Cues, concessions, and inheritance: dominance hierarchies in the paper wasp *Polistes dominulus*

Lorenzo Zanette[a] and Jeremy Field[b]
[a]School of Biological Sciences, University of East Anglia, Norwich, Norfolk NR4 7TJ, UK and
[b]Department of Biology and Environmental Science, School of Life Sciences, John Maynard Smith
Building, University of Sussex, Brighton BN1 9QG, UK

Hierarchies constitute the base of many social groups. Hence, understanding how they are established is critical. Here we examine how hierarchies are formed in foundresses associations of the common paper wasp *Polistes dominulus*. By comparing field data with computer simulations, we evaluate order of arrival at the nest, body size, facial color patterns, and within-group kinship structure as determinants of inheritance rank. Hierarchies (ranks 1–5) were experimentally inferred for 53 nests. Overall, the order in which foundresses arrived at the nest and their body size were not significantly correlated with rank. A foundress's rank was negatively correlated with the number of full sisters it had in its group. Highly ranked wasps (ranks 1 and 2) were less likely to share a nest with their full sisters than wasps of lower rank. A wasp's rank was not determined by the relative rank of its nest-mate sisters. A foundress's rank was significantly correlated with the size of its black clypeal marks, but the number of foundresses with clypeal marks in each nest was small. On 15 of 20 nests where wasps with marks were present, only 1 wasp had such marks. Overall, our results suggest that within-group relatedness structure is important in the establishment of dominance hierarchies in *P. dominulus* foundress associations. *Key words:* arrival order, group formation, inheritance, *Polistes*. *[Behav Ecol 20:773–780 (2009)]*

D ominance hierarchies are a very common feature of group living in organisms ranging from bumblebees to mandrills (Wilson 1975). By definition, they reflect the effects of asymmetries between group members on the partitioning of resources and reproductive success (present and future). Because higher ranked individuals obtain a larger share of the benefits of group living, constant competition to attain dominant status may be expected (Wilson 1971; Röseler 1991; Cant, English, et al. 2006). Conversely, group stability and the synergistic benefits associated with it, that is, higher group efficiency, depend on the stability of the dominant-subordinate(s) relationship (Bourke and Franks 1995; Cant and Field 2001; Cant, English, et al. 2006; Cant, Llop, and Field 2006). Understanding the importance of this trade-off in the evolution of group living has been the subject of considerable theoretical and empirical work (reviewed in Johnstone 2000; Queller et al. 2000; Camazine et al. 2001; Reeve and Keller 2001).

Since first being described by Pardi (1942), dominant-subordinate interactions in primitively eusocial wasps have received considerable attention (West 1967; Röseler 1991; Camazine et al. 2001). In temperate species, before worker emergence, multiple foundress groups are usually small. All individuals are potentially capable of independent reproduction and are of similar size and age (Reeve 1991). Moreover, different foundresses' chemical profiles are not distinguishable at the beginning of the nesting season (Sledge et al. 2001; Dapporto et al. 2007). This lack of well-defined asymmetries begs the question of what determines dominance status.

In theory, hierarchies can be established either through direct conflict between individuals or through predetermined conventions based on some arbitrary cue (e.g., order of arrival),

which may prevent costly fights between individuals of similar quality (Maynard Smith and Parker 1976). Physical contests between foundresses at the early stages of the nesting cycle are common in several polistine species (*Polistes dominulus*—Pardi 1942, 1946; *Polistes metricus*—Gamboa and Dropkin 1979; *Ropalidia marginata*—Premnath et al. 1996; *Polistes fuscatus*—Gamboa and Stump 1996; and *Belonogaster juncea juncea*—Tindo and Dejean 2000). However, the importance of these fights as determinants of dominance order remains to be confirmed (Cant, English, et al. 2006). It is not clear whether the aggression between cofoundresses is a cause or consequence of the rank order within the group: Does aggression establish the hierarchy or does it represent conflict over status? Evidence that apparently arbitrary cues such as order of arrival and age are used to determine dominance has been found in some primitively eusocial wasps (e.g., order of arrival in *Polistes carolina*: Seppä et al. 2002; age in *Liostenogaster flavolineata*: Field et al. 1999; Bridge and Field 2007). Because new arrivals do not affect the order of arrival of residents already present, a convention based on arrival order may promote group stability (Seppä et al. 2002). In contrast, if morphological traits (e.g., body size) were used to determine rank, outsiders (e.g., late joiners) could out-rank existing group members and consequently destabilize formed hierarchies. Indirect evidence that arrival order affects the outcome of dominant-subordinate interactions has been found in experimental nest-mate pairs of *P. dominulus* (Pratte and Gervet 1992).

Unlike many vertebrate groups, where dominance order can be directly correlated with body size (in extremis, Buston 2003), evidence that body size determines rank order in paper wasps is limited (Reeve 1991; Cervo et al. 2008). Few studies have found a strong correlation between rank and body size (e.g., Nonacs and Reeve 1995 in *P. dominulus*). It has, however, recently been shown that color patterns—black marks on the clypeus—affect dominant-subordinate interactions and potentially dominance status in *P. dominulus*, and individual recognition in *P. fuscatus* (Tibbetts 2002; Tibbetts and Dale 2004).

The importance of these visual cues as signals of quality has yet to be confirmed, however (but see Cervo et al. 2008).

Interestingly, within-group genetic relatedness in foundresses associations of *P. dominulus* is often low and unrelated individuals are common within groups, yet dominant (rank 1) individuals largely monopolize reproduction (Queller et al. 2000; Liebert and Starks 2006; Zanette and Field, 2008; but see Cant, English, et al. 2006). Differences in dyadic relatedness between a dominant and different subordinates are likely to generate an uneven distribution of indirect fitness benefits between group members (Reeve and Keller 1996). The possibility of obtaining direct fitness benefits through nest inheritance has been suggested as a possible solution to the conundrum of unrelated foundresses (Queller et al. 2000). Understanding how inheritance rank is established in these paper wasps is therefore critical (Cant and Field 2001; Cant, Llop, and Field 2006).

In this paper, we examine possible factors that could determine rank in *P. dominulus* cofoundress associations. We focus on inheritance rank: the order in which foundresses inherit the rank 1, egg-laying position in the hierarchy. This metric is correlated with or causally related to within-group variation in behaviors such as helping effort, aggression, and nest defense in primitively eusocial wasps including our *P. dominulus* study population (e.g., Cant and Field 2001; Cant, English et al. 2006; Cant, Llop, and Field 2006; Field and Cant 2006; Field et al. 2006; Cronin and Field 2007). Inheritance rank is therefore also strongly correlated with "social rank," which has in the past typically been measured using patterns observed in these behaviors (Reeve 1991; Pratte 1993). However, because of its obvious connection with prospects for obtaining direct fitness, we believe that inheritance rank is a more useful metric for our purposes. We first examine the hypothesis that rank is correlated with the order of arrival at newly founded spring nests. To test this, we examine whether the observed correlation between order of arrival and rank is different from the correlation obtained in simulated populations where rank order is randomized. We then use the same approach to test whether rank is associated with 2 potential indicators of general quality, body size and the size of clypeal marks.

In addition, we examine the potential importance of intragroup relatedness in the establishment of dominance. Concession models from reproductive skew theory predict that *Polistes* dominants should have to concede a larger share of reproduction to unrelated subordinates than is empirically observed, as an incentive for them to remain in the group (Johnstone 2000; Reeve and Keller 2001; Nonacs et al. 2006). Independent of relatedness, skew is usually in fact very high, that is, dominant (rank 1) individuals monopolize most or all reproduction (Queller et al. 2000; Liebert and Starks 2006; but see Cant, English, et al. 2006). Here, we examine the hypothesis that unrelated subordinates obtain higher than average positions in the inheritance queue, thus providing an alternative incentive for them to remain in the group. We also test whether an individual's rank is correlated with the proportion of its nest mates that are full sisters.

METHODS

Natural history of *P. dominulus*

Polistes dominulus has a colony cycle typical of polistine wasps in temperate regions (Reeve 1991). Gynes (mated females) overwinter in aggregations ranging from a few to more than a 100 individuals. Winter diapause refuges are often reused by females in different years and although their location varies considerably, they are commonly observed behind large nests from the previous year (Pardi 1942; Turillazzi et al. 2006). In late winter/early spring (February–March, in Spain), foundresses leave their winter refuges and start to found new nests, either singly or jointly with auxiliary foundresses (2–23 females in our study population). In Southern Spain, this founding (preworker) period is relatively long, usually starting at the end of February and ending between late April and mid-May.

All foundresses are potentially capable of reproducing (Röseler 1991), but in polygynic nests, 1 individual is behaviorally dominant and lays most of the eggs, whereas the subordinates take on most of the riskier activities away from the nest: foraging to feed the offspring and collecting material for nest construction (Pardi, 1942; Reeve 1991). Toward the middle of the founding period (early spring: March–April), changes in group composition are frequent, with new foundresses (joiners) arriving at established nests. Nest switching and usurpation are also relatively common in this period (but see Queller et al. 2000). Usurpers usually destroy the previous foundress's eggs and early instar larvae (Nonacs and Reeve 1995; Starks 2001). At our study site, *P. dominulus* is also subject to attack by a social parasite, *Polistes semenowi* (see Zacchi et al. 1996; Shreeves et al. 2003). The nest foundation period finishes in early summer, just before the first brood (mainly workers) ecloses, and changes in group composition then became less frequent. As the season progresses, more of the female offspring become new reproductives (gynes). Males and gynes do not participate in nest activities and generally mate away from their natal nest (Pardi 1942). The reproductive phase lasts until the early autumn when the colonies dissolve and gynes disperse to winter refuges (Pardi 1942).

Determining the order of arrival

Nests of *P. dominulus* were found on hedges of prickly pear cactus (*Opuntia* sp.) in 2 nearby rural sites in south-western Spain (Conil de la Frontera, Province of Cadiz; Site 1—36°17′11N, 06°04′28W and Site 2—36°17′11N, 06°03′57W). From February to April in 2004 and 2005, sites were monitored every other day (between 10 and 14 h) to locate newly founded nests. When they were first discovered, nests had 1–8 foundresses (mean = 2.5 ± 1.6SE) and 1–18 cells (mean = 5.7 ± 5.2SE). All nests were numbered and their locations mapped. The morning after a nest's discovery, before wasps were active (07.00–08.00), all wasps were gently collected with long forceps, placed into plastic bags and stored at 4 °C for 15–30 min. Wasps were then individually marked using combinations of 4 enamel paint dots (2004) or honey bee tags (2005; queen marking kit: Thorne, UK) and subsequently released on their original nests to minimize any possible effect of removal. Every other day, all nests were censused early in the morning to detect changes in group composition. All marked wasps were recorded, and newly arrived unmarked wasps were collected, marked, and released on the same day. The order of arrival on a nest was then recorded according to the date that each foundress first appeared in the group. Wasps that were recorded only once were disregarded.

Identifying inheritance ranks

The dominant female on polygynic *P. dominulus* nests can easily be identified from daytime censuses because it leaves the nest less frequently than the remaining females in the group, which spend most of their time foraging (Röseler 1991; Cant and Field 2001). During April and May 2004–2005, when group composition had become stable and before the first adult offspring emerged, daytime censuses were conducted to measure the proportion of time that each foundress spent on the nest. Censuses were carried out only on sunny days (≥22 °C) between 10.00 and 15.00, the time when wasps are most active. Two to 8 censuses were carried out each day

(interval between census: 30 min–1 h). Following previous studies, wasps were classed as dominants if they were present on the nest for more than 70% of daytime censuses (mean time in nest of dominants $\pm$ SE $= 88.2 \pm 0.017\%$) (Cant and Field 2001; Cant, English, et al. 2006).

After dominants had been identified, hierarchies were inferred for 41 experimental nests (down to rank 5, 2004 only) by removals of successive dominants (Cant and Field 2001). This method allowed us to establish the order in which wasps inherited the dominant position. Starting the day after the removal of the original dominant foundress (rank 1), repeated daytime censuses were conducted every other day until a new dominant was identified using the criterion above. This required a minimum of 15 censuses and an average of 6 days after dominant removal. New dominants were easily identified because only 1 wasp per nest markedly changed its behavior after the removal, that is, spent much less time off the nest. Once identified, the replacement dominant (rank 2) was also removed and the nests censused to identify the next wasp to inherit dominance. The process was repeated until only 2 wasps were left on the nest (no further removals were necessary) or until workers started to emerge (early May). From this point, new dominants could no longer be identified because more than 1 foundress was present on the nest during most censuses ($\geq 70\%$). Overall, between 1 and 4 dominants were removed successively from each experimental nest in 2004. In 2005, rank 1 and 2 females were identified in 12 nests also using the criterion above. At the end of the experiment, marked foundresses and their nests were collected and stored at -80 °C (Laboratorio de Ecología Aquàtica, University of Cadiz, Spain).

Morphological data collection

In the laboratory, wasps were divided into 4 segments: head, anterior thorax, posterior thorax, and abdomen. Wings were carefully removed, unfolded, mounted between glass slides, and measured under a 16× binocular microscope. The internal length of the longitudinal cell (Discoidal I) of the right wing was used as a size measure and is known to be correlated with overall body size (Sullivan and Strassmann 1984).

Wasp heads were mounted on a ruler and photographed with a 10× Macro Fuji digital camera. Images were used to measure the size of clypeal marks. The contour of the clypeal marks was traced, and the area of the resulting polygon used as an estimate of size. Image analyses were performed with the software Image/J (version 1.33u: http://rsb.info.nih.gov/ij/). The accuracy of these measurements was assessed by measuring the clypeal marks using a 22× monocular microscope and the software NIH Image (version 1.55, http://rsb.info.nih.gov/nih-image). The correlation between measurements was strong ($R = 0.903$, df $= 73$, $P \ll 0.001$).

DNA extraction, amplification, and visualization

Total DNA was extracted from the anterior section of the thorax of all 177 ranked wasps using 300 µl of grinding solution (0.1 M NaCl; 0.1 M Tris–HCl—pH $= 8.0$, 0.05 M ethylenediaminetetraacetic acid; 0.05% sodium dodecyl sulfate), following Strassman et al. (1996) with minor modifications. DNA extractions were diluted 1:10 with ultrafiltered distilled water.

Multiplex polymerase chain reactions (PCR) were performed using 5 previously described primers, fluorescently labeled (Pdom 7, Pdom 20, Pdom 127b, Pdom 139, and Pdom 140; Henshaw 2000). PCR was carried out in a Peltier Thermal Cycler using 10 µl reaction mixtures with: 2 µl of DNA sample, 2 µl of reaction buffer ($[NH_4]_2SO_4$), 0.6 µl of $MgCl_2$, 0.2 µl of each deoxyribonucleotide triphosphate, 0.8 µl of each primer,

and 0.05 µl of Taq polymerase. The PCR products were visualized using an Applied Biosystems 3100 sequencer (Foster City, CA). Allele sizes were scored against an internal size standard (Applied Biosystems GeneScan ROX 500) using ABI GENESCAN Analysis software (v. 3.7). Genotypes that appeared inconsistent with those of nest mates were rescored (through reexamination of the chromatogram) or retyped (with a repeat PCR). In addition, 20 random samples were retyped, of which all were confirmed correct.

Relatedness estimation

Pairs of foundresses were assigned to the categories of full sister or nonsisters (cousins or unrelated) using a likelihood-based method implemented by in the program Kinship (Goodnight and Queller 1999; http://www.gsoftnet.us/GSoft.html). Assuming single mating, no inbreeding, and no linkage disequilibrium, the program uses population allele frequencies to estimate the likelihood that the genotypes of each pair of individuals would occur if they were full sisters versus the likelihood of the same genotypes arising if the females were maternal cousins. Cousins are the next closest possible relationship after sisters, because cofoundresses are of the same generation and multiple mating has not been detected in *P. dominulus* so that cofoundresses are very unlikely to be half-sisters (Queller et al. 2000; Strassmann 2001). At $\alpha = 0.05$, 99% of true sisters should have been correctly assigned to the full-sister category (number of Kinship simulations $= 2000$).

Statistical analysis

Foundresses in the same group cannot be considered as completely independent data points. Hence, the relationship between rank (dominance status) and order of arrival at nests was evaluated by comparing the observed mean within-nest correlation with that obtained using simulated groups in which rank was randomized. First, Kendall's tau (τ) rank correlation between foundresses arrival order and rank was calculated for each nest, and the overall observed mean correlation across nests obtained. Then, Kendall's tau (τ) was recalculated after rank order was randomly permuted in each nest, and an overall simulated mean correlation obtained. This simulation procedure was repeated 10 000 times to obtain a null distribution of means (Legendre and Legendre 1998). The proportion of simulated mean correlation values that were equal to or stronger than the observed mean was used as an estimate of the probability (P) of obtaining the observed correlation by chance. Correlations were considered to be significant if this probability was smaller than 0.05. The same procedure was used to evaluate the relation between dominance rank and foundress body size, size of black clypeal marks and the proportion of full sisters in the group. To explore the possibility that factors determining rank status are different for different parts of the hierarchy, this analysis was also performed considering only part of the inheritance queue, that is, ranks 1 and 2, or ranks 1–3.

In order to account for any correlations between the 4 potential determinants of rank that we investigated, we examined their combined effect on rank using Generalized Linear Models (GLMs) assuming quasi-poisson error distributions. (Crawley 2005). Because nest mates are not independent data points, 1 foundress was randomly sampled from each group. Rank was considered as the response variable and order of arrival, the proportion of sisters in the nest, body size, and size of clypeal mark(s) as potential explanatory variables. Group size, site, and year were also considered as potential covariates. This procedure (resampling followed by GLM) was repeated 2000 times to determine the probability of obtaining

significant terms in the GLM by chance. Explanatory terms were considered to be significant if their associated P values were smaller than 0.05 in at least 10% of the resampling runs, twice what would be obtained by chance alone. Two-way interactions and the effect of site and year were not included in the results unless significant.

All analyses were carried out using the software R (version 2.4.1; http://www.r-project.org). The functions used to perform the simulations are described in the supplementary material.

RESULTS

Behavioral and genetic data were obtained for 53 foundresses associations (number of wasps: 177; 41 nests in 2004 and 12 in 2005). At the time of the first dominant removals, that is, when group composition had stabilized, the total number of foundresses per nest ranged between 2 and 10 (mean = 5 ± 2SD, Figure 1).

Rank and order of arrival

Overall, the order in which foundresses arrived at the nest was not correlated with rank (mean 0.152, $P = 0.119$). This result was maintained when arrival times were rounded up to intervals of 3 days, to account for gaps between censuses, so that the differences in order of arrival were reduced (mean $\tau = 0.139$, $P = 0.152$: Figure 2). When considering only rank 1, 2, and 3 wasps however, arrival order tended to be correlated with rank (mean $\tau = 0.221$, $P = 0.061$, number of nests = 39). The correlation became significant when considering just the 2 individuals of highest rank (mean $\tau = 0.396$, $P = 0.005$). Rank 1 arrived before rank 2 on 32 of 47 nests, but not necessarily before all other wasps in its group. These results were maintained when excluding wasps with black clypeal marks from the analysis.

Rank and kinship

Closely related foundresses, that is, full sisters, were present in 26 of 53 nests. Overall, a foundress's rank was negatively correlated with the proportion of its nest mates that were its full sister (mean $\tau = 0.252$, $P = 0.017$). Highly ranked wasps (ranks 1 and 2) were less likely to share a nest with their full sisters than wasps of lower rank (Figure 3A). Rank 1 wasps, in particular, shared a nest with their close relatives less frequently than wasps of any other rank (14 of 53 nests; Figure 3A). Because the proportion, rather than the number of sisters was used, this result is unlikely to be an artifact of group size.
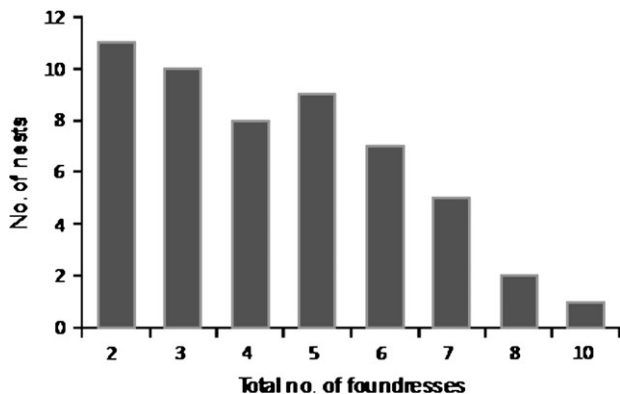

Figure 1
Overall distribution of group sizes in our study population, at the time of the first removals (41 nests in 2004 and 12 nests in 2005).

Moreover, the proportion of sisters for each individual was calculated independently of the rank of its sister(s), that is, both lower and higher ranked wasps were included. Hence, the number of wasps in the dominance queue had no direct effect on this result.

For an individual foundress of intermediate rank, the proportion of its sisters that were nest mates of higher rank was not significantly different from the proportion of sisters of lower rank (rank 2: $\chi^2 = 2.161$, df = 1, $P = 0.141$; rank 3: $\chi^2 = 0.001$, df = 1, $P = 0.976$; and rank 4: $\chi^2 = 0.836$, df = 1, $P = 0.361$, Figure 3B). For wasps of ranks 1 and 5, respectively, the highest and lowest rank considered, the distribution of their close relatives in their group was also balanced (Figure 3B). The number of nest mate sisters of rank 1 wasps that were ranks 2, 3, 4, or 5 was not significantly different ($\chi^2 = 0.579$, df = 3, $P = 0.901$). The number of nest mate sisters of rank 5 wasps that were rank 1, 2, or 3 tended to be smaller than the number of rank 5 sisters that were rank 4 ($\chi^2 = 7.0$, df = 3, $P = 0.071$). This was mainly because none of the sisters of rank 5 wasps were at rank 3.

When considering only nest mates that were full sisters (i.e., excluding unrelated nest mates from the analysis), rank was not correlated with the order of arrival (mean $\tau = -0.145$, $P = 0.209$, number of nests = 26), or the size of foundresses (mean $\tau = 0.442$, $P = 0.169$, number of nests = 26). The significance of the correlation with the size of clypeal black marks was not calculated because only 7 nests had wasps with these marks.

Rank and morphological differences

Foundresses with larger black clypeal marks were more likely to be the dominants (rank 1). Rank was significantly correlated with the size of clypeal marks (mean $\tau = -0.379$, $P = 0.022$; Figure 4A). This result was maintained when considering only smaller fractions of dominance hierarchies (e.g., ranks 1, 2, and 3, or ranks 1 and 2).

Nevertheless, the number of foundresses with clypeal mark(s) in each nest was small. No wasps had clypeal marks on 33 of 53 nests. Only 1 foundress had a mark on 15 of the 20 nests where wasps with marks were present (Figure 4B). Females with marks were rank 1 on 8 of these 15 groups. On the 5 nests where 2 or more females had marks, the dominant had the largest clypeal mark in 3 nests and was rank 3 in the other 2 nests.

Rank was not significantly correlated with foundresses body size (mean $\tau = 0.041$, $P = 0.622$). This result was maintained when considering only parts of the hierarchy (ranks 1 and 2; ranks 1–3).

GLM results

When considering all 4 potential determinants of rank together, the order of arrival of foundresses in the nest has no significant effect on rank (mean $z = -0.463$, $P < 0.05$ in only 3.7% of the simulations). GLM results also confirmed that wasp body size has no significant effect on rank (mean $z = -0.371$, $P < 0.05$ in only 2.3% of the simulations).

Interestingly, in contrast to the analysis above ("Rank and morphological differences"), where the correlation between rank and clypeal mark size was considered in isolation, the size of clypeal marks had no significant effect on rank in our GLM (mean $z = -0.172$, $P < 0.05$ in only 3.6% of the simulations). However, the very small number of wasps with clypeal marks present in our population may have limited our power to detect any effect.

GLM results confirmed that the proportion of full sisters that a foundress had in its group has a significant effect on its rank (mean $z = 0.786$, $P < 0.05$ for 15% of the simulations). Group
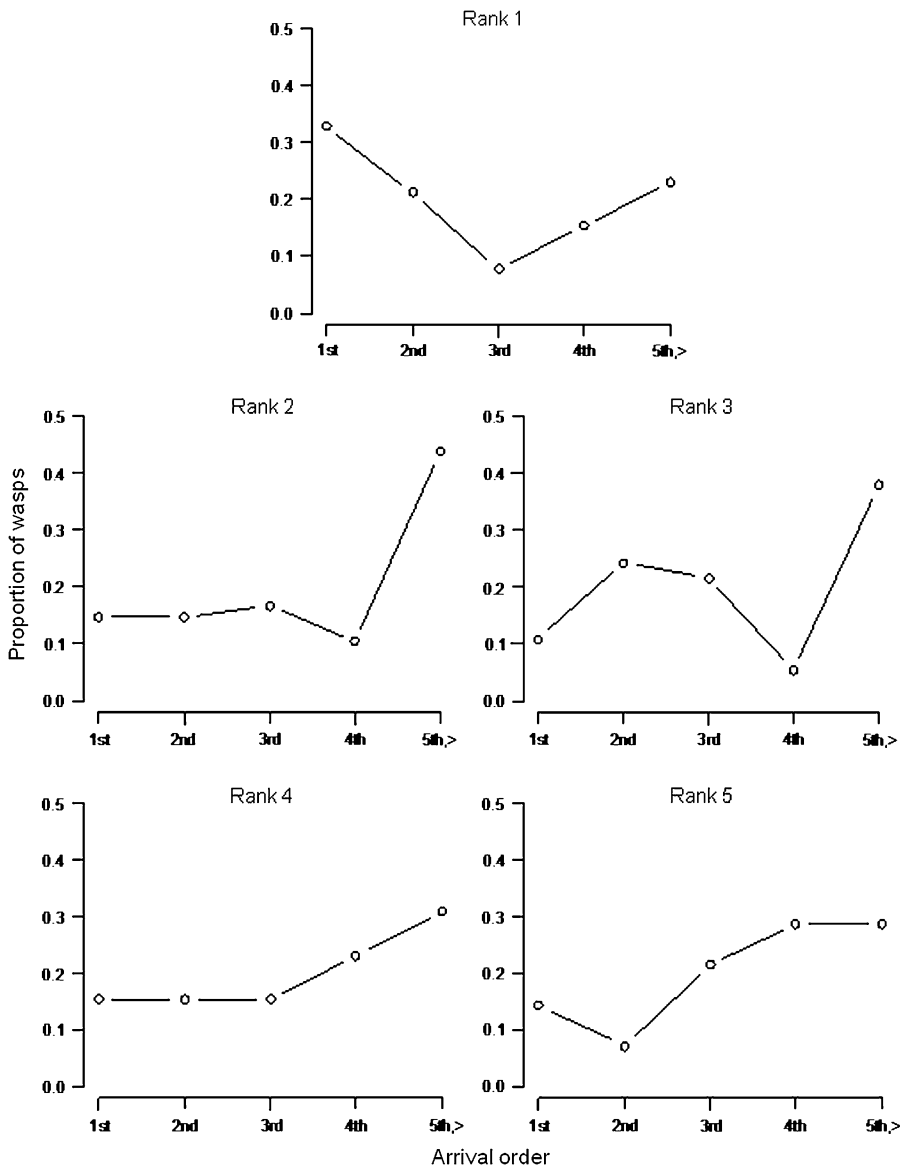
Figure 2
Distributions of the arrival orders of wasps at each rank.

size was the only other variable that had a significant effect on rank (mean $z = 2.112$, $P < 0.05$ for 53% of the simulations). The effect of group size is expected because the number of wasps in each nest directly determines the ranks that can be observed. The interaction between the proportion of full sisters and group size had no significant effect on rank (mean $z = -0.172$, $P < 0.05$ for 6% of the simulations). These results confirm that the observed correlation between rank status and the proportion of full sister is not an artifact of group size.

DISCUSSION

Rank and order of arrival

Contrary to our initial predictions, our results suggest that arrival order may have only a limited importance in the establishment of dominance in associations of *P. dominulus* foundresses. The observed correlation between order of arrival and rank was restricted to the top of the hierarchy (ranks 1 and 2). Moreover, when all ranked wasps and all potential rank determinants were considered together (GLMs), order of arrival had no significant effect on rank. Seppä et al. (2002) reported

a similar pattern for *P. carolina*, where the rank of the first 3 foundresses in the hierarchy was frequently correlated with their order of arrival on the nest. Note that Seppä et al. defined rank using the number of aggressive interactions between wasps, whereas we used inheritance rank.

The importance of order of arrival has been tested indirectly in *P. dominulus* by Pratte and Gervet (1992), who showed that prior residence can affect the outcome of dominance interactions. Resident rank 1 wasps were significantly more likely to retain their rank when potential usurpers (also rank 1 in their original nest) were introduced. Pratte and Gervet (1992) speculated that this residence effect may be associated with the resident rank 1 having a stronger chemical signature However, it has recently been shown that at the early stages of the nest foundation period, wasps cannot be distinguished by their chemical signatures (Dapporto et al. 2005). Thus, order of arrival may not necessarily be correlated with differences in chemical profiles.

Although conventions based on truly arbitrary cues are theoretically possible, it is logical to expect that these cues should instead reflect underlying differences between contenders (Nonacs 2001). One possibility is that precedence is a direct indicator of reproductive capacity. Variation in ovary size and
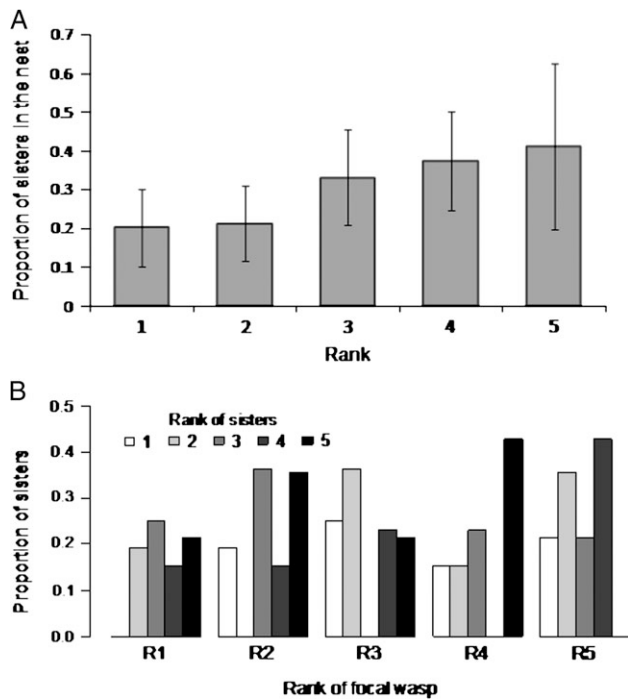
Figure 3
(A) Average proportion of nest mates that are sisters for focal wasps at each rank ($\pm$ 95% Confidence interval). (B) Distribution among other ranks of nest mates that are sisters for each of the 5 ranks (proportion of sisters was calculated as the total number of nest-mate sisters at each rank divided by the total number of foundresses at that rank).

juvenile hormone synthesis, both positively correlated with dominance, are observed in hibernating foundresses before nests are started (Röseler 1991). Foundresses of *Polistes* experimentally exposed to higher temperature and light levels at the end of the hibernation period had more developed corpora allata and ovaries and were more likely to become rank 1 (Röseler et al. 1985, 1986). Thus, if wasps exposed to early spring warmth leave their winter aggregations earlier, the order of arrival in the nest could reflect differences in reproductive capacity.

It is difficult to explain why arrival order may be important in determining only the highest positions of the hierarchy (ranks 1–2). One possibility is that the top 2 wasps are the ones that will effectively compete for the dominant position at the beginning of the nesting cycle (Cant, English, et al. 2006; Cant, Llop, and Field 2006). Hence, a convention based on order of arrival may prevent costly conflicts between the top ranked individuals, but may not be important for foundresses of lower rank.

Nevertheless, conventions based on arbitrary cues are expected to be established more frequently when differences between group members are small (Maynard Smith and Parker 1976; Nonacs 2001). In our study population, foundress associations are frequently a mixture of full sisters and nonsisters. Because differences between full sisters are likely to be smaller than those between unrelated individuals, we might expect that a convention based on arrival order would be more important between closely related individuals. Our results, however, suggest the opposite, in that arrival order may be important for only the top 2 ranks, which are often only distantly related or unrelated individuals.

Rank and kinship

Our prediction that subordinate wasps unrelated to the dominant would have higher ranks than wasps closely related to the

dominant was not confirmed. Full sisters of dominant wasps (rank 1) were evenly distributed over the remaining subordinate ranks. Moreover, rank 1 wasps frequently nested exclusively with distantly or nonrelated individuals. Thus it is unlikely that dominants are offering higher ranks as an incentive to unrelated subordinates in order to retain them in the group.

Nevertheless, our results (simple correlations and GLMs) show that social structure in foundresses associations of *P. dominulus* is correlated with kinship. Unrelated foundresses may form groups based on the prospects of nest inheritance (Queller et al. 2000). Because the likelihood of inheriting the nest decreases with decreasing rank, highly ranked wasps are more likely to obtain direct fitness benefits through inheritance (Shreeves and Field 2002). Hence, this could explain why foundresses with few or no close relatives in their groups tended to be at higher ranks. Furthermore, our results also show that low ranked wasps, although unrelated to the dominant, are frequently full sisters of rank 2 or 3 wasps, and so may obtain indirect fitness benefits when these inherit the dominant position. However, information on dominance turnover in naturally formed groups remains limited so that the full importance of nest inheritance cannot yet be determined (Shreeves and Field 2002).

Future studies may consider not only how frequently subordinates inherit the dominant position, but also whether the presence of its full sister(s) affect a foundress's chances of inheriting the dominant position through kin-based coalitions. Such coalitions are common in many vertebrates, particularly
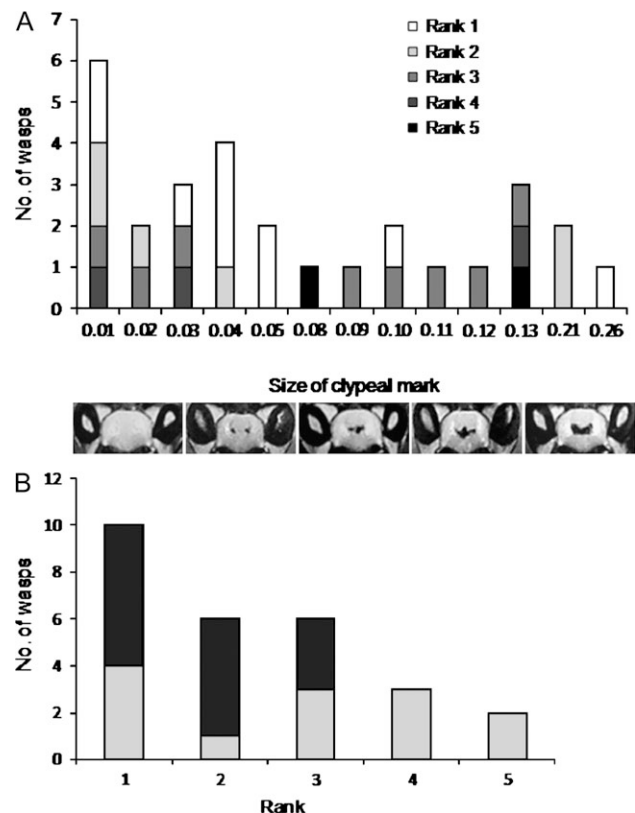

Figure 4
(A) Distribution of clypeal mark sizes among ranks (see inset legend). Images show the range of variation among marks in our study population. (B) Total number of wasps with clypeal marks at each rank (Black bars: only 1 wasp in the nest had a mark; Light-gray bars: 2 or more wasps in the nest had marks).

in primates, where they often have a direct effect on group stability, inheritance and reproductive skew (e.g., Perry et al. 2008). However, there is so far little evidence for kin-based coalitions in primitively eusocial wasps such as *Polistes*.

Rank and morphological differences

Our results suggest that body size has no effect on the establishment of dominance hierarchies. Evidence that body size could be important to the establishment of dominance hierarchies in paper wasps foundresses remains limited (e.g., Nonacs and Reeve 1995 and Cervo et al. 2008 for *P. dominulus*). In contrast, the observed correlation between rank and the size of black clypeal marks supports the idea that facial color patterns may be important in dominance subordinate interactions. It has been experimentally demonstrated that foundresses with more disrupted clypeal marks are frequently the winners of pairwise contests in *P. dominulus* (Tibbetts and Dale 2004). The results presented here provide the first evidence that clypeal marks may also be important under field conditions.

Nevertheless, females with clypeal marks were relatively uncommon in our study population and usually only 1 foundress with mark(s) was present in each group. The vast majority of groups were formed exclusively by foundresses with a completely yellow clypeus. Moreover, the GLM results have shown that when within-group kinship structure and group size were analyzed simultaneously, the size of clypeal marks had no significant effect on rank. Overall, our results are consistent with the possibility that clypeal marks represent a signal of quality, but they cannot be essential cues for the establishment of dominance hierarchies. Another possibility is that clypeal marks are a trait associated with the "sit-and-wait" nesting strategy, that is, with foundresses that instead of starting their own nest, "wait" and join established groups or adopt abandoned nests later in the season (Starks 2001). Females that join nests later in the season are faced with established social structures. Foundresses that clearly display their quality (e.g., fighting capacity) may have an increased chance of achieving high rank in such groups. In our study population, females with black clypeal marks are significantly more frequent at the end of the nest foundation period, suggesting that marks could be important for late joiners (Zanette LRS, Field J, in preparation).

CONCLUSIONS

The establishment of hierarchies in foundresses associations of *P. dominulus* is likely to be affected by within-group kinship structure. A foundress's rank is significantly correlated with the proportion of group members that is its full sisters. However, concessions in terms of rank by the dominant individual, based on relatedness alone, are unlikely to be important in the establishment of the social structure. Rank 1 wasps often have no close relative in their groups. Conventions based on precedence, that is, the order of arrival on the nest, are also unlikely to be essential determinants of rank order. Potential morphological indicators of overall quality, such as facial color patterns, may have only a limited importance in our population. Overall, a combination of direct and indirect fitness benefits through nest inheritance might explain the social structure observed in foundresses associations.

FUNDING

REFERENCES

Bourke AFG, Franks NR. 1995. Social evolution in ants. Princeton (NJ): Princeton University Press.

Bridge C, Field J. 2007. Queuing for dominance: gerontocracy and queue-jumping in the hover wasp *Liostenogaster flavolineata*. Behav Ecol Sociobiol. 61:1253–1259.

Buston P. 2003. Social hierarchies size and growth modification in clownfish. Nature. 424:145–146.

Camazine S, Deneubourg JL, Franks NR, Sneyd J, Theraulaz G, Bonabeau E. 2001. Self-organization in biological systems. Princeton (NJ): Princeton University Press.

Cant MA, English S, Reeve H, Field J. 2006. Escalated conflict in a social hierarchy. Proc R Soc B Biol Sci. 273:2977–2984.

Cant MA, Field J. 2001. Helping effort and future fitness in cooperative animal societies. ProcR Soc Lond Ser B Biol Sci. 268: 1959–1964.

Cant MA, Llop JB, Field J. 2006. Individual variation in social aggression and the probability of inheritance: theory and a field test. Am Nat. 167:837–852.

Cervo R, Dapporto L, Beani L, Strassmann JE, Turillazzi S. 2008. On status badges and quality signals in the paper wasp *Polistes dominulus*: body size, facial colour patterns and hierarchical rank. ProcR Soc B Biol Sci. 275(1639):1189–1196.

Crawley MJ. 2005. Statistics: an introduction using R. Chichester (UK): John Wiley & Sons.

Cronin A, Field J. 2007. Rank and colony defence in the facultatively eusocial hover wasp *Liostenogaster flavolineata*. Behav Ecol. 18: 331–336.

Dapporto L, Romana Dani F, Trillazzi S. 2007. Social dominance molds cuticular and egg chemical blends in a paper wasp. Curr Biol. 17(13):R504–R505.

Dapporto L, Sledge FW, Turillazzi S. 2005. Dynamics of cuticular chemical profiles of *Polistes dominulus* workers in orphaned nests (Hymenoptera, Vespidae). J Insect Physiology. 51:969–973.

Field J, Cant MA. 2006. Helping effort in primitively eusocial wasps. Ann Zool Fenn. 43(5–6):481–487.

Field J, Cronin A, Bridge C. 2006. Future fitness and helping in social queues. Nature. 441:214–217.

Field J, Shreeves G, Sumner S. 1999. Group size, queuing and helping decisions in facultatively eusocial hover wasps. Behav Ecol Sociobiol. 45:378–385.

Gamboa GJ, Dropkin JA. 1979. Comparisons of behaviors in early vs late foundress associations of the paper wasp, *Polistes metricus* (Hymenoptera, Vespidae). Can Entomol. 111:919–926.

Gamboa GJ, Stump KA. 1996. The timing of conflict and cooperation among cofoundresses of the social wasp *Polistes fuscatus* (Hymenoptera: Vespidae). Can J Zool-Rev Can Zool. 74:70–74.

Goodnight KF, Queller DC. 1999. Computer software for performing likelihood tests of pedigree relationship using genetic markers. Mol Ecol. 8(7):1231–1234.

Henshaw MT. 2000. Microsatellite loci for the social wasp *Polistes dominulus* and their application in other polistine wasps. Mol Ecol. 9:2155–2157.

Johnstone RA. 2000. Models of reproductive skew: a review and synthesis. Ethology. 106:5–26.

Legendre P, Legendre L. 1998. Numerical ecology. 2nd ed. Amsterdam (the Netherlands): Elsevier.

Liebert AE, Starks PT. 2006. Taming of the skew: transactional models fail to predict reproductive partitioning in the paper wasp *Polistes dominulus*. Anim Behav. 71:913–923.

Maynard Smith J, Parker GA. 1976. The logic of asymmetric contests. Anim Behav. 24:159–175.

Nonacs P. 2001. A life-history approach to group living and social contracts between individuals. Ann Zool Fenn. 38:239–254.

Nonacs P, Liebert AE, Starks PT. 2006. Transactional skew and assured fitness return models fail to predict patterns of cooperation in wasps. Am Nat. 167:467–480.

Nonacs P, Reeve HK. 1995. The ecology of cooperation in wasps - causes and consequences of alternative reproductive decisions. Ecology. 76:953–967.

Pardi L. 1942. Ricerche sui polistine. V. La poliginia iniziale in *Polistes gallicus*. (L.). BollIstitEntomol UnivStudi Bologna. 14:1–106.

Pardi L. 1946. Ricerche sui polistine VII: La "dominazione" e il ciclo ovario annuale in *Polistes gallicus* (L). Boll Istit Entomol Univ Studi Bologna. 15:25–84.

Perry S, Manson JH, Muniz L, Gros-Louis J, Vigilant L. 2008. Kin-biased social behaviour in wild adult female white-faced capuchins, *Cebus capucinus*. Anim Behav. 76(1):187–199.

Pratte M. 1993. Experimental changes of hierarchical rank in *Polistes dominulus* Christ foundresses. Ethology. 95:97–104.

Pratte M, Gervet J. 1992. Effects of prior residence and previous cohabitation on the *Polistes dominulus* Christ dominance hierarchy. Ethology. 90:72–80.

Premnath S, Sinha A, Gadagkar R. 1996. Dominance relationship in the establishment of reproductive division of labour in a primitively eusocial wasp (*Ropalidia marginata*). Behav Ecol Sociobiol. 39:125–132.

Queller DC, Zacchi F, Cervo R, Turillazzi S, Henshaw MT, Santorelli LA, Strassmann JE. 2000. Unrelated helpers in a social insect. Nature. 405:784–787.

Reeve HK. 1991. Polistes. In: Ross KG, Matthews RW, editors. The social biology of wasps. Ithaca (NY): Cornell University Press. p. 99–148.

Reeve HK, Keller L. 1996. Relatedness asymmetry and reproductive sharing in animal societies. Am Nat. 148(4):764–769.

Reeve HK, Keller L. 2001. Tests of reproductive-skew models in social insects. Ann Rev Entomol. 46:347–385.

Röseler PF. 1991. Reproductive competition during colony establishment. In: Ross KG, Matthews RW, editors. The social biology of wasps. Ithaca (NY): Cornell University Press. p. 309–335.

Röseler PF, Röseler I, Strambi A. 1985. Role of ovaries and ecdysteroids in dominance hierarchy establishment among foundresses of the primitively social wasp, *Polistes gallicus*. Behav Ecol Sociobiol. 18:9–13.

Röseler PF, Röseler I, Strambi A. 1986. Studies of the dominance hierarchy in the paper wasp, *Polistes gallicus* (L) (Hymenoptera-Vespidae). Monit ZoolItal Ital J Zool. 20:283–290.

Seppä P, Queller DC, Strassmann JE. 2002. Reproduction in foundress associations of the social wasp, *Polistes carolina*: conventions, competition, and skew. Behav Ecol. 13:531–542.

Shreeves G, Cant MA, Bolton A, Field J. 2003. Insurance-based advantages for subordinate co-foundresses in a temperate paper wasp. Proc Roy SocLond Ser B Biol Sci. 270:1617–1622.

Shreeves G, Field J. 2002. Group size and direct fitness in social queues. Am Nat. 159:81–95.

Sledge MF, Boscaro F, Turillazzi S. 2001. Cuticular hydrocarbons and reproductive status in the social wasp *Polistes dominulus*. Behav Ecol Sociobiol. 49:401–409.

Starks PT. 2001. Alternative reproductive tactics in the paper wasp Polistes dominulus with specific focus on the sit-and-wait tactic. Ann Zool Fenn. 38:189–199.

Strassmann J. 2001. The rarity of multiple mating by females in the social Hymenoptera. Insectes Soc. 48:1–13.

Strassman JE, Solis CR, Peters JM, Queller DC. 1996. Strategies for finding and using highly polymorphic DNA microsatellite loci for studies of genetic relatedness and pedigrees. In: Ferraris JD, Palumbi SR, editors. Molecular Zoology Advances, Strategies, and Protocols. New York: Wiley-Liss. p. 163–178.

Sullivan JD, Strassmann JE. 1984. Physical variability among nest foundresses in the polygynous social wasp, Polistes-Annularis. Behav Ecol Sociobiol. 15:249–256.

Tibbetts EA. 2002. Visual signals of individual identity in the wasp *Polistes fuscatus*. Proc Roy Soc Lond Ser B Biol Sci. 269:1423–1428.

Tibbetts EA, Dale J. 2004. A socially enforced signal of quality in a paper wasp. Nature. 432:218–222.

Tindo M, Dejean A. 2000. Dominance hierarchy in colonies of *Belonogaster juncea juncea* (Vespidae, oPlistinae). Insectes Soc. 47:158–163.

Turillazzi S, Mastrobuoni G, Dani FR, Moneti G, Pieraccini G, la Marca G, Bartolucci G, Perito B, Lambardi D, Cavallini V, et al. 2006. Dominulin A and B: two new antibacterial peptides identified on the cuticle and in the venom of the social paper wasp *Polistes dominulus* using MALDI-TOF, MALDI-TOF/TOF, and ESI-ion trap. J Am Soc Mass Spectr. 17:376–383.

West MJ. 1967. Foundress associations in polistine wasps: dominance hierarchies and the evolution of social behavior. Science. 157:1584–1585.

Wilson EO. 1971. The insect societies. Cambridge (MA): Harvard University Press.

Wilson EO. 1975. Sociobiology: the new synthesis. Cambridge (MA): Harvard University Press.

Zacchi F, Cervo R, Turillazzi S. 1996. How *Polistes semenowi*, obligate social parasite, invades the nest of its host *Polistes dominulus* (Hymenoptera, Vespidae). Insect Soc Life. 1:125–130.

Zanette LRS, Field J. 2008. Genetic relatedness in early associations of *Polistes dominulus*: from related to unrelated helpers. Mol Ecol. 17(11):2590–2597.