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Plant and associate, genetic distance and STRUCTURE

Comparative population structure of Chinese sumac aphid *Schlechtendalia chinensis* and its primary host-plant *Rhus chinensis*

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Abstract Most of our current understanding of comparative population structure has been come from studies of parasite–host systems, whereas the genetic comparison of gallnut-aphids and their host-plants remain poorly documented. Here, we examined the population genetic structure of the Chinese sumac aphid *Schlechtendalia chinensis* and its unique primary host-plant *Rhus chinensis* in a mountainous province in western China using inter-simple sequence repeat (ISSR) markers. Despite being sampled from a mountainous geographic range, analysis of molecular variance (AMOVA) showed that the majority of genetic variation occurred among individuals within populations of both the aphid and its host. The aphid populations were found to be structured similarly to their primary host populations (F_{ST} values were 0.239 for the aphid and 0.209 for its host), suggesting that there are similar patterns of gene flow between the populations of the aphid and between populations of its host-plant. The genetic distances ($F_{ST}/1 - F_{ST}$) between the aphid populations and between its host-plant populations were uncorrelated, indicating that sites with genetically similar host-plant populations may not always have genetically similar aphid populations. The lack of relationships between genetic and geographical distance matrices suggested that isolation by distance (IBD) played a negligible role at this level. This may be mainly attributed to the founder effect, genetic drift and the relative small spatial scale between populations.

Keywords Coevolution · ISSR marker · Population genetic structure · *Rhus chinensis* · *Schlechtendalia chinensis*

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Introduction

The genetic structure of populations is important in understanding evolution and ecology, as it reflects the extent of gene flow between populations, and hence, the evolutionary potential of an organism (Price 1980; Thompson 1994). Geographic structure influences not only the population biology of single species, but also interspecific interactions (Burdon 1997; Hanksi and Gilpin 1997). Depending on the nature of the interaction, much insight can also be gained by comparing the population genetic structures of species that inhabit sympatric range, which can help one to draw inferences about the population subdivision and offer important information and insight into the ecology of, for instance, a host and its parasite, such as effective population sizes, dispersal distance, evidence of local adaptations, gene flow rates and the potential of herbivores to coevolve with their hosts (McCoy et al. 2005). Specifically, these processes are important in

understanding the evolutionary relationships between plants and insects (Michalakis et al. 1993).

There have been many comparative studies of interacting species, mainly in host–parasite systems (Nadler et al. 1990; Michalakis et al. 1993; Dybdahl and Lively 1996; Delmotte et al. 1999; Hoffmann and Baker 2003). The studies of herbivorous insects in particular have played a major role in understanding how ecological divergence can facilitate genetic differentiation (Sword et al. 2005). Herbivorous insects are extraordinarily diverse in tropical and temperate biomes, with roughly a quarter of all eukaryotes being insects that feed on plants (Bernays 1998). The degree of interconnection between populations of phytophagous insects and their host plants may determine the potential for the herbivores to coevolve with their hosts (Michalakis et al. 1993). Furthermore, insect–plant associations are specialized, showing a high degree of phylogenetic conservation, i.e., related insect lineages tend to feed on related plants (Janz and Nylin 1998; Funk et al. 2002; Lopez-Vaamonde et al. 2003; Wimp et al. 2005; Sword et al. 2005). However, few studies have compared host–parasite co-structure and even fewer have included both the same populations and the same type of markers.

Aphids (Homoptera: Aphididae), the insect group for which the most frequent formation of host-adapted races or biotypes has been reported, are particularly interesting models in the study of genetic and demographic components of plant adaptation (Blackman et al. 1990; Wool et al. 1995; Margartopoulos et al. 2000; Lushai et al. 2002; Haack et al. 2000; Faten et al. 2002; Haley et al. 2004; Jyoti and Michaud 2005). Most aphids parasitizing their host-plant form gallnuts, which are abnormal growths produced by aphids parasitizing the midribs of leaves and other tissues. Chinese gallnuts are formed on several *Rhus* species (Anacardiaceae) (Lee et al. 1997). Native to East Asia, Chinese gallnuts grow on *Rhus* species in lowland, hills and mountain areas in China, Japan, Indochina, Java, Malaysia, Sumatra, etc. There are 14 kinds of Chinese gallnuts, which are formed by 14 species of gallnut aphids. The horned gallnut, also termed Galla rhois, is the most abundant, making up about 80% of Chinese gallnuts. It is caused by the Chinese sumac aphid, *Schlechtendalia chinensis*. Like other aphids, they are phloem feeders and have complex life cycles, with alternating sexual and parthenogenetic generations as well as alternating hosts. In the early spring, winged sexuparae fly to main branches of their host-plant, *Rhus chinensis*, where they produce sexual males and females, which have no mouthparts and do not feed. They mature, mate and then the male dies. The female aphid produces a single wingless fundatrix, whose only capability is to initiate a gallnut. The fundatrix climbs to the young and growing leaf tissues to suck the sap, where it causes the gallnut to form (The fundatrix reproduces parthenogenetically for three generations in one gallnut and the fundatrigeniae aphids live in it for about 3 months). Thus, each gallnut is initiated by a single aphid and her offspring (about 5–8 thousand individuals) are genetically identical to her. In late autumn, after the gallnuts have matured and dehisced, winged fundatrigeniae are formed and disperse from the gallnut to winter mosses as secondary hosts (about 14 species, mainly of *Plagiannium*). There they asexually produce larvae which overwinter on the mosses. Next spring, the larvae moult to become winged sexuparae and fly to the primary host-plant to begin their next life cycle (Qiu 1994; Li et al. 2003). *Rhus chinensis*, the primary host of *S. chinensis*, is a woody species with both sexual reproduction and clonal spread. The plant grows at a moderate to high rate, is not self-fertile and in flowers in August. Like most sumacs, *R. chinensis* is dioecious and pollinated by bees. Fruits (drupes, 8–9 mm long) remain attached year-round and may attract frugivorous birds. The species is widely distributed in temperate, subtropical, and tropical regions in East Asia including India and most provinces except the northernmost-Heilongjiang, Jilin, Neimenggu and Xinjiang in China. However, *R.chinensis* is only parasitized by *S. chinensis* in a small part of its range to form horned gallnuts because the parasitizing aphid must shift from primary to second host (mosses) to finish their life cycle, its flight capability is very weak and the mosses only develop in damp woodland or dank hillside. Hence, the horned gallnut is formed locally, and mainly in boundary mountainous areas (forests, e.g. Mt. Qing, Mt. Emei, and Mt. Miao) of Sichuan, Guizhou, Yunnan, Hubei and Hunan province, with the largest production of gallnuts being in Guizhou province in the central of this area. Because the horned gallnut aphids must form horned gallnut to finish its life cycle, the aphid *S. chinensis* distributes only in these areas too.

It has been found that *S. chinensis* selects its position on the leaf of the host, *R. chinensis*, by reference to the leaf structure. The aphid parasitizes the plant, by sucking sap from the leaf, destroying the morphological structure of young tissues and finally forming a gallnut. The plant provides food resource for the aphid, whereas the aphid's sucking promotes the generation of secondary metabolites by the plant, which may increase its resistance to aphid feeding (Li et al. 2003). *S. chinensis* completes most of its life cycle on *R. chinensis*, so the primary host should be more important for the aphid than the winter host in the co-evolutionary relationship. However, little attention has been paid to comparisons of the genetic structures of gallnut aphids and their host-plant populations at a molecular level.

The inter-simple sequence repeat (ISSR) technique is based on the amplification of regions (100–3000 bp)

between inversely oriented, closely spaced microsatellites. The technique employs a single PCR primer (5′ or 3′ anchored usually by 2–4 arbitrarily selected nucleotides), binding to di-or tri-nucleotide repeat motifs (microsatellites), which are abundant in eukaryotic genomes and result in a large number of polymorphic bands (Rakoczy and Bolibok 2004). As a potential molecular marker system, its uses in the analysis of phylogeny and in population genetics have been documented in a wide variety of organisms (Praukttan et al. 2005; Hardig et al. 2000; Deshpande et al. 2001; Gupta et al. 1994; Nagy et al. 2003; Wolfe and Liston 1998; Zietkiewicz et al. 1994).

In this study, we examined the population genetic structures of *S. chinensis* and *R. chinensis*, tested the correlation between their population structures, and the role played by isolation by distance (Rohlf and Schnell 1971) using ISSR markers. Because of the obligate relationship between the aphid and its primary host-plant and the fact that both species are likely to experience similar barriers to gene flow (e.g., in the form of landscape features), it is suggested that the two species are likely to have similar patterns of genetic structure. To test if the Chinese sumac aphid and its unique primary host-plant have correlated population structures, we compared the matrix of aphid pairwise F_{ST} to the matrix of host pairwise F_{ST}. We also compared the matrix of genetic distances and geographic distances for testing the isolation levels by distance for the aphid and host.

Materials and methods

Sample collection

The samples of horned gallnut Galla rhois were collected in October 2004 from eight populations of two districts (administration zones) in Guizhou Province, China (Fig. 1). The gallnuts were cut open, and the aphids collected and kept in absolute ethanol prior to DNA extraction. At the same time, the leaves were collected from the same trees and kept with silica gel in zip-lock plastic bags until DNA isolation.

Since it was common to have several gallnuts on one tree, we sometimes obtained more than one gallnut from the same tree. Sampling information is shown in Table 1.

DNA extraction and ISSR PCR amplification

Prior to the DNA isolation, individual aphids were immersed in distilled water for at least 36 h. Then the individual was ground using a small pestle in 1.5 ml Eppendorf tubes, lysis buffer (pH 8.0) was added and they were incubated at 55°C for 8–12 h (The buffer contained 200 mM Tris–HCl, 25 mM EDTA, 300 mM NaCl, 1% SDS and 200 μg/ml proteinase-K). Total genomic DNA of single individual was then extracted using phenol/chloroform methods (Ren et al. 2002). Host-plant DNA was extracted using the modified cetyltrimethylammonium bromide (CTAB) protocol (Nan et al. 2003). Dried leaf material was ground and transferred to 1.5 ml Eppendorf tubes holding 800 μl preheated 2× CTAB extraction buffer containing 0.2% mercaptoethanol and incubated at 64°C for 1 h. Subsequently, 600 μl cold chloroform:isoamylalcohol (24:1, v/v) was added, and DNA was extracted according to phenol/chloroform protocol. The supernatant was reserved and mixed with ice-cold isopropanol. DNA was washed in 70% ethanol, dried and dissolved in TE buffer (pH 8.0). DNA concentrations were estimated by agarose gels electrophoresis (1.0% gel containing 0.5 μg/ml ethidium bromide) using known concentrations of uncut lambda DNA as standards.

ISSR primers from the University of British Columbia (ISSR kit #9) were used and the sequences were obtained by Shenggong Inc., Shanghai, China (Table 2). PCR amplification was done in a MJ Research thermal cycle, PTC200, using 25 μl reaction mixture containing 10 mM Tris–HCl (pH 8.3), 50 mM KCl, 3.0 mM $MgCl_2$, 0.25 mM dNTPs, 0.6 μM primer, 2% formamide, 20–50 ng template DNA, and 2.5 unit *Taq* DNA polymerase. PCR amplification for the aphid was performed using the following program: 94°C, 2 min, for 1 cycle, 94°C, 30 s, 68°C, 30 s, 72°C, 1 min, for 1 cycle, the annealing temperature was dropped 0.7°C for each of the subsequent 12 cycles, then, for 25 cycles: 94°C, 30 s, 55°C, 30 s, 72°C, 1 min. For the host-plant, the program was: 5 min at 94°C for 1 cycle, followed by 40 s at 94°C, 40 s annealing at 52, 54 or 56°C (depending on the different primers), 1.5 min extension at 72°C for 43 cycles, and a single 7 min at 72°C for the final extension cycle. The products were visualized on 2.0% agarose gels containing ethidium bromide, using 1× TBE buffer. Molecular weights were estimated using a 200 bp DNA ladder (Shenggong Inc., Shanghai).

Data analyses

Data analyses were performed separately for each species. For each polymorphic amplification product or polymorphic band (locus), there were two possible states (alleles), amplified fragment present (1), or absent (0). Multilocus profiles are hereafter referred to haplotypes. Analysis of ISSR data is not as straightforward as allozyme or SSR data because of the dominant nature of the markers. As with RAPD or AFLP data, the presence of a band can denote either a dominant homozygote or a heterozygote, so that it is generally not possible to dis-

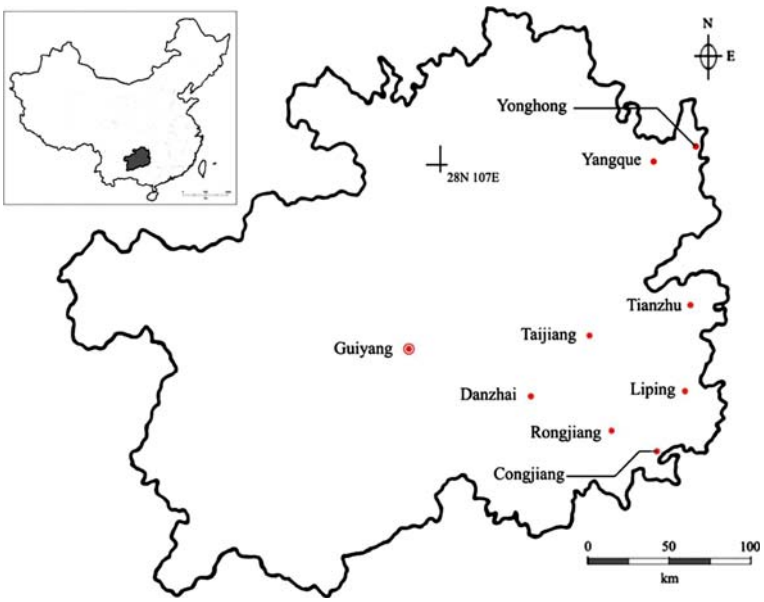

Fig. 1 Sketch maps showing the locations of eight populations of Chinese sumac aphid *Schlechtendalia chinensis* and its primary host plant *Rhus chinensis* from two districts of Guizhou Province, China. Sample numbers are described in materials and methods

Table 1 Sampling information and genetic diversity of ISSR analysis for the aphid and its host-plant

Population	No. of individuals		Altitude (m)	Latitude	Longitude	Topography	P.L		P (%)	
	Aphid	Host					Aphid	Host	Aphid	Host
Congjiang	16	16	500	25.46	108.54	Transitional zone between plateau, mountains and hilly area	81	131	60.9	76.61
Rongjiang	18	15	380	25.56	108.30	Mount Leigong, upstream of Duliu River	82	127	61.65	74.27
Danzhai	16	7	950	26.23	107.48	West of Mount Leigong, sloping ground between plateau and hilly area	76	112	57.14	65.5
Taijiang	19	18	730	26.34	108.19	Mount Leigong, mountain area	82	137	61.65	80.12
Tianzhu	18	12	450	26.54	109.13	Hilly area with valley and basin	88	130	66.17	76.02
Liping	17	17	500	26.15	109.10	Mount Fenghuang, mountain area	93	153	69.92	89.47
Yonghong	13	8	800	28.08	109.18	Mount Fanjing, mountain area	81	133	60.9	77.78
Yangque	22	13	1200	28.02	108.55	Mount Fanjing, mountain area	86	139	64.66	81.29
Total	139	106					133	171	100	100

Notes: P.L—No. of polymorphic loci; P (%)— Percentage of polymorphism

tinguish between genotypes (Culley and Wolfe 2001). Hardy–Weinberg equilibrium is often assumed so that all populations and loci can be tested in Popgene version 1.32 (Yeh et al. 1997).

For each population and each primer combination of aphids and host-plants, the numbers of polymorphic loci and the percentages of polymorphic loci at population and species levels were calculated, respectively. Genetic data (F_{ST}) were examined using an analysis of molecular variance (AMOVA) in ARLEQUIN version 2.0 (Schneider et al. 2000) with significance tests estimated using 1023 permutations. Partial correlations between matrices were calculated using two-level Mantel tests (Mantel 1967) implemented in NTSYS-PC version 2.02 (Rohlf 1998), with the significance of the autocorrelation coefficient tested by 1,000 resamplings, for examining the relationship of aphid genetic distance [(represent as $F_{ST}/(1 - F_{ST})$) versus either geographic distance (calculated as log distance in km) or host genetic distance [(represent as $F_{ST}/(1 - F_{ST})$) (Smouse et al. 1986; Rousset 1997, 1999; Jerome and Ford 2002). The comparison between matrices of genetic distances and log geographical distances was also evaluated using the two-level Mantel test. Prior to the Mantel tests, the aphid and host populations were divided into two groups based on geographical districts, i.e., the administration zones in Guizhou Province. One group includes Yonghong and Yangque populations and the other includes the other six populations. Scatterplots of the pairwise distances for pairs of populations were used to describe aphid genetic distance versus host genetic distance, as well as aphid–host genetic distance versus geographical distance.

Table 2 Codes and sequences of primers used to reveal polymorphism for Chinese sumac aphid and its primary host plant

Primer code	Sequence	No. of markers	
		Aphid	Host-plant
807	$(AG)_8T$	7	10
808	$(AG)_8C$	16	–
809	$(AG)_8G$	6	13
811	$(GA)_8C$	8	12
818	$(CA)_8G$	11	14
826	$(AC)_8C$	12	17
827	$(AC)_8G$	16	14
857	$(AC)_8YG$	11	19
866	$(CTC)_6$	12	17
868	$(GAA)_8$	11	18
873	$(GACA)_4$	15	17
881	$GGGT(GGGGT)_2G$	–	20
888	BDB(CA)7	8	–
Total		133	171

Results

Twelve primers for Chinese sumac aphid and 11 primers for its primary host-plant were evaluated for their ability to produce polymorphic bands (Table 2). The band profiles generated by ISSR primers showed considerable polymorphism among the aphid and host populations. Most of the amplified primers for ISSR analysis were the same for the aphid and its host-plant, although the number of polymorphic markers differed and were more in hosts than in aphids across all the same primers with the exception of primer UBC-827. There were no population-specific markers present in one population but absent in the others. The primers used in the analysis yielded a total of 133 scorable loci for the aphid and 171 for the host plant, and Popgene 1.32 analysis indicated that 100% of these loci were polymorphic for both species even though primers were not intentionally selected for high variability. The bands were in the range 200–3000 bp. The number of bands generated by individual primers varied from 6 (UBC-809) to 16 (UBC-808 and UBC-827) for the aphid and from 10 (UBC-807) to 20 (UBC-827) for the host (Table 2). The polymorphism among populations also varied and the highest diversity appears in Liping and the lowest in Danzhai population both in aphid and in host plant populations (Table 1).

For aphids and hosts, the greatest genetic variation occurred among individuals within populations. Significant genetic differentiation was detected at the level of populations ($P < 0.001$) (Table 3). The population-level AMOVA indicated that considerably more of the variance was found within aphid populations (75.87%) than between populations (24.13%) ($P < 0.001$) (Table 3). When all populations of the aphid and its host-plant were considered as two groups according to geographical districts and were examined using a nested AMOVA analysis, 76.06% of the variance was found within the populations, and 24.38% was found among the populations within these groups ($P < 0.001$). AMOVA analysis showed similar results for host-plant population, i.e., higher variance within than between populations (Table 3). Overall differentiation was calculated among the aphid populations ($F_{ST} = 0.239$) and host-plant populations ($F_{ST} = 0.209$), which indicated that the eight aphid and host populations possessed a similar genetic structure.

Correlation analysis of aphid genetic structure with host genotype was performed. There was no significant correlation between estimates of genetic distance of the two species ($P > 0.05$) (Fig. 2A). When the correlations for the genetic diversity of aphid (or host) populations and the geographical distance between colonies were examined, the same results were obtained, i.e., no significant relationship was found between genetic and geographical distances for aphid or host populations ($P > 0.05$) (Fig. 2B, C). Since several aphid individuals from the same host may bias results, we also randomly selected a single aphid individual per host and performed the same genetic analysis. The same results (similar F_{ST} and no significant correlationship between pairwise comparisons) were obtained (data not shown).

Discussion

In this study, we examined the population genetic structure of the Chinese sumac aphid using ISSR markers and compared it to that of its unique primary host-plant species. The genetic data indicated that the aphid and host-plant

Table 3 AMOVA analysis of ISSR variation for Chinese sumac aphid *Schlechtendalia chinensis* and its primary host plant *Rhus chinensis* populations

	d.f.	SSD	Variance components	% total	*P*-value
Population level					
Among aphid population	7	658.4533	4.59508	24.13	<0.001
Within aphid population	131	1893.1294	14.45137	75.87	<0.001
Among host population	7	703.0667	5.65166	17.99	<0.001
Within host population	99	2550.9146	25.76681	82.01	<0.001
Nested level (two groups)					
Among aphid groups	1	94.9981	–0.08400	–0.44	<0.001
Among aphid populations within groups	6	563.4552	4.63135	24.38	<0.001
Within aphid populations	131	1893.1294	14.45137	76.06	<0.001
Among host groups	1	146.7332	1.82099	5.59	<0.001
Among host populations within groups	6	556.3335	4.98700	15.31	<0.001
Within host populations	99	2550.9146	25.76681	79.10	<0.001

The *P*-value was calculated by 1,000 replication bootstrap between populations

populations showed high and similar levels of population structure (the global F_{ST} values were 0.239 for the aphid and 0.209 for its host-plant).

There are more than 500 species in the genus *Rhus*, but *S.chinensis* selects to live on only one species, *R. chinensis*, suggesting that it is adapted to the host-plant. The high structuring can be the result of co-evolution. In addition, Chinese gallnut is a very important economic resource, and stocks of the species have been occasionally exchanged between regions. Although the exchanges were mainly over small ranges, this may have caused sufficient migration in different parts to result in high differentiation among local populations, but with most variability within populations.

The present results show similar global F_{ST} for the aphid and its host-plant, indicating similar patterns of gene flow between populations of the aphid and between populations of its host-plant. The genetic differentiation between populations of herbivorous organisms has mainly resulted from the nature of their association with host plants (Futuyma and Peterson 1988), but other factors also contribute to genetic structure in the absence of host selection pressures. The similarities in the genetic structure of herbivore and host can conveniently be examined in an aphid-host system, where aphid populations are closely linked to, or dependent on their host-plant populations to finish their life history. However, the genetic distances between pairs of aphid populations were not correlated with the genetic distances between the corresponding pairs of host populations.

The specialization of herbivorous insects is more likely to occur when a relationship has been established with a host plant that has a long vegetative cycle, leading finally to the emergence of races (Haack et al. 2000). Host-adapted races and biotypes have been characterized within numerous species parasitizing host-plants with long vegetative cycles (Edmunds and Alstad 1978; Rausher 1983; Thompson 1994; Jerome and Ford 2002; Blair et al. 2005).

Aphids are of particular interest in the study of plant–herbivorous relationships because of their breeding system which combines parthenogenesis and sexual reproduction, and amongst these, species with cyclical parthenogenesis are especially interesting. They are the most abundant insect group, and many host-adapted races or biotypes have been reported (Blackman et al. 1990; Wool et al. 1995; Sunnucks et al. 1997; Vanlerberghe-Masutti and Chavigny 1998; Via 1999; Miller et al. 2003; Haley et al. 2004; Jyoti and Michaud 2005). The life cycles of aphid and its host plant were described above. From those, it is obvious that the propagation of the host plant is independent of the aphid, whereas the aphid must rely on the host-plant to complete its life cycle, implying that the host-plant genetics and dynamics should have a strong influence on the genetics of the aphid.

Ecological and evolutionary factors that affect one species of such a pair will often affect the other, and the joint consideration of such species may provide useful information about the relative importance of factors in shaping current distributions and population characteristics (Pellmyr et al. 1998; Avise 2000; McCoy et al. 2005). The population structure of a parasite should be a reflection not only of its life history and genetic characteristics but also of its relationship with host species, since the latter are a spatially varying resource (Price 1980). There are numerous examples of comparative studies of species with shared ranges, which have mostly focused on the degree of genetic structuring in parasites relative to their hosts (Michalakis

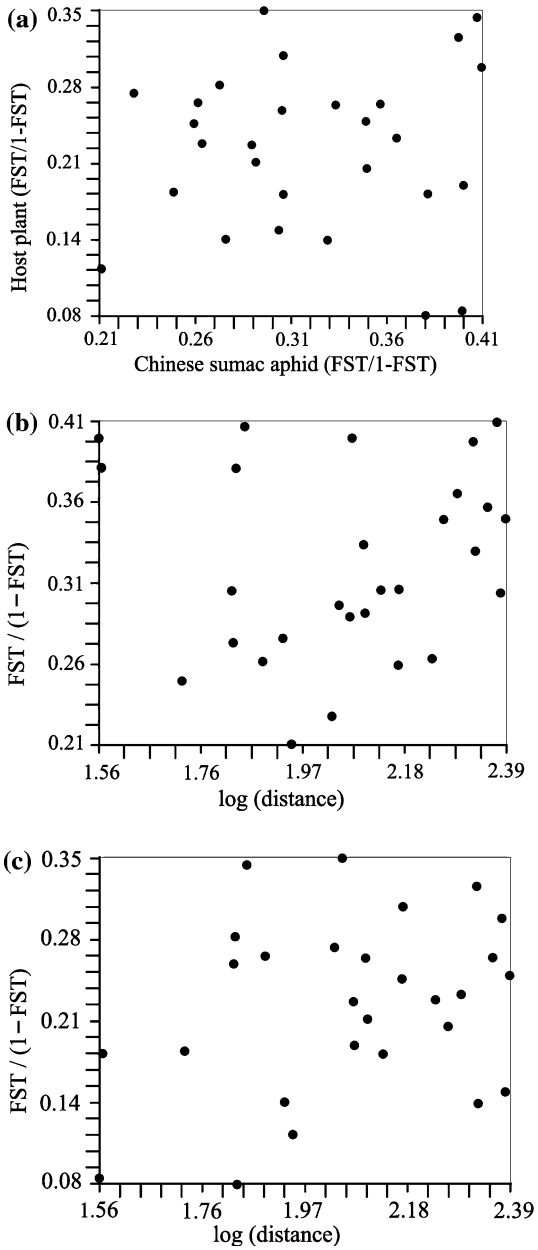

Fig. 2 Scatterplots of pairwise distances for eight populations in the Chinese sumac aphid *Schlechtendalia chinensis*-host plant *Rhus chinensis* system: (**A**) the aphid genetic distance $[F_{ST}/(1 - F_{ST})]$ versus host-plant genetic distance $[F_{ST}/(1 - F_{ST})]$, (**B**) the aphid genetic distance $[F_{ST}/(1 - F_{ST})]$ versus geographical distance [log (km)], (**C**) host-plant genetic distance $[F_{ST}/(1 - F_{ST})]$ versus geographical distance [log (km)]

et al. 1993; Dybdahl and Lively 1996; Martinez et al. 1999; Davies et al. 1999; Mutikainen and Koskela 2002; Burban and Petit 2003; Zink 2002; Hoffmann and Baker 2003). Parasites have often been found to have more structured populations than their hosts. For example, Delmotte et al (1999) found that a fungal parasite was much more differentiated than its *Silene* host-plant and this finding was attributed to different mating system because the fungal parasite undergoes routine selfing, whereas the host plant outcrosses. Great spotted cuckoo parasites had more strongly structured populations than their magpie hosts and it is suggested that cuckoo parasites impose a selection pressure on their magpie hosts to disperse relatively long distance from their birth sites (Martinez et al. 1999). Jerome and Ford (2002) showed that parasite populations were found to be 3–6 times more structured than any of their three principal hosts, which may partially be attributed to selection pressures imposed by different host taxa and/or local host genotypes, environmental conditions or differences in breeding and dispersal mechanisms. McCoy et al. (2005) also showed a similar pattern of genetic structure between parasite ticks and their seabird hosts and it is suggested that kittiwakes disperse more frequently than their parasites and thus should readily adapt to local parasites. On the other hand, the reverse has also been found. In these cases, the host populations were found to be more strongly structured than their parasites. For example, Michalakis et al. (1993) found that the populations of thistle hosts were much more subdivided than the populations of parasitizing weevils and this may mainly be attributed to the large migration rate between weevil populations. Dybdahl and Lively (1996) considered that the difference in structuring between a freshwater snail and its trematode parasite should be attributed to dispersal mechanisms: the primary host disperses itself over a short distance, while the parasites are dispersed over long distances by their hosts. Mutikainen and Koskela (2002) reported a similar result with the parasitic plant (great dodder) having more genetic drift than its host plant (stinging nettle). In a few cases host and parasite showed similar degrees of population structuring (Nadler et al. 1990; Mulvey et al. 1991; Parker and Spoerke 1998). The similarity in structuring was attributed to long-term continuity of mutualistic interactions or a close parasitic association between two species. Only in those systems where host and parasite dispersal are linked has population structure been found to be correlated (Nadler et al. 1990; Dybdahl and Lively 1996; Parker and Spoerke 1998). All these studies have provided important clues to the genetic relationship of interacting species.

Price (1980) predicted that adaptation to and strict dependence of a parasite on a host would lead to strong structuring of parasite populations. Spatial genetic structure within plant populations is primarily determined by the effects of such factors as limited seed and pollen dispersal, isolation in small patches, differential mortality, and microhabitat selection (Levin and Kerster 1974; Epperson 1993). Relative host-parasite dispersal rates are particularly important as they will help define the scale at which coevolution occurs and may alter the potential of each species to adapt locally (Gandon et al. 1996).

In the present study, the similar genetic structure between the aphid and its host may partially be attributed to the similar restrictions to gene flow for both species (e.g. mountainous terrain). The aphid must parasitize its unique primary host-plant to finish its life cycle, so the populations of both species must have the same environment conditions. Many geographical features can restrict gene flow between sets of populations, such as rivers, highways, mountain ranges, etc (Whitlock, 1999). Overall Guizhou is a mountainous province, more hilly in the west and relatively flat in the east and south. The western part of the province, with several water systems across, forms part of the Yunnan-Guizhou plateau, where the samples of eight populations were collected for ISSR analysis. With these natural barriers, the gene flow between populations will be greatly restricted.

The present results suggested that if this structure reflects current population functioning, aphids are dispersing with a similar frequency to their hosts and therefore they would quickly adapt to hosts. It is also likely that the similar structuring in the aphid populations relative to their host-plants is caused by their generation time. Generally, the aphids (winged fundatrigeniae) have less flight capability than other insects and usually need to rest for a while after they have flown for about 10 m, until they either find their winter-host mosses or lose the ability to fly (Qiu 1994). However, they can occasionally be carried by air currents for longer distances, resulting in wider gene flow among populations. Host-plant migration can occur either by seeds or pollen. Seed dispersal distance appears to be large in *R. chinensis* and pollen flow by bees may also disperse the genes. The host therefore has a greater dispersal potential than the aphids, particularly in mountainous regions. However, the plant has a relatively long generation time compared to aphids, which would slow down the accumulation of neutral genetic differences among populations of plants more than in aphids. Furthermore, differential selection pressures imposed by environmental conditions throughout the range of the aphid and host-plant may also contribute to the similar genetic structure.

The lack of relationship between population pairs of host and parasite genetic distances suggests that sites with genetically similar host-plant populations may not always have genetically similar aphid populations. The results may be affected by two factors: the interacting species belong to different taxa with completely different biological characteristics, and the horned gallnut aphid must transfer from one host to another to complete its life cycle. One must take into account that the winter host (moss) is an essential element for the completion of the aphid life cycle and may influence the genetic structure of aphid populations.

Correlation between genetic distance and geographic distance for the aphid and its host population was also examined. The genetic distances between pairs of the aphid (or host-plant) populations were not correlated with geographical distance ($P > 0.05$). Geographic distance between populations is often used as a reliable indicator of gene flow, and management decisions are often based on the presumption that geographically close populations are genetically the most similar (Bond et al. 2005). In aphid populations, however this does not seem to be the case. Massonnet et al. (2002) investigated isolation by distance (IBD) and population genetic structure in the aphid *Macrosiphoniella tanacetaria* and found that most pairs of populations were significantly different but there was no IBD pattern. However, adding populations from a larger spatial scales, on average 470 km apart resulted in a weak but significant suggestion of isolation by distance. They suggested that for the host-specific *M. tanacetaria*, patterns of genetic variation among populations were, on an ecologically meaningful scale, governed by colonization/extinction dynamics and genetic drift rather than by a drift-dispersal equilibrium. In a study of *Pemphigus bursarius*, a cyclically parthenogenetic aphid (Miller et al. 2003), microsatellite markers were used to examine the population structure, and the results showed that the degree of allele frequency divergence between populations was not correlated with their geographical separation, indicating that isolation by distance was not the sole cause of spatial genetic structuring.

By contrast with aphids, a wide range of plant species show isolation by distance (Hess et al. 2000). However, some factors can disrupt isolation by distance including physical barriers to pollen or seed movement, such as mountains or large water bodies (Williams and Arnold 2001; Bond et al. 2005), population bottlenecks and historical patterns of recolonization (Hewitt 1999).

The present results showed the lack of a relationship between genetic and geographic distance matrices, which suggests that isolation by distance may play a negligible role on the scale of these investigations. This might be mainly to the result of the relatively small spatial scale, 40–245 km between populations and the mountainous isolation among these populations, e.g. by Wuling Mount. *S. chinensis* originated in China (Zhang et al. 1999), so the founder effect and genetic drift may play an important role in population divergence. In addition, the effect of geographical isolation may have been diminished because of the influence of factors such as occasional longer distance flight for aphids, and water flow, wind or random seed dispersal by animal vectors for plants or the artificial utilization and adaptation to non-geographically patterned environmental conditions.

In conclusion, we compared the population structure of Chinese sumac aphid and its primary host-plant and found that their genetic structures are strong and similar to what we expected. On the other hand, the aphid and host genetic distance matrices were uncorrelated and there was no significant correlation between geographic and genetic distance (estimated as pairwise $F_{ST)}$, either for the aphid or for the host. We also suggest that the *Schlechtendalia-Rhus* association may be a good model system for the study of local co-evolution. However, the present results applied only to our sampled populations, which are in the middle of the Chinese horned gallnuts' distributions, and further studies are needed: (i) to sample from extremes of the distributions, which may provide a clearer picture of effects of differential selection and migration, (ii) to add more molecular markers, i.e., AFLP as a marker with more stability and greater numbers of bands than ISSR, and (iii) to investigate the distribution of the winter host moss species and its influence on the distributions and the relationships of the three species.

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