Life history and development - A framework for understanding developmental plasticity...

Article *in* Biological Reviews · September 2008

DOI: 10.1111/j.1469-185X.2008.00044.x · Source: PubMed

Klaus Hartfelder
University of São Paulo - Ribeirão Preto School of Medicine

doi:10.1111/j.1469-185X.2008.00044.x

Life history and development - a framework for understanding developmental plasticity in lower termites

Judith Korb[1*†] and Klaus Hartfelder[2]

[1] *Biologie I, Universität Regensburg, D-93040 Regensburg, Germany*
[2] *Departamento de Biologia Celular e Molecular e Bioagentes Patogênicos, Faculdade de Medicina de Ribeirão Preto, Universidade de São Paulo, Ribeirão Preto, Brazil* (*E-mail: klaus@fmrp.usp.br*)

(*Received* 17 *September* 2007; *revised* 16 *April* 2008; *accepted* 08 *May* 2008)

ABSTRACT

Termites (Isoptera) are the phylogenetically oldest social insects, but in scientific research they have always stood in the shadow of the social Hymenoptera. Both groups of social insects evolved complex societies independently and hence, their different ancestry provided them with different life-history preadaptations for social evolution. Termites, the 'social cockroaches', have a hemimetabolous mode of development and both sexes are diploid, while the social Hymenoptera belong to the holometabolous insects and have a haplodiploid mode of sex determination. Despite this apparent disparity it is interesting to ask whether termites and social Hymenoptera share common principles in their individual and social ontogenies and how these are related to the evolution of their respective social life histories. Such a comparison has, however, been much hampered by the developmental complexity of the termite caste system, as well as by an idiosyncratic terminology, which makes it difficult for non-termitologists to access the literature.

Here, we provide a conceptual guide to termite terminology based on the highly flexible caste system of the "lower termites". We summarise what is known about ultimate causes and underlying proximate mechanisms in the evolution and maintenance of termite sociality, and we try to embed the results and their discussion into general evolutionary theory and developmental biology. Finally, we speculate about fundamental factors that might have facilitated the unique evolution of complex societies in a diploid hemimetabolous insect taxon. This review also aims at a better integration of termites into general discussions on evolutionary and developmental biology, and it shows that the ecology of termites and their astounding phenotypic plasticity have a large yet still little explored potential to provide insights into elementary evo-devo questions.

Key words: social insect, caste, polyphenism, developmental plasticity, pseudergate, neotenic, wing development, moult, metamorphosis, juvenile hormone.

CONTENTS

* Address for correspondence: (Tel: 0049 541 9693496; Fax: 0049 541 9692830; E-mail: judith.korb@biologie.uni-osnabrueck.de).
† Present address: Verhaltensbiologie, Universität Osnabrück, D-49080 Osnabrück, Germany (E-mail: judith.korb@biologie.uni-osnabrueck.de).

I. INTRODUCTION

Caste differentiation in termites is one of the most conspicuous examples of facultative polyphenism in animals, in which individuals show various phenotypes despite the same genetic background. Within a termite colony - with few exceptions where a genetic component might be involved (Hayashi *et al.*, 2007) - the offspring of a single king and queen can develop into workers, soldiers, and two sexual morphs depending on environmental and social stimuli. Each caste generally exhibits a particular behavioural repertoire and often caste-specific morphological characters. As in social Hymenoptera (ants, and some bees and wasps), this caste system of termites is considered to be the basis for the evolutionary and ecological success of these eusocial insects (Oster & Wilson, 1978).

Obviously, such elaborate societies evolved from solitary ancestors. Consequently, a focus of current research is to explain how complex phenotypes can evolve from ancestral solitary forms. Often differences in complex traits among species are not the result of the presence or absence of particular genes, but arise from changes in the mechanisms of gene regulation affecting when and where a gene or an entire regulatory module is expressed. As noted by (Brakefield, 2006; p. 362): 'There is a limited genetical tool kit and much of the morphological diversity evolution is about old genes performing new tricks. Although existing genetic pathways can be co-opted and subsequently elaborated upon to do something different, and specific genes can take on additional tasks at new times during development and in different tissues *via* gene duplication and divergence, *de novo* evolution of new pathways appears to be rare'.

Besides the reproductive polyphenisms in social insects and that of male weaponry in dung beetles, other major types of polyphenisms in insects are sequential (seasonal) polyphenisms in wing length (in its extreme form, winged and wingless morphs) or coloration; these are also often connected to differences in reproductive strategies (for review see Hartfelder & Emlen, 2005). In both termites and ants, the caste syndrome is coupled to a wing dimorphism. This clearly represents convergent evolution since the wingless workers of ants are adults of a holometabolan clade, whereas the wingless workers in termites are either immatures (in the wood-nesting termite families Termopsidae and Kalotermitidae, and *Prorhinotermes*) or have conserved an immature phenotype [the worker caste of the Mastotermitidae, Rhinotermitidae (except *Prorhinotermes*), Hodotermitidae, Serritermitidae, and Termitidae]. Additional wingless morphs in termites are the neotenic reproductives and soldiers, for which there is no real equivalent in ants. Termites evolved complex societies more than 130 million years ago, probably during the upper Jurassic, (Thorne, Grimaldi & Krishna, 2000) from a cockroach-like ancestor similar to the wood-roaches (Nalepa & Bandi, 2000) which form the sister group to present termites (Eggleton, 2001; Inward, Beccaloni & Eggleton, 2007*a*). Cockroaches as well as termites are characterized by a highly flexible development (Roth, 1981; Nalepa, 1994).

Even though termites are phylogenetically distant from ants, and the two groups have been considered either to be engaged in arms races (Longhurst, Johnson & Wood, 1978; Deligne, Quennedy & Blum, 1981) or in more peaceful co-evolutionary interactions (Dejean, Durand & Bolton, 1996), they share several characters as a result of convergent evolution. Both ants and termites have clearly evolved independently from winged solitary or primitively social ancestors (Wilson, 1971); winglessness is an adaptation to burrowing activities and a nest structure which functions as a fortress against predators. The question, thus, becomes which mechanisms underlie the flexible expression of wing phenotypes in ants and termites, where wings develop in the alate reproductives and where wing development is shut down in wingless reproductives and in the worker and soldier castes.

Pattern formation in wings, including their transformation into very different forewing and hindwing types, is best described in *Drosophila melanogaster*; not only the individual signaling pathway elements, but even entire gene regulatory networks are found to be highly conserved across species, orders and even phyla (Carroll, Grenier & Weatherbee, 2005). The *D. melanogaster* wing formation network has been successfully applied to a study on the loss of wings in workers of several ant species (Abouheif & Wray, 2002) which showed that wing disc development in workers is not brought to a halt at a single unique interruption point in the network, but rather can stop at different points among ant genera. Caste polyphenism and its relation to wing bud development has not yet been studied to this extent in termites, but recent progress on endocrine regulation and associated gene expression differences is now gradually shedding light on regulatory mechanisms underlying caste development, especially the fascinating (and notoriously confusing) plasticity of caste fate in the lower termites.

We will summarise here the state of termite research regarding this developmental question. First, we will explain termite caste systems and their distribution among taxa. This is supplemented by summaries of the classification of

Table 1. Classification of termite species

Based on their ecology and particularly nesting and feeding habits, termite species can be grouped into two life types:

One-piece nesting termites	Multiple-pieces nesting termites (incl. Abe's intermediate type; Abe, 1990)
• live inside single piece of dead wood serving both as nest and food source	• well-defined nest separate from foraging grounds
• with exception of winged sexual's individuals never leave nest	• workers exploit new food resources outside the nest
• colony life limited by food availability	• colony life not limited by food availability
• basal life history	
• highly flexible individual development (see *false workers*)	• true, morphologically differentiated worker caste with reduced reproductive potential
• Termopsidae, Kalotermitidae, *Prorhinotermes*	• Mastotermitidae, Hodotermitidae, Serritermitidae, Rhinotermitidae (except *Prorhinotermes*), Termitidae

An alternative traditional classification of termites is based on their gut symbionts:

Lower termites	Higher termites
• bacteria and flagellates in gut	• bacteria only
• all termites except Termitidae	• Termitidae

Table 2. Developmental terms applied to termites

Lower termites are characterized by a unique flexibility in development which is generated through three molting types:

- *Progressive moult* - a moult characterizing the gradual development from egg *via* several instars into an adult. Associated with progressive moults is an increase in body size and morphological development. This is the default developmental program in all hemimetabolous and holometabolous insects.
- *Stationary moult* - an intermittent moult that is associated with a lack of increase in body size and morphological development. This type of development occurs in several insect species and is frequently associated with periods of food shortage, when a larva or nymph is not capable of passing a critical mass threshold in an instar. In some termites it might also be linked to the wear of mandibles.
- *Regressive moult* - a moult that is characterized by a decrease in body size and/or regression of morphological development, generally a reduction of wing bud size in nymphal instars. This type of development is unique to termites.

In contrast to other hemimetabolous insects where postembryonic development is characterized by a progression of nymphal instars, termites show two distinct types of instars:

- *Larval instar(s)* - instar(s) without externally visible wing buds
- *Nymphal instars* - instars with externally visible wing buds; these instars characterize the gradual, progressive development into winged sexuals.

Larval instars are sometimes further split into:

- *Dependent larvae* - the first up to the second or third larval instar, when larvae supposedly still depend on brood care by 'workers' (but see 'false workers' in Table 3)
- *Independent larvae* - all other larval instars, when larvae care for themselves. They comprise the *false workers* and the *pseudergates sensu lato* (see Table 3).

termite species (Table 1), developmental terms (Table 2) and classification of castes (Table 3) to provide a comprehensive introduction to termite terminology, essential for wider recognition and comprehension of termite research. We will mainly concentrate on phylogenetically basal termite taxa, but occasional notes on higher termite taxa are given where appropriate. Second, we will summarise what is known about ultimate causes influencing cooperation and altruism in termite societies. Third, we will describe current views about proximate mechanisms underlying social complexity in termites starting with environmental and social triggers that affect different developmental trajectories, then outlining the underlying endocrinology of termite caste development, and presenting the current understanding of differential gene expression during termite caste differentiation. Finally, by embedding our knowledge within a general framework of insect development, we will argue that termite studies can add novel facets to our understanding of the evolution of holometabolous development from hemimetabolous ancestors and we speculate about fundamental facilitators that may help to explain the exceptional position of termites as the only diploid group within the highly social insects.

There is a plethora of literature published on termites and on how best to erradicate them. Undeniably, we have come a long way since the pioneering descriptions on termite life cycles by Eugène Nielen Marais (1937), but there remain enormous gaps in our knowledge on this group of insects; we can still share his observation that 'The entomologist who made the acquaintance of the termite for the first time, would be justified in thinking it to be an immigrant from a different planet' (Marais, 1937, chap. 9, par 1).

Table 3. Classification of termite castes

Reproductives

Individuals that reproduce within a colony, generally one female (queen) and one male (king). These can be:

- *Primary reproductives* - reproductives that found a new colony after a nuptial dispersal flight. They develop gradually *via* several nymphal instars into winged sexuals (alates) that shed their wings (dealates) after the nuptial flight. They are characterized by stark sclerotization, the presence of compound eyes and wing marks (remnants of the wings' articulation after they have been shed).
- *Adultoids* - alates that shed their wings and reproduce within the natal nest (they are not neotenics).
- *Neotenic reproductives* - wingless reproductives that develop within the natal colony *via* a single moult from any instar after the third larval instar. At this neotenic moult, their gonads grow and they develop some imaginal characters while maintaining an otherwise larval appearance; some characters, like wing pads, may regress. Neotenic reproductives are characterized by the absence of wings and usually by the lack of compound eyes. The cuticle is less sclerotized than in primary reproductives. They are subdivided into:
  - *Replacement reproductives* if they develop after the death of the same-sex reproductive of a colony.
  - *Supplementary reproductives* if they develop in addition to other same-sex reproductive(s) already present within a colony.

Depending on the termite life type and the instar from which they develop the neotenics can be further classified into:

  - *Neotenics* (*sensu stricto*): They can be either *apterous neotenics* developing from a larval instar or *brachyterous neotenics* developing from a nymphal instar. They are found in lower termites. Neotenics developing from nymphs are sometimes also called *secondary reproductives*, while those developing from workers are called *tertiary reproductives*.
  - *Ergatoids*: neotenics developing from workers in higher termites
  - *Nymphoids*: neotenics developing from nymphs in higher termites

'Workers'

The majority of individuals within a colony belong to the so-called 'worker' caste, although they do not necessarily have to work (see 'false workers'). With a few exceptions among some higher termites, they are not restricted to a specific sex.

A clear separation should be drawn between the 'workers' of the one- and multiple-pieces nesting termites (see Table 1) as they are not equivalent in function and development:

- *False workers* - the majority of the individuals within a colony of one-piece nesting termites. They differ from the (true) workers of multiple-pieces nesting termites as they are totipotent larvae that lack morphological differentiations. Correspondingly, they are less involved in truly altruistic working tasks, such as foraging, brood care, or building behaviours. Therefore, they may rather be regarded as large immatures that delay reproductive maturity ('hopeful reproductives').
- *True workers* - workers in colonies of the multiple-pieces nesting termites. They can be considered altruistic individuals as they perform most tasks within a colony (e.g. foraging, brood care, and building behaviour) except for reproduction and specialized defence. Although they sometimes, especially in lower termites, still have some reproductive options (for instance as neotenic reproductives), their morphological differentiations (especially their sclerotisation) largely restrict their developmental capability (a notable exception is *Mastotermes darwiniensis*). In functional terms, these true workers, often just called workers, are equivalent to the workers of the social Hymenoptera, even though the latter are imagoes, whereas the true workers here are preimaginal stages.

An alternative technical term can be found that distinguishes workers with a flexible development and options for direct reproduction from workers with restricted developmental trajectories:

- *Pseudergates* - 'workers' of many lower termites (including one- and multiple-pieces nesting species) that have broad developmental options, generally including progressive, stationary and regressive moults. Current use of this term often lacks the precision of its original definition (Grasse & Noirot,1947) for individuals that develop regressively from nymphal instars to 'worker' instars without wing buds.

We propose here the following definitions which we use consistently throughout our review:

  - *Pseudergates sensu stricto* - individuals that develop *via* regressive moults from instars with wing buds ('nymphal instars') to instars without wing buds ('larval instars'), as defined by Grasse & Noirot (1947).
  - *Pseudergates sensu lato* - 'workers' of the lower termites that have the potential to undergo progressive, stationary, and regressive moults. They include the 'false workers' of the one-piece nesting termites and those 'true workers' of the multiple-pieces nesting termites that belong to the lower termites and have a flexible development. They comprise larval and nymphal instars.

Soldiers

This caste is unique among social insects in function and development. Soldiers are ancestral in termites and evolved prior to a true worker caste. Unlike the soldiers found in other social insects, this caste is monophyletic in termites.

- *Soldiers* - a clearly altruistic caste that is always sterile and that is morphologically and behaviourally specialized for defence of the colony against predators and competitors.
- *Presoldiers* - a single transitional instar during development from 'worker' to soldier.

II. CASTE PATTERNS IN TERMITES

In termites two reproductive and two non-reproductive castes can be distinguished: primary and neotenic reproductives on the one hand, and soldiers and true workers on the other, with the latter two forming the majority of the individuals of a colony (Noirot, 1990; Roisin, 2000). The occurrence of the castes differs among families (Roisin, 2000) (Fig. 1). While soldiers are present in all families with exception of a few genera in the Termitidae (higher

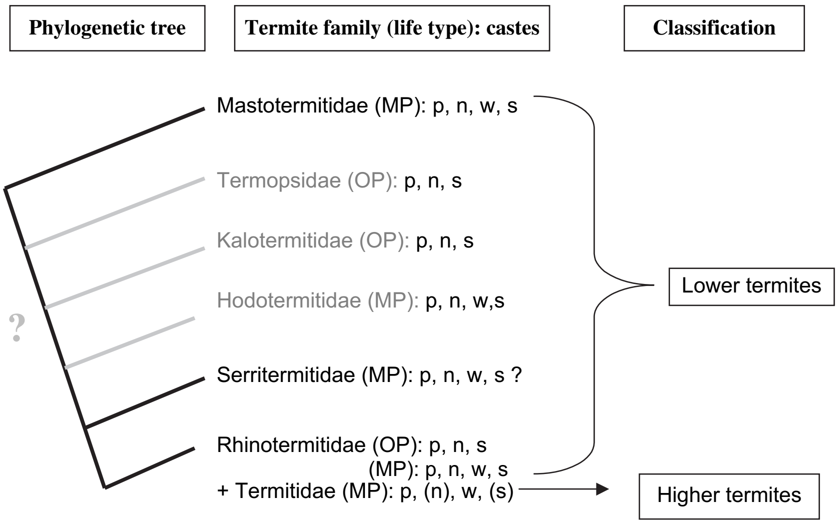

Fig. 1. Phylogenetic tree with life types and the occurrence of different castes in termites. OP- one-piece life type termites which nest in a piece of wood that serves both as shelter and food; MP- multiple-pieces life type termites where nest and food are separated. Unresolved positions are shown in grey or marked ?. Traditionally, termites are classified into lower and higher termites according to the presence or the absence of protozoan gut symbionts. For the monotypic Serritermitidae the caste system is unknown (Roisin, 2000). Castes: p = primary reproductives, n = neotenic reproductives, w = (true) workers, s = soldiers; castes in parentheses indicate that the caste is not present in all species of this family. In taxa lacking workers, totipotent large immatures (often also called pseudergates, helpers, or false workers; see Table 3) occur. As they do not represent a developmental endpoint, they are not equivalent to the other termite castes and therefore are not listed as a caste here.

termites), true workers are absent in the families Termopsidae (dampwood termites) and Kalotermitidae (drywood termites), and in the genus *Prorhinotermes* within the Rhinotermitidae. In these taxa the majority of individuals are large immatures (also called workers, helpers, false workers or pseudergates) which differ from true workers in several aspects. As termite terminology can be very confusing, we recommend the use of 'false workers' for the large immatures of the Termopsidae, Kalotermitidae and *Prorhinotermes*: the term 'pseudergate' originally had a more restricted definition (individuals that developed regressively from nymphs; Grasse & Noirot, 1947; see also Table 3), and there is accumulating evidence that they are not engaged in brood care or raising of siblings to the extent that the term 'helper' might imply (Korb, 2007b; B. Rosengaus personal communication). The workers of all other termites are 'true workers' (see Table 3). True workers with flexible development including regressive moults together with false workers have often been called 'pseudergates'. We propose here to use the terms 'pseudergate *sensu lato*' for workers with flexible development including false workers, and 'pseudergate *sensu stricto*' exclusively for individuals that develop *via* regressive moult(s) from nymphs (i.e. pseudergates as defined by Grasse & Noirot, 1947; see Table 3).

The false workers of the Termopsidae, Kalotermitidae and *Prorhinotermes* lack morphological differentiations and are not a final caste. They are a mixed group of several instars which are not arrested in their development, but rather retain the option to develop into sterile soldiers (a few individuals within each colony) or one of the two types of reproductives (Fig. 2A). Thus, in contrast to what was recently reported for a *Reticulitermes* species (Hayashi *et al.*, 2007), caste development in these taxa is not genetically determined. In the pathway leading to reproductives' false workers can either develop into winged sexuals that found a new nest (primary reproductives) or they can become neotenic replacement reproductives in the natal nest if the same-sex reproductive of the colony is unhealthy or dies. Neotenic reproductives in Termopsidae, Kalotermitidae and *Prorhinotermes* are characterized by the absence of wings and compound eyes and a less sclerotized cuticle than in alates. They may originate *via* a single moult from any instar after the third. At this neotenic moult, their gonads grow and they develop some imaginal characters while maintaining an otherwise larval appearance; some characters, like wing pads, may even regress. This stands in contrast to the development of winged sexuals which is gradual and generally occurs *via* several nymphal instars with increasing wing pad development.

Associated with the lack of a true worker caste in Termopsidae, Kalotermitidae and *Prorhinotermes* is a characteristic life type. These termites live inside a piece of wood that serves both as food and nest and which they, except occasionally in a few species, never leave to exploit new resources (one-piece nesting termites; Abe, 1987, 1990). Consequently, when the food is depleted, the colony dies. In contrast to the typical drywood and dampwood termites, however, it has recently been shown for some *Prorhinotermes* species that they can move to a new nest site when food supplies are low (Roisin & Parmentier, 2006). Interestingly, neotenic development can be triggered by adding new potential nest sites (i.e. pieces of wood) at a distance of a few centimetres, and some data indicate that these neotenics

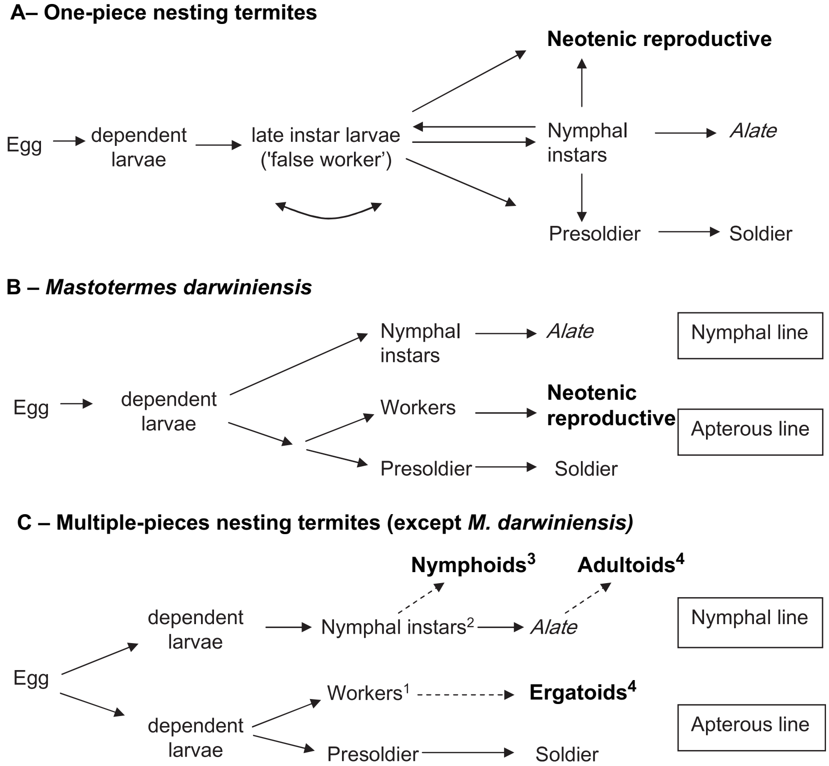

Fig. 2. Developmental pathways in termites. Italics indicate winged reproductives; bold type indicates wingless reproductives; →: progressive moult; ←: regressive moult; ↔: stationary moults. [1]: workers develop from different instars depending on the species; they are partly polymorphic: i.e. major and minor workers; [2]: in some species nymphal instars can have regressive moults; [3–5]: reproductives that stay in the nest to reproduce, present in some species (Myles, 1999); [3] nymphoids: neotenic reproductives developing from nymphs; [4] adultoids: non-dispersing reproductives developing from alates, mainly in Termitidae; [5] ergatoids: neotenic reproductives developing from (true) workers.

become the kings and queens of the new nest in a budding process (Roisin, 2006). Thus, although these neotenics do not reproduce in the natal nest, as occurs in the Termopsidae and Kalotermitidae, immatures of *Prorhinotermes* species can become reproductives without a costly winged dispersal process.

Strikingly, this option to become an unwinged reproductive that avoids a costly nuptial flight also exists in the most basal termite family, the monotypic Mastotermitidae; *Mastotermes darwiniensis* lacks totipotent large immatures and has true workers (Fig. 2B). Recent results show that similar to derived termite taxa, such as the Termitidae, there is an early separation into two developmental pathways, the apterous and nymphal lines, in *M. darwiniensis* (Watson & Abbey, 1985; Watson & Sewell, 1985; Parmentier, 2006). These have been called neuter and alate lines respectively, but the former terms seem to be more appropriate (see Roisin, 2000). Individuals can either develop *via* nymphal instars into winged sexuals (nymphal line) or they can become workers (apterous line) (Fig. 2B). Interestingly, these true workers have the option to develop into wingless neotenic reproductives (ergatoids; neotenics derived from workers) which have been shown to head most of the field colonies that originate primarily *via* colony budding (Goodisman & Crozier, 2002; M. Lenz, personal communication). Although alates do occur, they are apparently less successful in colony foundation; field colonies with primary reproductives were not found in recent studies (Goodisman & Crozier, 2002; M. Lenz, personal communication). Thus, *M. darwiniensis* displays mosaic evolution of ancestral and highly derived traits. Convergently to derived taxa, like some Rhinotermitidae and the Termitidae (Fig. 2C), it evolved a separate apterous worker line, although from a developmental point of view, this worker line is not equivalent to those in the derived taxa; both the nymphal and apterous lines in *M. darwiniensis* have direct reproductive options, the nymphal line in the form of winged sexuals and the apterous worker line in the form of unwinged neotenic reproductives. In *M. darwiniensis* the neotenics, on which colony reproduction largely relies, originate from the workers, whereas in most other termite species nymph-derived neotenics are found (M. Lenz, personal communication). Clearly, in all basal termite clades neotenic reproduction is common (Fig. 1) and presents an alternative to winged dispersing sexuals.

Neotenic reproduction, which only has been lost secondarily in some derived clades, thus, can be considered a synapomorphy that characterizes termites. Its ancestral origin goes hand in hand with the transition to eusociality and has long been claimed to be fundamental for the evolution of eusociality in termites (Myles, 1988; Thorne, 1997; but see also Roisin, 1999). Reproduction as wingless neotenics might be regarded as an alternative breeding tactic that avoids the cost of winged dispersal. Early separation during development into a nymphal and apterous line in species with true workers [Rhinotermitidae (except *Prorhinotermes*), Serritermitidae, Hodotermopsidae, Termitidae] which forage outside the nest (multiple-pieces nesting termites, *sensu* Abe, 1987) (Fig. 2C) might further imply that the termite caste system, including true workers, arose from a wing-polyphenism reflecting alternative breeding tactics (*sensu* Gross, 1996).

III. ULTIMATE CAUSES INFLUENCING CASTE DEVELOPMENT IN LOWER TERMITES

Their developmental flexibility combined with a basal phylogenetic position make one-piece nesting termites ideal subjects to study the ultimate causes for the development of immatures into soldiers and two types of reproductives (wingless neotenics and dispersing winged sexuals). In solitary insects, wing polyphenisms reflect alternative breeding options: winged individuals disperse to breed elsewhere, while wingless adults reproduce at the natal nest (philopatric breeding) or close to it. The 'developmental decision' whether to stay or leave in these solitary insects generally depends on density and food availability at the natal nest, which will both influence reproductive opportunities and the direct fitness of philopatric breeding (Roth, 1981; Müller, Williams & Hardie, 2001; Braendle *et al*., 2006). The default option in solitary insects is to become a winged sexual, as dispersal in all organisms is generally selected for to avoid competition with relatives and inbreeding (Hamilton & May, 1977). Abundant food resources at the nest, opportunities to meet unrelated mating partners close to the nest, or a lack of incest avoidance (e.g. through secondary mechanisms that protect against inbreeding depression) combined with high dispersal costs may, however, select for philopatric breeding and the evolution of wingless morphs (e.g. Alexander, 1974; Braendle *et al*., 2006).

In the ancestors of termites, it can be hypothesised that the first individuals to remain at the nests 'chose' philopatric breeding to avoid costly dispersal, while helping evolved only secondarily. This is illustrated by an extant drywood termite species, *Cryptotermes secundus*. We will concentrate on this species here because it not only exhibits an ancestral one-piece nesting life type (Korb, 2007*a*) but also is one of the most thoroughly studied. References to other lower termites will be given where appropriate. In *Cryptotermes secundus*, like in all one-piece nesting termites, totipotent individuals have the option to develop into winged sexuals or to stay in the nest with a chance of becoming a wingless neotenic replacement sexual when the same-sex reproductive of the colony dies. There is no local resource competition, as the nest constitutes a bonanza food resource that generally outlasts the lifetime of the founding primary reproductives (Korb, 2008). If the nest quality declines, individuals are predicted to show an increased tendency to develop into winged sexuals. Experiments have confirmed this, showing that reduced food availability (Korb & Lenz, 2004; Korb & Schmidinger, 2004) or a high parasite load (Korb & Fuchs, 2006) at the nest together with large group sizes lead to increased development of dispersing sexuals. Furthermore, the number of individuals developing into winged sexuals that leave the nest can be explained by the relative probability of inheriting the nest *versus* successfully founding a new colony (Korb, 2008). As has been suggested for termites in general (Nutting, 1969), the probability of successfully founding a nest is extremely low in *C. secundus* (< 1 %). The chances of inheriting the colony are on the same order of magnitude and depend on colony size, age of the present reproductives, and the potential longevity of the nest (Korb & Schneider, 2007). These three variables explain the variation in the number of individuals developing into dispersing sexuals in field colonies. This suggests that individuals remaining at the nest as neotenics gain direct fitness benefits as has also been proposed by Myles (1988): dispersal is risky, while the nest presents a safe haven (*sensu* Kokko & Ekman, 2002). Similarly, in *Zootermopsis nevadensis* inheritance of the natal breeding position after intercolonial encounters seems to favour the development of soldier-like neotenics (i.e. neotenics with soldier-like traits, misleadingly also called reproductive soldiers) which are a peculiarity of the Termopsidae (Thorne, Breisch & Muscedere, 2003). At least in *C. secundus*, indirect fitness benefits gained through raising siblings seem to be less important (Korb, 2007*b*): the 'decision' to develop into a winged sexual is independent of the number of young present in the nest. If individuals were staying in order to raise young, one would predict a negative correlation between these variables: individuals would be less likely to leave the colony when there are more offspring to raise. The lack of a correlation was explained by subsequent observations of an absence of brood care in this species (Korb, 2007*b*).

Although *C. secundus* is the only species for which we have such detailed results derived from field as well as laboratory experiments, these results might apply to one-piece nesting termites in general (Korb, 2008) for several reasons: (i) all one-piece nesting termites have totipotent false workers that can develop into winged sexuals and neotenic reproductives; (ii) reports exist for many one-piece nesting termites that a reduction in food availability triggers the development of winged sexuals (Buchli, 1958; Lenz, 1976, 1994; La Fage & Nutting, 1978; Korb & Lenz, 2004); (iii) they live within a bonanza-type food resource, removing the value of food provisioning for nestmates as all individuals have easy access to food, and a lack of specialized brood care has been recorded for at least four other species [*Zootermopsis nevadensis* (Howse, 1968); *Zootermopsis angusticollis* (Rosengaus & Traniello, 1993); *Cryptotermes domesticus* and *Cryptotermes cynocephalus* (J. Korb, personal observations)]; (iv) they live in a wooden nest that is well protected against predators, while dispersal is relatively risky (Nutting, 1969; Myles, 1988). Taken together, it appears that breeding opportunities as neotenic reproductives offer high incentives for staying at the nest in one-piece nesting termites. Hence, as in solitary insects with wing polymorphism, there seem to be two alternative tactics: stay and breed as wingless sexuals, or leave as winged sexuals to reproduce elsewhere.

It can therefore be suggested that the safety of the nest, together with the chance of inheriting the natal breeding position, led to delayed dispersal and the formation of family groups. In *C. secundus,* as well as in other termites (Haverty, 1977, 1979; Haverty & Howard, 1981; Shellman-Reeve, 1997), the first soldier only develops after such a group has become established. In *C. secundus* this commonly is at the end of the first year after colony foundation, when about 20 false workers are present. Thereafter, the number of soldiers within a colony is adjusted to colony size, so that a rather constant and species-specific proportion of soldiers will be present within a nest (Haverty, 1977, 1979; Haverty & Howard, 1981). These sterile soldiers defend their family against predators and competitors, hence gaining indirect fitness benefits. This was shown in a field experiment with *C. secundus* in which soldiers were removed and their re-development hormonally suppressed: soldier-less colonies had a lower fitness than similar-sized control colonies (Roux & Korb, 2004).

IV. PROXIMATE MECHANISMS UNDERLYING CASTE DEVELOPMENT

(1) Social and environmental triggers of polyphenisms in termites and other hemimetabolous insects

For most organisms displaying alternative phenotypes, neither phenotype exhibits higher fitness overall. Rather, there is a trade-off, with the relative fitness of the different phenotypes being contingent upon environmental conditions. The evolution and maintenance of polyphenisms, therefore, requires and is a consequence of variation in the environment. For the evolution of polyphenisms several conditions must be met. First, environmental conditions must influence development to generate different phenotypes. Second, the resulting phenotypes must exhibit higher than average fitness in their respective environments. The factors acting as triggering cues may be the same as the selective agent, or they may be different. As the developmental environment of a phenotype often precedes the selective environment for the adult organism, an environmental cue must at least be correlated with future selective factors (West-Eberhard, 2003). Environmental control of alternative phenotypes can, therefore, evolve in organisms living in spatially or temporally variable environments in which cues can be used to predict reliably the future selective environment (Moran, 1992).

In termites, the cues triggering wing development are identical with the selective agents that favour dispersal of individuals. Reduced food availability or increased parasite load (both causing reduced fitness benefits for individuals staying at the nest) immediately induce a change in behaviour of *C. secundus* linked to development into winged sexuals (Korb & Schmidinger, 2004): false workers increase food acquisition behaviours and spend less time moving. More strikingly, while proctodeal trophallaxis (anal feeding) is reduced at the colony level, those individuals which are the most active feeders of other individuals develop progressively into nymphal instars. These results support a long-standing, but so far unproven suggestion that inhibitory substances are transmitted within the colony *via* proctodeal trophallaxis (Lüscher, 1974). Similarily, large group sizes which decrease an individual's chance of inheriting the nest, function as triggers of winged sexual development.

Assuming that termite societies evolved as a consequence of immatures (false workers) following conditionally two alternative reproductive tactics (dispersing or staying at the nest), and that such alternative tactics ("should I lay or should I go") are common in solitary hemimetabolous insects and are frequently associated with wing length polyphenisms, one would expect that the mechanisms underlying the development of alternative phenotypes are evolutionarily conserved. Indeed, similar to one-piece nesting termites, food quality or quantity and population density (i.e. group sizes) are factors known to affect wing development and reproductive physiology in a large number of hemimetabolous insects, including aphids, locusts and crickets (reviewed by Zera, 2003; Hartfelder & Emlen, 2005; Braendle *et al.*, 2006). Thus, the plethora of results on reproductive physiology in cockroaches (for review see Raikhel, Brown & Bellés, 2005) may heuristically guide future in-depth studies on this major aspect of sociality in termites.

The second non-reproductive caste in termites, the soldiers, which are the only individuals in one-piece nesting termites that lose the ability to reproduce, also result from an environmentally induced polyphenism (Lüscher, 1958; Lenz, 1976; Korb, Roux & Lenz, 2003). Their development is triggered by the presence of reproductives, food availability and the size of a colony, while the presence of soldiers inhibits further soldier differentiation (Miller, 1942; Lüscher, 1969; Springhetti, 1969; Haverty & Howard, 1981; Bordereau & Han, 1986; Liu *et al.*, 2005*a*). This results in colonies having a more or less constant proportion of soldiers in relation to colony size (Haverty, 1977; Noirot & Darlington, 2000). Whether predation pressure also may influence soldier numbers is still controversially debated (Noirot & Darlington, 2000).

(2) Endocrine control of caste development in lower termites

The coordination of growth and tissue differentiation within modular systems, such as the segmented body plan of insects, requires both long-range and short-range signaling by hormones. Ecdysteroids and juvenile hormones are the key factors that drive an insect through larval and nymphal moults and metamorphosis, and their chemistry, haemolymph titres and mode of action in target tissues have been investigated in great detail in a wide variety of hemimetabolous and holometabolous species (for reviews see Nijhout, 1994; Riddiford, 1996; Hartfelder, 2000; Goodman

& Granger, 2005; Henrich, 2005; Lafont *et al.*, 2005). Furthermore, models have been developed from hormone titre analyses and hormone application experiments that explain the transition from hemimetabolous to holometabolous development through the exploitation of the pronymphal stage and successive recruitment of epidermal cells into imaginal discs (Truman & Riddiford, 1999, 2002). This already highly successful life-history transition has been further extended by the introduction of alternative phenotypes (polyphenisms), which represent adaptive responses to environmental changes without disrupting successful genotypic combinations.

As detailed above, caste development in the lower termites is highly plastic (with certain restrictions in multiple-pieces nesting termites) (Fig. 2), making them a challenge to any model on endocrine regulation in hemimetabolous development, because moults are not only progressive or stationary, but can even be regressive. Below we review the literature on morphogenetic hormones and their actions in termite caste development and try to integrate these still fragmentary findings with the much better studied cockroaches, within which the termites are nested (Inward *et al.*, 2007*a*; see also Lo *et al.*, 2007), and with general ideas on the evolution of insect metamorphosis. The focus will be on the lower termites because caste fate in the higher termites (Termitidae) is generally determined rather early in development and may even involve embryonic predisposition *via* maternally deposited hormones (Lanzrein, Gentinetta & Fehr, 1985*b*). Furthermore, moults in higher termites are generally progressive and there are no records of regressive moults in Termitidae.

(3) Juvenile hormone in progressive moults of lower termites

As illustrated in Fig. 2, progressive moults of particular importance to caste fate in lower termites are (i) the late instar larvae to presoldier/soldier transition, (ii) the transition from late instar larvae to nymph (first nymphal moult, to the nymphal line), and (iii) from late instar larvae/nymphs to a neotenic replacement reproductive. Of these, the presoldier-soldier transition is at present the best understood.

Although the induction of soldier differentiation by juvenile hormone (JH) and juvenile hormone analogues (JHAs) was one of the earliest findings in JH research (Lüscher, 1969; Howard & Haverty, 1979; for a recent summary see Hrdy *et al.*, 2006), the regulation of JH titres and its mode of action is only now becoming clear, one of the main problems being the role of the social environment.

When monitoring JH and ecdysteroid titres in isolated larvae of the rhinotermitid *Reticulitermes flavipes*, Okot-Kotber *et al.* (1993) noted a gradual increase in levels of ecdysteroids followed by a steeper increase in JH titre, both peaking at day 9 after isolation, shortly before a presoldier moult normally initiates in isolated pseudergates *sensu lato* of this species. A radiochemical assay for measuring JH biosynthesis rates was employed to monitor corpora allata activity in pseudergates *sensu lato* of *Reticulitermes flavipes* that were kept isolated from their nest in groups of 12–50 individuals (Elliott & Stay, 2008). The results showed an increase in JH synthesis around the time point when some of the pseudergates *sensu lato* were expected to develop into neotenic reproductives or presoldiers. Also, while JH synthesis rates were generally higher in presoldiers than in pseudergates *sensu lato* or soldiers, corpora allata activity was considerably lower in presoldiers than in pseudergates *sensu lato* or neotenics in the pharate stage, that is during the subsequent moulting phase. The authors interpreted this as similar to the terminal stage of development in cockroaches.

A caveat in the interpretation of these results is that the developmental profiles of JH are strongly modulated by season, food availability and colony composition. In the rhinotermitid species, *Coptotermes formosanus*, meticulous studies revealed a strong seasonal cycling of JH titres in soldiers and pseudergates *sensu lato* (Liu *et al.*, 2005*b*), similar to earlier observations on corpora allata (CA) volumes in the kalotermitid *Kalotermes flavicollis* (Lüscher, 1972). JH titre levels in pseudergates *sensu lato* were either negatively affected by increasing the percentage of soldiers in experimental groups (Mao *et al.*, 2005; Park & Raina, 2005), or were positively affected by improved food or temperature conditions (Liu *et al.*, 2005*a*). Furthermore, the time course of JH synthesis rates for *Reticulitermes flavipes* pseudergates *sensu lato* kept isolated from their nest was shown to be dependent on group size (Elliott & Stay, 2008). Such social influences on factors controling caste development are widespread and not limited to soldier development in termites, but rather are a facet of the pleiotropic functions of JH in insect development and reproduction. For example, JH both prevents wing shedding and precocious ovarian activity in immature alates of the termopsid *Zootermopsis angusticollis*, yet stimulates oogenesis in mated queens in this species (Brent, Schal & Vargo, 2005). This pleiotropic role of JH also became apparent in a study on corpora allata activity in apterous and brachypterous neotenic *Reticulitermes flavipes* females, where an increase in the number of vitellogenic ovarioles was accompanied by an increase in corpora allata activity (Elliott & Stay, 2007). The critical questions, therefore, are (a) how is the hormone titre regulated, and (b) which are the molecular targets for JH action in termites?

The first inhibitors of JH biosynthesis were discovered in cockroaches and were termed allatostatins (Woodhead *et al.*, 1989; Stay *et al.*, 1991). Members of this large family of neuropeptides have now been identified in all major orders of insects (for review see Stay & Tobe, 2007). Allatostatin immunoreactivity has recently been described in the rhinotermid, *Reticulitermes flavipes*, showing that lateral and medial neurosecretory cells in the brain innervate the corpus allatum (Yagi *et al.*, 2005). Furthermore, *in vitro* incubation of termite corpora allata (CA) in the presence of two cockroach allatostatins significantly inhibited JH production (Yagi *et al.*, 2005). Allatostatins, however, do not only regulate JH synthesis but, due to their widespread occurrence and origin as gut-brain peptides, can affect a large suite of body functions, including food intake by modulating gut contraction (Aguilar *et al.*, 2003; Aguilar, Maestro & Belles, 2006). The effects of food quality on soldier induction (Liu *et al.*, 2005*a*) may partially be mediated through these routes. In conjunction with their allatostatic effect, these peptides would then link reduced individual food intake due to deteriorating colony conditions to a reduction in the percentage of larvae that enter the presoldier-soldier pathway. The pleiotropic functions of such endogenous regulatory peptides could, thus, provide a switch mechanism that links the social with the internal environment. Links between the social and the internal environment are still little understood, not only in termites but also in the caste development of other social insects. Even though inhibitory pheromones have long been thought to be involved in the development of reproductives and in the adjustment of caste ratios in termites (Lüscher, 1964; Lefeuve & Bordereau, 1984; Noirot, 1991), the neural substrates for their perception and transmission to the endocrine system are unknown, except for the architecture of the allatostatin-expressing neurons (Yagi *et al.*, 2005).

After its release into the haemolymph, the extremely lipophilic JH molecule must be bound to transport proteins in order to reach target organs and to remain protected against degradation by JH esterases and/or JH epoxide hydrolases (reviewed by Goodman & Granger, 2005). JH binding proteins have been identified by photoaffinity labeling in haemolymph of the rhinotermitids *Reticulitermes flavipes*, *Coptotermes formosanus* and the termopsid *Zootermopsis nevadensis* (Okot-Kotber & Prestwich, 1991*a*, *b*). While these proteins bind JH with high affinity, two general transport and storage proteins, hexamerin 1 (Hex-1) and hexamerin 2 (Hex-2) have recently emerged as major candidates for regulators of soldier development in the rhinotermitid *Reticulitermes flavipes*. As in other insects, fat body expression levels and haemolymph titres of these proteins are strongly modulated during development and seem to be affected by JH, especially in the case of Hex-2 (Scharf *et al.*, 2005*a*, *b*; Zhou *et al.*, 2006*b*; Zhou, Faith & Scharf, 2006*a*; Zhou, Traver & Scharf, 2007*b*). In turn, JH regulation of hexamerin expression has a marked feedback effect on JH availability to target tissues. Whereas hexamerins of other insects are known to bind JH with low affinity (Tawfik *et al.*, 2006), they apparently do this in a peculiar way in *R. flavipes.* Based on RNA interference (RNAi) results and Western blotting with a JH-specific antiserum, Zhou *et al.* (2006*b*) conclude that Hex-1 might covalently bind JH and that these two proteins interact and form a sink for JH that, at the colony level, would allow the fine-tuning of worker to soldier caste ratios. This termite therefore seems to have exploited an abundant haemolymph protein with pleiotropic functions and co-opted it into a regulatory network of social organization (Zhou *et al.*, 2006*a*). A similar feedback network involving a hormone of pleiotropic functions (JH) and a phylogenetically equally old transport/storage protein (vitellogenin) has recently been demonstrated to regulate age polyethism in honey bee workers (Amdam *et al.*, 2003, 2006).

While these examples illustrate the co-option of ground plan components of insect developmental and reproductive physiology, there are still a number of questions to be answered. Firstly, the proposed covalent binding of JH is peculiar and raises the following questions: (a) is Hex-1-bound JH accessible to degradation by JH esterase or JH epoxide hydrolase in haemolymph, (b) is it sequestered in Hex-1-bound form into the fat body, and if so, what happens there, (c) can JH-sensitive target tissues also sequester Hex-1-bound JH, and what are the subsequent effects and, most importantly, (d) is this mechanism of JH sequestration a general property of lower termites or is it restricted to *Reticulitermes flavipes*?

Additional questions have been raised by Hrdy *et al.* (2006) who showed that the racemic JH-III used in the above experiments on hexamerin function is not a particularly strong inducer of soldier development in *Reticulitermes* species, when compared to other JHAs. A feasible explanation for the low activity of JH-III would be its well-known lower metabolic stability in insect haemolymph, where it is efficiently degraded by insect JH-esterases (Oakeshott *et al.*, 2005). In addition, the fraction of JH-III that is covalently bound to Hex-1 in *Reticulitermes flavipes* haemolymph would essentially be unavailable for physiological functions. An important question to answer would, thus, be whether JHAs modulate hexamerin expression similar to JH, that is, do they transcriptionally mimic JH-III and do they also bind to Hex-1?

Regarding intracellular JH clearing, the finding of a JHA-induced expression of a cytochrome P450 transcript in the fat body of the termopsid *Hodotermopsis sjostedti* (Cornette *et al.*, 2006) is interesting. This enzyme could be a candidate for intracellular JH degradation, since cytochrome P450s are not only general detoxifying enzymes, but also have been specifically implicated in the metabolism of methyl farnesoate to JH-III (Helvig *et al.*, 2004). Similarly, Zhou *et al.* (2007*a*) showed for the rhinotermitid *Reticulitermes flavipes* that several fat-body-related P450s (CYP4) were differentially expressed after JH treatments, and we found that a cytochrome P450 enzyme was overexpressed in female *Cryptotermes secundus* neotenics compared to false workers (Weil, Rehli & Korb, 2007; see below).

(4) Molecular underpinnings of caste in lower termites

Attempts to unravel the molecular basis of caste development have also focused on the soldier differentiation pathway, because of its relative ease of induction by JH and JHA applications, and also because of the marked morphological differences in soldiers. The first differential gene expression screens performed on the termopsid *Hodotermopsis sjostedti* (formerly *H. japonica*) (Miura *et al.*, 1999; Miura, 2001) led to the identification of a gene with soldier-specific expression (SOL1) in the mandibular gland of this dampwood termite. It encodes a putative member of the lipocalin family that may be a soldier-specific secretory product of this gland. In the termitid *Hospitalitermes medioflavus*, this gland develops from a disc-like structure once a presoldier-differentiating moult has been induced by a high JH titre (Miura & Matsumoto, 2000).

In a follow-up study performed as a differential display reverse-transcription polymerase chain reaction (DDRT-PCR) screen on RNA extracted from mandibles of

Hodotermopsis sjostedti, Koshikawa *et al.* (2005) investigated tissue-specific differentiation processes induced by the application of a JHA. They confirmed the expected overexpression of cuticle proteins in the mandibles of developing soldiers and also revealed a set of putative transcription and translation regulators (including a *staufen* orthologue), an actin-binding protein possibly involved in cellular morphogenesis and also a member of the aldehyde dehydrogenase (Adh) family.

Macroarray screens on whole-body pseudergate *sensu lato*, presoldier, soldier and nymphal RNA of the rhinotermitid *Reticulitermes flavipes* (Scharf *et al.*, 2003) detected 25 differentially expressed sequence tags (ESTs), 16 of which represented orthologues (E-values $< e^{-6}$). Pseudergates *sensu lato* showed a strong overexpression of endosymbiont cellulase genes and soldiers overexpressed cytoskeletal proteins, especially ones related to skeletal muscle, and a cytochrome oxidase I encoding gene. While these were not surprising findings considering the functions of these two castes, the overexpression of vitellogenin in presoldiers is perplexing. Another remarkable point is that most of the unknown genes (no BLAST matches) were overexpressed in pseudergates *sensu lato* or in soldiers and, thus, may represent novel genes typical for either of these castes, like the SOL1 transcript of the termopsid *H. sjostedti*.

The molecular basis of development from pseudergates *sensu lato* to reproductives (especially into neotenic replacement reproductives, since this requires only a single moult) is now under investigation in the kalotermitid *Cryptotermes secundus* (Weil *et al.*, 2007). Using a highly sensitive suppression subtractive hybridization strategy (Representational Difference Analysis), this study led to the complete or partial cloning of five differentially expressed genes, as validated by quantitative real-time PCR. These genes (a member of the esterase-lipase family, a putative beta-glycosidase, a cytochrome P450 gene, vitellogenin, and an unknown gene) were markedly overexpressed in female neotenics. Surprising findings for vitellogenin transcripts were the lack of sex-specificity (highly expressed both in neotenic females and males) and lack of compartmentalization (expressed in the head, thorax, and abdomen). Vitellogenin has long been considered a sex-specific protein exclusively required for oogenesis. This paradigm has lately undergone considerable change, especially in social insects. In the honey bee, vitellogenin has been shown to be involved in the regulation of task performance, *via* repression of the JH titre (Guidugli *et al.*, 2005*a*), and in longevity (Amdam *et al.*, 2005; Corona *et al.*, 2007), and vitellogenin expression has also been detected in larval stages (Guidugli *et al.*, 2005*b*). In cockroaches, the induction of vitellogenin by JH is also not restricted to adult females, but has been reported to occur in preadult stages (Lanzrein, 1974; Cruz *et al.*, 2003) and in males (Mundall, Tobe & Stay, 1979).

Such differential expression of several metabolism-related genes in termite caste development as well as the overexpression of a cytochrome C oxidase subunit III in nymphs and pseudergates *sensu lato* of the rhinotermitid *Reticulitermes santonensis* (Lienard *et al.*, 2006), may shed light on a largely overlooked aspect of caste development, the importance of metabolic regulation. In this respect, termites parallel the honey bee, where similar observations have emerged from several expression screens that can now be explored in depth on the basis of genomic information (Cristino *et al.*, 2006).

Whereas these studies are informative on metabolic regulation and on the differential expression of structural genes they tell us relatively little about patterning mechanisms involved in the shaping of caste-specific structures. In particular, the shutting down of wing development in termite castes (true workers, neotenics and soldiers) has not yet been investigated at the molecular level, although studies have been initiated to reveal underlying cellular mechanisms (Miura *et al.*, 2004).

V. PERSPECTIVES ON HORMONAL REGULATION, CASTE DEVELOPMENT AND TERMITE EVOLUTION

(1) Ecdysteroids and insulin signaling

Whereas the role of JH in termite caste development has received much attention due to its potential in pest control strategies, ecdysteroids have only rarely been studied, despite their importance in moulting cycles and metamorphosis. JH and ecdysteroid titres rise concomitantly in isolated pseudergates *sensu lato* of the rhinotermitid *Reticulitermes flavipes* (Okot-Kotber *et al.*, 1993), and the deposition and sclerotization of cuticle in the mandible of soldiers is dependent on an interaction between these two hormones (Okot-Kotber, 1983).

For molecular studies, the ecdysteroid signaling pathway may be more productive since, in distinction to JH signaling, the ecdysone receptor(s) (EcRs) and their dimerization partners, the orphan nuclear receptors of the rexinoid/ultraspiracle (RXR/USP) family have been identified in a large number of species (Thummel, 1996; Henrich, 2005). An EcR-A isoform has recently been identified in the cockroach *Blattella germanica,* and RNAi experiments have revealed that it is involved in adult-specific developmental processes, including wing development (Cruz *et al.*, 2006). Similar functional RNAi analyses have also been performed for the *RXR/USP* orthologue of this cockroach showing that *BgRXR* knockdown arrests the nymphal to adult moult (Martin *et al.*, 2006). The identification of *Drosophila EcR* and *RXR/USP* orthologues in cockroaches and the results of the functional assays demonstrate that this nuclear receptor pathway is highly conserved in hemimetabolous and holometabolous insects. Furthermore, besides being a dimerization partner for EcR, the RXR/USP protein has also been suggested as a possible JH receptor in *Drosophila melanogaster* (Jones & Sharp, 1997; Xu *et al.*, 2002; Jones *et al.*, 2006) and also in the honey bee (Barchuk, Maleszka & Simoes, 2004). Alternatively, with their low-affinity binding characteristics for specific ligands, the RXR/USP nuclear receptors could function as lipid sensors, and thus provide a direct link between nutritional status and moulting or metamorphosis induction (Chawla *et al.*, 2001).

This link between the classic morphogenetic hormones, JH and ecdysteroids, and nutritional status could also involve another phylogenetically old signaling pathway which functions as a local growth regulator, the insulin-/-insulin-like signaling (IIS) and the associated target of rapamycin (TOR) pathway (Brogiolo *et al.*, 2001; Edgar, 2006). These flexible response systems to the endogenous nutrient milieu regulate cell growth and cell division, and thus affect organ and body size. Furthermore, they have been shown to interact directly with the function of the prothoracic gland (Colombani *et al.*, 2005; Mirth, Truman & Riddiford, 2005). Through this gateway, an important aspect of metamorphosis, the critical size threshold, becomes directly amenable to natural selection.

In the honey bee, the availability of the complete genome sequence has facilitated the annotation of insulin/insulin-like peptides, insulin receptors, TOR, as well as most of the downstream elements of these signaling pathways (The Honey Bee Genome Sequencing Consortium, 2006). The expression of some of the IIS components has been shown to be strongly affected by the diet provided to honey bee larvae (Wheeler, Buck & Evans, 2006), in correspondence with the caste-specific differential expression profiles found for an insulin receptor (Azevedo & Hartfelder, 2008); interference with TOR signaling by rapamycin administration or RNAi has been shown to affect queen/worker differentiation directly (Patel *et al.*, 2007). The high conservation of IIS components should make this pathway an interesting target for studies on termite caste differentiation. This group of social insects is of singular interest because of its dietary specialization, especially in those lower termite species which continuously reside in and feed on a single piece of dry or damp wood and for which food availability has been shown to trigger different caste developmental trajectories (see above). The question of critical size in the context of nutrition and caste divergence and its consequences for direct and indirect fitness should, thus, be of singular importance to the highly flexible systems of caste determination in the lower termites.

(2) What is the status of the larval stages?

A major problem in termite developmental biology is actually a terminological one. While they are unquestionably hemimetabolous insects, their early postembryonic instars are nevertheless denominated as larvae and not as nymphs, the typical terminology for growth stages of solitary hemimetabolans. In termites, the term nymph is reserved for later developmental stages that exhibit progressively growing wing pads and, thus, are gradually committed to winged dispersal (Noirot, 1990). In some termites only the early stages (first to third instars) are termed larvae, but *de facto*, the false workers of the one-piece nesting lower termites can also be considered larvae, as long as they have an unsclerotized cuticle, simple mandibles and do not show external wing pads or compound eyes. The traditional terminology of termite developmental stages has been called into question by the discovery of an early larval stage with wing buds in the genus *Termitogeton* (Rhinotermitidae). Their wing buds regress in successive moults until they reappear in the single nymphal stage (Parmentier & Roisin, 2003). A second major problem is how to describe stationary and regressive moults in the lower termites.

From their morphological characters and possibly also their developmental status, the early postembryonic stages of termites (Fig. 3) are more similar to the grub-like larvae of lepidopterans or coleopterans than to the more adult-like cockroach or locust nymphs. They are holometabolan-like with respect to the developmental status of their wing and eye primordia and the cuticle structure, and they are hemimetabolan-like with respect to the body tagmata (more pronounced thorax-abdomen division). In terms of evolutionary trajectories, the postembryonic stages of termites (probably including parts of the false worker/pseudergates *sensu lato* stage, which is not a single developmental stage but a composite resulting from progressive, stationary and regressive moulting events) could actually be considered as equivalent to the pronymphal stage of Orthoptera and Blattodea.

This hypothesis would be consistent with the scenario proposed by Truman & Riddiford (1999, 2002) on the transition from hemimetabolous to holometabolous development (but see also Heming, 2003; Minelli *et al.*, 2006; for a wider discussion and alternative viewpoints). In termites external wing buds only start to grow during the nymphal moults (but are present in *Termitogeton*; Parmentier & Roisin, 2003), and larval termites do not have compound eyes but only primordia (Miura, 2005). In holometabolous insects, wings develop from imaginal discs and the eye primordia of termites resemble the eye imaginal discs of *Manduca sexta* (Champlin & Truman, 1998; MacWhinnie *et al.*, 2005). The pseudergates *sensu lato*, which can comprise several instars,

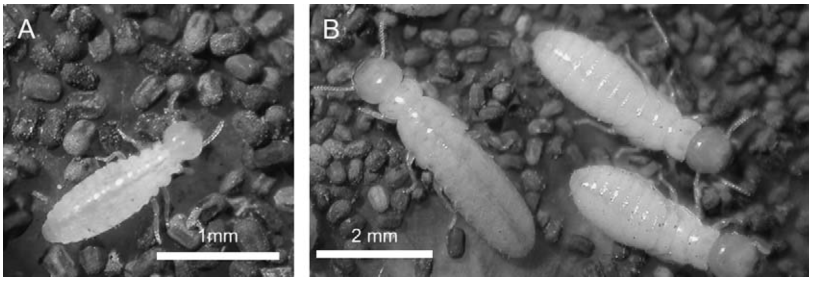

Fig. 3. Developmental stages of the drywood termite *Cryptotermes secundus*. (A) 'Dependent larva', (B) false workers (larval and nymphal instars).

could, thus, be conceived as a platform for flexibility in developmental decisions, leading to soldiers, alates, or neotenic reproductives.

So far, this hypothesis remains to be tested. Some data hint at this scenario, such as a change in mandible shape and an increased degree of sclerotization observed in rhinotermitids following JHA application (Lenz, 1976; Lelis & Everaerts, 1993), similar to results obtained in locust pronymphs (Truman & Riddiford, 1999), and the formation of soldier-alate intermediates (Lelis & Everaerts, 1993; Koshikawa, Matsumoto & Miura, 2002; Miura, 2005) in *Hodotermopsis sjostedti* (Termopsidae), which can be interpreted as a trade-off between wing/eye development and that of defence structures on the head.

A revision of termite embryogenesis is clearly required. There are excellent descriptions on the embryonic development of *Kalotermes flavicollis* and *Zootermopsis nevadensis* (Striebel, 1960) and *Crypotermes brevis* (Kawanishi, 1975), but these do not address the question of whether termites undergo embryonic moults. In locusts, such moults are important for the appearance of a pronymphal stage, before eclosion of the larva from the egg. Identification of a pronymphal stage would require detailed JH and ecdysteroid titre measurements to determine whether there are differences in the titre profiles between the larval and nymphal/presoldier stages and whether and how these titre patterns differ from those established for a well-studied basal hemimetabolan insect, the cockroach *Nauphoeta cinerea* (Lanzrein *et al*., 1985*a*). An alternative would be to investigate candidate genes for gene regulatory networks underlying moulting and metamorphosis, in an approach similar to that taken by Abouheif & Wray (2002) for understanding the prevention of wing development in ant workers. Both approaches should enable us to clarify the position of the stationary and especially the peculiar regressive moults in lower termites.

(3) Regressive moults in lower termites, a major enigma

The regulatory network for the stationary moults of pseudergates *sensu lato* may be equivalent to that of supernumerary moults of solitary hemimetabolous insects, which are associated with little growth and no differentiation. The driving force for stationary moults in the lower termite pseudergates *sensu lato* may actually be the wear of their mandibles (Roisin & Lenz, 1999). Renewing this structure in a moult would allow large immatures (i.e. pseudergates *sensu lato*) to remain for an extended period in the nest with a chance of eventually inheriting the natal breeding position.

Regressive moults are observed in the nymphal-alate transition, where nymphs that did reach the alate moult in one year regress to the false worker stage, accompanied by wing pad reduction (pseudergates *sensu stricto*) (Korb & Katrantzis, 2004). So why do these individuals not remain as nymphs in the nest and develop into alates early in the next swarming season? This is especially interesting as there seem to be developmental deadlines for each nymphal instar; individuals that fail to reach these deadlines cannot become alates.

There might be two explanations. First, a regressive moult in the nymphal-alate transition could be a consequence of wing pad mutilation. Mutilated wing buds were found in several lower termite species and it was hypothesized that they were the result of manipulations by siblings or parents (Zimmermann, 1983; Myles, 1988; Roisin, 1994, 2006; Miura *et al*., 2004) similar to the mutilation of gemmae in some queenless ponerine ants (Peeters & Higashi, 1989; Ramaswamy *et al*., 2004). However, as nobody has actually observed the process of mutilation in nature, its causes are unknown. Results for the drywood termite *Cryptotermes secundus* suggest that such damage is an artefact of handling conditions (Korb, 2005). From a theoretical point of view there should be no selective advantage in monogamous colonies to sibling or parental manipulation (Korb, 2005), especially when false workers do not provide much help in raising siblings, as recent data for *C. secundus* suggest (Korb, 2007*b*). Why then should parents or siblings manipulate their nestmates to stay in the nest, when these nestmates do not provide costly brood care and when breeding opportunities outside the nest are not limited by intraspecific competition? Roisin (2006) suggested that intracolonial competition might exist for some high-quality resources or brood care. But why then can individuals, which do not disperse under abundant food conditions, can become winged sexuals when food availability is reduced? If individuals are capable to develop into alates under reduced food conditions they should be even more capable to become alates under abundant food conditions. These considerations suggest that individuals are staying voluntarily under abundant food conditions.

Conflicts among totipotent individuals over dispersal might occur in fused termite colonies with a within-colony relatedness below 0.5 and if dispersal is a better option then staying at the nest. Further research must show whether these latter conditions are met in any lower termite species under natural conditions. So far, although scant, the available field data suggest that staying in the nest with a chance to inherit the colony is not an inferior reproductive tactic compared to leaving the colony as winged sexual (Korb, 2007*b*, 2008).

An alternative explanation is that regression might be part of an 'honest signal' in a test for alate competence. All individuals have to start from the same stage as apterous individuals. From this stage, only those individuals that reach the developmental deadlines are the most competent for alate development. Individuals that are less competent would stay for another year and try to gain enough resources for the subsequent nuptial flight.

At the proximate level, the general paradigm for hormonal regulation in metamorphic moults provides a series of predictions for regressive moults in the nymphal-alate transition, since the nature of the subsequent moult is always determined during the preceding intermoult period (Riddiford, 1994). A critical factor in the last preimaginal stage is the JH level at the rise of the moult-inducing ecdysteroid peak. We predict that only nymphs with a sufficiently suppressed level of JH will become alates, while an above-threshold JH level during the ecdysteroid peak could block wing bud expansion and reverse the moulting type in terms of cuticle protein expression.

VI. CONCLUSIONS

(1) "Pseudergates *sensu lato*" are a fascinating example of ample developmental plasticity with far-reaching consequences in terms of the ecological and evolutionary success of termites. We compile here current knowledge on the ecology and life history of lower termites, and set this into a framework of developmental biology, especially the endocrine regulation underlying caste differentiation.

(2) The so-called one-piece nesting termites with their wood-nesting life style are considered a basal group (Roisin, 2000; Thorne & Traniello, 2003; Korb, 2007*a*). From phylogenetic data on termite families it is not possible to reconstruct the evolution of true workers unambiguously (Thompson *et al*., 2000, 2004; Grandcolas & D'Haese, 2004; Inward, Vogler & Eggleton, 2007*b*). Other results leave less doubt about the basal position of false workers, and thus of the one-piece nesting life style (reviewed in Korb, 2007*a*): (i) a recently published comprehensive phylogenetic analysis on Dictyopterans places the termites within the cockroaches, Blattodea, as a sister group to the woodroaches, Cryptocercidae (Inward *et al*., 2007*a*; see also Lo *et al*., 2007) which have a similar life style to the one-piece nesting termites; (ii) the presence of true workers in the basal group *Mastotermes darwiniensis*, which are not equivalent to the true workers of other multiple-pieces nesting termites (Parmentier, 2006), suggests at least two independent origins of true workers, implying a basal position for one-piece nesting termites.

(3) The one-piece nesting termites show the greatest flexibility in developmental decisions related to caste. Developmental plasticity is, thus, a basal character in termites. Termite caste allometries (as e.g. shown by Koshikawa *et al*., 2002) are results of phenotypic plasticity that does not involve genetic change. The demonstration of major epigenetic effects on honey bee caste fate through differential genome methylation (Kucharski *et al*., 2008) furthermore shows that social insect polyphenisms can be interpreted as split developmental reaction norms in terms of Schlichting & Pigliucci's (1998) approach to phenotypic evolution.

(4) We propose that a pronymphal stage similar to that of the holometabolan clade (Truman & Riddiford, 1999, 2002) may have been translocated from within the egg to a prolonged postembryonic stage at the beginning of termite evolution, in association with the suppression of wing development due to a burrowing feeding habit in a woodroach-like ancestor. Under this scenario, the prolonged postembryonic stage, exemplified by the false worker/pseudergates *sensu lato* stage, provides a platform for developmental trajectories into wingless and winged reproductives, which present alternative breeding tactics, and, probably as a subsequent evolutionary step, also development into soldiers. The extension of the pronymphal stage of basal hemimetabolans into a sequence of postembryonic instars in such a termite ancestor would then be considered a pre-adaptation for developmental plasticity contingent on the nutritional and social environment. A next step could then have been the assimilation of this contingency into a morphogenetic program that directs the development of alternative phenotypes *via* endocrine control.

(5) The role of JH has long been explored in proposals for termite pest control strategies and based on such data and hormone titre measurements we propose a scenario of endocrine regulation of caste development that incorporates critical periods and response thresholds (Nijhout, 1994; Hartfelder & Emlen, 2005). Recent results on the differential expression of hexamerin genes in the genus *Reticulitermes* can be interpreted as a co-option of a general storage protein that belongs to an ancient family of arthropod proteins (Burmester, 2002).

(6) Even though the focus of the current review is on lower termites, the castes of higher termites can be easily integrated into this framework. The restricted plasticity in the apterous line and their much earlier commitment to reproductive castes can be conceived as a change in the timing of hormone-dependent determination steps. The higher JH levels observed in *Macrotermes michaelseni* eggs dedicated to become reproductives (Lanzrein *et al*., 1985*b*) indicate that the lines split at at least two stages, an early one in the embryonic phase, for the apterous/nymphal decision, and a later one in the early nymphal stages, for the worker/soldier decision (Fig. 2).

VII. ACKNOWLEDGMENTS

We would like to thank Michael Lenz, Paul Eggleton and two anonymous referees for their helpful comments on this manuscript. We also acknowledge financial support from the Deutsche Forschungsgemeinschaft (DFG, KO 1895/6) and from a Brazilian/German (CAPES/DAAD) cooperation program (PROBRAL 261/07).

VIII. REFERENCES

Abe, T. (1987). Evolution of life types in termites. In *Evolution and Coadaptation in biotic communities* (ed. S. Kawano, J. H. Connell and T. Hidaka), pp. 125–148. University of Tokyo Press, Tokyo.

Abe, T. (1990). Evolution of worker caste in termites. In *Social Insects and the Environment* (eds. G. K. Veeresh, B. Mallik and C. A. Viraktamath), pp. 29–30. Oxford & IBH, New Delhi.

Abouheif, E. & Wray, G. A. (2002). Evolution of the genetic network underlying wing polyphenism in ants. *Science* **297**, 249–252.

Aguilar, R., Maestro, J. L. & Belles, X. (2006). Effects of myoinhibitory peptides on food intake in the German cockroach. *Physiological Entomology* **31**, 257–261.

Aguilar, R., Maestro, J. L., Vilaplan, L., Pascual, N., Piulachs, M. D. & Belles, X. (2003). Allatostatin gene expression in brain and midgut, and activity of synthetic allatostatins on feeding-related processes in the cockroach *Blattella germanica*. *Regulatory Peptides* **115**, 171–177.

Alexander, R. D. (1974). The evolution of social behavior. *Annual Reviews of Ecology and Systematics* **5**, 325–383.

Amdam, G. V., Aase, A. L. T. O., Seehuus, S. C., Fondrk, M. K., Norberg, K. & Hartfelder, K. (2005). Social reversion of immunosenescence in honey bee workers. *Experimental Gerontology* **40**, 939–947.

Amdam, G. V., Csondes, A., Fondrk, M. K. & Page, R. E. (2006). Complex social behaviour derived from maternal reproductive traits. *Nature* **439**, 76–78.

Amdam, G. V., Norberg, K., Hagen, A. & Omholt, S. W. (2003). Social exploitation of vitellogenin. *Proceedings of the National Academy of Sciences of the United States of America* **100**, 1799–1802.

Azevedo, S. V. & Hartfelder, K. (2008). The insulin signaling pathway in honey bee (*Apis mellifera*) caste development-differential expression of insulin-like peptides and insulin receptors in queen and worker larvae. *Journal of Insect Physiology*, (in press; doi 10.1016/j.jinsphys.2008.04.009).

Barchuk, A. R., Maleszka, R. & Simoes, Z. L. P. (2004). *Apis mellifera ultraspiracle*: cDNA sequence and rapid up-regulation by juvenile hormone. *Insect Molecular Biology* **13**, 459–467.

Bordereau, C. & Han, S. H. (1986). Stimulatory influence of the queen and king on soldier differentiation in the higher termites *Nasutitermes lujae* and *Cubitermes fungifaber. Insectes Sociaux* **33**, 296–305.

Braendle, C., Davis, G. K., Brisson, J. A. & Stern, D. L. (2006). Wing dimorphism in aphids. *Heredity* **97**, 192–199.

Brakefield, P. M. (2006). Evo-devo and constraints on selection. *Trends in Ecology and Evolution* **21**, 362–368.

Brent, C. S., Schal, C. & Vargo, E. L. (2005). Endocrine changes in maturing primary queens of *Zootermopsis angusticollis. Journal of Insect Physiology* **51**, 1200–1209.

Brogiolo, V., Stocker, H., Ikeya, T., Rintelen, F., Fernandez, R. & Hafen, E. (2001). An evolutionarily conserved function of the *Drosophila* insulin receptor and insulin-like peptides in growth control *Current Biology*, **11**, 213–221.

Buchli, H. R. (1958). L'origine des castes et les potentialités ontogéniques des termites européens du genre *Reticulitermes* Holmgren. *Annales des Sciences Naturelles Zoologie et Biologie Animale* **11**, 267–429.

Burmester, T. (2002). Origin and evolution of arthropod hemocyanins and related proteins. *Journal of Comparative Physiology, Series B* **172**, 95–107.

Carroll, S. B., Grenier, J. K. & Weatherbee, S. D. (2005). *From DNA to Diversity - Molecular Genetics and the Evolution of Animal Design*, 2 edition. Blackwell, Oxford.

Champlin, D. T. & Truman, J. W. (1998). Ecdysteroids govern two phases of eye development during metamorphosis of the moth, Manduca sexta. *Development* **125**, 2009–2018.

Chawla, A., Repa, J. J., Evans, R. M. & Mangelsdorf, D. J. (2001). Nuclear receptors and lipid physiology: opening the X-files. *Science* **294**, 1866–1870.

Colombani, J., Bianchini, L., Layalle, S., Pondeville, E., Dauphin-Villemant, C., Antoniewski, C., Carré, C., Noselli, S. & Leopold, P. (2005). Antagonistic actions of ecdysone and insulins determine final size in *Drosophila. Science* **310**, 667–670.

Cornette, R., Koshikawa, S., Hojo, M., Matsumoto, T. & Miura, T. (2006). Caste-specific cytochrome P450 in the dampwood termite *Hodotermopsis sjostedti* (Isoptera, Termopsidae). *Insect Molecular Biology* **15**, 235–244.

Corona, M., Velarde, R. A., Remolina, S., Moran-Lauter, A., Wang, Y., Hughes, K. A. & Robinson, G. E. (2007). Vitellogenin, juvenile hormone, insulin signaling, and queen honey bee longevity. *Proceedings of the National Academy of Science of the United States of America* **104**, 7128–7133.

Cristino, A. S., Nunes, F. M., Lobo, C. H., Bitondi, M. M. G., Simões, Z. L. P., Fontoura Costa, L., Lattorff, H. M. G., Moritz, R. F. A., Evans, J. D. & Hartfelder, K. (2006). Caste development and reproduction: a genome-wide analysis of hallmarks of insect eusociality. *Insect Molecular Biology* **15**, 703–714.

Cruz, J., Mane-Padros, D., Belles, X. & Martin, D. (2006). Functions of the ecdysone receptor isoform-A in the hemimetabolous insect *Blattella germanica* revealed by systemic RNAi *in vivo*. *Developmental Biology* **297**, 158–171.

Cruz, J., Martin, D., Pascual, N., Maestro, J. L., Piulachs, M. D. & Belles, X. (2003). Quantity does matter: Juvenile hormone and the onset of vitellogenesis in the German cockroach. *Insect Biochemistry and Molecular Biology* **33**, 1219–1225.

Dejean, A., Durand, J. L. & Bolton, B. (1996). Ants inhabiting *Cubitermes* termitaries in African rain forests. *Biotropica* **28**, 701–713.

Deligne, A., Quennedy, A. & Blum, M. S. (1981). The enemies and defense mechanisms of termites. In *Social Insects* vol. 2 (ed. H. R. Hermann), pp. 1–76. New York Academic Press, New York.

Edgar, B. A. (2006). How flies get their size: genetics meets physiology. *Nature Review Genetics* **7**, 907–916.

Eggleton, P. (2001). Termites and trees: a review of recent advances in termite phylogenetics. *Insectes Sociaux* **48**, 187–193.

Elliott, K. L. & Stay, B. (2007). Juvenile hormone synthesis as related to egg development in neotenic reproductives of the termite *Reticulitermes flavipes*, with observations on urates in fat body. *General and Comparative Endocrinology* **152**, 102–110.

Elliott, K. L. & Stay, B. (2008). Changes in juvenile hormone synthesis in the termite *Reticulitermes flavipes* during development of soldiers and neotenic reproductives from groups of isolated workers. *Journal of Insect Physiology* **54**, 492–500.

Goodisman, M. A. D. & Crozier, R. H. (2002). Population and colony genetic structure of the primitive termite *Mastotermes darwiniensis. Evolution* **56**, 70–83.

Goodman, W. G. & Granger, N. A. (2005). The juvenile hormones. In *Comprehensive Insect Molecular Science*, vol. 3 (ed. L. I. Gilbert, K. Iatrou and S. Gill), pp. 319–408. Elsevier, Oxford.

Grandcolas, P. & D'Haese, C. (2004). The origin of a 'true' worker caste in termites: phylogenetic evidence is not decisive. *Journal of Evolutionary Biology* **15**, 885–888.

Grassé, P. P. & Noirot, C. (1947). Le polymorphisme social du termite a cou jaune (*Kalotermes flavicollis* F.) Les faux-ouvriers ou pseudergates et les mues regressive. *Comptes Rendus de Academie des Sciences* **214**, 219–221.

Gross, M. R. (1996). Alternative reproductive strategies and tactics: diversity within sexes. *Trends in Ecology and Evolution* **11**, 92–98.

Guidugli, K. R., Nascimento, A. M., Amdam, G. V., Barchuk, A. R., Omholt, S. W., Simões, Z. L. P. & Hartfelder, K. (2005*a*). Vitellogenin regulates hormonal dynamics in the worker caste of a eusocial insect. *FEBS Letters* **579**, 4961–4965.

Guidugli, K. R., Piulachs, M. D., Belles, X., Lourenco, A. P. & Simoes, Z. L. P. (2005*b*). Vitellogenin expression in queen ovaries and in larvae of both sexes of *Apis mellifera. Archives of Insect Biochemistry and Physiology* **59**, 211–218.

Hamilton, W. D. & May, R. M. (1977). Dispersal in stable habitats. *Nature* **269**, 578–581.

Hartfelder, K. (2000). Arthropoda - Insecta: Caste differentiation. In *Progress in Developmental Endocrinology*, vol. X part B. *Reproductive Biology of Invertebrates* (ed. A. Dorn), pp. 185–204. Wiley, Chichester.

HARTFELDER, K. & EMLEN, D. J. (2005). Endocrine control of insect polyphenism. In *Comprehensive Insect Molecular Science*, vol. 3 (ed. L. I. Gilbert, K. Iatrou and S. Gill), pp. 651–703. Elsevier, Oxford.

HAVERTY, M. I. (1977). The proportion of soldiers in termite colonies: a list and a bibliography (Isoptera). *Sociobiology* **2**, 199–216.

HAVERTY, M. I. (1979). Soldier production and maintenance of soldier proportions in laboratory experimental groups of *Coptotermes formosanus* Shiraki. *Insectes Sociaux* **26**, 69–84.

HAVERTY, M. I. & HOWARD, R. W. (1981). Production of soldiers and maintenance of soldier proportions by laboratory experimental groups of *Reticulitermes flavipes* (Kollar) and *Reticulitermes virginicus* (Banks) (Isoptera: Rhinotermitidae). *Insectes Sociaux* **28**, 32–39.

HAYASHI, Y., LO, N., MIYATA, H. & KITADE, O. (2007). Sex-linked genetic influence on caste determination in a termite. *Science* **318**, 985–987.

HELVIG, C., KOENER, J. F., UNNIHATHAN, G. C. & FEYEREISEN, R. (2004). CYP15A1, the cytochrome P450 that catalyzes epoxidation of methylfarnesoate to juvenile hormone III in cockroach corpora allata. *Proceedings of the National Academy of Sciences of the United States of America* **101**, 4024–4029.

HEMING, B. S. (2003). *Insect Development and Evolution*. Cornell University Press, Ithaca and London.

HENRICH, V. C. (2005). The ecdysteroid receptor. In *Comprehensive Insect Molecular Science*, vol. 3 (ed. L. I. Gilbert, K. Iatrou and S. Gill), pp. 243–285. Elsevier, Oxford.

HOWARD, R. W. & HAVERTY, M. I. (1979). Termites and juvenile hormone analogues: a review of methodology and observed effects. *Sociobiology* **4**, 269–278.

HOWSE, P. E. (1968). On the division of labour in the primitive termite *Zootermopsis nevadensis* (Hagen). *Insectes Sociaux* **15**, 45–50.

HRDY, I., KULDOVA, J., HANUS, R. & WIMMER, Z. (2006). Juvenile hormone III, hydroprene and a juvenogen as soldier caste differentiation regulators in three *Reticulitermes* species: potential of juvenile hormone analogues in termite control. *Pest Management Science* **62**, 848–854.

INWARD, D., BECCALONI, G. & EGGLETON, P. (2007*a*). Death of an order: a comprehensive molecular phylogenetic study confirms that termites are eusocial cockroaches. *Biology Letters* **3**, 331–335.

INWARD, D. J. G., VOGLER, A. P. & EGGLETON, P. (2007*b*). A comprehensive phylogenetic analysis of termites (Isoptera) illuminates key aspects of their evolutionary biology. *Molecular Phylogenetics and Evolution* **44**, 953–967.

JONES, G., JONES, D., TEAL, P., SAPA, A. & WOZNIAK, M. (2006). The retinoid-X receptor ortholog, ultraspiracle, binds with nanomolar affinity to an endogenous morphogenetic ligand. *FEBS Journal* **273**, 4983–4996.

JONES, G. & SHARP, P. A. (1997). Ultraspiracle: An invertebrate nuclear receptor for juvenile hormones. *Proceedings of the National Academy of Sciences of the United States of America* **94**, 13499–13503.

KAWANISHI, C. Y. (1975). Embryonic development of the drywood termite *Cryptotermes brevis. Hawaii Agricultural Experimental Station, College of Agriculture, UNiversity of Hawaii, Technical Bulletin* **95**, 35 pp.

KOKKO, H. & EKMAN, J. (2002). Delayed dispersal as a route to breeding: Territorial inheritance, safe havens, and ecological constraints. *American Naturalist* **160**, 462–484.

KORB, J. (2005). Regulation of sexual development in termites: mutilation, pheromonal manipulation or honest signal? *Naturwissenschaften* **92**, 45–49.

KORB, J. (2007*a*). Termites. *Current Biology* **17**, R995–999.

KORB, J. (2007*b*). Workers of a drywood termite do not work. *Frontiers in Zoology* **4**, 7 pp.

KORB, J. (2008). The ecology of social evolution in termites. In *Ecology of social evolution* (ed. J. Korb and J. Heinze), pp. 151–174. Springer, Heidelberg.

KORB, J. & FUCHS, A. (2006). Termites and mites - adaptive behavioural responses to infestation? *Behaviour* **143**, 891–907.

KORB, J. & KATRANTZIS, S. (2004). Influence of environmental conditions on the expression of the sexual dispersal phenotype in a lower termite: implications for the evolution of workers in termites. *Evolution & Development* **6**, 342–352.

KORB, J. & LENZ, M. (2004). Reproductive decision-making in the termite, *Cryptotermes secundus* (Kalotermitidae), under variable food conditions. *Behavioral Ecology* **15**, 390–395.

KORB, J., ROUX, E. A. & LENZ, M. (2003). Proximate factors influencing soldier development in the basal termite *Cryptotermes secundus* (Hill). *Insectes Sociaux* **50**, 299–303.

KORB, J. & SCHMIDINGER, S. (2004). Help or disperse? Cooperation in termites influenced by food conditions. *Behavioral Ecology and Sociobiology* **56**, 89–95.

KORB, J. & SCHNEIDER, K. (2007). Does kin structure explain the occurrence of workers in a lower termite? *Evolutionary Ecology* **21**, 817–828.

KOSHIKAWA, S., CORNETTE, R., HOJO, M., MAEKAWA, K., MATSUMOTO, T. & MIURA, T. (2005). Screening of genes expressed in developing mandibles during soldier differentiation in the termite *Hodotermopsis sjostedti*. *FEBS Letters* **579**, 1365–1370.

KOSHIKAWA, S., MATSUMOTO, T. & MIURA, T. (2002). Morphometric changes during soldier differentiation of the damp-wood termite *Hodotermopsis japonica* (Isoptera, Termopsidae). *Insectes Sociaux* **49**, 245–250.

KUCHARSKI, R., MALESZKA, J., FORET, S. & MALESZKA, R. (2008). Nutritional control of reproductive status in honeybees via DNA methylation. *Science* **319**, 1827–1830.

LA FAGE, J. P. & NUTTING, W. L. (1978). Nutrient dynamics of termites. In *Production ecology of ants and termites* (ed. M. V. Brian), pp. 165–232. Cambridge University Press, Cambridge.

LAFONT, R., DAUPHIN -VILLEMANT, C., WARREN, J. T. & REES, H. (2005). Ecdysteroid chemistry and biochemistry. In *Comprehensive Insect Molecular Science*, vol. 3 (ed. L. I. Gilbert, K. Iatrou and S. Gill), pp. 125–195. Elsevier, Oxford.

LANZREIN, B. (1974). Influence of a juvenile hormone analog on vitellogenin synthesis and oogenesis in larvae of *Nauphoeta cinerea*. *Journal of Insect Physiology* **20**, 1871–1885.

LANZREIN, B., GENTINETTA, V., ABEGGLEN, H., BAKER, F. C., MILLER, C. A. & SCHOOLEY, D. A. (1985*a*). Titers of ecdysone, 20-hydroxyecdysone and juvenile hormone III throughout the life cycle of a hemimetabolous insect, the ovoviviparous cockroach *Nauphoeta cinerea*. *Experientia* **41**, 913–917.

LANZREIN, B., GENTINETTA, V. & FEHR, R. (1985*b*). Titres of juvenile hormone and ecdysteroids in reproduction and eggs of *Macrotermes michaelseni*: Relation to caste determination. In *Caste Differentiation in Social Insects* (ed. J. A. L. Watson, B. M. Okot-Kotber and C. Noirot), pp. 307–327. Pergamon Press, Oxford.

LEFEUVE, P. & BORDEREAU, C. (1984). Soldier formation regulated by a primer pheromone from the soldier frontal gland in a higher termite, *Nasutitermes lujae*. *Proceedings of the National Academy of Sciences of the United States of America* **81**, 7665–7668.

LELIS, A. T. & EVERAERTS, C. (1993). Effects of juvenile-hormone analogs upon soldier differentiation in the termite *Reticulitermes* *santonensis* (Rhinotermitidae, Heterotermitinae). *Journal of Morphology* **217**, 239–261.

Lenz, M. (1976). The dependence of hormone effects in caste determination on external factors. In *Phase and Caste Determination in Insects. Endocrine Aspects* (ed. M. Lüscher), pp. 73–90. Pergamon Press, Oxford.

Lenz, M. (1994). Food resources, colony growth and caste development in wood-feeding termites. In *Nourishment and Evolution in Insect Societies* (ed. J. Hunt and C. A. Nalepa), pp. 159–210. Westview Press, Boulder.

Liénard, M. A., Lassance, J.-M. X. S., Paulmier, I., Picimbon, J.-F. & Löfstedt, C. (2006). Differential expression of cytochrome c oxidase subunit III gene in castes of the termite *Reticulitermes santonensis*. *Journal of Insect Physiology* **52**, 551–557.

Liu, Y. X., Henderson, G., Mao, L. X. & Laine, R. A. (2005*a*). Effects of temperature and nutrition on juvenile hormone titers of *Coptotermes formosanus* (Isoptera: Rhinotermitidae). *Annals of the Entomological Society of America* **98**, 732–737.

Liu, Y. X., Henderson, G., Mao, L. X. & Laine, R. A. (2005*b*). Seasonal variation of juvenile hormone titers of the formosan subterranean termite, *Coptotermes formosanus* (Rhinotermitidae). *Environmental Entomology* **34**, 557–562.

Lo, N., Engel, M. S., Cameron, S., Nalepa, C. A., Tokuda, G., Grimaldi, D., Kitade, O., Krishna, K., Klass, K. D., Maekawa, K., Miura, T. & Thompson, G. J. (2007). Save Isoptera: A comment on Inward et al. *Biology Letters* **3**, 562–563.

Longhurst, C., Johnson, R. A. & Wood, T. G. (1978). Predation by *Megaponera foetens* (Fabr.) (Hymenoptera: Formicidae) on the termites in the Nigerian Southern Guinea savanna. *Oecologia* **32**, 101–107.

Lüscher, M. (1958). Über die Entstehung der Soldaten bei Termiten. *Revue Suisse Zoologie* **65**, 372–377.

Lüscher, M. (1964). Die spezifische Wirkung männlicher und weiblicher Ersatzgeschlechtstiere auf die Entstehung von Geschlechtstieren bei der Termite *Kalotermes flavicollis* (Fab.). *Insectes Sociaux* **11**, 79–90.

Lüscher, M. (1969). Die Bedeutung des Juvenilhormons für die Differenzierung der Soldaten bei der Termite *Kalotermes flavicollis*. In *Proceedings VI. Congress of IUSSI*, pp. 165–170, Bern.

Lüscher, M. (1972). Environmental control of juvenile hormone (JH) secretion and caste differentiation in termites. *General and Comparative Endocrinology* Suppl. 3, 509–504.

Lüscher, M. (1974). Kasten und Kastendifferenzierung bei niederen Termiten. In *Sozialpolymorphismus bei Insekten* (ed. G. H. Schmidt), pp. 694–739. Wissenschaftliche Verlagsgesellschaft, Stuttgart.

MacWhinnie, S. G. B., Allee, J. P., Nelson, C. A., Riddiford, L. M., Truman, J. W. & Champlin, D. T. (2005). The role of nutrition in creation of the eye imaginal disc and initiation of metamorphosis in *Manduca sexta*. *Developmental Biology* **285**, 285–297.

Mao, L. X., Henderson, G., Liu, Y. X. & Laine, R. A. (2005). Formosan subterranean termite (Isoptera: Rhinotermitidae) soldiers regulate juvenile hormone levels and caste differentiation in workers. *Annals of the Entomological Society of America* **98**, 340–345.

Marais, E. N. (1937). *The Soul of the White Ant*. Methuen, London

Martin, D., Maestro, J. L., Cruz, J., Mane-Padros, D. & Belles, X. (2006). RNAi studies reveal a conserved role for RXR in molting in the cockroach *Blattella germanica*. *Journal of Insect Physiology* **52**, 410–416.

Miller, E. M. (1942). The problem of castes and caste differentiation in *Prorhinotermes simplex* (Hagen). *Bulletin of the University of Miami* **15**, 3–27.

Minelli, A., Brena, C., Deflorian, G., Maruzzo, D. & Fusco, G. (2006). From embryo to adult - beyond the conventional periodization of arthropod development. *Development, Genes and Evolution* **216**, 373–383.

Mirth, C., Truman, J. W. & Riddiford, L. M. (2005). The role of the prothoracic gland in determining critical weight for metamorphosis in *Drosophila melanogaster. Current Biology* **15**, 1–12.

Miura, T. (2001). Morphogenesis and gene expression in the soldier-caste differentiation of termites. *Insectes Sociaux* **48**, 216–223.

Miura, T. (2005). Developmental regulation of caste-specific characters in social-insect polyphenism. *Evolution & Development* **7**, 122–129.

Miura, T., Kamikouchi, A., Sawata, M., Takeuchi, H., Natori, S., Kubo, T. & Matsumoto, T. (1999). Soldier caste-specific gene expression in the mandibular glands of *Hodotermopsis japonica* (Isoptera: Termopsidae). *Proceedings of the National Academy of Sciences of the United States of America* **96**, 13874–13879.

Miura, T., Koshikawa, S., Machida, M. & Matsumoto, T. (2004). Comparative studies on alate wing formation in two related species of rotten-wood termites: *Hodotermopsis sjostedti* and *Zootermopsis nevadensis* (Isoptera, Termopsidae). *Insectes Sociaux* **51**, 247–252.

Miura, T. & Matsumoto, T. (2000). Soldier morphogenesis in a nasute termite: discovery of a disk-like structure forming a soldier nasus. *Proceedings of the Royal Society of London, Series B* **267**, 1185–1189.

Moran, N. A. (1992). The evolutionary maintenance of alternative phenotypes. *American Naturalist* **139**, 971–989.

Müller, C. B., Williams, I. S. & Hardie, J. (2001). The role of nutrition, crowding and interspecific interactions in the development of winged aphids. *Ecological Entomology* **26**, 330–340.

Mundall, E. C., Tobe, S. S. & Stay, B. (1979). Induction of vitellogenin and growth of implanted oocytes in male cockroaches. *Nature* **282**, 97–98.

Myles, T. G. (1988). Resource inheritance in social evolution from termite to man. In *Ecology of Social Behavior* (ed. C. N. Slobodchikoff), pp. 379–425. Academic Press, New York.

Myles, T. G. (1999). Review of secondary reproduction in termites (Insecta: Isoptera) with comments on its role in termite ecology and social evolution. *Sociobiology* **33**, 1–91.

Nalepa, C. A. (1994). Nourishment and the origin of termite eusociality. In *Nourishment and Evolution in Insect Societies* (ed. J. H. Hunt and C. A. Nalepa), pp. 57–104. Westview Press, Boulder.

Nalepa, C. A. & Bandi, C. (2000). Characterizing the ancestors: paedomorphosis and termite evolution. In *Termites: Evolution, Sociality, Symbioses, Ecology* (ed. T. Abe, L. D. E. Bignel and M. Higashi), pp. 53–76. Kluwer Academic Press, Dordrecht.

Nijhout, H. F. (1994). *Insect Hormones*. Princeton University Press, Princeton.

Noirot, C. (1990). Sexual castes and reproductive strategies in termites. In *Social Insects - an Evolutionary Approach to Caste and Reproduction* (ed. W. Engels), pp. 5–35. Springer Verlag, Heidelberg.

Noirot, C. (1991). Caste differentiation in Isoptera - basic features, role of pheromones. *Ethology, Ecology & Evolution*, 3–7.

Noirot, C. & Darlington, J. P. E. C. (2000). Termite nests: architecture, regulation and defence. In *Termites: Evolution, Sociality, Symbioses, Ecology.* (ed. T. Abe, D. E. Bignell and M. Higashi), pp. 121–140. Kluwer Academic Press, Dordrecht.

Nutting. (1969). Flight and colony foundation. In *Biology of Termites*, vol. 1 (ed. K. Krishna and F. M. Weesner), pp. 233–282. Academic Press, New York.

Oakeshott, J. G., Claudianos, C., Campbell, P. M., Newcomb, R. D. & Russell, R. J. (2005). Biochemical genetics and genomics of insect esterases. In *Comprehensive Molecular Insect Science*, vol. 5 (ed. L. I. Gilbert, K. Iatrou and S. S. Gill), pp. 309–381. Elsevier, San Diego.

Okot-Kotber, B. M. (1983). Ecdysteroid levels associated with epidermal events during worker and soldier differentiation in *Macrotermes michaelseni* (Isoptera: Macrotermitinae). *General and Comparative Endocrinology* **52**, 409–417.

Okot-Kotber, B. M. & Prestwich, G. D. (1991*a*). Identification of a juvenile-hormone binding protein in the castes of the termite, *Reticulitermes flavipes*, by photoaffinity labeling. *Insect Biochemistry* **21**, 775–784.

Okot-Kotber, B. M. & Prestwich, G. D. (1991*b*). Juvenile hormone binding proteins of termites detected by photoaffinity labeling: comparison of *Zootermopsis newadensis* with two rhinotermitids *Coptotermes formosanus* and *Reticulitermes flavipes*. *Archives of Insect Biochemistry and Physiology* **17**, 119–128.

Okot-Kotber, B. M., Prestwich, G. D., Strambi, A. & Strambi, C. (1993). Changes on morphogenetic hormone titers in isolated workers of the termite *Reticulitermes flavipes* (Kollar). *General and Comparative Endocrinology* **90**, 290–295.

Oster, G. F. & Wilson, E. O. (1978). *Caste and Ecology of Social Insects*. Princeton University Press, Princeton.

Park, Y. I. & Raina, A. K. (2005). Regulation of juvenile hormone titers by soldiers in the Formosan subterranean termite, *Coptotermes formosanus*. *Journal of Insect Physiology* **51**, 385–391.

Parmentier, D. (2006). *Developmental Flexibility and Evolution of the Worker Caste in Termites*. PhD thesis, Université Libre de Bruxelles, Bruxelles.

Parmentier, D. & Roisin, Y. (2003). Caste morphology and development in *Termitogeton nr. planus* (Insects, Isoptera, Rhinotermitidae). *Journal of Morphology* **255**, 69–79.

Patel, A., Fondrk, M. K., Kaftanoglu, O., Emore, C., Hunt, G., Frederick, K. & Amdam, G. V. (2007). The Making of a queen: TOR pathway is a key player in diphenic caste development. *PLoS One* **2** (6), e509.

Peeters, C. & Higashi, S. (1989). Reproductive dominance controlled by mutilation in the queenless ant *Diacamma australe*. *Naturwissenschaften* **76**, 177–180.

Raikhel, A. S., Brown, M. R. & Bellés, X. (2005). Hormonal control of reproducive processes. In *Comprehensive Molecular Insect Science*, vol. 3 (ed. L. I. Gilbert, K. Iatrou and S. S. Gill), pp. 433–491. Elsevier, Oxford.

Ramaswamy, K., Peeters, C., Yuvana, S. P., Varghese, T., Pradeep, H. D., Dietemann, V., Karpakakunjaram, V., Cobb, M. & Gadagkar, R. (2004). Social mutilation in the Ponerine ant *Diacamma*: cues originate in the victims. *Insectes Sociaux* **51**, 410–413.

Riddiford, L. M. (1994). Cellular and molecular actions of juvenile hormone. I. General considerations and premetamorphic actions. *Advances in Insect Physiology* **24**, 213–274.

Riddiford, L. M. (1996). Juvenile hormone: the status of its "status quo" action. *Archives of Insect Biochemistry and Physiology* **32**, 271–286.

Roisin, Y. (1994). Intragroup conflicts and the evolution of sterile castes in termites. *American Naturalist* **143**, 751–765.

Roisin, Y. (1999). Philopatric reproduction, a prime mover in the evolution of eusociality? *Insectes Sociaux* **46**, 297–305.

Roisin, Y. (2000). Diversity and evolution of caste patterns. In *Termites: Evolution, Sociality, Symbioses, Ecology* (ed. T. Abe, D. E. Bignell and M. Higashi), pp. 95–120. Kluwer Academic Publishers, Dordrecht.

Roisin, Y. (2006). Life history, life types and caste evolution in termites. In *Life Cycles in Social Insects: Behaviour, Ecology and Evolution* (ed. V. E. Kipyatkov), pp. 85–95. St. Petersburg University Press, St. Peterburg.

Roisin, Y. & Lenz, M. (1999). Caste developmental pathways in colonies of *Coptotermes lacteus* (Froggatt) headed by primary reproductives (Isoptera, Rhinotermitidae). *Insectes Sociaux* **46**, 273–280.

Roisin, Y. & Parmentier, D. (2006). Foraging by termites without workers: implications for the evolution of castes and life types. In *XV International Congress of IUSSI*, pp. 101, Washington, D.C.

Rosengaus, R. B. & Traniello, J. F. A. (1993). Temporal polyethism in incipient colonies of the primitive termite *Zootermopsis angusticollis*: a single multiage caste. *Journal of Insect Behavior* **6**, 237–252.

Roth, L. M. (1981). Introduction. In *The American Cockroach* (ed. W. J. Bell and K. G. Adiyodi), pp. 1–14. Chapman and Hall, London.

Roux, E. A. & Korb, J. (2004). Evolution of eusociality and the soldier caste in termites: a validation of the intrinsic benefit hypothesis. *Journal of Evolutionary Biology* **6**, 342–352.

Scharf, M. E., Ratliff, C. R., Wu-Scharf, D., Zhou, X. G., Pittendrigh, B. R. & Bennett, G. W. (2005*a*). Effects of juvenile hormone III on *Reticulitermes flavipes*: changes in hemolymph protein composition and gene expression. *Insect Biochemistry and Molecular Biology* **35**, 207–215.

Scharf, M. E., Wu-Scharf, D., Pittendrigh, B. R. & Bennett, G. W. (2003). Caste and development-associated gene expression in a lower termite. *Genome Biology* **4**, R:62.

Scharf, M. E., Wu-Scharf, D., Zhou, X., Pittendrigh, B. R. & Bennett, G. W. (2005*b*). Gene expression profiles among immature and adult reproductive castes of the termite *Reticulitermes flavipes*. *Insect Molecular Biology* **14**, 31–44.

Schlichting, C. D. & Pigliucci, M. (1998). *Phenotypic Evolution - a Reaction Norm Perspective*. Sinauer Associates, Sunderland, Massachusetts.

Shellman-Reeve, J. S. (1997). The spectrum of eusociality in termites. In *The Evolution of Social Behaviour in Insects and Arachnids* (ed. J. C. Choe and B. J. Crespi), pp. 52–93. Cambridge University Press, Cambridge.

Springhetti, A. (1969). Influenza dei reali sulla differenziazione dei soldati di *Kalotermes flavicollis* Fabr. (Isoptera). *Proceedings of the 6th Congress of IUSSI, Bern*, 267–273.

Stay, B. & Tobe, S. S. (2007). The role of allatostatins in juvenile hormone synthesis in insects and crustaceans. *Annual Review of Entomology* **52**, 277–299.

Stay, B., Woodhead, A. P., Joshi, S. & Tobe, S. S. (1991). Allatostatins, neuropeptide inhibitors of juvenile hormone biosynthesis in brain and corpora allata of cockroaches, *Diploptera punctata*. In *Insect Neuropeptides* (ed. J. J. Menn, T. J. Kelly and E. P. Masler), pp. 164–176. American Chemical Society, Washington, D.C.

Striebel, H. (1960). Zur Embryonalentwicklung der Termiten. *Acta Tropica* **17**, 193–260

Tawfik, A. I., Kellner, R., Hoffmann, K. H. & Lorenz, M. W. (2006). Purification, characterisation and titre of the haemolymph juvenile hormone binding proteins from *Schistocerca gregaria* and *Gryllus bimaculatus*. **52**, 255–268.

The Honey Bee Genome Sequencing Consortium (2006). Insights into social insects from the genome of the honeybee *Apis mellifera*. *Nature* **443**, 931–949.

Thompson, G. J., Kitade, O., Lo, N. & Crozier, R. H. (2000). Phylogenetic evidence for a single, ancestral origin of a 'true' worker caste in termites. *Journal of Evolutionary Biology* **13**, 869–881.

Thompson, G. J., Kitade, O., Lo, N. & Crozier, R. H. (2004). On the origin of termite workers: weighing up the phylogenetic evidence. *Journal of Evolutionary Biology* **17**, 217–220.

Thorne, B. L. (1997). Evolution of eusociality in termites. *Annual Review of Ecology and Systematics* **28**, 27–54.

Thorne, B. L., Breisch, N. L. & Muscedere, M. L. (2003). Evolution of eusociality and the soldier caste in termites: Influence of intraspecific competition and accelerated inheritance. *Proceedings of the National Academy of Sciences of the United States of America* **100**, 12808–12813.

Thorne, B. L., Grimaldi, D. A. & Krishna, K. (2000). Early fossil history of the termites. In *Termites: Evolution, Sociality, Symbioses, Ecology* (ed. T. Abe, D. E. Bignell and M. Higashi), pp. 77–94. Kluwer Academic Publishers, Dordrecht.

Thorne, B. L. & Traniello, J. F. A. (2003). Comparative social biology of basal taxa of ants and termites. *Annual Review of Entomology* **48**, 283–306.

Thummel, C. (1996). Flies on steroids - *Drosophila* metamorphosis and the mechanisms of steroid hormone action. *Trends in Genetics* **12**, 306–310.

Truman, J. W. & Riddiford, L. M. (1999). The origin of insect metamorphosis. *Nature* **401**, 447–452.

Truman, J. W. & Riddiford, L. M. (2002). Endocrine insights into the evolution of metamorphosis in insects. *Annual Review of Entomology* **47**, 467–500.

Watson, J. A. L. & Abbey, H. M. (1985). Development of neotenics in *Mastotermes darwiniensis* Froggatt: an alternative strategy. In *Caste Differentiation in Social Insects* (ed. J. A. L. Watson, B. M. Okot-Kotber and C. Noirot), pp. 107–124. Pergamon Press, Oxford.

Watson, J. A. L. & Sewell, J. J. (1985). Caste development in Mastotermes and Kalotermes: which is primitive? In *Caste Differentiation in Social Insects* (ed. J. A. L. Watson, B. M. Okot-Kotber and C. Noirot), pp. 27–40. Pergamon Press, Oxford.

Weil, T., Rehli, M. & Korb, J. (2007). Molecular basis for the reproductive division of labour in a lower termite. *BMC Genomics* **8**, e198.

West-Eberhard, M. J. (2003). *Developmental Plasticity and Evolution*. Oxford University Press, Oxford, New York.

Wheeler, D. E., Buck, N. & Evans, J. S. (2006). Expression of insulin pathway genes during the period of caste determination in the honey bee, *Apis mellifera*. *Insect Molecular Biology* **15**, 597–602.

Wilson, E. O. (1971). *The Insect Societies*. The Belknap Press of Harvard University Press, Cambridge, MA.

Woodhead, A. P., Stay, B., Seidel, S. L., Khan, M. A. & Tobe, S. S. (1989). Primary structure of 4 allatostatins - neuropeptide inhibitors of juvenile hormone synthesis. *Proceedings of the National Academy of Sciences of the United States of America* **86**, 5997–6001.

Xu, Y., Fang, F., Chu, Y., Jones, D. & Jones, G. (2002). Activation of transcription through the ligand-binding pocket of the orphan nuclear receptor ultraspiracle. *European Journal of Biochemistry* **269**, 6026–6036.

Yagi, K. J., Kwok, R., Chan, K. K., Setter, R. R., Myles, T. G., Tobe, S. S. & Stay, B. (2005). Phe-Gly-Leu-amide allatostatin in the termite *Reticulitermes flavipes*: Content in brain and corpus allatum and effect on juvenile hormone synthesis. *Journal of Insect Physiology* **51**, 357–365.

Zera, A. J. (2003). The endocrine regulation of wing polymorphism in insects: State of the art, recent surprises, and future directions. *Integrative and Comparative Biology* **43**, 607–616.

Zhou, X., Faith, M. O. & Scharf, M. E. (2006*a*). Social exploitation of hexamerin: RNAi reveals a major caste-regulatory factor in termites. *Proceedings of the National Academy of Sciences of the United States of America* **103**, 4499–4504.

Zhou, X., Song, C., Grzymala, T. L., Oi, F. M. & Scharf, M. E. (2007*a*). Juvenile hormone and colony conditions differentially influence cytochrome P450 gene expression in the termite *Reticulitermes flavipes*. *Insect Molecular Biology* **15**, 749–761.

Zhou, X., Tarver, M. R., Bennett, G. W., Oi, F. M. & Scharf, M. E. (2006*b*). Two hexamerin genes from the termite *Reticulitermes flavipes*: Sequence, expression, and proposed functions in caste regulation. *Gene* **376**, 47–58.

Zhou, X., Tarver, M. R. & Scharf, M. E. (2007*b*). Hexamerin-based regulation of juvenile hormone-dependent gene expression underlies phenotypic plasticity in a social insect. *Development* **134**, 601–610.

Zimmermann, R. B. (1983). Sibling manipulation and indirect fitness in termites. *Behavioral Ecology and Sociobiology* **12**, 143–145.