



# Patterns of reproductive differentiation and reproductive plasticity in the major evolutionary transition to superorganismality

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Major evolutionary transitions in individuality are characterised by the formation of new levels of biological complexity from the cooperation of previously independent lower-level units. The evolution of superorganismality in insects is one such major transition, and is characterised by an extreme division of reproductive labour between ancestrally autonomous units, in the form of queen and worker castes. Here, we discuss the nature of plasticity in the emergence of castes across the major transition to superorganismality in insects. We identify key changes in plasticity which act at different levels of selection: a loss of reproductivity plasticity at the individual level is matched by a gain in plasticity at the colony level. Taking multi-level selection into consideration has important implications for formulating testable hypotheses regarding the nature of plasticity in a major transition from a lower to a higher level of biological complexity.

## Addresses

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## Introduction

The evolution of life on earth has been shaped by a number of major transitions in individuality [1,2]. Each of these transitions has involved the formation of new, more complex individuals from the cooperation of previously independently replicating units [1,2]. Canonical major evolutionary transitions include the transition from independently living unicells to multicellular organisms [3,4], the formation of the eukaryotic cell from the conjunction of prokaryotic cells [5,6], and the evolution of highly

cohesive superorganismal insect societies from solitary ancestors [7<sup>••</sup>,8<sup>••</sup>].

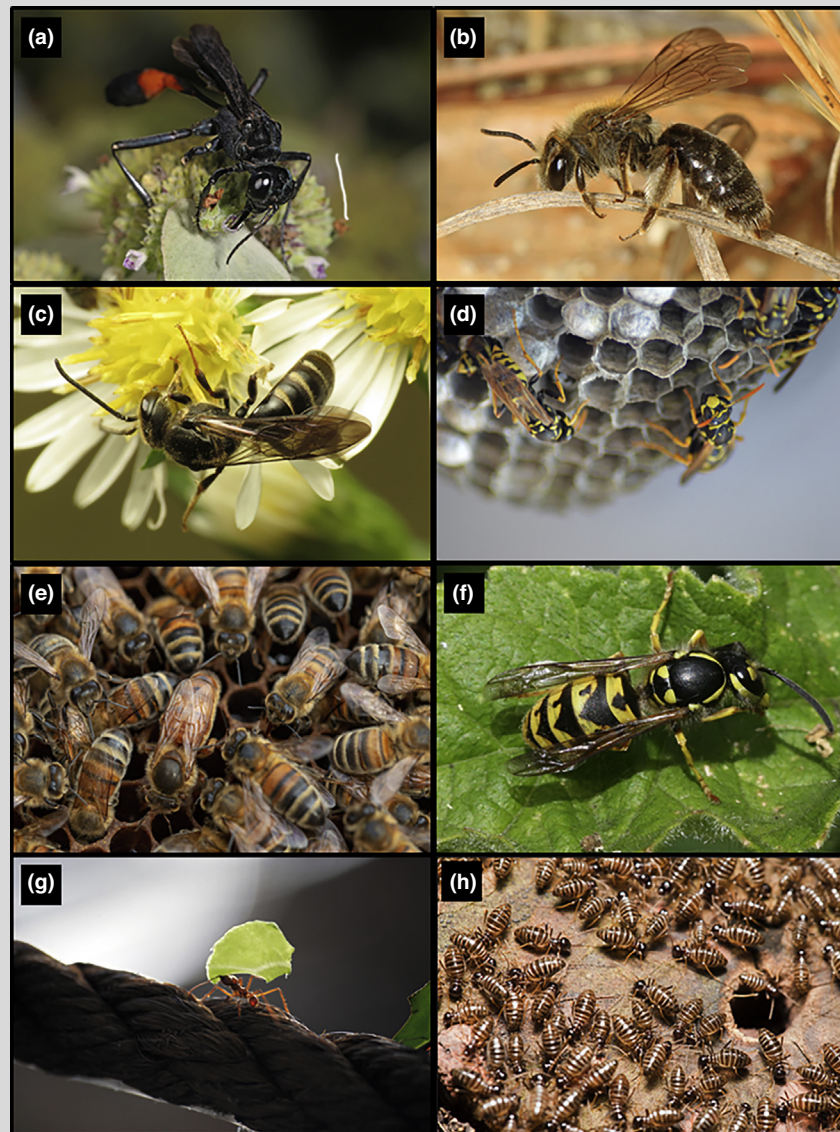
Despite having played a large role in the evolutionary history of life, major evolutionary transitions in individuality have proven difficult to study. Transitions such as the formation of the genome and the formation of the eukaryote cell occurred in the distant past, and probably only once each. By contrast, the transition to complex insect sociality has occurred more recently, and multiple times independently, in the corbiculate bees, termites, ants and vespine wasps [7<sup>••</sup>,9]. Superorganismal colonies are easily decomposed and manipulated, and, therefore, provide excellent models with which to study major evolutionary transitions and unravel the proximate and ultimate foundations that underlie major shifts in individuality [8<sup>••</sup>].

Here, we review the role that reproductive plasticity has played in the transition from a solitary to a superorganismal lifestyle. A fundamental aspect of each major transition is the functional differentiation and division of labour among the lower-level units that together constitute a new higher-level unit of individuality [1,2]. The evolution of multicellularity, for example, has involved a transition from phenotypically flexible unicells to higher-level organisms with an obligate division of reproductive labour between germ and somatic cells [3]. This is mirrored in the transition to insect superorganismality, which is defined by a reduction in phenotypic plasticity and a division of reproductive labour between reproductive queens and non-reproductive workers [7<sup>••</sup>,8<sup>••</sup>].

We argue that the emergence of a fixed reproductive division of labour does not so much represent a loss of reproductive plasticity among lower-level units but a transfer of plasticity between levels of selection — from the individual to the colony. We review the proximate mechanisms that may underlie this shift and the ultimate pressures that may have driven it.

## Social organisation and reproductive plasticity

Insect sociality covers a broad spectrum of social systems of varying levels of social complexity. While a range of classification systems have been proposed [7<sup>••</sup>,10,11], most insect species broadly fall into one of three categories (Box 1). Solitary breeding is

**Box 1 Levels of insect sociality**

**Solitary insect species (a-b)**, in which individuals reproduce independently and rely heavily upon direct reproduction to pass on their genes, comprise the majority of all insects. Solitary breeding is the basal state for all insect lineages.

**Cooperatively breeding insect species** (commonly referred to as 'primitively eusocial' species, **c-d**) are those in which individuals form cohesive groups with division of labour between reproductive and non-reproductive individuals. Within such species, some proportion of individuals are reliant upon indirect fitness gains from altruistic behaviour directed towards relatives. However, commitment to reproductive roles within such societies is not complete: individuals retain the plasticity to switch between non-reproductive and reproductive roles. Model examples of cooperatively breeding insects include halictid bees, paper wasps and many 'lower' termites.

**Superorganismal insect species** (commonly referred to as 'complex eusocial' species, **e-h**) are those in which reproductive roles are irreversibly determined during development, resulting in extremely cohesive groups in which the majority of individuals are morphologically distinct workers. Superorganismal species include honeybees, vespine wasps, most 'higher' termites, and the large majority of ant species.

Not all species will fit squarely within these categories: some insects may facultatively switch between solitary and cooperatively breeding social systems dependent upon prevailing ecological conditions, for example in the case of carpenter bees and sweat bees [44]. Nonetheless, the majority of species possess a social structure that is approximately described by one of these three broad categories and there are clear adaptations that differentiate the three groups from one another, most notably the presence of reproductive division of labour and the subsequent obligate fixation of that division of labour [38,45]. As such it is possible to make meaningful predictions about the proximate and ultimate factors that have contributed to the evolutionary transition between each category.

It is well-established that the transition to superorganismality represents a major evolutionary transition in individuality [1,2,6], but disagreement exists regarding the exact nature of this transition. It is possible to conceptualise the major transition as spanning the entire range of sociality from solitary to superorganismal species [45], with cooperatively breeding species representing stable but approximately intermediate stages within this transition. A recent set of publications have argued against this view, proposing instead that there is a unique and qualitative evolutionary discontinuity between cooperatively breeding and superorganismal species [7\*\*,46,47]. Within this model, the transition to cooperative living is only a difference of degree, whereas the transition to superorganismality is a difference of kind: as such, only the latter is a true major evolutionary transition in individuality.

In this paper, we have employed terminology that reflects this latter model, referring to 'cooperatively breeding' and 'superorganismal' species to emphasise a qualitative distinction between the two systems. Nonetheless, it should be noted that the continuous model of insect social evolution remains significantly more prominent in the social insect literature than the discontinuous model that we have advocated here. Descriptors that imply a continuous scale of social complexity, such as simple (or primitive) and complex (or advanced) eusociality, are, therefore, widely employed in the literature at present. Regardless of which model best describes the true nature of this major evolutionary transition, species must necessarily pass through a cooperatively breeding stage with plastic castes before they can reach the point of obligate, irreversible caste differentiation, so we consider all three stages here.

Photo credits, clockwise from top left: A: *Ammophila nigricans* © Judy Gallagher; B: *Andrena sp.* © Judy Gallagher; C: *Lasioglossum sp.* © Katja Schulz; D: *Polistes dominula* © Dimitri Dresde; E: *Apis mellifera* © Phin Hall; F: *Vespula vulgaris* © S. Rae; G: *Atta sp.* © Gordon Milligan; H: *Nasutitermus luzonicus* © Diaz Geo.

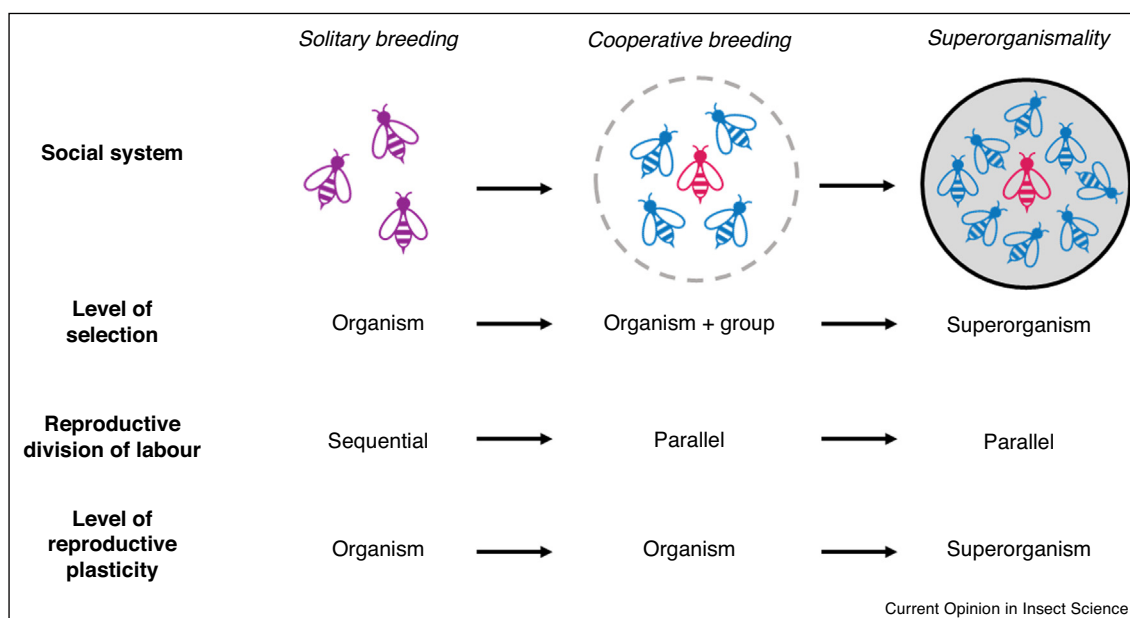
characterised by individuals that are able to reproduce independently and represents the likely ancestral state for all social lineages. Cooperative breeding involves the formation of social groups with division of reproductive labour but in which most individuals retain a significant degree of reproductive flexibility. Finally, superorganismal species exhibit an irreversible division of reproductive labour, cementing the colony as the unit of selection. Here, we outline the broad trends in

reproductive differentiation and reproductive plasticity across the major transition from cooperative breeding to superorganismality (Figure 1).

### Shifts in reproductive plasticity across solitary and cooperative breeders

Solitary breeding is the likely basal state (or 'ground-plan') for all social insect lineages. Solitary insects perform reproductive tasks sequentially, switching

Figure 1



The evolutionary transition from solitary to cooperative breeding involves a shift from sequential execution of reproductive and non-reproductive tasks by individuals to parallel execution of tasks by groups of cooperating individuals, but with little or no loss of reproductive plasticity: at this stage in the major transition, most or all group members are able to switch between tasks if the opportunity arises. The transition from cooperative breeding to superorganismality is defined by a dramatic and putatively irreversible loss of reproductive plasticity at the level of the lower-level organism, but the simultaneous generation of a higher-level superorganism with its own colony-level plasticity. Arrows indicate the likely ancestral trajectory of modern superorganismal species; note we do not wish to imply an inevitable evolutionary trajectory for extant solitary or cooperatively breeding species.

frequently between them in response to environmental cues such as seasonal fluctuations in the availability of resources [12,13] or expected mortality risk [14,15], as well as in response to their physiological state. The sequential execution of reproductive and non-reproductive tasks by the same individual, and the capacity to modify their expression in response to external and internal conditions, imply high reproductive plasticity at the level of the individual. This, however, comes at the cost of trade-offs when the optimisation of the phenotype to the opposing selective demands of each task is limited by the time and energetic costs of continuously remodelling traits [16,17]. These costs will be particularly large in unpredictable environments and for traits that take a long time to modify [18]. The need to facultatively

switch between reproductive modes should be reflected in the mechanisms of reproductive differentiation in solitary species (Box 2).

Intriguingly, among the solitary insects with the most pronounced sequential division of reproductive labour are progressively provisioning Eumenid (e.g. *Synagris cornuta* and *Zethus miniatus*) or Sphecid wasps (e.g. *Ammophila pubescens*), which are likely to represent the ancestral state of the social wasps. After laying eggs, females in these species go through extended periods of brood care and provisioning while having diminished ovaries, in effect cycling between distinct periods of reproductive and non-reproductive labour [19–21,22\*]. It has been proposed that the uncoupling of reproductive and non-reproductive

## Box 2 Proximate mechanisms of plasticity across the spectrum of sociality

Although every social lineage has a unique evolutionary history, there are likely to be broad commonalities in the mechanisms that underpin reproductive differentiation at each level of insect social evolution. As reproductive roles become less flexible across the transition from solitary breeding to superorganismality, we predict that more stable ('committed') mechanisms of differentiation (such as epigenetic modifications) will become increasingly important relative to more changeable mechanisms (such as hormonal regulation). The development of more sophisticated bioinformatic and molecular assays is now making it possible to test this prediction [8\*]. Several possible mechanisms of reproductive differentiation and molecular commitment, and the evidence for their roles across the transition to superorganismality, are reviewed below.

### Hormonal regulation

As hormones can be produced quickly and are relatively short-lived, hormonal regulation of reproductive differentiation might be expected to be most prominent in solitary species that must switch frequently between reproductive modes. In practice, however, the weight of evidence suggests that endocrine regulation remains highly important at all levels of sociality. Juvenile hormone (JH) is the primary regulator of reproduction in basal solitary insects [48,49\*], including in progressively provisioning wasps that may represent the basal state for wasp sociality [50,51]. This role appears to be maintained in both cooperatively breeding and superorganismal species. JH is associated with reproductive differentiation in cooperatively breeding bees [52], termites [53] and wasps [54,55]; and likewise in ants [56,57] and honeybees [58,59]. It thus appears that JH is a mediator of reproductive differentiation across insect lineages of all degrees of sociality. However, we predict that in less reproductively plastic lineages additional mechanisms will be necessary to stabilise the expression of reproductive and non-reproductive phenotypes.

### DNA methylation

Methylation offers a stable means of altering gene expression levels. Drawing comparisons with mammalian development (in which levels of epigenetic modification increase with increasing cell differentiation) Patalano *et al.* [60] have proposed that reduced reproductive plasticity in the transition to insect sociality ought to be associated with increasing evidence of methylation, while Maleszka *et al.* [61] have emphasised the potential role of methylomic degeneracy in producing functionally divergent castes from a single genome. In the last decade, the role of methylation in social insect differentiation has been the topic of extensive research [62–64], but it has proven difficult to establish a consistent pattern of association between methylation and social complexity [65\*,66]. In cooperatively breeding species, methylation appears to be associated with caste in some species of wasps [65\*] and bees [67]. Two separate studies, however, have found no significant reproductive role for methylation in the cooperatively breeding paper wasps *P. canadensis* [68] and *Polistes dominula* [69], and indeed both these species appear to have lost a key methyltransferase gene. Whether this loss of epigenetic machinery is a lineage-specific trait or rather a more general signature of social complexity remains unknown. In superorganismal ants, several papers have linked caste to levels of methylation [70–72], but the methodology of these and other methylation studies has been called into question [73].

### Histone acetylation

Histone acetylation alters the accessibility of chromatin, and thus joins DNA methylation as a possible stable source of reproductive differentiation. To our knowledge there is no evidence of a role for histone acetylation in reproductive differentiation in solitary or cooperatively breeding insect species. The discovery of histone deacetylase inhibitor (HDACi) as a component in the royal jelly of honey bees suggests a role for this mechanism in superorganismal reproductive caste determination [74], as does the recent finding that caste-specific chromatin modification patterns are present early in honey bee development [75\*]. Royal jelly also contains noncoding microRNAs (miRNAs), another possible reproductive regulator [76]. Histone acetylation additionally appears to play a role in the determination of ant worker subcastes [77,78], strengthening the evidence that this mechanism is suited to the generation of stable phenotypes.

### Genetic differentiation

In general, allelic variants that bias individuals towards either a reproductive or a non-reproductive role should become fixed or lost, since reproductives have a clear advantage over non-reproductives in passing on their genes directly. Several exceptions to this rule have, however, become known in both ants [79] and superorganismal termites [80]. Such examples probably represent a form of evolutionary 'cheating' by queen-biasing variants and are unlikely to be widespread mechanisms of caste differentiation.



phenotypes in solitary insects such as progressively provisioning wasps has acted as a ground plan for the division of labour that occurs in cooperative insect societies [21,22\*,23,24].

A prerequisite for this major transition is the transition from solitary to cooperative breeding, which involves a shift from sequential to parallel task execution in the form of a division of reproductive labour: cooperative groups are able to divide reproductive and non-reproductive tasks between multiple individuals. Thus, cooperatively breeding insect groups typically contain one (or a few) dominant individual(s) that performs the vast majority of reproductive activity on the nest, and a (usually larger) number of non-reproductive individuals that perform most foraging, nest building and offspring care. This parallelisation of roles likely represents an efficient solution to the trade-offs between reproductive and non-reproductive investment [25,26], although it also entails a source of potentially costly conflict in fitness interests between reproductive and non-reproductive specialists.

Reproductive and non-reproductive phenotypes within typical cooperatively breeding insect societies are significantly more stable than those expressed within the lifetime of a solitary individual; yet, substantial task plasticity persists in these societies. Non-reproductives are capable of upregulating reproductive traits in response to the loss of a dominant reproductive across cooperatively breeding bees [27,28], wasps [29,30] and termites [31]. Nonetheless, the rate at which individuals are able to transition between roles appears to be limited. While behavioural changes may be apparent within days [27,28], time to physiological upregulation of reproductive tissues appears to be on the order of weeks or months [30,31]. Additionally, there is evidence that non-reproductive individuals may experience a loss of reproductive plasticity as they age, for example in *Polistes canadensis* [32] and *Metapolybia cingulata* [33] paper wasps. The transition from solitary to cooperative breeding thus appears to involve some loss in reproductive plasticity, but of a minor degree: the majority of individuals in a cooperatively breeding society will remain reproductively totipotent for the majority of their adult lives. This should be reflected in the proximate mechanisms underlying plasticity at this level of sociality (Box 2).

### Transfer of plasticity from the individual level to the colony level in the superorganismal society

The major transition from cooperative breeding to superorganismality is itself defined by the evolution of obligate and irreversible differentiation of reproductive phenotypes and non-reproductive phenotypes, cementing the reproductive division of labour [7\*\*,8\*\*]. Superorganismal castes are determined during development, after which non-reproductive individuals are unable to transition to a

fully reproductive role. This reduction in reproductive plasticity commits non-reproductives to indirect fitness strategies and shifts the target of selection from the individual to the colony. Selection will then favour traits that maximise indirect fitness gains, opening the door for strong task specialisation. Accordingly, superorganismal castes are typically morphologically differentiated, exhibit a reversal of the usual negative trade-off between reproductive activity and longevity [34], and may even include specialised phenotypic subdivisions of the non-reproductive caste. Relative to reproductively plastic organisms, the mechanisms that underpin reproductive differentiation in superorganismal species are expected to include a significantly larger role for high-stability molecular mechanisms (Box 2).

Adult individuals in superorganismal colonies are much less reproductively plastic than adult individuals in cooperative species, which themselves are somewhat less reproductively plastic than solitary individuals. It is, therefore, tempting to infer a relatively straightforward pattern of decreasing reproductive plasticity across the spectrum of insect sociality from solitary to cooperatively breeding to superorganismal insect species. Such a pattern only holds, however, when one focuses exclusively on a single level of individuality, namely that of the individual adult organism. The nature of a major evolutionary transition is that it results in the generation of a higher level of individuality: reproductive plasticity must, therefore, be considered at both the level of the individual organisms that constitute the superorganismal society and the higher level of the superorganism itself.

There are significant analogies between the organisation of complex multicellular organisms and the organisation of superorganisms: the queen is the reproductive tissue of a superorganism, while workers are its somatic tissue [35,36]. Just as a solitary multicellular organism can be said to possess reproductive plasticity if it is able to facultatively vary its investment into reproductive and non-reproductive traits, the superorganism is reproductively plastic if it has the capacity to facultatively vary investment into the production of workers and sexual offspring. The ability of a superorganism to engage in this kind of plastic response is a function of its sociogenome, a composite of the genetic traits of all individuals that together constitute the superorganism and the interactions of those traits to produce colony-level feedback loops [37,38].

Sociogenome-mediated interactions between constituent individuals allow superorganisms to plastically mediate reproductive investment in much the same way that multicellular organisms do. Though the individuals within a superorganism are not perfectly analogous to the cells of a multicellular individual – the latter being fully clonal and, therefore, almost entirely insulated from

internal reproductive conflict, while the former retain the potential for conflict since workers are usually not clonal and are often capable of producing males despite having limited fertility – the high degree of task parallelisation and specialisation has facilitated similar adaptations in each system. Like multicellular organisms, superorganismal colonies may upregulate investment into reproduction (i.e. the production of sexuals) in response to seasonality and may store energy within non-reproductives to later transfer to sexual brood [39,40]. Both multicellular organisms and superorganisms typically transition from somatic to reproductive investment having reached some growth threshold [41]. Ant colonies appear to possess a form of terminal investment, switching to the production of sexuals in response to queen mortality [25,42]. Honeybee colonies respond to the loss of a queen by rearing replacement queens [43]. Thus, while superorganismal colonies lack the individual-level reproductive plasticity necessary for non-reproductives to directly replace reproductives, at the superorganism level these colonies are able to plastically respond to the loss of a queen.

Viewed at a single level of selection, the transition to superorganismality might be generalised as a simple reduction in reproductive plasticity, producing individuals that cannot adapt their reproductive roles in response to changing environments. However, such an approach would overlook the opposing patterns of plasticity at the different levels of selection. Specifically, we highlight here that the transition should be viewed as a transferral of plasticity from the lower-level organism to the higher-level superorganism. Just as it would be meaningless to argue that an adult mammal lacks phenotypic plasticity because it largely comprises unipotent cells, so too must we recognise that the transition to superorganismality can only be understood by considering the superorganism as a unified whole that is the greater than the sum of its parts.

## Conclusions

The major evolutionary transition to insect superorganismality is one of our best models for understanding the way in which lower-level units are able to come together to form new (higher) levels of biological complexity. Here, we highlight that the current view of the process of the major transition to superorganismality overlooks the effects of multi-level selection. We have briefly summarised how the shifts in reproductive division of labour have made this radical shift in complexity possible. The patterns of differentiation involved in this transition are likely to be of general relevance to other shifts in individuality, such as the transition to multicellularity. In order to fully appreciate these commonalities, however, social insect researchers must embrace the status of the superorganismal colony as an individual in its own right.

## Conflict of interest statement

Nothing declared.

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## References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

- of special interest
- of outstanding interest

1. Szathmáry E, Smith JM: **The major evolutionary transitions.** *Nature* 1995, **374**:227-232.
2. West SA, Fisher RM, Gardner A, Kiers ET: **Major evolutionary transitions in individuality.** *Proc Natl Acad Sci U S A* 2015, **112**:10112-10119.
3. Hanschen ER, Shelton DE, Michod RE: **Evolutionary transitions in individuality and recent models of multicellularity.** *Evolutionary Transitions to Multicellular Life*. Dordrecht: Springer; 2015, 165-188.
4. Sebé-Pedrós A, Degnan BM, Ruiz-Trillo I: **The origin of Metazoa: a unicellular perspective.** *Nat Rev Genet* 2017, **18**:498-512.
5. Vellai T, Vida G: **The origin of eukaryotes: the difference between prokaryotic and eukaryotic cells.** *Proc R Soc B Biol Sci* 1999, **266**:1571-1577.
6. Szathmáry E: **Toward major evolutionary transitions theory 2.0.** *Proc Natl Acad Sci U S A* 2015, **112**:10104-10111.
7. Boomsma JJ, Gawne R: **Superorganismality and caste differentiation as points of no return: how the major evolutionary transitions were lost in translation.** *Biol Rev* 2018, **93**:28-54 <http://dx.doi.org/10.1111/brv.12330>.  
This paper lays out a comprehensive history of the terms 'eusocial' and 'superorganism', forcefully arguing that only the latter constitutes a biologically meaningful category. The authors posit that social biology cannot become a hard science unless it possesses hard categories.
8. Kennedy P, Baron G, Qiu B, Freitak D, Helanterä H, Hunt ER, Manfredini F, O'Shea-Wheller T, Patalano S, Pull CD *et al.*: **Deconstructing superorganisms and societies to address big questions in biology.** *Trends Ecol Evol* 2017, **32**:861-872.  
A synthesis of the opinions of 120 biologists to identify key areas in which social insect research is contributing to our broader understanding of biology. Particular emphasis is placed upon methodological advances which are making it possible to better understand the evolution and functioning of superorganismal societies
9. Hughes WOH, Oldroyd BP, Beekman M, Ratnieks FLW: **Ancestral monogamy shows kin selection is key to the evolution of eusociality.** *Science* 2008, **320**:1213-1216.
10. Sherman PW, Lacey EA, Reeve HK, Keller L: **Forum: the eusociality continuum.** *Behav Ecol* 1995, **6**:102-108.
11. Costa JT, Fitzgerald TD: **Social terminology revisited: where are we ten years later?** *Ann Zool Fennici* 2005, **42**:87-98.
12. Košťál V: **Eco-physiological phases of insect diapause.** *J Insect Physiol* 2006, **52**:113-127.
13. Hahn DA, Denlinger DL: **Energetics of insect diapause.** *Annu Rev Entomol* 2011, **56**:103-121.
14. Javoš J, Tammaru T: **Reproductive decisions are sensitive to cues of life expectancy: the case of a moth.** *Anim Behav* 2004, **68**:249-255.
15. Cotter SC, Ward RJS, Kilner RM: **Age-specific reproductive investment in female burying beetles: independent effects of state and risk of death.** *Funct Ecol* 2011, **25**:652-660.
16. Flatt T, Heyland A (Eds): *Mechanisms of life history evolution: the genetics and physiology of life history traits and trade-offs*. OUP Oxford; 2011 <http://dx.doi.org/10.1093/acprof:oso/9780199568765.001.0001>.

17. Murren CJ, Auld JR, Callahan H, Ghalambor CK, Handelsman CA, Heskell MA, Kingsolver JG, Maclean HJ, Masel J, Maughan H *et al.*: **Constraints on the evolution of phenotypic plasticity: limits and costs of phenotype and plasticity.** *Heredity (Edinb)* 2015, **115**:293-301.
  18. Gabriel W: **Selective advantage of irreversible and reversible phenotypic plasticity.** *Arch Hydrobiol* 2006, **167**:1-20.
  19. Baerends GP: **On the life-history of *Ammophila campestris* Jur.** *Proc K Ned Akad Wet* 1941, **44**:483-488.
  20. Field J: **Patterns of nest provisioning and parental investment in the solitary digger wasp *Ammophila sabulosa*.** *Ecol Entomol* 1992, **17**:43-51.
  21. West-Eberhard MJ: **Wasp societies as microcosms for the study of development and evolution.** *Natural History and Evolution of Paper-Wasps*. 1996:290-317.
  22. Kelstrup HC, Hartfelder K, Lopes TF, Wossler TC: **The behavior and reproductive physiology of a solitary progressive provisioning vespid wasp: evidence for a solitary-cycle origin of reproductive castes.** *Am Nat* 2018, **191**:E27-E39.
- One of the strongest attempts yet to test the ovarian ground plan hypothesis. The authors present support for the hypothesis in the form of evidence for a strong linkage between ovarian development and queen-like and worker-like behaviours in a progressively provisioning wasp. They find no support for the mechanistic predictions of the hypothesis, however.
23. West-Eberhard MJ: **Wasp societies as microcosms for the study of development and evolution.** *Natural History and Evolution of Paper-Wasps*. 1996:290-317.
  24. Amdam GV, Csondes A, Fondrk MK, Page RE: **Complex social behaviour derived from maternal reproductive traits.** *Nature* 2006, **439**:76-78.
  25. Hölldobler B, Wilson EO: *The Ants*. 1990.
  26. Peeters C, Molet M: **Colonial reproduction and life histories.** *Ant Ecology*. 2010.
  27. Michener CD, Brothers DJ: **Were workers of eusocial hymenoptera initially altruistic or oppressed?** *Proc Natl Acad Sci U S A* 1974, **71**:671-674.
  28. Breed MD, Gamboa GJ: **Behavioral control of workers by queens in primitively eusocial bees.** *Science* 1977, **195**:694-696.
  29. Gadagkar R: **Social structure and the determinants of queen status in the primitively eusocial wasp *Ropalidia cyathiformis*.** *Chemistry and Biology of Social Insects, Proceedings of X International Congress IUSSI* 1987:377-378.
  30. Strassmann JE, Fortunato A, Cervo R, Turillazzi S, Damon JM, Queller DC, Strassmann JE, Queller DC: **The cost of queen loss in the social wasp *Polistes dominulus* (Hymenoptera: Vespidae).** *J Kansas Entomol Soc* 2004, **77**:343-355.
  31. Roisin Y: **Queen replacement in the termite *Microcerotermes papuanus*.** *Entomol Exp Appl* 1990, **56**:83-90.
  32. Sumner S, Kelstrup H, Fanelli D: **Reproductive constraints, direct fitness and indirect fitness benefits explain helping behaviour in the primitively eusocial wasp, *Polistes canadensis*.** *Proc R Soc London B Biol Sci* 2010, **277**:1721-1728.
  33. West-Eberhard MJ: **Temporary queens in metapolybia wasps: nonreproductive helpers without altruism?** *Science* 1978, **200**:441-443.
  34. Blacher P, Huggins TJ, Bourke AFG: **Evolution of ageing, costs of reproduction and the fecundity-longevity trade-off in eusocial insects.** *Proc R Soc B Biol Sci* 2017, **284**:20170380.
  35. Hölldobler B, Wilson EO: *The Superorganism: The Beauty, Elegance, and Strangeness of Insect Societies*. 2008.
  36. Gadau J: **Genetic architecture of key social trait differs significantly between primitive and advanced eusocial species.** *Proc Natl Acad Sci U S A* 2015, **112**:13755-13756.
  37. Linksvayer TA, Fondrk MK, Page RE Jr: **Honeybee social regulatory networks are shaped by colony-level selection.** *Am Nat* 2009, **173**:E99-E107.
  38. Johnson BR, Linksvayer TA: **Deconstructing the superorganism: social physiology, groundplans, and sociogenomics.** *Q Rev Biol* 2010, **85**:57-79.
  39. Tschinkel W: **Sociometry and sociogenesis of the harvester ant *Pogonomyrmex badius*.** *Insectes Soc* 1998, **45**:385-410.
  40. Hart LM, Tschinkel WR: **A seasonal natural history of the ant, *Odontomachus brunneus*.** *Insectes Soc* 2012, **59**:45-54.
  41. Smith ML, Koenig PA, Peters JM: **The cues of colony size: how honey bees sense that their colony is large enough to begin to invest in reproduction.** *J Exp Biol* 2017, **220**:1597-1605.
- Simple experimental manipulation of honeybee colonies yields powerful insights into the means by which a superorganism can self-regulate its development. The authors find that it is contact rates between workers, rather than pheromone levels or nest temperature, that colonies use to optimise the beginning of sexual investment.
42. Heinze J, Schrempf A: **Terminal investment: individual reproduction of ant queens increases with age.** *PLoS One* 2012, **7**:e35201.
  43. Hatch S, Tarpy DR, Fletcher DJC: **Worker regulation of emergency queen rearing in honey bee colonies and the resultant variation in queen quality.** *Insectes Soc* 1999, **46**:372-377.
  44. Shell WA, Rehan SM: **Behavioral and genetic mechanisms of social evolution: insights from incipiently and facultatively social bees.** *Apidologie* 2018, **49**:13-30.
- Reviewing data from highly behaviourally plastic sweat bees and carpenter bees, this paper argues convincingly in favour of the importance of simple societies for advancing our understanding of the genetic basis of cooperative behaviour.
45. Rehan SM, Toth AL: **Climbing the social ladder: the molecular evolution of sociality.** *Trends Ecol Evol* 2015, **30**:426-433.
  46. Boomsma JJ: **Lifetime monogamy and the evolution of eusociality.** *Philos Trans R Soc B Biol Sci* 2009, **364**:3191-3207.
  47. Boomsma JJ: **Beyond promiscuity: mate-choice commitments in social breeding.** *Philos Trans R Soc Lond B Biol Sci* 2013, **368**:20120050.
  48. Nijhout H: **Insect hormones.** *Nature* 1995, **375**:200.
  49. Roy S, Saha TT, Zou Z, Raikhel AS: **Regulatory pathways controlling female insect reproduction.** *Annu Rev Entomol* 2018, **63**:489-511 <http://dx.doi.org/10.1146/annurev-ento-020117-043258>.
- A comprehensive review of the pathways by which female reproduction is regulated in insects. Of particular interest is the variation among species, which suggests that different lineages have converged upon parallel solutions to the molecular control of vitellogenesis and oogenesis.
50. Giray T, Giovanetti M, West-Eberhard MJ: **Juvenile hormone, reproduction, and worker behavior in the neotropical social wasp *Polistes canadensis*.** *Proc Natl Acad Sci U S A* 2005, **102**:3330-3335.
  51. Tibbetts EA, Mettler A, Donajkowski K: **Nutrition-dependent fertility response to juvenile hormone in non-social *Euodynerus foraminatus* wasps and the evolutionary origin of sociality.** *J Insect Physiol* 2013, **59**:339-344.
  52. Hartfelder K, Makert GR, Judice CC, Pereira GAG, Santana WC, Dallacqua R, Bitondi MMG: **Physiological and genetic mechanisms underlying caste development, reproduction and division of labor in stingless bees.** *Apidologie* 2006, **37**:144-163.
  53. Korb J: **Juvenile hormone: a central regulator of termite caste polyphenism.** *Adv Insect Phys* 2015, **48**:131-161.
  54. Jandt JM, Tibbetts EA, Toth AL: ***Polistes* paper wasps: a model genus for the study of social dominance hierarchies.** *Insectes Soc* 2014, **61**:11-27.
  55. Kelstrup HC, Hartfelder K, Wossler TC: ***Polistes smithii* vs. *Polistes dominula*: the contrasting endocrinology and epicuticular signaling of sympatric paper wasps in the field.** *Behav Ecol Sociobiol* 2015, **69**:2043-2058.
  56. Penick CA, Prager SS, Liebig J: **Juvenile hormone induces queen development in late-stage larvae of the ant *Harpegnathos saltator*.** *J Insect Physiol* 2012, **58**:1643-1649.

57. Libbrecht R, Corona M, Wende F, Azevedo DO, Serrao JE, Keller L: **Interplay between insulin signaling, juvenile hormone, and vitellogenin regulates maternal effects on polyphenism in ants.** *Proc Natl Acad Sci U S A* 2013, **110**:11050-11055.
  58. Schmidt Capella IC, Hartfelder K: **Juvenile-hormone-dependent interaction of actin and spectrin is crucial for polymorphic differentiation of the larval honey bee ovary.** *Cell Tissue Res* 2002, **307**:265-272.
  59. Bomtorin AD, Mackert A, Rosa GCC, Moda LM, Martins JR, Bitondi MMG, Hartfelder K, Simões ZLP: **Juvenile hormone biosynthesis gene expression in the corpora allata of honey bee (*Apis mellifera* L.) female castes.** *PLoS One* 2014, **9**:e86923.
  60. Patalano S, Hore TA, Reik W, Sumner S: **Shifting behaviour: Epigenetic reprogramming in eusocial insects.** *Curr Opin Cell Biol* 2012, **24**:367-373.
  61. Maleszka R, Mason PH, Barron AB: **Epigenomics and the concept of degeneracy in biological systems.** *Brief Funct Genomics* 2014, **13**:191-202.
  62. Kronforst MR, Gilley DC, Strassmann JE, Queller DC: **DNA methylation is widespread across social Hymenoptera.** *Curr Biol* 2008, **18**:R287-R288.
  63. Weiner SA, Toth AL: **Epigenetics in social insects: a new direction for understanding the evolution of castes.** *Genet Res Int* 2012 2012:1-11.
  64. Yan H, Simola DF, Bonasio R, Liebig J, Berger SL, Reinberg D: **Eusocial insects as emerging models for behavioural epigenetics.** *Nat Rev Genet* 2014, **15**:677-688.
  65. Weiner SA, Galbraith DA, Adams DC, Valenzuela N, Noll FB, Grozinger CM, Toth AL: **A survey of DNA methylation across social insect species, life stages, and castes reveals abundant and caste-associated methylation in a primitively social wasp.** *Naturwissenschaften* 2013, **100**:795-799.
- An impressively detailed attempt to identify patterns of DNA methylation across different insect clades, with a particular focus on social species. The authors are unable to find any support for an association between sociality and methylation after phylogenetic correction. Also, of note is the authors' observation that methylation is underlain by diverse, nonneutral genetic machinery, indicating that multiple different methylation pathways are in effect across insects.
66. Bewick AJ, Vogel KJ, Moore AJ, Schmitz RJ: **Evolution of DNA methylation across insects.** *Mol Biol Evol* 2017, **34**:654-665.
  67. Li B, Hou L, Zhu D, Xu X, An S, Wang X: **Identification and caste-dependent expression patterns of DNA methylation associated genes in *Bombus terrestris*.** *Sci Rep* 2018, **8**:2332.
  68. Patalano S, Vlasova A, Wyatt C, Ewels P, Camara F, Ferreira PG, Asher CL, Jurkowski TP, Segonds-Pichon A, Bachman M *et al.*: **Molecular signatures of plastic phenotypes in two eusocial insect species with simple societies.** *Proc Natl Acad Sci U S A* 2015, **112**:13970-13975.
  69. Standage DS, Berens AJ, Glastad KM, Severin AJ, Brendel VP, Toth AL: **Genome, transcriptome and methylome sequencing of a primitively eusocial wasp reveal a greatly reduced DNA methylation system in a social insect.** *Mol Ecol* 2016, **25**:1769-1784.
  70. Kucharski R, Maleszka J, Foret S, Maleszka R: **Nutritional control of reproductive status in honeybees via DNA methylation.** *Science* 2008, **319**:1827-1830.
  71. Bonasio R, Li Q, Lian J, Mutti NS, Jin L, Zhao H, Zhang P, Wen P, Xiang H, Ding Y *et al.*: **Genome-wide and caste-specific DNA methylomes of the ants *Camponotus floridanus* and *Harpegnathos saltator*.** *Curr Biol* 2012, **22**:1755-1764.
  72. Foret S, Kucharski R, Pellegrini M, Feng S, Jacobsen SE, Robinson GE, Maleszka R: **DNA methylation dynamics, metabolic fluxes, gene splicing, and alternative phenotypes in honey bees.** *Proc Natl Acad Sci U S A* 2012, **109**:4968-4973 <http://dx.doi.org/10.1073/pnas.1202392109>.
  73. Libbrecht R, Oxley PR, Keller L, Kronauer DJC: **Robust DNA methylation in the clonal raider ant brain.** *Curr Biol* 2016, **26**:391-395.
  74. Spannhoff A, Kim YK, Raynal NJM, Gharibyan V, Su MB, Zhou YY, Li J, Castellano S, Sbardella G, Issa JPJ *et al.*: **Histone deacetylase inhibitor activity in royal jelly might facilitate caste switching in bees.** *EMBO Rep* 2011, **12**:238-243.
  75. Wojciechowski M, Lowe R, Maleszka J, Conn D, Maleszka R, Hurd PJ: **Phenotypically distinct female castes in honey bees are defined by alternative chromatin states during larval development.** *Genome Res* 2018, **28**:1532-1542.
- By mapping chromatin structure in developing honey bees, this study demonstrates the role of chromatin modifications in establishing caste-specific gene expression profiles. The authors pay particular attention to the developmental timing of differentiation, thereby adding to our knowledge of the precise developmental stages at which superorganismal castes can become irreversibly fixed.
76. Guo X, Su S, Skogerboe G, Dai S, Li W, Li Z, Liu F, Ni R, Guo Y, Chen S *et al.*: **Recipe for a busy bee: microRNAs in honey bee caste determination.** *PLoS One* 2013, **8**:e81661.
  77. Simola DF, Ye C, Mutti NS, Doleza K, Bonasio R, Liebig J, Reinberg D, Berger SL: **A chromatin link to caste identity in the carpenter ant *Camponotus floridanus*.** *Genome Res* 2013, **23**:486-496.
  78. Glastad KM, Hunt BG, Goodisman MAD: **DNA methylation and chromatin organization in insects: insights from the ant *Camponotus floridanus*.** *Genome Biol Evol* 2015, **7**:931-942.
  79. Anderson KE, Linksvayer TA, Smith CR: **The causes and consequences of genetic caste determination in ants (Hymenoptera: Formicidae).** *Ecology* 2008, **11**:119-132.
  80. Lo N, Hayashi Y, Kitade O: **Should environmental caste determination be assumed for termites?** *Am Nat* 2009, **173**:848-853.