THE ROYAL
SOCIETY

# Regulation of queen–worker conflict in bumble-bee (*Bombus terrestris*) colonies

**Guy Bloch**†

*Department of Zoology, G. S. Wise Faculty of Life Sciences, Tel Aviv University, 69978 Tel Aviv, Israel*
*(gbloch@life.uiuc.edu)*

In annual colonies of bumble-bees overt queen–worker conflict is limited to a distinct 'competition phase' (CPh). In unmanipulated *Bombus terrestris* colonies, the queen's switch to male production (the 'switch point', SP) accounted for only 22% of the variation in the onset of the CPh. In some colonies, the CPh even began before the SP. The CPh was more strongly correlated with the transition in queen production ($r = 0.79$). Replacing the queen eggs with male eggs or doubling the number of workers in young colonies resulted in a significantly earlier onset of the CPh and a significantly earlier transition to queen production. Replacing queen eggs with female eggs did not have this effect. These manipulations did not affect the timing of the queen's switch from female to male production. These findings show that the mechanism underlying the queen–worker conflict in insect societies is more complex than previously appreciated. The onset of queen–worker conflict cannot be attributed simply to a single factor such as the queen's switch to male production or a decrease in queen inhibition. Rather, multiple cues are important.

**Keywords:** social evolution; bumble-bees; reproduction; queen–worker conflict; caste determination

## 1. INTRODUCTION

Unlike queens, most workers in colonies of social insects remain sterile and help the queen raise her offspring. This apparent altruism on the part of the workers is difficult to explain under simple 'selfish gene' models of natural selection. A plausible explanation for worker sterility in the social Hymenoptera is provided by kin-selection theory. Worker fitness is expected to be higher when assisting the queen in raising her daughters than when raising the worker's own offspring. However, overt conflict over reproduction is predicted to occur if the queen switches to male production. This is because a worker's fitness is higher when raising her own sons rather than when raising the queen's sons (Hamilton 1964, 1972; Trivers & Hare 1976). Most experimental studies pertaining to the proximate mechanisms of regulation of worker reproduction posit that workers are inhibited by the queen. The onset of overt queen–worker conflict is explained by a decrease in queen inhibition either due to changes in her pheromones or due to an increase in worker number (reviewed in Wilson 1971; Fletcher & Ross 1985; Röseler & Van Honk 1990; Spradbery 1991; Heinze *et al.* 1994). This gap between the kin-selection theory-based prediction and the observed mechanism is not easily resolved into one comprehensive model (Keller & Nonacs 1993; Bourke 1994; Bourke & Franks 1995). Moreover, although queen–worker conflict in hymenopteran societies has gained impressive theoretical attention, only a few studies (e.g. Ratnieks & Visscher 1989; Mueller 1991) have tested the predictions of the different theories by manipulating colonies in a way which affected the timing or resolution of the conflict.

The bumble-bee *Bombus terrestris* is an excellent model for testing theories of queen–worker conflict. Overt conflict is restricted to a well-characterized 'competition phase' (CPh) (Duchateau & Velthuis 1988). During this stage a subgroup of dominant workers reproduce and destroy eggs laid by the queen and other laying workers. Annual colonies are founded in the spring by a single queen who apparently mates once (Estoup *et al.* 1995; Schmid-Hempel 1998). Thus, the asymmetry in relatedness among colony members is expected to be pronounced (Hamilton 1964; Trivers & Hare 1976). At the initial stage of colony development the queen lays fertilized (diploid) eggs which develop into females; she later switches to produce unfertilized (haploid) eggs which develop into males (the 'switch point', SP) (Duchateau & Velthuis 1988). Thus, during a season a predictable change in worker relatedness to the brood occurs. While all first batches of diploid eggs develop into workers, the later batches often (but not always) develop into queens. This typical pattern of colony development is maintained under laboratory conditions, indicating that environmental factors such as photoperiod, ambient temperatures or floral resource abundance have limited effects (e.g. Van der Blom 1986; Van Doorn & Heringa 1986; Duchateau & Velthuis 1988; Bloch & Hefetz 1999*a*).

In normally developing colonies, the processes of queen ageing, increased worker number, and the switch from female to male rearing are all coupled together and associated with seasonal environmental changes (Wilson 1971; Michener 1974). Thus, it is difficult to unravel the relative importance of each of these factors in the onset of queen–worker conflict. Here, I describe experiments which

---

†Present address: Department of Entomology, University of Illinois, 320 Morrill Hall, Urbana, IL 61801, USA.

*Proc. R. Soc. Lond.* B (1999) **266**, 2465–2469
*Received* 9 August 1999 *Accepted* 22 September 1999

explore the mechanism of the regulation of reproduction by manipulating colonies in a way which affects the timing of the conflict. To do this, I first characterized the development of normal colonies in the laboratory. I then simultaneously manipulated the number of workers and the sex of the brood in young colonies. If the switch to male production is a cause of the onset of the conflict, then artificial alternation of the sex of the brood, but not of worker number, is expected to affect the timing of the onset of the CPh. On the other hand, if the increase in worker number is an important cue (possibly because there are more workers than the queen can control), then artificial alternation of worker number but not of the sex of the brood is expected to affect the timing of the onset of the CPh.

## 2. MATERIAL AND METHODS

### (a) *The duration of development from egg to adult*

All colonies used in this study were obtained from Bio-Bee Sde-Eliahu Industry, Bet Shean, Israel, one to a few days after the emergence of the first worker (for details on colony maintenance see Bloch & Hefetz (1999*a*)).

To monitor egg to adult development, the combs of 15 different colonies were sketched on a daily basis. The development of eggs to larvae and pupae was identified by the typical changes in the brood cups (Wilson 1971; Michener 1974). Gender was determined upon adult emergence.

### (b) *Characterization of colony development*

In order to explore the relationships between the onset of the CPh and the production of reproductives, I monitored unmanipulated colonies ($n=92$) for signs of competition and for adult gyne and male emergence. The onset of the CPh was identified when one or more of the following events were observed: (i) worker oviposition, (ii) egg eating (oophagy), (iii) clear signs of egg cup destruction, and (iv) two or more open egg cups over two or more successive days. Because bumble-bee queens typically construct one egg cup at a time, the occurrence of two open egg cups is a sign that workers are also constructing egg cups or that an already sealed cup has been destroyed (Duchateau & Velthuis 1988; Bloch & Hefetz 1999*a*).

Because the combs of these colonies were not sketched on a daily basis, the time of the deposition of the eggs which developed into the first male and queen was estimated retrospectively by subtracting the duration of development from the day of first male and queen emergence. The duration of development was corrected for colony age because I found that the duration of development changes as the colony ages (see equations (1) and (2) in § 3).

### (c) *The effects of the number of workers and sex of the brood on colony development*

To explore the significance of population size and the switch to male production on the onset of the CPh, the sex of the brood and the number of workers were simultaneously manipulated in young colonies. Three trials were performed, each including eight colonies of the same age. Half of the colonies in each trial were set to have a mean number of workers (×1 colonies). The other four colonies were set to have twice the mean number of workers (×2 colonies). Each day the mean number of workers was calculated by counting the number of workers in ×1 colonies. Worker numbers were adjusted by adding or removing the appropriate number of callow workers. To control the sex of the brood in these colonies, all the queen-laid eggs were replaced by eggs obtained from 'donor queens'. Two different types of donor queens were used: (i) young queens from colonies up to seven days after the emergence of the first worker, and (ii) old queens from colonies during the CPh. Because bumble-bee queens typically lay female eggs during the initial stage of colony development and later switch to male production (e.g. Van der Blom 1986; Van Doorn & Heringa 1986; Duchateau & Velthuis 1988), young donor queens were expected to lay female eggs and old donor queens were expected to lay male eggs. The manipulation procedure achieved the expected changes in the sex of the brood. The introduction of eggs from old donor queens into recipient colonies resulted in a significantly earlier transition from female to male brood rearing relative to colonies which received eggs from young donor queens (first egg which developed into a male $10.8 \pm 1.2$ and $n=12$ and $5.4 \pm 0.4$ and $n=9$, respectively and last egg developed into a female $17.8 \pm 2.4$ and $n=12$ and $9.1 \pm 2.0$ and $n=8$, respectively; two-way ANOVA, $F_{1,19}=12.9$ and $p=0.002$ and $F_{1,18}=6.5$ and $p=0.02$, respectively). The changes in the sex of the brood are the result of brood introduction because all eggs laid by the queen were removed. Colonies ($n=3$) in which the introduced eggs were not accepted or which did not have the expected gender were omitted from later analyses. The survival in egg cups from old donor queens (male eggs) and young donor queens (female eggs) was similar in the recipient colonies for the five days after the first introduction (day 10 when most of the eggs had not yet hatched 81.1 and 89.9%, respectively and *G*-test of independence $p=0.1$; Sokal & Rohlf 1995). A lower percentage of the old donor queens' brood (51.3 versus 87.7%, $p<0.0001$) survived to the last day of the manipulations (day 15) or to the onset of the CPh (in colonies for which the CPh started before day 15).

Manipulations of the number of workers began on day 1 after the emergence of the first worker. Manipulations of the sex of the brood began on day 5. This was done in order to avoid possible interference of the removal of the egg cup in the normal development of the colony. Manipulations were performed daily, except for day 10, until day 15. Colony development was monitored by direct observations and by comb tracing (see § 2(a)).

To verify the sex of the eggs laid by the queens heading the experimental colonies, the eggs were removed and introduced into young 'nursing colonies' where they were reared to pupation. The pupae were then transferred into separate containers where the emerging adults were sexed. Only colonies in which the changes in the sex of the brood were determined unequivocally were included in analyses.

Because young larvae remain in a common brood cup for several days after hatching, it was not always possible to count the number of larvae in a colony directly (Wilson 1971; Michener 1974). Two different methods were therefore used to estimate the number of larvae. First, the number of larvae was deduced from the number of pupae produced from larva cups present at the focal day and, second, the area of the larvae on the comb sketches (see above) was calculated by using an image analyser (Olympus, CUE-3). Accordingly, the worker:larva ratio was calculated either as the worker:larva numerical ratio or worker:larva area ratio. Estimations were made either at the end of the manipulations (day 15) or at the onset of the CPh if the CPh began before the end of manipulations.

## 3. RESULTS

### (a) *The duration of development from egg to adult*

The duration of development depended on the colony age (time after the emergence of the first worker). Male development lasted 24–31 days (mean ± s.e. $= 27.1 \pm 0.4$ days) and the regression equation ($p < 0.0001$, $r^2 = 0.49$ and $n = 28$) was

$$D_\mathrm{d} = 0.19 \times E_\mathrm{m} + 18.88, \qquad (1)$$

where $D_\mathrm{d}$ is the duration of development (days) and $E_\mathrm{m}$ is the colony age (days) at the emergence of the first male. Queen development lasted 28–34 days (mean ± s.e. $= 30.7 \pm 0.4$ days) and the regression equation ($p < 0.002$, $r^2 = 0.46$ and $n = 18$) was

$$D_\mathrm{d} = 0.24 \times E_\mathrm{q} + 19.39, \qquad (2)$$

where $E_\mathrm{q}$ is the colony age (days) at the emergence of the first queen.

### (b) *Characterization of colony development*

The CPh began before the SP in 7.7% of the unmanipulated colonies. If we consider the maximal and minimal durations of development for the first male in each colony (using the 95% confidence limits of the regression line in equation (1)), the onset of the CPh preceded the SP in 3.8–9% of the colonies. In colonies in which the SP preceded the onset of the CPh, the SP accounted for a significant proportion of the variation in the time to onset of the CPh (linear regression analysis, $Y = 0.53X + 19.53$, $n = 71$, $p < 0.0001$ and $r^2 = 0.22$, where $X$ is the number of days to the SP and $Y$ is the number of days to the onset of the CPh). These results show that in most (but not in all) colonies the onset of the CPh occurred after the queen's switch to male production.

There was a strong correlation between the onset of the CPh and the transition to queen production ($r = 0.79$, $n = 62$ and $p < 0.0001$).

To assess the relationship between colony size and the initiation of the CPh, the number of workers was counted at the onset of the CPh in 14 colonies. The number varied between 16 and 54, with a mean of 28.7 (s.d. = 10.9).

### (c) *The effects of the number of workers and sex of the brood on colony development*

The queens' reproduction in the recipient colonies was not affected by the manipulations. The time at which the queens switched to male production, as well as their fecundity, did not differ between treatments (table 1). In contrast, both the introduction of male eggs and the doubling of the number of workers resulted in a significant and independently earlier onset of the CPh (figure 1; two-way ANOVA, brood sex $F_{1,17} = 8.0$ and $p = 0.01$, worker number $F_{1,17} = 11.3$ and $p = 0.004$ and interaction $F_{1,17} = 0.45$ and $p = 0.51$). Likewise, both factors had similarly significant and independent effects on the timing of the deposition of the egg which eventually developed into the first queen (note that not all the colonies produced queens) (figure 1; two-way ANOVA, brood sex $F_{1,10} = 10.2$ and $p = 0.01$, worker number $F_{1,10} = 8.0$ and $p = 0.02$ and interaction $F_{1,10} = 0.07$ and $p = 0.8$). In a separate comparison it was found that both the onset of the CPh and the transition to queen production in female ×1 colonies were similar to that in normally developing colonies (CPh in normal colonies $24.0 \pm 0.7$ days and $n = 92$ and CPh in manipulated colonies $24.0 \pm 2.1$ days and $n = 6$, $t$-test, $t_{96} = 0.99$ and no significance; transition to queen production in normal colonies $13.2 \pm 0.6$ days and $n = 71$ and transition to queen production in manipulated colonies $14.0 \pm 1.0$ days and $n = 3$, $t$-test, $t_{72} = 0.76$ and no significance). These last comparisons show that the manipulation procedure by itself (replacement of egg cups and adjustment of the number of workers) did not affect colony development. In male ×2 colonies there was an average of 20.8 workers (s.e. = 5.6 and $n = 5$) and maximum of 26 workers at the onset of the CP. This maximum is lower than the average in normally developing colonies (28.7, see above).

Table 1 *Time elapsed from the emergence of the first worker to the queen's switch from female to male production (days) and queen's fecundity (egg cups per day) in* B. terrestris *colonies for which the number of workers and sex of the brood were manipulated*

(Means ± s.e. (number of queens). ×1, average number of workers; ×2, double the average number of workers; male, introduced male brood; female, introduced female brood. There were no significant differences between the different treatments for all three parameters presented (two-way ANOVA, $p > 0.1$).)

| treatment | queen fecundity | queen first haploid egg | queen last diploid egg |
|---|---|---|---|
| female, ×1 | 1.62 ± 0.20 (6) | 13.00 ± 1.35 (4) | 14.50 ± 0.96 (4) |
| female, ×2 | 2.31 ± 0.25 (5) | 11.00 ± 1.87 (4) | 10.00 ± 3.21 (3) |
| male, ×1 | 2.00 ± 0.14 (3) | 10.67 ± 1.33 (3) | 12.50 ± 2.02 (4) |
| male, ×2 | 2.19 ± 0.41 (3) | 9.67 ± 2.19 (3) | 9.50 + 3.50 (2) |

To evaluate the relationship between the switch to male brood and onset of the CPh further, separate regression analyses were performed for the manipulated ×1 and ×2 colonies. In both cases, the first deposition of male eggs (experimentally introduced or naturally laid by the queen after the end of manipulations) accounted for a significant proportion of the variance in the onset of the CPh (×1 colonies, $r^2 = 0.57$, $n = 10$ and $p = 0.01$ and ×2 colonies, $r^2 = 0.39$, $n = 11$ and $p = 0.04$). These coefficients of determination ($r^2$-values) are higher than in normally developing colonies ($r^2 = 0.22$, see above).

The lower survival rate of brood obtained from old donor queens resulted in trends of higher worker:larva numerical ratios ($1.4 \pm 0.3$ and $0.8 \pm 0.2$, respectively; two-way ANOVA, $F_{1,16} = 3.4$ and $p = 0.08$) and significantly higher worker:larva area ratios ($0.13 \pm 0.03$ and $0.06 \pm 0.01$, respectively; two-way ANOVA, $F_{1,16} = 6.5$ and $p = 0.02$) for colonies in which male brood was introduced compared to those that received female brood. In colonies that received female eggs, the worker:larva ratio accounted for around 50% of the variation in the onset of the CPh (linear regression analysis, numerical ratio $n = 12$, $r^2 = 0.48$ and $p = 0.01$ and area ratio $n = 12$, $r^2 = 0.49$ and $p = 0.01$). In contrast, when the analysis was limited to colonies which received male brood, the worker:larva ratio did not account for a significant proportion of the variation in the time to onset of the CPh (linear

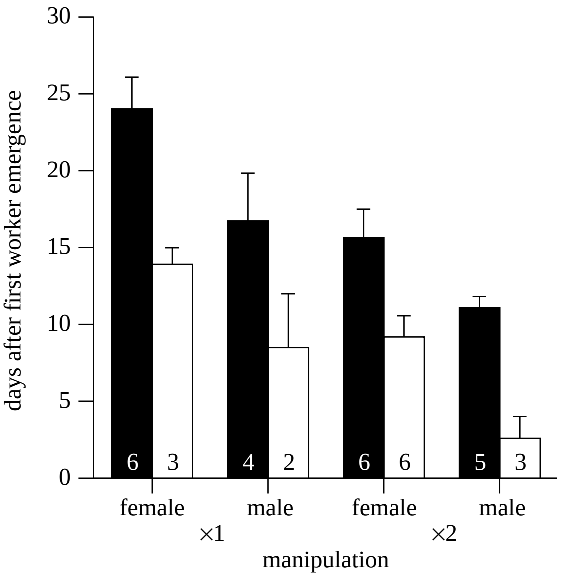

Figure 1. Time elapsed (days) (mean ± s.e.) from the emergence of the first worker to the initiation of the CPh (filled bars) and to the transition in queen production (deposition of the egg developing into the first queen) (open bars) in *B. terrestris* colonies for which the number of workers and sex of the brood were manipulated. The legends for the treatments are as in table 1. The changes in both the sex of the brood and number of workers, but not the interaction between them, had significant effects on both the onset of the CPh and the transition in queen production ($p < 0.05$). The sample sizes (number of colonies) are given within the bars.

regression analysis, numerical ratio $n = 8$, $r^2 = 0.003$ and $p = 0.89$ and area ratio $n = 8$, $r^2 = 0.12$ and $p = 0.4$). These analyses suggest that the differences in the timing to onset of the CPh are due to the changes in the sex of the brood and not because of differences in the worker:larva ratio.

## 4. DISCUSSION

The principal importance of this study is that it demonstrates that both changes in the number of workers and in the sex of the brood alter the timing of the onset of queen–worker conflict. The results are also consistent with the hypothesis that common social mechanisms affect both the eruption of conflict over reproduction and the onset of queen production (Michener 1974). First, there was a high correlation between the onset of the CPh and the transition to queen production in normally developing colonies (see also Duchateau & Velthuis 1988). Second, and more importantly, I have demonstrated that introducing male brood and increasing the number of workers resulted in a similar acceleration of both the onset of the CPh and the transition to queen production. Because the CPh also occurs in colonies which do not produce queens, it seems unlikely that queen production by itself causes initiation of the CPh.

It seems unlikely that genetic or physiological differences between the queens can explain the results because the different manipulations were randomly assigned to colonies. It also is unlikely that environmental differences can account for the results because the colonies were not exposed to the external environment but housed under similarly controlled conditions. Likewise, it seems that treatment-dependent changes in queen reproduction cannot account for the observed results because there were no detectable changes in behaviour, fecundity or time of switching to male production between queens of colonies which received different treatments (table 1).

The mechanism underlying queen–worker conflict in bumble-bees seems to be more complex than previously appreciated. The results above clearly show that neither a decline in queen inhibition (the prevailing proximal explanation) nor the switch to male production (predicted by kin-selection theory) is sufficient to explain the eruption of conflict. In terms of queen inhibition, there is no simple explanation for how changes in the sex of the brood or an increase in the worker:larva ratio could affect the distribution, quality or quantity of the queen pheromone. The decline in queen inhibition hypothesis is also inconsistent with earlier studies showing that live or body extracts of queens from colonies before and during the CPh had a similar inhibitory effect on the behaviour and reproduction of callow workers (Bloch *et al.* 1996; Bloch & Hefetz 1999*b*). Even if the queen pheromone does not change, queen inhibition can decline because she cannot control the increasing number of workers as the colony matures (Van Honk & Hogeweg 1981; Van Doorn & Heringa 1986). This hypothesis also predicts that the onset of conflict will occur earlier in colonies for which the number of workers is doubled. However, in all the male ×2 colonies, the CPh started when 26 or fewer workers were present. This number of workers is lower than the average in normally developing colonies at the onset of the CPh. In addition, earlier studies have shown that an increase in colony size does not necessarily result in fewer encounters with the queen and in reduced worker inhibition. For example, those workers who meet the queen most frequently and, thus, are expected to be most strongly inhibited commonly become the principal egg layers (Van Doorn & Heringa 1986; Van Honk & Hogeweg 1981). Finally, ovarian development and the rates of biosynthesis of JH (the presumed gonadotropic hormone in *B. terrestris*; Röseler & Van Honk 1990; Bloch *et al.* 2000) were low for workers in colonies during the CPh when queen inhibition is expected to be reduced (Bloch & Hefetz 1999*a*). Thus, although the results above clearly show that an increase in the number of workers causes an earlier onset of the CPh, they do not show that the mechanism underlying these changes is a decrease in the queen's ability to control workers in large colonies.

Kin-selection theory predicts that an overt conflict between the workers and the queen should erupt when the queen switches to male production (Hamilton 1964; Trivers & Hare 1976). The results presented above suggest that the sex of the brood is important. (i) Introducing male eggs into young colonies resulted in a significantly earlier onset of the CPh. (ii) In most normally developing and manipulated colonies, the switch to male production occurred before the onset of the CPh. (iii) In normally developing colonies, the switch to male production accounted for a significant proportion of the variation in the timing to the onset of the CPh; the proportion was higher in the manipulated colonies for which colony size was controlled.

However, the results above show that the switch to male production is not the only cause of the onset of an overt conflict between the queen and the workers. In 7.7% of the normally developing colonies, the onset of the CPh occurred before the queen switched to male production; in the rest of the colonies the SP accounted for only 22% of the variation in the time to the onset of the CPh. More importantly, the hypothesis that the CPh starts because the queen switches to male production cannot explain why the onset of the CPh occurred significantly earlier in colonies for which the number of workers was doubled.

This study shows that both the sex of the brood and the number of workers are key social determinants of the timing of queen–worker conflict and the transition to queen production in bumble-bees. The experimental data presented here, along with recent theoretical evolutionary analyses, call for recognition that the number of workers along with kin structure is an important social factor underlying the regulation of reproduction and caste determination. In a wider perspective, this study is consistent with the emerging acknowledgement that multiple factors are important in the evolution and organization of animal societies (Alexander 1991; Bourke 1994, 1999).

I thank Abraham Hefetz for supporting this study and for critical discussions, Kenneth G. Ross, Susan E. Fahrbach, Samuel Beshers, Gene E. Robinson, Zhi-Yong Huang, and two anonymous reviewers for reading and commenting on previous versions of this manuscript, and Ori Oren for expert help with the image analyses. This research was carried out through the generous support of the Josef Buchmann Doctoral Fellowship Fund (to G.B.) and a Binational Agriculture Research and Development (BARD) grant no. IS-2306–93 (to A.H.).

## REFERENCES

Alexander, R. D., Noonan, K. M. & Crespi, B. J. 1991 The evolution of eusociality. In *The biology of the naked mole-rat* (ed. P. W. Sherman, J. U. M. Jarvis & R. D. Alexander), pp. 3–44. Princeton University Press.

Bloch, G. & Hefetz, A. 1999*a* Regulation of reproduction by dominant workers in bumble bee (*Bombus terrestris*) queenright colonies. *Behav. Ecol. Sociobiol.* **45**, 125–135.

Bloch, G. & Hefetz, A. 1999*b* Reevaluation of the role of mandibular glands in regulation of reproduction in bumblebee colonies. *J. Chem. Ecol.* **25**, 881–896.

Bloch, G., Borst, D. W., Robinson, G. E., Huang, Z.-Y. & Hefetz, A. 1996 Effects of social conditions on juvenile hormone mediated reproductive development in *Bombus terrestris* workers. *Physiol. Entomol.* **21**, 257–267.

Bloch, G., Borst, D. W., Huang, Z.-Y., Robinson, G. E., Cnaani, J. & Hefetz, A. 2000 Juvenile hormone titers, juvenile hormone biosynthesis, ovarian development and social environment in *Bombus terrestris*. *J. Insect Physiol.* **46**, 47–57.

Bourke, A. F. G. 1994 Worker matricide in social bees and wasps. *J. Theor. Biol.* **167**, 283–292.

Bourke, A. F. G. 1999 Colony size, social complexity and reproductive conflict in social insects. *J. Evol. Biol.* **12**, 245–257.

Bourke, A. F. G. & Franks, N. R. 1995 *Social evolution in ants*. Princeton University Press.

Duchateau, M. J. & Velthuis, H. H. W. 1988 Development and reproductive strategies in *Bombus terrestris* colonies. *Behaviour* **107**, 186–207.

Estoup, A., Scholl, A., Pouvreau, A. & Solignac, M. 1995 Monoandry and polyandry in bumble bees (Hymenoptera: Bombinae) as evidenced by highly variable microsatellites. *Mol. Ecol.* **4**, 89–93.

Fletcher, D. J. C. & Ross, K. G. 1985 Regulation of reproduction in eusocial Hymenoptera. *A. Rev. Entomol.* **30**, 319–343.

Hamilton, W. D. 1964 The genetical evolution of social behaviour. I and II. *J. Theor. Biol.* **7**, 1–52.

Hamilton, W. D. 1972 Altruism and related phenomena in social insects. *A. Rev. Ecol. Syst.* **3**, 193–232.

Heinze, J., Hölldobler, B. & Peeters, C. 1994 Conflict and cooperation in ant societies. *Naturwissenschaften* **81**, 489–497.

Keller, L. & Nonacs, P. 1993 The role of queen pheromones in social insects: queen control or queen signal? *Anim. Behav.* **45**, 787–794.

Michener, C. D. 1974 *The social behavior of the bees*. Cambridge, MA: Belknap Press of Harvard University Press.

Mueller, U. G. 1991 Haplodiploidy and the evolution of facultative sex ratios in primitively eusocial bee. *Science* **254**, 442–444.

Ratnieks, F. L. W. & Visscher, P. K. 1989 Worker policing in the honey bee. *Nature* **342**, 796–797.

Röseler, P. F. & Van Honk, C. G. J. 1990 Castes and reproduction in bumblebees. In *Social insects, an evolutionary approach to castes and reproduction* (ed. W. Engels), pp. 147–166. Berlin: Springer.

Schmid-Hempel, R. 1998 Female mating frequencies in *Bombus* spp. from central Europe. In *The Proceedings of the 13th International Congress of the International Union for the Study of Social Insects* (ed. M. P. Schwarz & K. Hogendoorn), p. 422. Adelaide, Australia: Finders University Press.

Sokal, R. R. & Rohlf, F. J. 1995 *Biometry*, 3rd edn. New York: Freeman & Co.

Spradbery, J. P. 1991 Evolution of queen number and queen control. In *The social biology of wasps* (ed. K. G. Ross & R. W. Matthews), pp. 336–388. Ithaca, NY: Cornell University Press.

Trivers, R. L. & Hare, H. 1976 Haplodiploidy and the evolution of the social insects. *Science* **191**, 249–263.

Van der Blom, J. 1986 Reproductive dominance within colonies of *Bombus terrestris* (L.). *Behaviour* **97**, 37–49.

Van Doorn, A. & Heringa, J. 1986 The ontogeny of a dominance hierarchy in colonies of the bumble bee *Bombus terrestris* (Hymenoptera: Apidae). *Insectes Soc.* **33**, 3–25.

Van Honk, C. G. J. & Hogeweg, P. 1981 The ontogeny of the social structure in a captive *Bombus terrestris* colony. *Behav. Ecol. Sociobiol.* **9**, 111–119.

Wilson, E. O. 1971 *The insect societies*. Cambridge, MA: Belknap Press of Harvard University Press.