Sambia that is stored in Brighton, and 2.4 % of all the insects in the small section of a representative collection of Rovno amber (see Perkovsky *et al.* 2012). The modern counterparts of the longirostred ant-attended aphids of the genus *Germaraphis* (Perkovsky 2011; LaPolla *et al.* 2013) are aphids of the genus *Stomaphis* Walker, 1870, which are obligate ant symbionts (Heie & Wegierek 2009).

## 1. Aphids: geographical distribution and the evolution of their life cycle – prevailing hypotheses

The patterns of geographical and geological distribution are among the main riddles of aphids. Scientists have been puzzled

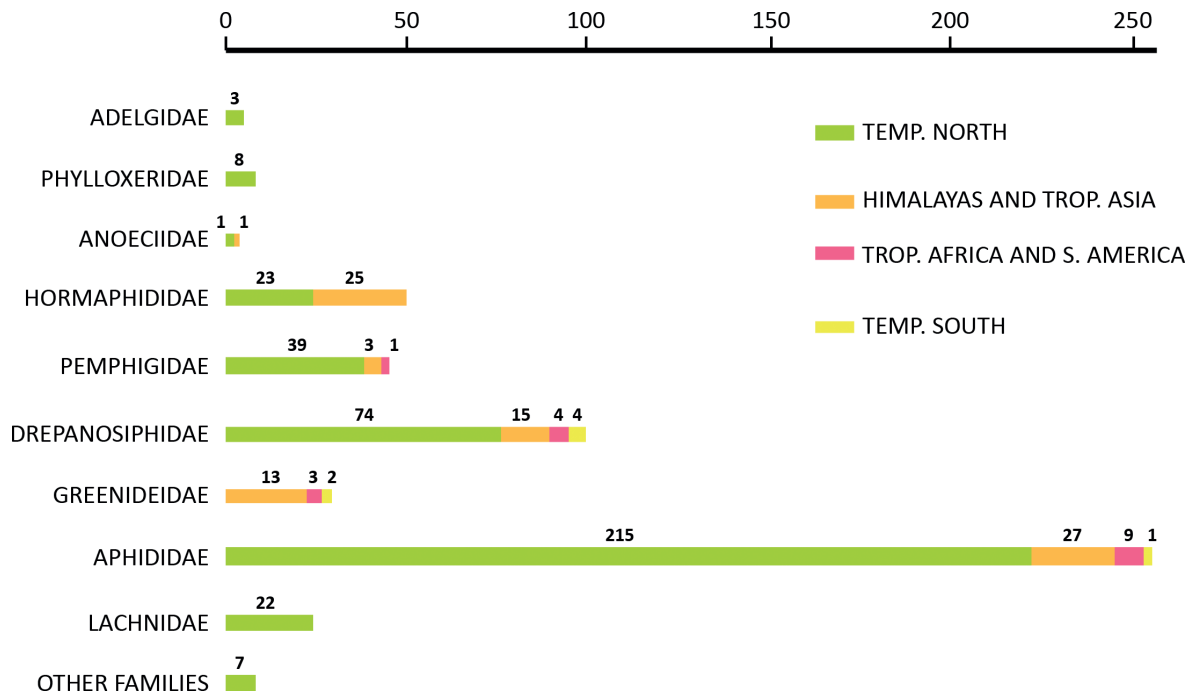


Figure 1 The recent distribution of aphids: geographical origins of 500 aphid genera. (After Heie 1994).

by the fact that the world distribution of aphids does not seem to follow a typical pattern (there are numerous species in temperate zones of the northern hemisphere, but only a few species in the southern tropics). It was Heie (1994) who discussed a previously formulated hypothesis about the adaptation of aphid life cycles to seasons of temperate zones (Bodenheimer & Swirski 1957). He also commented on the hypothesis focusing on difficulties aphids encounter in the tropics (Dixon 1985, 1987; Dixon *et al.* 1987), indicating that the low species richness in tropics is due to high plant diversity, making the chance of successful migration between conspecific hosts very small. Challenging the latter, Heie stated that aphids in tropics are monophages. He was equally skeptical of Dixon's other proposals, such as the method of comparing areas rich and poor in fauna (Heie 1994). Moreover, Heie pointed out that most aphids occurring in tropical and southern temperate countries are introduced, cosmopolitan and, most often, polyphagous species and they cope quite well with the climate. He also showed that the tendency to avoid the tropics is not equally strong in different aphid families (Fig. 1).

It should be noted that all other Hemiptera Sternorrhyncha (coccids, white flies and psyllids) do remarkably well in the tropics, although their sizes, mobility and feeding habits are comparable to those of aphids (Evans 2008; García Morales *et al.* 2016; Ouvrard 2016).

According to Heie (1994), aphids became cyclically parthenogenetic while adapting to the warm climate of the Mesozoic. This idea seems quite plausible, because during most of the long history of aphids' existence (more than 250 million years), the temperatures were considerably higher than those which have prevailed for the last 25 million years. Originally, life cycles were adapted to dry periods (Heie 1994). However, this does not deny the fact that aphids, except during the earliest stages of their history, prefer cooler climates (Kania & Wegierek 2008; Martin *et al.* 2016).

The geological and climatic changes of the second half of the Cenozoic resulted in the differentiation of climatic zones on the Earth. This, in turn, influenced the development of northern hemisphere vegetation. Herbs and open grassy areas became available and aphids adapted their life cycle (and

some morphs; Heie 1994) to maximise the use of those plant nutrients (Dixon 1985). It may also help to explain the fact that the Aphididae family, the aphid group richest in number of species, did not spread into the tropics. According to Heie (1994), Drepanosiphidae, Hormaphididae and Greenideidae had already adapted to a warmer climate in the Tertiary, and the south temperate aphid fauna is more similar to that of the Tertiary than to that of the northern temperate zones; great evolutionary events take place in northern hemisphere (but see below). The southern hemisphere aphid fauna became quite distinct because of the tropical barrier (Heie 1994). We disagree with Heie (1994) that Drepanosiphidae, Hormaphididae and Greenideidae "had already adapted to a warmer climate in the Tertiary". The majority of drepanosiphid subfamilies (all including myrmecophiles, see below) were never found in the tropics or temperate regions of the Southern Hemisphere.

Another hypothesis (e.g., Blackman & Eastop 2006) stressed that the stimuli that induce the sexual phase (a long scotophase and decreasing temperature; Kawada 1987) are lacking in the tropics, and the very lack of stimuli makes aphids lose the sexual phase of the life cycle, further resulting in their loss of the evolutionary potential dependent on the gene recombination. However, the occurrence of sexuales is a complicated phenomenon not triggered exclusively by a photoperiod. For example, *Mindarus* Koch, 1857 in Poland produces sexuales in June when the day is longest (Heie 1980, p. 85: "... sexuales are born in early summer or in mid summer..."). The aphids living under bark in complete darkness (e.g., *Stomaphis*) also produce sexuales in autumn (Depa *et al.* 2015). A similar situation was observed in the genus *Pterochloroides* Mordvilko, 1914 and the species *P. persicae* (Cholodkovsky, 1899) in which, at the same latitudes, sexuales occurred in the mountains; whereas anholocyclic forms prevail in wet lowlands. ("The place of collection [of sexuales] – Isfahan (Esfahan) is located at 1590 m above sea level, in the foothills of the Zagros mountain range. Isfahan has an arid climate with hot summer and cold winter. This type of climate is favourable for the occurrence of sexual forms of aphids. Although *P. persicae* species, a serious pest of orchards, is reported from numerous locations, records of sexuales are extremely rare." Wieczorek *et al.* 2013, p. 98).

Another example of the complicated control of aphid sexuality represents the fact that anholocyclic species transferred from warm climates do not restore sexual forms, and the attempts to produce them under controlled experimental conditions are mostly unsuccessful (Durak & Durak 2015: in *Cinara tujafilina* (del Guercio, 1909), breeding males were obtained only in the 126th generation in the climatic chamber). To cope with environmental uncertainty, aphids have evolved different types of polyphenism (Tagu *et al.* 2005), so finding alternative stimuli responsible for inducing the sexual phase is not impossible. We remember that the primary aphid adaptation has been the one towards using the dry *vs.* wet season alteration. Nevertheless, most aphids lose their sexual forms in the tropics, a fact deserving explanation (see discussion below). In a temperate climate, when days become shorter in the autumn, most aphid clones switch to the production of sexual forms, because only eggs and not nymphs can survive low temperatures (Simon *et al.* 2002). In contrast, the warmer, equable climate of the Cretaceous through to the Eocene should permit nymphal hibernation, which suggests different stimuli were used to induce the sexual phase at that time as compared to the present. This point of view confirms the fact that aphid males are unexpectedly frequent in Burmese amber (Poinar & Brown 2005).

All three mentioned hypotheses concentrated on external factors in trying to account for the distribution of aphids. In this paper, it also seems important to indicate the internal limitations which are due to the strong dependence of aphids on their symbionts. This has influenced the geographical distribution of aphids, at least since the mid-Cretaceous, as was suggested in our earlier paper (Perkovsky 2012).

## 2. Internal factors influencing aphid distribution

### 2.1. Importance of functional integration with symbionts and biggest extinction

Aphids and symbiont genotypes have evolved to form a functionally integrated whole, so that in many contexts both genomes need to be considered (Hales *et al.* 1997). At low latitudes, aphids usually switch to obligate parthenogenesis, and if their initial genetic diversity is low, they soon lose their evolutionary potential. It may prove to be one of the factors determining the world distribution of the group. Some aphid families (Hormaphididae and Greenideidae) managed to cross the climatic barrier, thanks to individual adaptations discussed below.

Fossil remains of aphids are known from at least the Middle Triassic (but see Szwedo *et al.* 2015); however, they have been mostly described on the basis of isolated wings. The only exception is the family Dracaphididae (Naibioidea), but its assignment to aphids is disputable (Heie & Wegierek 2011). Slightly more numerous aphids are known from the Jurassic, including the oldest undoubted aphid body fossils; but overall, the fossil record is rather scarce and a significant abundance of aphid fossils begins from the Early Cretaceous.

We believe that at least some of the answers are linked to yet another riddle of aphids, which as yet has attracted no attention: their mass extinction in the Late Cretaceous. Most Cretaceous families became extinct at the end of the epoch. The richest Cenozoic fauna is known from Baltic amber (Late Eocene) and, with the exception of Elektraphididae, represents exclusively Recent families, which originated at the beginning of the Cenozoic (with two families dating from the end of the Cretaceous; for stratigraphic distribution of families with number of species included, see Heie & Wegierek 2011, p. 71).

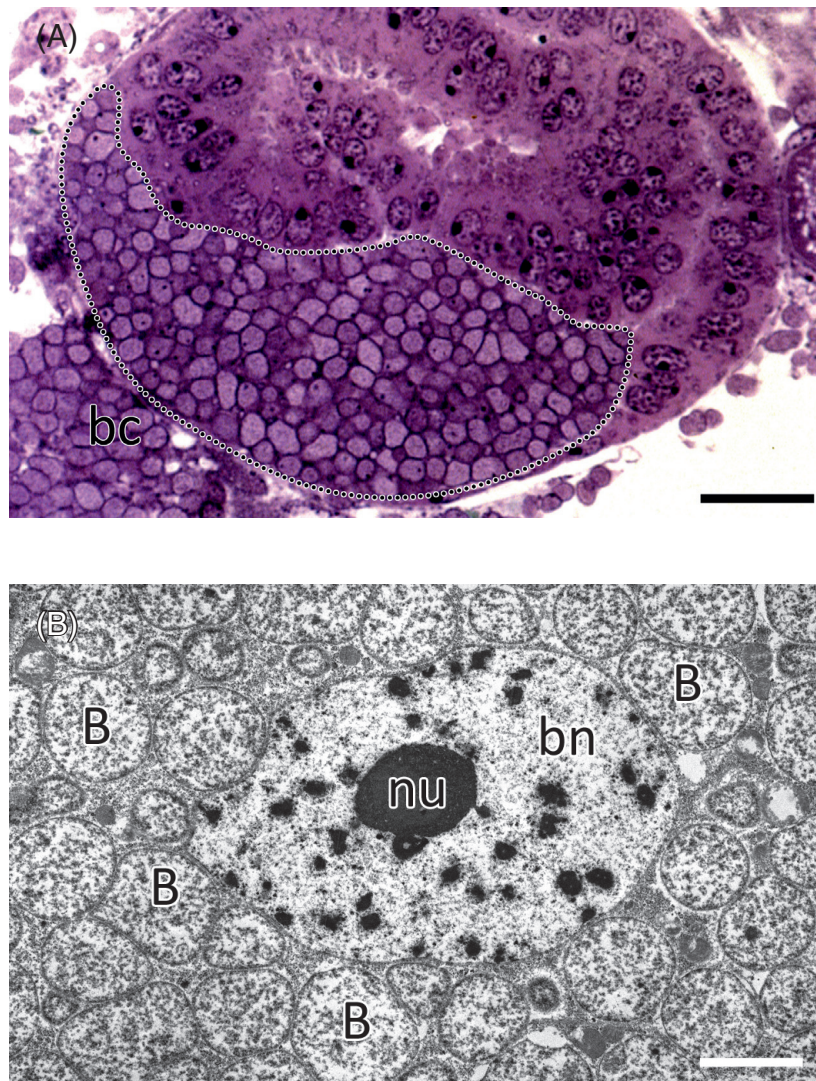
### 2.2. Susceptibility of primary obligate aphid symbionts to high temperature

Currently, the distribution of aphids (Heie 1994; Fig. 1) is largely limited by the low thermal tolerance of their primary obligate symbionts – the bacteria *Buchnera aphidicola* (Fig. 2 a, b) (phylum Proteobacteria, class Gammaproteobacteria, family Enterobacteriaceae) (Perkovsky 2012). The genome of *B. aphidicola* is one seventh of that of its close relative, *Escherichia coli*, and these endosymbionts are not able to live outside their host (Charles & Ishikawa 1999). It is believed that associations between insects and their primary endosymbionts are the result of a single ancient infection (Buchner 1965). Within Recent aphids, only representatives of the superfamily Phylloxeroidea do not contain endosymbiotic microorganisms; Adelgoidea may harbour gamma- and/or beta-proteobacterial endosymbionts, but not *Buchnera* (Michalik *et al.* 2014). The gamma-proteobacterium *Buchnera aphidicola* is present in all existing aphids of the superfamily Aphidoidea [classification after Heie & Wegierek Nieto (Nafria & Favret 2011)], except for some representatives of the family Hormaphididae.

Mutualism between aphids and *B. aphidicola* has reached such an extent that both partners have become completely interdependent and neither can reproduce in the absence of the other; the young adult pea aphid *Acyrtosiphon pisum* (Harris, 1766), maintained at 21°C, had  $1.2 \times 10^7$  *B. aphidicola* cells (Simonet *et al.* 2016). These intracellular symbionts synthesise most of the irreplaceable amino acids (Douglas 2006), as well as riboflavin and other compounds that are absent in the phloem sap (Chen *et al.* 2009). During their intracellular existence [it has been firmly proven that symbiotic *Buchnera* was already present in the common ancestors of all Aphidoidea (Moran *et al.* 1993)] for over 200 million years (Unterman *et al.* 1989; Moran & Baumann 1994; Martinez-Torres *et al.* 2001; Moran *et al.* 2008), *Buchnera aphidicola* underwent genetic degradation, which is the almost inevitable fate of all obligate intracellular symbionts. The genome of *Buchnera* became simpler and lost its plasticity as a result of the loss of mobile genetic elements and repetitions (Perkovsky 2012). Genetic recombinations are strongly limited in *Buchnera* populations, which are isolated in the host bacteriocytes and pass through a bottleneck with each new host generation (Charles & Ishikawa 1999). The ability to repair DNA is also limited in *Buchnera* (the part of the genome that provided much of this ability was lost during the shift to symbiosis); the genotype has accumulated too many harmful mutations (a classical example of Muller's ratchet; in some lachnids it has degraded to only 362 protein-coding genes, which is very close to the minimum number even for symbionts that have lost most of their metabolic functions and its repair machinery is more strongly reduced than in other strains (Pérez-Brocal *et al.* 2006).

It is a firmly established fact (Rasnitsyn & Quicke 2002; Kania & Wegierek 2008; Heie & Wegierek 2011; Perkovsky 2012) that as early as the Cretaceous (in contrast to the Late Jurassic), aphids were abundant in areas with a warm temperate climate (such as the Early Cretaceous Baissa Lagerstätte (125–129 Mya; nearly 50 species) and Cretaceous Siberian (85–107 Mya) and Canadian (78–79 Mya) ambers) and very rare south of 40°N palaeolatitude. They are absent in Charentese amber (99–100 Mya); very rare in Early Cretaceous Lebanese (130 Mya; three species, four specimens), Spanish (105 Mya; one specimen) and Late Cretaceous New Jersey (92 Mya; two specimens) ambers; and uniform and rather scant in Burmese amber (98–99 Mya, just above the Early–Late Cretaceous boundary) (Fig. 3). And yet we are not aware of any attempts to explain this fact before Perkovsky (2012). Interestingly, the two Cretaceous aphids from the Southern Hemisphere were





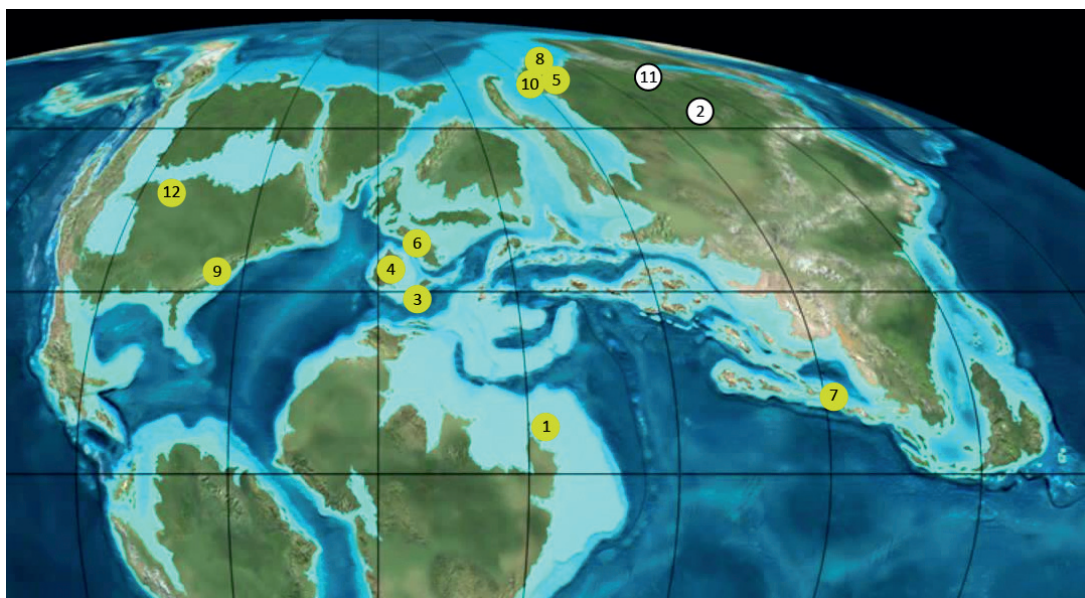
**Figure 2** *Pemphigus spyrothecae*, posterior pole of the young embryo (cross-section), containing numerous cells of bacterium *Buchnera aphidicola* (encircled with dotted line). bc = bacteriocyte. Methylene blue. Scale bar = 20  $\mu$ m. (B) *Pemphigus spyrothecae*, fragment of the bacteriocyte with bacterium *Buchnera aphidicola* (B). bn = bacteriocyte nucleus, nu = nucleolus. TEM. Scale bar = 4  $\mu$ m.

also recorded from two sites that supposedly had a temperate-seasonal climate (Rayner *et al.* 1997; Drinnan & Chambers 1986): in the Orapa Lagerstätte (90–94 Mya) in Botswana and Koonwarra (115–118 Mya); and well inside the Antarctic Circle of the day (Rich *et al.* 1989) in Victoria, Australia (Martin *et al.* 2016).

A major impact of heat on cellular function stems from the degradation of protein tertiary structures. Most cellular organisms have the capacity to respond to heat stress by directing transcription and/or translation to the production of chaperones (Feder & Hoffman 1999). They function by ‘forcibly’ giving other proteins the right configuration. *Buchnera* has four transcriptional promoters, which affect the expression of the five heat-shock genes (Wilcox *et al.* 2003) that are responsible for chaperone synthesis. Some genes, which are usually activated in other organisms under extreme heat, are constitutively highly expressed in *Buchnera*, a feature that has been interpreted as an adaptation that was made in order to maintain the function of proteins that had been destabilised by mutational accumulation (Wilcox *et al.* 2003). *Buchnera* is doomed to the continuous accumulation of its mutation load because it cannot perform homologous recombination, and because of the loss of many of the genes that are important for DNA repair and replication (Funk *et al.* 2001; van Ham *et al.* 2003). Moreover, it

also lacks the chaperones that smooth out the negative effects of mutations somewhat by folding the partly incorrect proteins in the correct way. It is believed that the chaperones of intracellular bacteria are subject to a relatively strong pressure of selection (van Ham *et al.* 2003). The density of *Buchnera* in the host decreases sharply if the temperature rises from as little as 25°–30°C, even in aphids that were collected on the subtropical island of Taiwan (Chen *et al.* 2009). This decrease in *Buchnera* density is accompanied by the fecundity of the host dropping to a small fraction of its former values.

When Dunbar *et al.* (2007) measured the activity of *Buchnera* genes in three laboratory lines of *A. pisum* (Aphididae) at a normal temperature and when overheated, it turned out that in one of the three lines the heat shock in *Buchnera* does not activate the expression of *ibpA*, which encodes a small heat-shock protein that is not only present in bacteria but in almost all living organisms. The *ibpA* heat-shock protein is detrimental in cool conditions, perhaps due to its binding with other *Buchnera* proteins (Burke *et al.* 2010). The line that had this anomaly was produced only five years ago from a ‘normal’ line in which heat shock activates *ibpA*. It was revealed that the anomalous response to overheating was caused by a mutation (most probably single) that appeared less than five years ago. In these bacteria, adenine is absent in the chromosome at



**Figure 3** Geographic distribution of the studied Cretaceous assemblages (from Rasnitsyn *et al.* 2016, with changes). Map modified from Blakey 2011. 1 = Levantine amber belt; 2 = Russia, Eastern Siberia, Baissa; 3 = Spain Escucha Formation; 4 = Spain Utrillas Group; 5 = Russia, Taimyr Peninsula, Begichevo Formation; 6 = France, Charentese amber; 7 = Myanmar, Hukawng Valley; 8 = Russia, Taimyr Peninsula, Dolgan Formation; 9 = United States, Raritan Formation ‘New Jersey amber’; 10 = Russia, Taimyr Peninsula, Kheta Formation; 11 = Russia, Far East, Obeshchayushchiy; 12 = Canada, Alberta and Manitoba, Foremost Formation.

the site to which the promoter  $\sigma_{32}$ , which activates the gene *ibpA* (and other heat-shock genes), attaches when the animal is under stress (Dunbar *et al.* 2007).

Investigations with other laboratory lines of the pea aphid revealed another case of the independent appearance of the same mutation in one of them and indicated that the mutation appeared in two lines in 2001 and 2005 (Dunbar *et al.* 2007). It turned out that the mutation is rather common (present in 13–21 % of individuals) in *Acyrtosiphon pisum* from the northern US states of New York and Wisconsin, which have rather cold summers with daytime temperatures sometimes never reaching 35° for several years; but which were absent in Arizona and Utah, where daytime temperatures in the summer normally reach 35°C.

Experiments have confirmed that the gene *ibpA* is not activated by overheating in all of the mutants, although it is activated by overheating in all of the lines that lack this mutation. The other heat-shock genes were activated in exactly the same way in both the mutant and normal *Buchnera*. It has also been proven experimentally that a mutation arising in the bacterial symbiont affects the viability and reproductive potential of the symbiotic superorganism. Aphids that had both mutant and normal *Buchnera* were grown under different temperature conditions – some at a stable temperature of 15°C or 20°C and some heated for three hours to 35.5°C two days after hatching. It turned out that after overheating, the aphids that had mutant symbionts became almost infertile, while the aphids with normal symbionts bred quite well (only 17 % were infertile). However, at a constant temperature of 15°C or 20°C, the aphids that had mutant symbionts had clear advantages. They began breeding earlier and produced more offspring on average.

Additional experiments have shown why brief overheating is so fatal to aphids with mutant *Buchnera*. When the temperature rises to 35.5°C or higher, many bacterial symbionts die rapidly and one aphid ends up having only about 1000 bacteria (about 0.001 of the norm) and these insects weigh only 40 % as much as the control group (‘normal’ aphids weigh 80 % as much as the control group after overheating;

Dunbar *et al.* 2007). Thus, the mutant *Buchnera* gives advantages to aphids at constant and rather low temperatures, while ‘normal’ *Buchnera* can be beneficial only when overheating is possible (chaperones make up 10 % of all of their protein).

The direction of selection in natural populations of aphids may probably change depending on the climate, and even on the season; sometimes aphids with normal symbionts are favoured by selection and sometimes aphids with mutant symbionts are favoured. As a result, both genetic variants are preserved in natural populations and none of them can completely replace the other. However, this polymorphism is especially effective if overheating happens rather seldom. Recently, it was found (Moran & Yun 2015) that *Buchnera* in most *A. pisum* matrilineal lines evolve to the heat-sensitive genotype after extended laboratory culture at 15–20°C, reflecting a high mutation rate at this allele and selection favouring this genotype under cool conditions. The replacement of *Buchnera* not producing *ibpA* by a heat-tolerant mutant (injection after heat treatment was used) greatly increased the heat tolerance of the *A. pisum* aphid, demonstrating a major effect of the symbiont genotype on host fitness (Moran & Yun 2015).

### 2.3. The dependence of aphid’s sexual generation on *Buchnera* susceptibility to overheating

Approximately 30–40 % of the aphid species that have been described include both cyclical-parthenogenic and asexual clones (Fig. 4) (Moran 1992; Dixon 1998; Simon *et al.* 2010). The strictly asexual species (or clones) tend to be distributed in low-latitude regions. Strictly asexual generations have only been described in approximately 3 % of all aphid species (Simon *et al.* 2002). Dramatically varying population sizes make parthenogenesis much more beneficial. The same species (*Macrosiphum rosae* (Linnaeus, 1758) have cyclic parthenogenesis in Norway, cyclic in some populations and obligate in other populations in England and exclusively obligate in the Canary Islands (Wöhrmann & Tomiuk 1988); similar differences are also known for many other species (Hales *et al.* 1997; Dedryver *et al.* 2001).



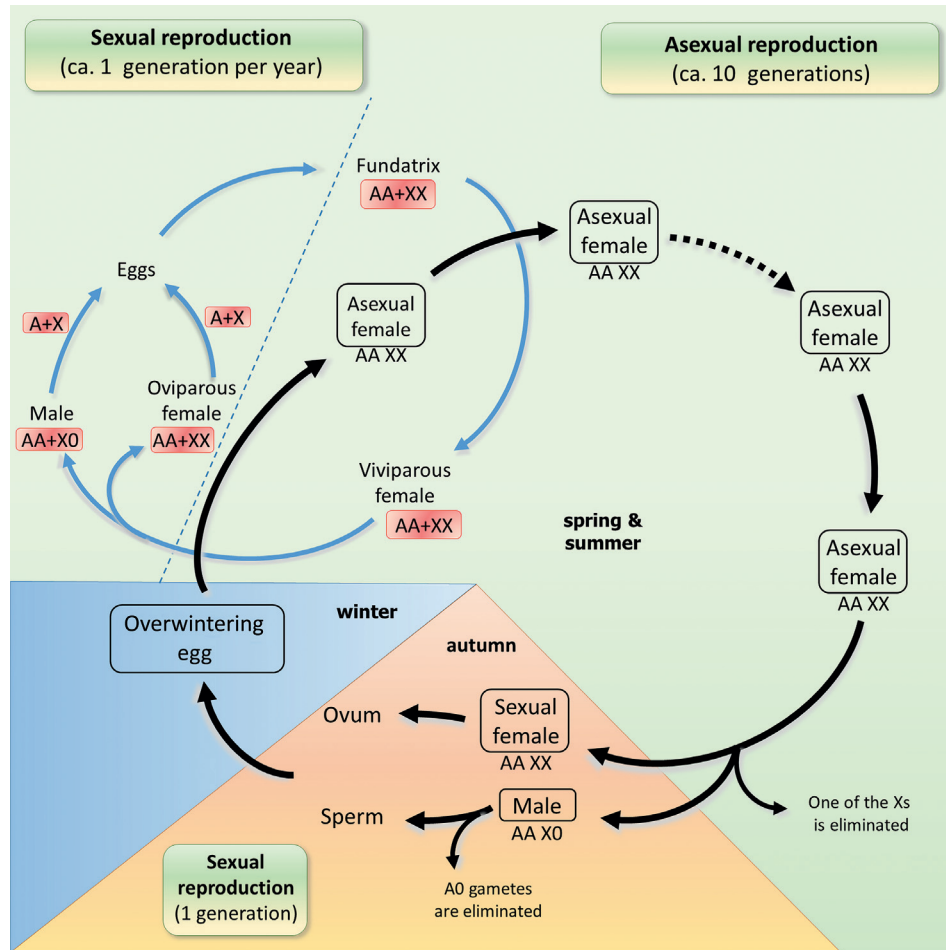


Figure 4 Life cycles of the aphid.

The genetic diversity of *M. rosae* also sharply decreases from north to south (Wöhrmann & Tomiuk 1988), and has a negative correlation with the average temperature of July. The cold resistance of overwintering eggs is considered to be very important, but live parthenogenic individuals can resist temperatures above  $-8^{\circ}\text{C}$  or  $-10^{\circ}\text{C}$  (Simon *et al.* 2010).

In species that had been introduced into the tropics, only obligate parthenogenesis is found. Penetrating into the tropics, aphids simply lose the sexual phase of their life cycle, as well as their evolutionary potential to the extent of its dependence on gene recombination. Those that omit sexual reproduction from their life cycle become encumbered by the negative effects of exclusively asexual reproduction; e.g., mutation load and sluggish adaptation (Smith & Maynard-Smith 1978; Otto 2009; McDonald *et al.* 2016). It should be noted that in nonaphidid aphids (*sensu* Heie & Wegierek 2009), sexual reproduction is very closely linked to the primary host plant (Hardy *et al.* 2015). Hardy *et al.* (2015) revealed a very strong correlation between heteroecy and holocycly (i.e. sexual reproduction) in nonaphidid aphids, which is indicative of the presence of formidable developmental constraints that oppose adaptation to changing host-plant communities.

We suggest that the loss of the full life cycle of aphids in the tropics is due to *Buchnera*'s low tolerance to overheating, and by the peculiar life cycle and sex determination of aphids. The fact that 10–30 parthenogenic generations of aphids are followed by only one sexual generation means that the prerequisites are high for switching to obligate parthenogenesis when the probability of the overheating of the sexual generation is also high; whilst the decreased viability of males additionally decreases their chances of surviving overheating.

Male aphids have a single X chromosome and all of the mutations associated with this chromosome manifest themselves. As a result, the number of malformations is huge (Depa *et al.* 2015) and the general viability of males is decreased. This is the likely explanation of the two-fold mortality rate of male embryos from the recessive mutations that are lethal at any temperature. Let us also remember that even in parthenogenic females without mutations of *ibpA* in *Buchnera*, 17 % of the individuals became infertile after three hours of overheating; in males the proportion should be much higher. Warm temperate regions demonstrate an instable equilibrium of holocyclic and anholocyclic clones of comparable abundance. The sexual generation develops here in rather cool environments with a low risk of overheating, in contrast to the parthenogenic ones which experienced some 40 % loss, even in northern Germany (Ma *et al.* 2004).

Further south, the loss becomes inevitable in sexual generations, as well resulting in the overwhelming prevalence of obligate parthenogenic populations. In our opinion, the loss of sexual generation due to overheating accelerates genetical drift and reduces the already low genetic diversity in populations. The occasional complete death of sexual generations due to genetic problems in the symbiotic bacteria reduces genetic recombination dramatically beyond the level at which loss of sexual generations cannot be compensated by the two-fold fecundity preference of the thelytoky (female-producing parthenogenesis).

There are also aphids with sexual forms occurring concurrently with parthenogenic females (e.g., many species in the family Greenideidae; Tomiuk *et al.* 1991); we discuss this phenomenon below.

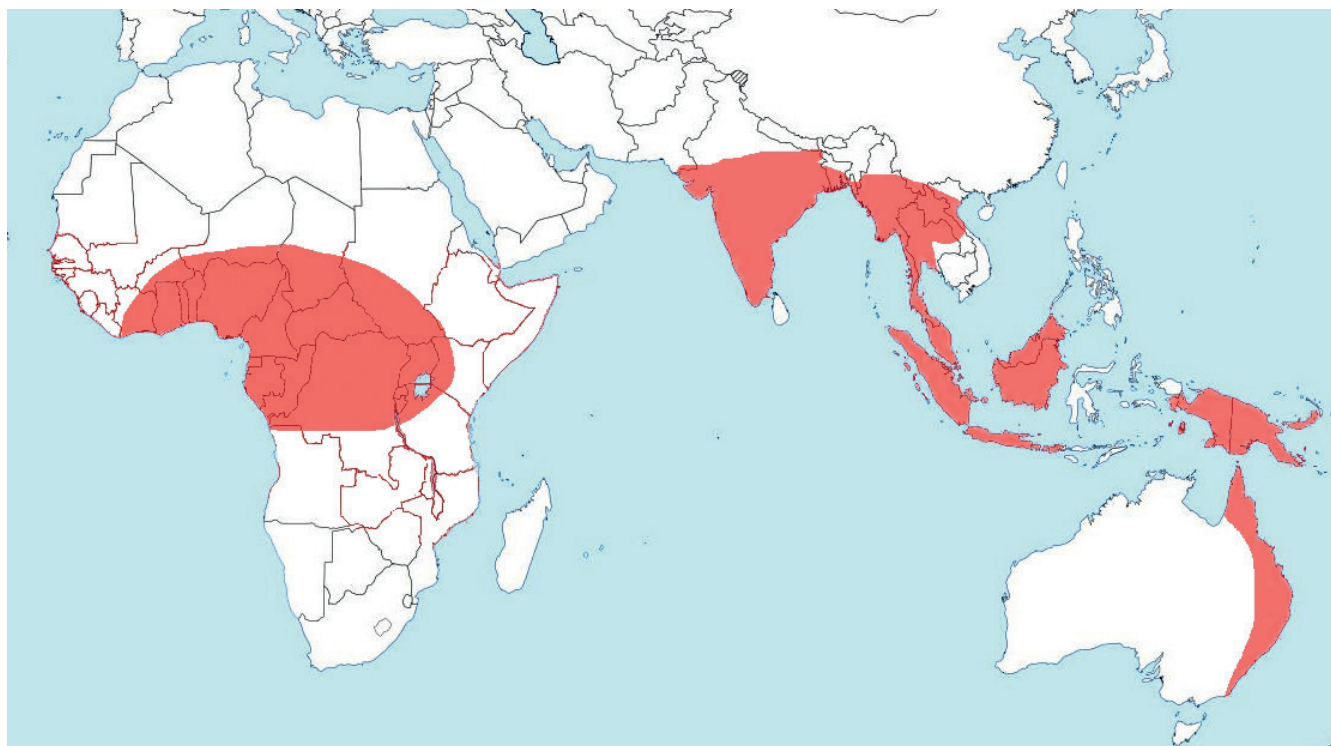


Figure 5 The world distribution of *Schoutedenia ralumensis* Rübssaamen, 1905 (data from Blackman & Eastop 2006).

#### 2.4. Modifications of aphids in tropics to save a sexual generation

In our opinion, the cyclic parthenogenesis can be preserved in the tropics when the males are freed from their dependence on *Buchnera*, when sexual forms occur concurrently with parthenogenetic females and/or when the sex-determination system changes.

Let us discuss the families Hormaphididae and Greenideidae, which have, respectively, half or all of their genera distributed in the tropics. Hormaphidid aphids are gall-forming species that often possess soldier nymphs. Males and the egg-laying females that mate with them are wingless and dwarfish in Hormaphididae and aphagous in many species. The best known, most successful and best studied hormaphidid aphids are cerataphidines. Their colonies include up to a million individuals that cause galls in their primary hosts. The diversity and relative abundance of cerataphidines in the tropics are determined by their sociality, which made it possible for them to evolve aphagous males (Kurosuo *et al.* 2006) which, in turn, makes them independent from *Buchnera*. The life cycle of gall-forming cerataphidines makes the search for mates easier; hence, adult males of the species that have *Buchnera* can be short-lived and not dependent on additional feeding.

In those species of *Cerataphis* Lichtenstein, 1882 that have preserved *Buchnera*, the males and egg-laying females are aphagous, and so their survival is not affected by the death of *Buchnera* from overheating. This made it possible for some cerataphidines to replace *Buchnera* with yeast-like symbionts (Pyrenomyces; see Fukatsu *et al.* 1994). Three genera of Cerataphidinae are an example, with one of these genera still preserving two species of *Buchnera*. The nymphs of egg-laying females that have the fungus are not aphagous and have a long rostrum, while in the males or females without the fungus, the rostrum is always short or absent. The importance of male aphagy is confirmed by the fact that it also evolved independently in the second family of gall-forming social aphids, Eriosomatidae. Moreover, in social eriosomatids not only

males, but also soldiers, are aphagous, which could increase the evolutionary potential of the taxon.

Thus, the high mortality of *Buchnera* from overheating leads to the replacement of the bacterial symbiont with a fungal symbiont, the emergence of aphagous males or the increased role of secondary symbionts of the genus *Serratia* (Chen *et al.* 2000; Montllor *et al.* 2002; Russell & Moran 2006), which are also transmitted transovarially (Michalik *et al.* 2014).

There is no reason to infer the obligate association between cerataphidine males and *Buchnera* lost as early as the Miocene, since all cerataphidines, in contrast to the aphagous eriosomatid males, still have fully-fledged bacteriocytes (Braendle *et al.* 2003). The late breakup of the association between Cerataphidinae and *Buchnera* (it can be assumed that their sexual generation was not yet aphagous in the Miocene) can explain the extinction of American cerataphidines. Cerataphidines are widespread in the Old World in all areas in which their primary host plant, *Styrax*, is present. Although *Styrax* species are still abundant in the New World, cerataphidines became extinct after the Middle Miocene.

To preserve sexual reproduction, greenideids have not only switched to the concurrent existence of sexual and parthenogenetic generations that decrease inbreeding (Tomiuk *et al.* 1991), which is important because of greenideid poor flight capacity. They also changed the XX–X0 sex-determination system that is characteristic of all other aphids. The genetic sex-determination system of some greenideids, which is unique amongst Paraneoptera (Gavrilov-Zimin *et al.* 2015), was discovered by Hales (1989). She found multiple X chromosomes fused with autosomes (X1 + A, X1, X2 + A, X2) in *Schoutedenia* Rübssaamen, 1905 (Fig. 5), so as all four X chromosomes appear to be of different length (Hales *et al.* 1997). The study of male meiosis has shown that X1 or X2, apparently at random, gave up its attached autosome which was then lost in the inviable (not X-bearing) spermatocyte (Hales *et al.* 1997). This special system of sex determination should kill half of the gametes which, hence, should be counterbalanced with another, yet unknown, adaptation.

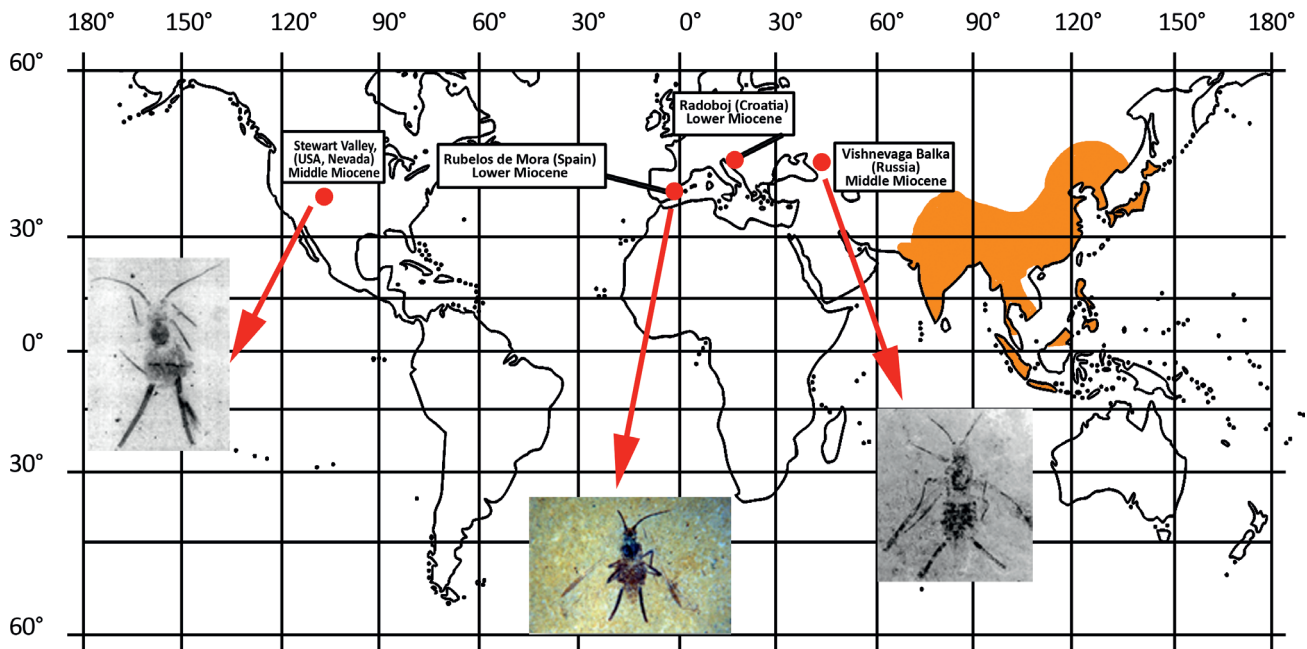


Figure 6 The recent (orange) and past (red) world distribution of Greenideinae.

The X chromosomes of aphids consist mostly of heterochromatin, and their composition, apart from rDNA (located in X chromosomes in all aphids), is not discussed. Thus, males have half as much rDNA as females (a single copy rather than two copies). This may be the explanation for the two-fold mortality rate of male embryos from mutations that are lethal at any temperature and for the very low growth rate of males (Dixon et al 1998). In *Schoutedenia*, rDNA is transferred to paired autosomes; i.e., the amount of rDNA is doubled and any lethal recessive mutations of rDNA have no effect on most males. Thus, at least the main advantage that has been acquired by *Schoutedenia* is fairly obvious. The shift of rDNA into autosomes is probably of principal importance; male lachnids of the genus *Stomaphis* also have two X chromosomes, but 32 % of adult males show various morphological aberrations, many of them very striking (Depa *et al.* 2015, figs 11–13), which have not yet been recorded in *Schoutedenia*.

Because of above adaptations, greenideids have advantages at high temperatures, but in cooler climates they are poor competitors. The family has appeared in the fossil record since the Early Miocene (Fig. 6) and do so in abundance (greenideines reached 18 % of all of the aphids recorded in Rubielos de Mora, Aragon), thus apparently reflecting the increasing seasonality of the climate beginning with the Oligocene. During the Middle Miocene Climatic Optimum, the number of recorded greenideines reached 25 % of all of the aphids recorded in Stewart Valley, Nevada; five of the six aphids recorded from the pre-optimum Radoboj, Croatia, also belong to Greenideinae; extant greenideids (all subfamilies) account for 18 % of aphids found only on Java; north of the Equator, they are more poorly represented (at most 13 % in India and 5 % in Sri Lanka). The family is absent in the fauna of North America and Western Eurasia after the Middle Miocene (Fig. 6). Greenideids are easily distinguishable and, therefore, the absence of records of this family from pre-Miocene sites, including Fushun and other Early and Middle Eocene localities that had high temperatures in July, actually supplies evidence of the relatively young age of this taxon.

As indicated above, an alternative explanation of the scarcity of aphids in the tropics is the huge diversity of plants, which makes it difficult to locate a particular host plant species (Dixon

1998). Dixon (1998) supported this claim by noting the abundance of greenideids in a plantation near Kolkata at 30°C and above. The fact that the aphids in the plantation had switched to obligate parthenogenesis (Agarwala & Dixon 1986) apparently did not bother him. We suppose that the sexual reproduction retained in the Himalaya secures the evolutionary potential of northern Indian populations.

Middle Miocene greenideines are known also in Stavropol, Russia (Fig. 6). However, the family make up only 3 % of all aphids there, although the fossil site also occurred not far from the Miocene optimum at that time. The genus *Stomaphis* was dominant in Stavropol (Wegierek & Mamontova 1993). Most representatives of this genus probably survive overheating rather well (many of them live under bark). They include dwarf arostrate non-feeding males (many of them living on underground parts of the trunk base or under bark; Depa *et al.* 2012) and also obligatory myrmecophiles, which was probably the main reason of their success. *Lasius* ants, the permanent symbionts of *Stomaphis*, also increased their abundance in Stavropol in parallel to that of *Stomaphis* (Dlussky 1981; Wegierek & Mamontova 1993). Greenideidae have never evolved strong association with ants.

The riddle of aphid biogeography has one more aspect, a poverty of the southern temperate realm in aphids. To try and decipher it, we need to consider the event of the Late Cretaceous aphid extinction (Fig. 7), which has never been discussed by aphidologists. The extinction at the Cretaceous–Palaeogene boundary is traditionally explained by floristic changes; moreover, Late Cretaceous aphids were considered to be similar to Early Cretaceous ones (Kononova 1976). Recently, Early Cretaceous aphids have been studied (Heie & Wegierek 2011b) much better than Late Cretaceous ones, and the results are very different. Eighty-four Early Cretaceous aphid species are now known from 414 specimens, whilst 44 Late Cretaceous species are known from only 61 specimens. Since thelaxids, which are not known from the Late Cretaceous (Fig. 7), exist to this day, it appears that none of the Early Cretaceous superfamilies, and only 58 % of families (seven out of 13), became extinct at the time of the Cretaceous Terrestrial Revolution. Three strictly Early Cretaceous families are known from only seven specimens, and if the Early and



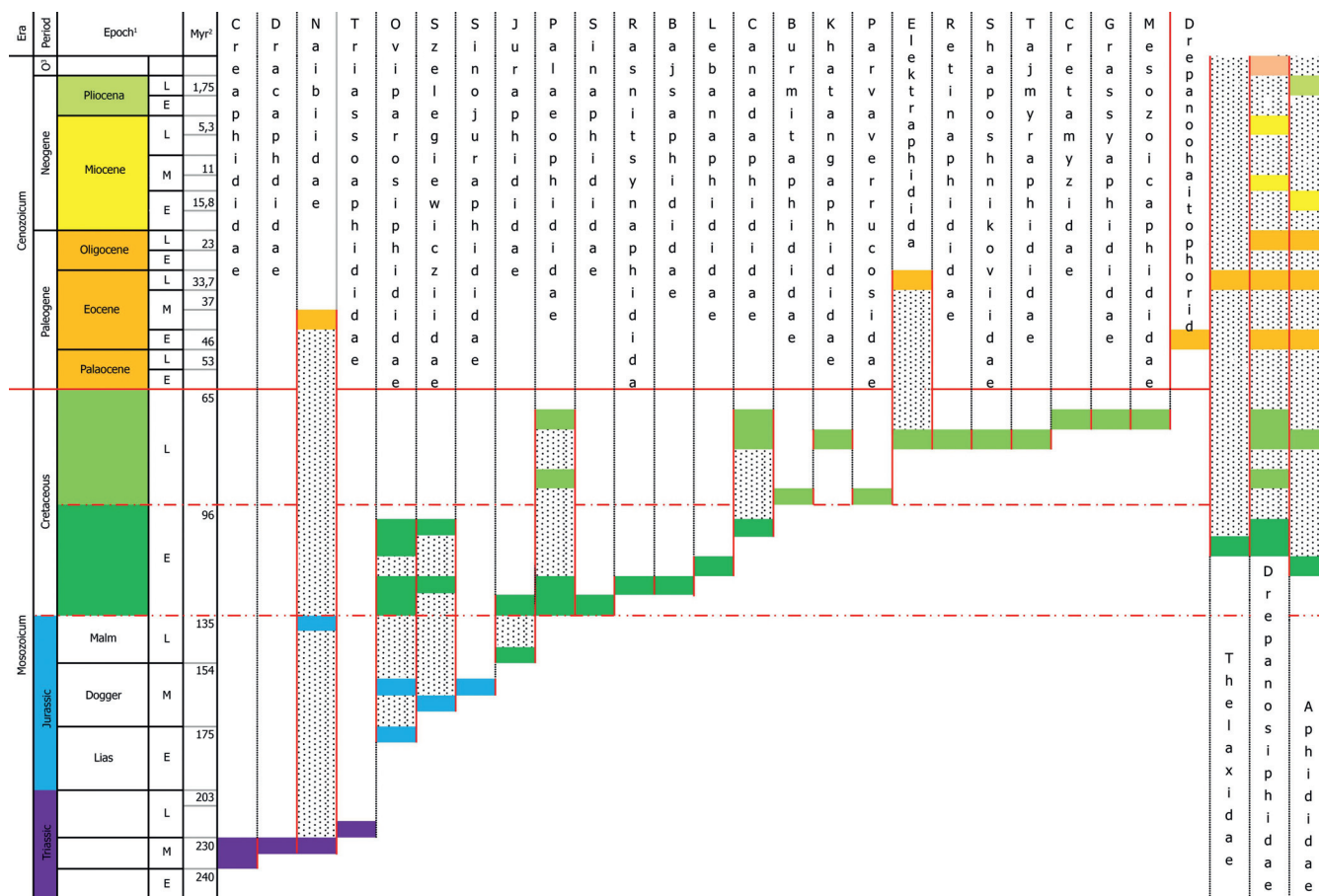


Figure 7 The distribution of aphid families in geological time.

Late Cretaceous faunas were studied at similar levels, the proportion of families that are known to have become extinct in the Early Cretaceous would be even smaller.

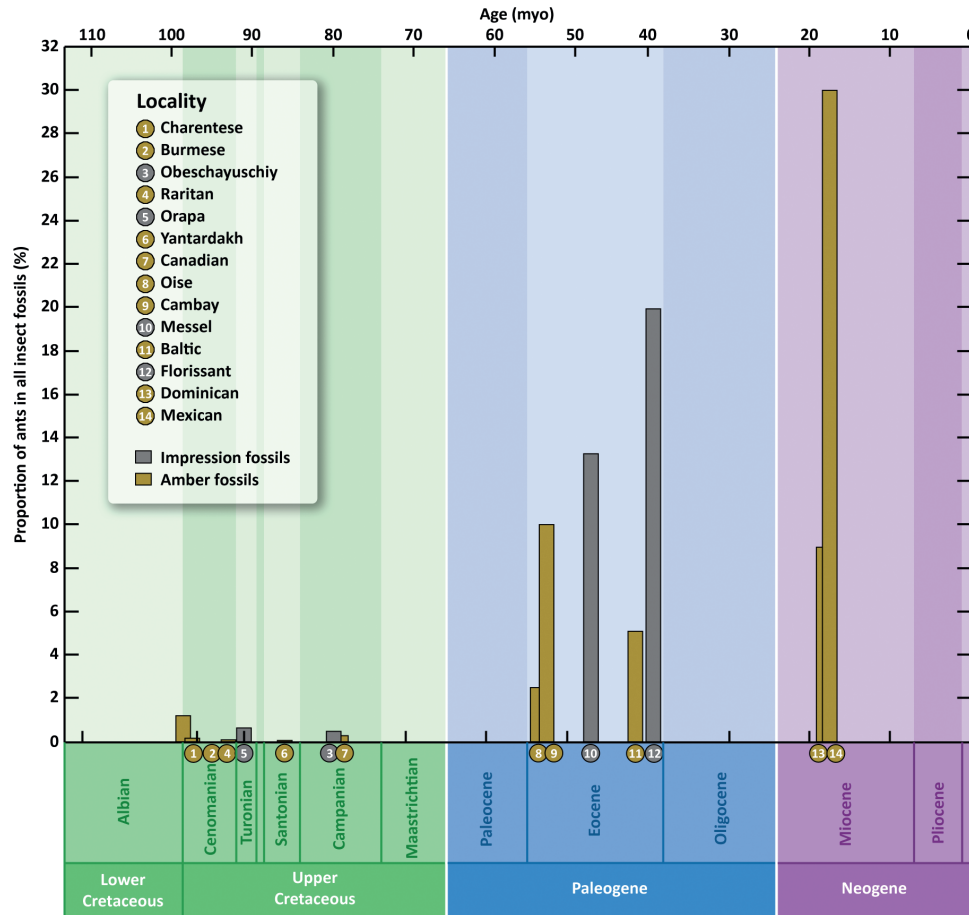
The Cretaceous Terrestrial Revolution is characterised by the rise of the angiosperms (Dilcher 2000; Davies *et al.* 2004; Magallon & Castillo 2009; Wang *et al.* 2009; Coiffard *et al.* 2012). It had huge ecological consequences, including the extinction of the closest counterparts of the angiosperms Bennettitales and Czekanowskiales, as well as most Ginkgo-phyta and most Gnetophyta (Ponomarenko 1998). This means changes can be expected in the family composition of Late Cretaceous aphids as compared to Early Cretaceous ones. However, the much deeper aphid extinction at the Cretaceous/Paleogene (K/Pg) boundary cannot be explained by the floristic changes. Half of the 30 plant morphotypes represented by more than 30 specimens were preserved on the best studied sites of North America (western North Dakota and western South Dakota), at the transition from the Cretaceous to the Palaeogene (Johnson 2002). The floristic associations of the Russian Far East and high latitudes of Asia and North America also provide no evidence of any considerable reorganisation of the terrestrial flora at the Cretaceous–Palaeogene boundary (Herman *et al.* 2009).

In contrast to the modest floristic turnover at the K/Pg boundary, the aphid faunal transformation was drastic there. In the Kheta Formation of Taimyr (Santonian), 78 % of the aphid families are extinct (Perkovsky & Wegierek 2015). For comparison, all of the Kheta Formation arthropod assemblages include 16 % extinct families, and non-aphids include only 11 % extinct families; 20 % of all arthropod families are unknown from the Early Cretaceous (Burmese and Charentese

amber dated near the boundary of the Early and Late Cretaceous are calculated as Early Cretaceous; Rasnitsyn *et al.* 2016). In Canadian amber (Campanian), the proportion of extinct aphid families is 83 % and in all of the aphid fauna of the Late Cretaceous, this proportion is 86 %.

To sum up, 79 % of Late Cretaceous aphid families became extinct at the Cretaceous–Palaeogene boundary. Almost all tajmyraphidoids (five families) and palaeoaphidoids (two families), as well as three of the five families of aphidoids known from the Late Cretaceous, also became extinct. Aphids of the first two superfamilies lacked the siphunculi that had a signalling function and without these siphunculi, it proved impossible for aphids to survive the increased pressure of predators. This was particularly fatal because almost all Late Cretaceous aphids had a long or very long rostrum. It is well known that even among members of the same genus, a longer rostrum increases both the time required to escape a predator and the rate of the destruction of one colony by predators by a considerable factor (Shingleton *et al.* 2005). This particularly concerns the longirostred aphids (Depa *et al.* 2015). In the Late Cretaceous, only three (*Cretamyzus* Heie, 1992 (Cretamyzidae), *Calgariaphis* Heie, 1992 in Heie & Pike 1992 (Mesozoicaphididae) and larvae II type) of the 12 morphotypes of wingless aphids had the proboscis not longer than the body (Kononova 1977; Heie & Pike 1992); and even in winged aphids, such as *Nordaphis* Kononova, 1977 (Drepanosiphidae, Lizeriinae) and *Palaeoforda* Kononova, 1977 (Aphidomorpha *incertae sedis*) the proboscis is longer than the body, and it almost reaches the apex of the abdomen in some representatives of other families.

At the same time, elongation of the rostrum was a pre-adaptation to symbiosis with ants (Shingleton *et al.* 2005;



**Figure 8** Ant abundance in different Lagerstätten (from all insects). (Redrawn from LaPolla *et al.* 2013, fig. 1, with many changes)

Perkovsky 2012). It is noteworthy that the only recent genus of aphids that has a rostrum longer than the body (*Stomaphis*) depends entirely on ants. Although the trophobiotic association of Late Eocene longirostred aphids with formicines and dolichoderines have also been proven (Perkovsky 2010), in the Late Cretaceous there were simply no ants that had colonies large enough to 'keep' aphids (Dlussky & Rasnitsyn 2007; LaPolla *et al.* 2013): even now, aphid milkers from small colonies (hundreds of specimens) never protect their symbionts (Novgorodova 2015), and homopteran-tending ants are more likely to be dominant, with large colonies of  $10^4$ – $10^5$  workers (Oliver *et al.* 2008).

### 3. The Cretaceous–Paleogene (K–Pg) extinction event and the development of a symbiotic relationship between aphids and ants

It is well known that aphids are one of the main sources of hydrocarbon food for ants, particularly for Formicinae and Dolichoderinae (Stadler & Dixon 2005; Oliver *et al.* 2008). At the same time, aphids form an important resource for a wide array of predators and parasites (Skaljac 2016). In reward, the ants protect the tended aphids from competitors including aphidophages (Stadler & Dixon 2005), acting both as a symbiont and a predator (Rosengren & Sundström 1991; Sakata, 1995; Offenberg 2001). Among almost 5000 aphid species (Blackman & Eastop 2006), some 60 % are visited by ants (Stadler 1997). Unless tended by ants, aphid colonies fail quickly, due to predation (Watanabe *et al.* 2016).

Worth mentioning is that prior to the large ant colonies having evolved with their efficient ant–aphid mutualism, the aphids remain unprotected before the growing ant predation. The origin of the aphid trophobiosis with large colonies of Formicinae and Dolichoderinae resulted in the steep decline of aphids left beyond that ant–aphid symbiotic network. By at least the basal Eocene, ant proportions in the entomofauna increased sharply (Fig. 8), up to 6.9 % in the Early Eocene of the Green River and 13.1 % in the Middle Eocene of Messel, as compared with 0.0001–1.5 % in the Late Cretaceous (Dlussky & Rasnitsyn 2007). In the Cretaceous, all of the representatives of extant subfamilies occupied positions that were subordinate to Armaniinae and Sphecomyrminae. Moreover, the scape is very short in armaniines and in most sphecomyrminae, which has even raised some doubts about their sociality (Dlussky 1983; Dlussky & Rasnitsyn 2007); whilst the long scapus of some Cretaceous fossils suggests group recruitment and socially advanced behaviour amongst stem-group ants (Perrichot *et al.* 2016). The fauna did not include any evident dominants (Dlussky & Rasnitsyn 2007).

In contrast, in the Eocene, the dolichoderines and formicines were absolute dominants in all fossil myrmecofaunas (except for the Messel, Eckfeld and Mo-clay assemblages, dominated, for taphonomic reasons only, by giant ants; Rust & Andersen 1999; Archibald *et al.* 2011). The evolutionary success of dolichoderines and formicines is considered to be directly linked to their symbiosis with homopterans (Wilson & Hölldobler 2005; Dlussky & Rasnitsyn 2007). Two evident dominants of the myrmecofauna emerged as early as the Early Eocene of the

Green River – the dolichoderine or formicine *Eoformica pinguis* (Scudder, 1877) (40 % of the ants) and dolichoderine *Dolichoderus kohlsi* Dlussky & Rasnitsyn, 2003 (25 % of the ants) (Dlussky & Rasnitsyn 2003; LaPolla & Greenwalt 2015). This means that highly social species with a large family size had appeared (Dlussky & Rasnitsyn 2007). The dominants of Eocene myrmecofauna belonged to extant aphidophilous ant subfamilies (with the above taphonomically-caused exceptions), while the dominants of aphidofauna were myrmecophilous. Unfortunately, the Paleocene ants are practically unknown: the Sakhalinian amber is currently dated as Middle Eocene (Kodrul 1999; Baranov *et al.* 2015), and its impoverished ant fauna (Dlussky & Rasnitsyn 2007; La Polla *et al.* 2013) is probably due to its origin amongst wetland.

Thus the extinction of Late Cretaceous aphid families should be associated with the emergence of new dominants among ants from among the representatives of the subfamilies Formicinae and Dolichoderinae, which were symbionts of homopterans and developed a characteristic dolichoderine–formicine proventriculus that allows the crop to store large amounts of liquid (Eisner & Wilson 1952); it regulates the amount of this liquid which passes into the midgut and in this way separates the individual supply of food in the midgut from the communal supply in the crop.

The predation of these ants and of other aphidophages that appeared by the basal Cenozoic, such as syrphids (not known from the Cretaceous as was believed earlier; Perkovsky & Makarkin 2015; Popov 2015), proved to be such a serious problem to aphids that the only survivors were those that either became poorly accessible to ants in some way (e.g., by having found shelters in galls) or could transform ant predation into symbiosis.

Adaptation to symbiosis with ants occurred in the extra-tropical regions of the Northern Hemisphere and because of the climatic limitations discussed above, this adaptation failed to cross the tropical belt and reach the south temperate realm, which otherwise could have served as another heaven for the ant–aphid mutualistic system. The Cenozoic dominant groups of aphids, especially Aphididae (Heie 1994), failed to cross into the tropics and colonise the temperate regions of the Southern Hemisphere, where only five relict genera with a short rostrum and well-developed siphunculi remain. These genera are also shown to be ancient due to the divisible variations in the number of chromosomes within one genus (in other aphids the number of chromosomes is usually stable within one genus, especially if the genus is small). The degree of the depletion of the aphid fauna of the temperate regions of the southern continents is so great that only a dozen species of the undoubtedly relict genus *Neophyllaphis* Takahashi, 1920 have been recorded on gymnosperms. None of these genera have any connection to ants. Similarly, it has been suggested that the diversification of ants was also the cause of the extinction of Limaiinae (Chrysopidae are homopteran feeding) (Khramov *et al.* 2016).

#### 4. Conclusions

At least since the Cretaceous Terrestrial Revolution, the geographical distribution of aphids, particularly in the Northern Hemisphere, has been strongly affected by the low thermal tolerance of their obligatory bacterial symbiont, *Buchnera aphidicola*, which was why the aphids switched to obligate parthenogenesis in low latitudes.

Hormaphidids and greenideids penetrated into the tropics only after the Oligocene strengthening of climate seasonality, and the specialisations of the tropical representatives of these

families did not allow them to spread further south (in the case of cerataphidines), or only allowed this spread in a few cases (in the case of greenideids).

Aphids suffered from the Mesozoic–Cenozoic boundary extinction event much more strongly than other insects. The extinction roughly coincided with the establishment of the tight symbiosis of aphids with formicine and dolichoderine ants, which was accompanied by the flourishing of all three groups. Because of the low overheating tolerance of *Buchnera*, the mutualistic ant–aphid system failed to cross the tropical belt during the Cenozoic. As a result, the native southern temperate aphid fauna consists now of seven genera only, five of which being Late Cretaceous relicts; some of them had relatives in Late Cretaceous amber of the Northern Hemisphere.

Ole Heie (1994) asked 22 years ago in his title “Why are there so few aphid species in the temperate areas of the southern hemisphere?” We hope that our paper proposes a new answer.

#### 5. Acknowledgements

We would like to thank A. P. Rasnitsyn (Paleontological Institute, Moscow), for discussing the problems and the invaluable comments on the manuscript; the late G. M. Dlussky (Moscow State University), for his continuous encouragement and support in the studies on the history of ant–aphid relationships; V. N. Makarkin (Institute of Biology and Soil Sciences, Vladivostok), for helpful discussion on aphid predators; M. Kanturski (University of Silesia) and T. M. Kodrul (Geological Institute, Moscow), for their help in the literature search; and T. Szklarzewicz (Jagiellonian University) and Ł. Depa (University of Silesia) for reading the manuscript and their suggestions, which we incorporated into the study. We would also like to thank T. Szklarzewicz for the photographs of *Buchnera*. The authors thank the two anonymous reviewers, whose comments improved our work considerably.

#### 6. References

- Agarwala, B. K. & Dixon, A. F. G. 1986. Population trends of *Cervaphis schouteniae* on *Microcos paniculata* and its relevance to the paucity of aphid species in India. *Indian Biologist* **18**, 37–39.
- Archibald, S. B., Johnson, K. R., Mathewes, R. W. & Greenwood, D. R. 2011. Intercontinental dispersal of giant thermophilic ants across the Arctic during early Eocene hyperthermals. *Proceedings of the Royal Society B* **278**, 3679–88.
- Baranov, V. A., Andersen, T. & Perkovsk?, E. E. 2015. Orthocladids from Eocene Amber from Sakhalin (Diptera: Chironomidae, Orthoclaadiinae). *Insect Systematics & Evolution* **46**(4), 359–78.
- Blackman, R. L. & Eastop, V. 2006. *Aphids on World's herbaceous plants and shrubs*. Chichester: Wiley. Available: <http://www.aphidsonworldsplants.info>
- Blakey, R. 2011. *Global Paleogeography* (accessed 15.12.14.). <http://cpgeosystems.com/paleomaps.html>.
- Bodenheimer, F. S. & Swirski, E. 1957. *The Aphidoidea of the Middle East*. Jerusalem: Weizmann Science Press. 378 pp.
- Braendle, C., Miura, T., Bickel, R., Shingleton, A. W., Kambhampati, S. & Stern, D. L. 2003. Developmental origin and evolution of bacteriocytes in the aphid – *Buchnera* symbiosis. *PLoS Biology* **1**, 70–76.
- Buchner, P. 1965. *Endosymbiosis of animals with plant microorganisms*. New York: John Wiley. 909 pp.
- Burke, G., Fiehn, O. & Moran, N. 2010. Effects of facultative symbionts and heat stress on the metabolome of pea aphids. *The ISME Journal: Multidisciplinary Journal of Microbial Ecology* **4**, 242–52.
- Charles, H. & Ishikawa, H. 1999. Physical and Genetic Map of the Genome of *Buchnera*, the Primary Endosymbiont of the Pea Aphid *Acyrtosiphon pisum*. *Journal of Molecular Evolution* **48**, 142–50.



- Chen, Ch.-Y., Lai, Ch.-Y. & Kuo, M.-H. 2009. Temperature effect on the growth of *Buchnera* endosymbiont in *Aphis craccivora* (Hemiptera: Aphididae). *Symbiosis* **49**, 53–59.
- Chen, D.-Q., Montllor, C. B. & Purcell, A. H. 2000. Fitness effects of two facultative endosymbiotic bacteria on the pea aphid *Acyrtosiphon pisum*, and the blue alfalfa aphid, *A. kondoi*. *Entomologia Experimentalis et Applicata* **95**, 315–23.
- Cholodkovsky, N. 1899. Aphidologische Mitteilungen. *Zoologischer Anzeiger* **22**(602), 468–77.
- Coiffard, C., Gomez, B., Daviero-Gomez, V. & Dilcher, D. L. 2012. Rise to dominance of angiosperm pioneers in European Cretaceous environments. *Proceedings of the National Academy of Sciences of the United States of America* **109**, 20955e20959.
- Davies, T. J., Barraclough, T. G., Chase, M. W., Soltis, P. S., Soltis, D. E. & Savolainen, V. 2004. Darwin's abominable mystery: insights from a supertree of the angiosperms. *Proceedings of the National Academy of Sciences of the United States of America* **101**, 1904–09.
- DeDryver, Ch.-A., Hulle, M., Le Gallic, J.-F., Caillaud, M. C. & Simon, J. C. 2001. Coexistence in space and time of sexual and asexual populations of the cereal aphid *Sitobion avenae*. *Oecologia* **128**, 379–88.
- del Guercio, G. 1909. Contribuzione alla conoscenza dei Lacnidi Italiani. Morphologia, sistematica, biologia generale e loro importanza economica. *Redia* **5**, 173–243.
- Depa, L., Mróz, E. & Szawaryn, K. 2012. Molecular identity of *Stomaphis quercus* (Hemiptera: Aphidoidea: Lachnidae) and description of a new species. *European Journal of Entomology* **109**(3), 435–44.
- Depa, L., Kanturski, M., Junkiert, Ł. & Wieczorek, K. 2015. Giant females vs. dwarfish males of the genus *Stomaphis* Walker (Hemiptera: Aphididae) – an aphid example of the ongoing course to permanent parthenogenesis? *Arthropod Systematics and Phylogeny* **73**(1), 19–40.
- Dilcher, D. 2000. Toward a new synthesis: major evolutionary trends in angiosperm fossil record. *Proceedings of the National Academy of Sciences of the United States of America* **97**, 7030–36.
- Dixon, A. F. G. 1985. *Aphid ecology an Optimization Approach*. Glasgow: Blackie and Sons. 157 pp.
- Dixon, A. F. G. 1987. The way of life of aphids: host specificity, speciation and distribution. In Minks, A. K. & Harrewijn, P. (eds) *Aphids their biology, natural enemies and control*, 197–207. Amsterdam: Elsevier Science Publishers. xx + 450 pp.
- Dixon, A. F. G. 1998. *Aphid ecology an Optimization Approach*. 2nd Edition. London: Chapman and Hall. 300 pp.
- Dixon, A. F. G., Kindlmann, P., Lepš, J. & Holman, J. 1987. Why there so few species of aphids, especially in the tropics. *American Naturalist* **129**, 580–92.
- Dixon, A. F. G., Holman, J. & Thieme, T. 1998. Sex and size in aphids. In Nieto Nafria, J. M. & Dixon, A. F. G. (eds) *Aphids in Natural and Managed Ecosystems*, 173–78. León, Spain: Universidad de León, Secretariado de Publicaciones. 688 pp.
- Dlussky, G. M. 1981. [Miocene ants (Hymenoptera, Formicidae) of the USSR]. *Trudy Paleontologicheskogo Instituta Akademii Nauk SSSR* **183**, 64–83. [In Russian.]
- Dlussky, G. M. 1983. [A new family of Upper Cretaceous Hymenoptera: an “intermediate link” between ants and Scolioidea.] *Paleontologicheskii Zhurnal* **3**, 65–78. [In Russian. English translation: *Paleontological Journal* **17**(3), 63–76.]
- Dlussky, G. M. & Rasnitsyn, A. P. 2003. Ants (Hymenoptera: Formicidae) of Formation Green River and some other Middle Eocene deposits of North America. *Russian Entomological Journal* **11**, 411–36.
- Dlussky, G. M. & Rasnitsyn, A. P. 2007. [Paleontological record and stages of ant evolution.] *Uspehi Sovremennoi Biologii* **127**(2), 118–34. [In Russian.]
- Douglas, A. E. 2006. Phloem-sap feeding by animals: problems and solutions. *Journal of Experimental Botany* **57**(4), 747–54.
- Drinnan, A. N. & Chambers, T. C. 1986. Flora of the Lower Cretaceous Koonwarra Fossil Bed (Korumburra Group), South Gippsland, Victoria. *Memoirs of the Association of Australasian Palaeontologists* **3**, 1–77.
- Dunbar, H. E., Wilson, A. C. C., Ferguson, N. R. & Moran, N. A. 2007. Aphid thermal tolerance is governed by a point mutation in bacterial symbionts. *PLoS Biology* **5**(5), e96.
- Durak, R. & Durak, T. 2015. Redescription of males of the aphid species *Cinara (Cupressobium) tujafilina* and *Cinara (Cupressobium) pressi* (Hemiptera, Lachninae). *Zootaxa* **4032**(2), 209–14.
- Eisner, T. & Wilson, E. O. 1952. The Morphology of the Proventriculus of a Formicine Ant. *Psyche* **59**(2), 47–60.
- Evans, G. A. 2008. *The whiteflies (Hemiptera: Aleyrodidae) of the World and their host plants and natural enemies*. Version 2008-09-23. <http://www.sel.barc.usda.gov.591/WF/world-whitefly-catalog.pdf>. Accessed 14 October 2014.
- Feder, M. E. & Hofmann, G. E. 1999. Heat-shock proteins, molecular chaperones, and the stress response: Evolutionary and ecological physiology. *Annual Review of Physiology* **61**, 243–82.
- Fukatsu, T., Aoki, S., Kurosu, U. & Ishikawa, H. 1994. Phylogeny of Cerataphidini Aphids Revealed by Their Symbiotic Microorganisms and Basic Structure of Their Galls: Implications for Host–Symbiont Coevolution and Evolution of Sterile Soldier Castes. *Zoological Science* **11**, 613–23.
- Funk, D. J., Wernegreen, J. J. & Moran, N. A. 2001. Intraspecific variation in symbiont genomes: bottlenecks and the aphid–*Buchnera* association. *Genetics* **157**(2), 477–89.
- García Morales, M., Denno, B. D., Miller, D. R., Miller, G. L., Bendov, Y. & Hardy, N. B. 2016. *ScaleNet: A literature-based model of scale insect biology and systematics*. Database. doi: 10.1093/database/bav118. <http://scalenet.info>.
- Gavrilov-Zimin, I. A., Stekolshchikov, A. V. & Gautam, D. C. 2015. General trends of chromosomal evolution in Aphidococca (Insecta, Homoptera, Aphidinea + Coccinea). *Comparative Cytogenetics* **9**(3), 335–422.
- Hales, D. F. 1989. The chromosomes of *Schoutedenia lutea* (Homoptera, Aphidoidea, Greenideinae), with an account of meiosis in the male. *Chromosoma* **98**, 295–300.
- Hales, D. F., Tomiuk, J., Wöhrmann, K. & Sunnucks, P. 1997. Evolutionary and genetic aspects of aphid biology: a review. *European Journal of Entomology* **94**, 1–55.
- Hardy, N. B., Peterson, D. A. & Dohlen, C. D. 2015. The evolution of life cycle complexity in aphids: ecological optimization, or historical constraint? *Evolution* **69**, 1423–32.
- Harris, M. 1766. *The Aurelian, or Natural History of English Insects; namely Moths and Butterflies. Together with the Plants on which they feed. A faithful Account of their respective Changes; their usual Haunts when in the winged State; and their standard names, as given and established by the worthy and ingenious Society of Aurelians. Drawn, engraved and coloured, from the natural subjects themselves. By Moses Harris, Secretary to the Aurelian Society*. London: printed for the author. 92 pp; 44 col. pics.
- Heie, O. E. 1967. Studies on fossil aphids (Homoptera: Aphidoidea), especially in the Copenhagen collection of fossils in Baltic amber. *Spolia zoologica Musei Hauniensis* **26**, 1–274.
- Heie, O. E. 1980. The Aphidoidea (Hemiptera) of Fennoscandia and Denmark. I General Part. The Families Mindaridae, Hormaphididae, Thelaxidae, Anoecidae and Pemphigidae. *Fauna Entomologica Scandinavica* **9**, 1–236.
- Heie, O. E. 1994. Why are there so few aphid species in the temperate areas of the southern hemisphere? *European Journal of Entomology* **91**, 127–33.
- Heie, O. E. 2009. Aphid mysteries not yet solved (Hemiptera: Aphidomorpha). *Aphids and Other Hemipterous Insects* **15**, 31–48.
- Heie, O. E. & Pike, E. M. 1992. New aphids in Cretaceous amber from Alberta (Insecta, Homoptera). *Canadian Entomologist* **124**(6), 1027–53.
- Heie, O. E. & Wegierek, P. 2009. A classification of the Aphidomorpha (Hemiptera Sternorrhyncha) under consideration of the fossil taxa. *Redia* **92**, 69–77.
- Heie, O. E. & Wegierek, P. 2011. A list of fossil aphids (Hemiptera, Sternorrhyncha, Aphidomorpha). *Monographs of the Upper Silesian Museum* **6**, 82 pp.
- Herman, A. B., Akhmetiev, M. A., Kodrul, T. M., Moiseeva, M. G. & Yakovleva, A. I. 2009. Flora development in Northeastern Asia and Northern Alaska during the Cretaceous–Paleogene transitional epoch. *Stratigraphy and Geological Correlation* **17**(1), 79–97.
- Ivanov, V. D., Melnitsky, S. I. & Perkovsky, E. E. 2016. [Caddisflies from Cenozoic resins of Europe.] *Paleontologicheskii Zhurnal* **5**, 33–61. [In Russian. English translation: *Paleontological Journal* **50**(5), 485–93.]
- Johnson, K. R. 2002. Megaflora of the Hell Creek and lower Fort Union Formations in the western Dakotas: vegetational response to climate change, the Cretaceous–Tertiary boundary event, and rapid marine transgression. *Geological Society of America, Special Paper* **361**, 329–91.
- Kania, I. & Wegierek, P. 2008. *Palaeoaphididae (Hemiptera, Sternorrhyncha) from Lower Cretaceous Baissa deposits. Morphology and Classification*. Kraków: Instytut Systematyki i Ewolucji Zwierząt Polskiej Akademii Nauk. 133 pp.
- Kawada, K. 1987. Polymorphism and morph determination. In Minks, A. K. & Harrewijn, P. (eds) *Aphids their biology, natural enemies*

- and control, 255–68. Amsterdam: Elsevier Science Publishers. 384 pp.
- Khranov, A. V., Liu, Q., Zhang, H. & Jarzembowski, E. A. 2016. Early green lacewings (Insecta: Neuroptera: Chrysopidae) from the Jurassic of China and Kazakhstan. *Papers in Palaeontology* **2**(1), 25–39.
- Koch, C. L. 1857. *Die Pflanzenläuse Aphiden getreu nach dem Leben abgebildet und beschrieben*. Nürnberg: J. L. Lotzbeck. 335 pp.
- Kodrul, T. M. 1999. Paleogenovaja fitostratigraphija Juzhnogo Sakhalina. [Paleogene phytostratigraphy of the South Sakhalin.] *Transactions of GIN RAS* **519**, 1–150. [In Russian.]
- Kononova, E. L. 1976. [Extinct aphid families (Homoptera, Aphidinea) of the Late Cretaceous.] *Paleontologicheskii Zhurnal* **3**, 117–26. [In Russian. English translation: *Paleontological Journal* **10**(3), 352–60.]
- Kononova, E. L. 1977. [New species of aphids (Homoptera, Aphidinea) from the Upper Cretaceous deposits of the Taimyr.] *Entomologicheskoe Obozrenie* **56**(3), 588–600. [In Russian.]
- Kurosu, U., Buranapanichpan, S. & Aoki, S. 2006. *Astegopteryx spinoccephala* (Hemiptera: Aphididae), a new aphid species producing sterile soldiers that guard eggs laid in their gall. *Entomological Science* **9**, 181–90.
- LaPolla, J. S., Dlussky, G. M. & Perrichot, V. 2013. Ants and the Fossil Record. *Annual Review of Entomology* **58**, 609–30.
- LaPolla, J. S. & Greenwalt, D. E. 2015. Fossil Ants (Hymenoptera: Formicidae) of the Middle Eocene Kishenehn Formation. *Sociobiology* **62**(2), 163–74.
- Lichtenstein, J. 1882. *Coccus lataniae* = *Boisduvalia lataniae* = *Cerataphis lataniae*. *Annales de la Société Entomologique de France* **2**, XVI.
- Linnaeus, C. 1758. *Systema naturae per regna tria naturae secundum classes, ordines, genera, species, cum characteribus, differentiis, synonymis, locis*, 10<sup>th</sup> ed., vol. 1. Holmiae: Impensis Laurentii Salvii. 824 pp.
- Ma, C.-S., Hau, B. & Poehling, H.-M. 2004. The effects of heat stress on the survival of the rose grain aphid, *Metopolophium dirhodum* (Hemiptera: Aphididae). *European Journal of Entomology* **101**, 327–31.
- Magallon, S. & Castillo, A. 2009. Angiosperm diversification through time. *American Journal of Botany* **96**, 349–65.
- Martin, S. K., Skidmore, L. I. & Stilwell, J. D. 2016. A first record of Cretaceous aphids (Hemiptera, Sternorrhyncha, Aphidomorpha) in Australia, from the Lower Cretaceous Koonwarra Fossil Bed, Victoria. *Zootaxa* **4137**, 95–107.
- Martinez-Torres, D., Buades, C., Latorre, A & Moya, A. 2001. Molecular Systematics of Aphids and Their Primary Endosymbionts. *Molecular Phylogenetics and Evolution* **20**(3), 437–49.
- McDonald, M. J., Rice, D. P. & Desai, M. M. 2016. Sex speeds adaptation by altering the dynamics of molecular evolution. *Nature* **531**, 233–36.
- Michalik, A., Szklarzewicz, T., Jankowska, W. & Wieczorek, K. 2014. Endosymbiotic microorganisms of aphids (Hemiptera: Sternorrhyncha: Aphidoidea): ultrastructure, distribution and transovarial transmission. *European Journal of Entomology* **111**(1), 91–104.
- Montllor, C. B., Maxmen, A. & Purcell, A. H. 2002. Facultative bacterial endosymbionts benefit pea aphids *Acyrtosiphon pisum* under heat stress. *Ecological Entomology* **27**(2), 189–95.
- Moran, N. A. 1992. The evolution of life cycles in aphids. *Annual Review of Entomology* **37**, 321–48.
- Moran, N. A., Munson, M. A., Baumann, P. & Ishikawa, H. 1993. A molecular clock in endosymbiotic bacteria is calibrated using the insect hosts. *Proceedings of the Royal Society, London B* **253**, 167–71.
- Moran, N. A., McCutcheon, J. P. & Nakabachi, A. 2008. Genomics and evolution of heritable bacterial symbionts. *Annual Review of Genetics* **42**, 165–90.
- Moran, N. A. & Baumann, P. 1994. Phylogenetics of cytoplasmically inherited micro-organisms of arthropods. *Trends in Ecology & Evolution* **9**, 15–20.
- Moran, N. A. & Yun, Y. 2015. Experimental replacement of an obligate insect symbiont. *Proceedings of the National Academy of Sciences of the United States of America* **112**(7), 2093–96.
- Mordvilko, A. K. 1914. Insectes Hémiptères (Insecta Hemiptera). I. Aphidoidea. *Faune de la Russie et des pays limitrophes fondée principalement sur les collectionnes du Musée Zoologique de l'Académie Impériale des Sciences de Petrograd Livraison* **1**, 1–CLXIV, 1–236, 1–9.
- Nieto Nafria, J. M. & Favret, C. (eds). 2011. *Register of 12 family-group and genus-group taxa of Aphidoidea (Hemiptera Sternorrhyncha)*. León, Spain: Universidad de León, Secretariado de Publicaciones. 465 pp.
- Novgorodova, T. A. 2015. Organization of honeydew collection by foragers of different species of ants (Hymenoptera: Formicidae): Effect of colony size and species specificity. *European Journal of Entomology* **112**(4), 688–97.
- Offenberg, J. 2001. Balancing between mutualism and exploitation: the symbiotic interaction between *Lasius* ants and aphids. *Behavioral Ecology & Sociobiology* **49**, 304–10.
- Oliver T. H., Leather S. R. & Cook J. M. 2008. Macroevolutionary patterns in the origin of mutualisms involving ants. *Journal of Evolutionary Biology* **21**, 1597–1608.
- Otto, S. P. 2009. The evolutionary enigma of sex. *The American Naturalist* **174**, Suppl. 1, S1–S14.
- Ouvrard, D. 2016. *Psyllist – The World Psylloidea Database*. <http://www.hemiptera-databases.com/psyllist> - searched on 18 October 2016 doi:10.5519/0029634
- Pérez-braci, V., Gil, R., Ramos, S., Lamelas, A., Postigo, M., Michelena, J. M., Silva, F. J., Moya, A. & Latorre, A. 2006. A small microbial genome: the end of a long symbiotic relationship? *Science* **314**, 312–13.
- Perkovsky, E. E. 2010. [Participation of *Germaraphis* aphids (Homoptera, Aphidinea) in weight fractions of the Rovno amber and their syninclusions with ants.] *Vestnik zoologii* **44**(1), 55–62. [In Russian, with English Abstract.]
- Perkovsky, E. E. 2011. Syninclusions of the Eocene winter ant *Prenolepis henshei* (Hymenoptera: Formicidae) and *Germaraphis* aphids (Hemiptera: Eriosomatidae) in Late Eocene Baltic and Rovno amber: some implications. *Russian Entomological Journal* **20**(3), 303–13.
- Perkovsky, E. E. 2012. [On influence of latitudinal changes in summer temperatures on the Late Eocene aphids *Germaraphis* (Homoptera, Aphidoidea) and on their symbiotic relationships with ants.] *Vestnik zoologii* **46**(1), 51–58. [In Russian, with English Abstract.]
- Perkovsky, E. E., Rasnitsyn, A. P., Vlaskin, A. P. & Taraschuk, M. V. 2007. A comparative analysis of the Baltic and Rovno amber arthropod faunas: representative samples. *African Invertebrates* **48**(1), 229–45.
- Perkovsky, E. E., Zosimovich, V. Yu. & Vlaskin, A. P. 2010. Rovno amber. In Penney, D. (ed.) *Biodiversity of Fossils in Amber from the Major World Deposits*, 116–36. Manchester: Siri Scientific Press. 304 pp.
- Perkovsky, E. E., Rasnitsyn, A. P., Vlaskin, A. P. & Rasnitsyn, S. P. 2012. [Contribution to the study of the structure of amber forest communities based on analysis of syninclusions in the Rovno Amber (Late Eocene of Ukraine).] *Paleontologicheskii Zhurnal* **3**, 70–78. [In Russian. English translation: *Paleontological Journal* **46**(3), 293–301.]
- Perkovsky, E. E. & Makarkin, V. N. 2015. First confirmation of spongillafies (Neuroptera: Sisyridae) from the Cretaceous. *Cretaceous Research* **56**, 363–71.
- Perkovsky, E. E. & Wegierek, P. 2015. [On causes of mass extinction of Late Cretaceous aphids.] *Palaeontology: Communities and crises*, 22–24. Conference dedicated to the 70<sup>th</sup> anniversary (memory) of V. V. Zherikhin. Abstracts. Moscow: Borissiak Paleontological Institute of the Russian Academy of Sciences. [In Russian.]
- Perrichot, V., Wang, B. & Engel, M. S. 2016. Extreme Morphogenesis and Ecological Specialization among Cretaceous Basal Ants. *Current Biology*. <http://dx.doi.org/10.1016/j.cub.2016.03.075>
- Poinar, G.O. Jr. & Brown, A. E. 2005. New Aphidoidea (Hemiptera: Sternorrhyncha) in Burmese amber. *Proceedings of the Entomological Society of Washington* **107**, 835–45.
- Ponomarenko, A. G. 1998. [Paleobiology of angiospermization.] *Paleontologicheskii Zhurnal* **4**, 3–10. [In Russian. English translation: *Paleontological Journal* **32**(4), 325–31.]
- Popov, G. V. 2015. Syrphidae from the Cretaceous – refuted? 8th International Symposium on Syrphidae, Monschau, Germany 4–8 June 2015, *Abstracts*, 47.
- Rasnitsyn, A. P., Bashkuev, A. S., Kopylov, D. S., Lukashevich, E. D., Ponomarenko, A. G., Popov, Yu. A., Rasnitsyn, D. A., Ryzhkova, O. V., Sidorchuk, E. A., Sukatsheva, I. D. & Vorontsov, D. D. 2016. Sequence and scale of changes in the terrestrial biota during the Cretaceous (based on materials from fossil resins). *Cretaceous Research* **61**, 234–55.
- Rasnitsyn, A. P. & Quicke, D. L. (eds) 2002. *History of Insects*. Dordrecht: Kluwer Academic Publishers. xii + 517 pp.
- Rayner, R. J., Bamford, M. K., Brothers, D. J., Dippenaar-Schoeman, A. S., Mckay, I. J., Oberprieler, R. G. & Waters, S. B. 1998 (1997). Cretaceous fossils from the Orapa diamond mine. *Palaeontologica Africana* **33**, 55–65.
- Rich, T. H., Rich, P. V., Wagstaff, B., McEwen-Mason, J., Douthitt, C. B. & Gregory, R. T. 1989. Early Cretaceous biota from the

- northern side of the Australo–Antarctic rift valley. *Geological Society, London, Special Publications* **47**, 121–30.
- Rosengren, R. & Sundström, L. 1987. The foraging system of a redwood ant colony (*Formica* s. str.) – collecting and defending food through an extended phenotype. *Experientia Supplementum*, **54**, 117–37.
- Rübsaamen, E. H. 1905. Beiträge zur Kenntnis Aussereuropäischer Zooecidien. I. Gallen von Bismark Archipel. *Marcellia* **4**, 5–25.
- Russell, J. A. & Moran, N. A. 2006. Costs and benefits of symbiont infection in aphids: variation among symbionts and across temperatures. *Proceedings of the Royal Society, London B* **273**, 603–10.
- Rust, J. & Andersen, N. M. 1999. Giant ants from the Paleogene of Denmark, with a discussion of the fossil history and early evolution of ants (Hymenoptera: Formicidae). *Zoological Journal of the Linnean Society* **125**, 331–48.
- Sakata, H. 1995. Density-dependent predation of the ant *Lasius niger* (Hymenoptera: Formicidae) on two attended aphids *Lachnus tropicalis* and *Myzocallis kuricola* (Homoptera: Aphididae). *Researches on Population Ecology* **37**, 159–64.
- Scudder, S. H. 1877. The first discovered traces of fossil insects in the American Tertiaries. *Geological Survey of Canada. Report of Progress* **3**, 741–62.
- Shingleton, A. W., Stern, D. L. & Foster, W. A. 2005. The origin of a mutualism: a morphological trait promoting the evolution of ant-aphid mutualisms. *Evolution* **59**(4), 921–26.
- Simon, J. C., Risper, C. & Sunnucks, P. 2002. Ecology and evolution of sex in aphids. *Trends in Ecology & Evolution* **17**(1), 34–39.
- Simon, J. C., Stoeckel, S. & Tagu, D. 2010. Evolutionary and functional insights into reproductive strategies of aphids. *Comptes Rendus Biologies* **333**, 488–96.
- Simonet, P., Dupont, G., Gaget, K., Weiss-Gayet, M., Colella, S., Febvay, G., Charles, H., Viñuelas, J., Heddi, A. & Calevro, F. 2016. Direct flow cytometry measurements reveal a fine-tuning of symbiotic cell dynamics according to the host developmental needs in aphid symbiosis. *Scientific Reports* **6**, 19967.
- Skaljac, M. 2016. Bacterial Symbionts of Aphids (Hemiptera: Aphididae). In Vilcinskis, A. (ed.) *Biology and Ecology of Aphids*, 100–26. Boca Raton: CRC Press. 282 pp.
- Smith, J. M. & Maynard-Smith, J. 1978. *The evolution of sex*. Cambridge: University Press. 236 pp.
- Sontag, E. 2003. Animal inclusions in a sample of unselected Baltic amber. *Acta Zoologica Cracoviensia* **46**, 431–40.
- Stadler, B. 1997. The relative importance of host plant, natural enemies and ants in the evolution of life-history characters of aphids. In Dettner, K., Bauer, G. & Völkl, W. (eds) *Vertical food web interactions*, 241–56. Berlin & Heidelberg: Springer-Verlag. xxii + 390 pp.
- Stadler, B. & Dixon, A. F. G. 2005. Ecology and evolution of aphid–ant interaction. *Annual Review of Ecology Evolution and Systematics* **36**, 345–72.
- Szwedo, J., Lapeyrie, J. & Nel, A. 2015. Rooting down the aphid's tree – the oldest record of the Aphidomorpha lineage from Palaeozoic (Insecta: Hemiptera). *Systematic Entomology*, **40**(1), 207–13. doi: 10.1111/syen.12099
- Tagu, D., Sabater-Muñoz, B. & Simon, J.-Ch. 2005. Deciphering reproductive polyphenism in aphids. *Invertebrate Reproduction and Development* **48**, 71–80.
- Takahashi, R. 1920. A new genus and species of aphid from Japan (Hem.). *Canadian Entomologist* **52**(1), 19–20.
- Tomiuk, J., Hales, D. F., Wöhrmann, K. & Morris, D. 1991. Genotypic variation and structure in Australian populations of the aphid *Schoutedenia lutea*. *Hereditas* **115**, 17–23.
- Unterman, B. M., Baumann, P. & McLean, D. L. 1989. Pea aphid symbiont relationships established by analysis of 16SrRNAs. *Journal of Bacteriology* **171**, 2970–74.
- van Ham, R. C. H. J., Kamerbeek, J., Palacios, C., Rausell, C., Abascal, F., Bastolla, U., Fernández, J. M., Jiménez, L., Postigo, M., Silva, F. J., Tamames, J., Viguera, E., Latorre, A., Valencia, A., Morán, F. & Moya, A. 2003. Reductive genome evolution in *Buchnera aphidicola*. *Proceedings of the National Academy of Sciences of the United States of America* **100**(2), 581–86.
- Walker, F. 1870. Notes on aphides. *The Zoologist, Second Series* **5**, 1996–2001.
- Wang, H., Moore, M. J., Soltis, P. S., Bell, C. D., Brockington, S. F., Alexandre, R., Davis, C. C., Latvis, M., Manchester, S. R. & Soltis, D. E. 2009. Rosid radiation and the rapid rise of angiosperm-dominated forests. *Proceedings of the National Academy of Sciences of the United States of America* **106**, 3853–58.
- Watanabe, S., Murakami, T., Yoshimura, J. & Hasegawa, E. 2016. Color polymorphism in an aphid is maintained by attending ants. *Science Advances* **2**(9), e1600606.
- Wegierek, P. & Mamontova, V. A. 1993. A new fossil species of the genus *Stomaphis* Walker (Aphidoidea: Lachnidae). *Annals of the Upper Silesian Museum, Entomology Suppl.* **1**, 37–50.
- Wieczorek, K., Kanturski, M. & Junkiert, L. 2013. The sexuales of giant black bark aphid, *Pterochloroides persicae* (Hemiptera, Aphidoidea: Lachninae). *Zootaxa* **3626**, 94–98.
- Wilcox, J. L., Dunbar, H. E., Wolfinger, R. D. & Moran, N. A. 2003. Consequences of reductive evolution for gene expression in an obligate endosymbiont. *Molecular Microbiology* **48**, 1491–1500.
- Wilson, E. O. & Hölldobler, B. 2005. The rise of the ants: a phylogenetic and ecological explanation. *Proceedings of the National Academy of Sciences of the United States of America* **202**, 7411–14.
- Wöhrmann, K. & Tomiuk, J. 1988. Life cycle strategies and genotypic variability in populations of aphids. *Journal of Genetics* **67**(1), 43–52.

MS received 6 June 2016. Accepted for publication 27 February 2017.