



**HAL**  
open science

# The foraging gene as a modulator of division of labour in social insects

Christophe Lucas, Yehuda Ben-Shahar

► **To cite this version:**

Christophe Lucas, Yehuda Ben-Shahar. The foraging gene as a modulator of division of labour in social insects. *Journal of Neurogenetics*, 2021, pp.1-11. 10.1080/01677063.2021.1940173 . hal-03266302

**HAL Id: hal-03266302**

**<https://hal.science/hal-03266302>**

Submitted on 12 Nov 2021

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

1 **The *foraging* gene as a modulator of division of labour in social insects**

2 Christophe Lucas<sup>a</sup> and Yehuda Ben-Shahar<sup>b</sup>

3 *<sup>a</sup>Institut de Recherche sur la Biologie de l’Insecte (UMR7261), CNRS – University of Tours,*  
4 *Tours, France; <sup>b</sup>Department of Biology, Washington University in St. Louis, St. Louis MO*  
5 *63130, USA*

6 [<sup>a</sup>christophe.lucas@univ-tours.fr](mailto:christophe.lucas@univ-tours.fr)

7 [<sup>b</sup>benshahary@wustl.edu](mailto:benshahary@wustl.edu)

## 8 **The *foraging* gene as a modulator of division of labour in social insects**

### 9 **Abstract**

10 The social ants, bees, wasps, and termites include some of the most ecologically-successful  
11 groups of animal species. Their dominance in most terrestrial environments is attributed to their  
12 social lifestyle, which enable their colonies to exploit environmental resources with remarkable  
13 efficiency. One key attribute of social insect colonies is the division of labour that emerges  
14 among the sterile workers, which represent the majority of colony members. Studies of the  
15 mechanisms that drive division of labour systems across diverse social species have provided  
16 fundamental insights into the developmental, physiological, molecular, and genomic processes  
17 that regulate sociality, and the possible genetic routes that may have led to its evolution from a  
18 solitary ancestor. Here we specifically discuss the conserved role of the *foraging* gene, which  
19 encodes a cGMP-dependent protein kinase (PKG). Originally identified as a behaviourally  
20 polymorphic gene that drives alternative foraging strategies in the fruit fly *Drosophila*  
21 *melanogaster*, changes in *foraging* expression and kinase activity were later shown to play a key  
22 role in the division of labour in diverse social insect species as well. In particular, *foraging*  
23 appears to regulate worker transitions between behavioural tasks and specific behavioural traits  
24 associated with morphological castes. Although the specific neuroethological role of *foraging* in  
25 the insect brain remains mostly unknown, studies in genetically tractable insect species indicate  
26 that PKG signalling plays a conserved role in the neuronal plasticity of sensory, cognitive and  
27 motor functions, which underlie behaviours relevant to division of labour, including appetitive  
28 learning, aggression, stress response, phototaxis, and the response to pheromones.

29

30 **Keywords (5 to 6):** *foraging* gene; sociality; insects; behaviour; division of  
31 labour

32

33 **Subject classification codes:** review

## 34 **Introduction**

35           Animal social grouping is common because it increases the fitness of individuals by,  
36 for example, maximizing their access to resources and providing improved defences against  
37 predators (Wilson, 1987). Some of the most derived forms of sociality are exhibited by  
38 species of eusocial insects in the orders Hymenoptera (ants, bees, wasps) and Isoptera  
39 (termites), which together are estimated to represent more than 30% of the global animal  
40 biomass (Hölldobler & Wilson, 2009). All social insect colonies exhibit reproductive division  
41 of labour, in which majority of a colony is comprised of sterile female workers and just one or  
42 few reproductive individuals (the “queen” in social Hymenoptera and the “royal couple” in  
43 termites) (Oster & Wilson, 1978). The sterile workers, in turn, self-organize to become  
44 specialized in different tasks that are necessary for the growth and maintenance of the colony  
45 such as foraging, brood care and colony defence (Corona et al., 2016; Gordon, 2016; Leitner  
46 & Dornhaus, 2019). Although division of labour among workers is common across many  
47 different social insect taxa, different species show varying levels of individual specializations.  
48 At one extreme, workers might exhibit almost no task specialization, while at the other  
49 extreme workers might differentiate into developmentally-determined morphological castes  
50 (Oster & Wilson, 1978) (Figure 1).

51           Despite the overwhelming complexity associated with insect sociality and the  
52 regulation of division of labour, the evolutionary emergence of division of labour systems  
53 across different social insect species most likely depended on the co-option of existing  
54 sensory pathways, and associated modulatory neural circuits, to generate differential  
55 behavioural syndromes across individuals that share a common social and physical  
56 environment. Subsequently, although sensory stimuli for all tasks are simultaneously present  
57 in the colony, division of labour systems maintain differential behavioural response thresholds  
58 to task specific cues in different individuals (Figure 1). The highly-tuned network of semi-  
59 autonomous individuals provides a robust, systems-level modulation of task allocation across

60 time and space based on the overall needs of the colony (Buttstedt et al., 2016; Haydak, 1970;  
61 Krista K Ingram, 2002; Kamakura, 2011; Romain Libbrecht & Keller, 2013; Nygaard et al.,  
62 2011; Patel et al., 1960; Schwander & Keller, 2008; Schwander et al., 2010; Wheeler, 1991).  
63 Phylogenetic studies indicate that division of labour systems likely evolved multiple times  
64 across, and within, different lineages of the social hymenopterans (Gordon, 2016; Rueffler et  
65 al., 2012). Therefore, division of labour in colonies of social insects has emerged as an  
66 excellent model for understanding how animal social systems have evolved and are regulated  
67 at the physiological, molecular, and cellular levels (Kapheim et al., 2015; Robinson et al.,  
68 2005; Smith et al., 2008; Toth & Robinson, 2007; A. L. Toth et al., 2010; Whitfield et al.,  
69 2006). Transcriptomic analyses of gene expression in the brains of workers that are engaged  
70 in specific tasks, for example, have indicated that division of labour depends on broad and  
71 complex changes in many different neuronal signalling pathways across different parts of the  
72 brain and other tissues (Figure 1). This appears to occur via differential promoter activity,  
73 regulatory non-coding miRNAs, and epigenetic processes (Cash et al., 2005; Greenberg et al.,  
74 2012; K. M. Kapheim et al., 2020; Lattorff & Moritz, 2013; Simola et al., 2016; Sinha et al.,  
75 2006; Toth et al., 2007; Whitfield et al., 2006). To date, changes in the expression levels of  
76 several genes have been associated with division of labour across diverse social insect  
77 species. However, only a few have been studied in any depth. The highly conserved cGMP-  
78 dependent protein kinase gene *foraging* (*for*), in particular, has been well-studied for its role  
79 in regulating division of labour. Previous work has established a causal association between  
80 *for* and task specialization, which is conserved across multiple independently-evolved  
81 lineages. Here we review the role of *for*, first identified as a behaviourally polymorphic gene  
82 in *Drosophila* associated with larval and adult foraging decisions (J. S. de Belle et al., 1989;  
83 K. A. Osborne et al., 1997), as a conserved signalling pathway that has been co-opted for the  
84 regulation of division of labour across diverse social insect species.

## 85 **The study of sociality in the era of molecular genetics**

86 Over 50 years ago, the pioneering ethologist Niko Tinbergen published his seminal  
87 paper entitled ‘On aims and methods of ethology’ (Burkhardt, 2014; Tinbergen, 1963). With  
88 brilliant simplicity, Tinbergen argued that if biologists want to really understand “behaviour”  
89 then they need to ask the following four questions (rephrased): 1) What is the studied trait  
90 good for (its impact on fitness)? 2) How does it develop during the lifetime of an individual  
91 (development/ontogeny)? 3) How did it evolve over the history of the species (trait  
92 phylogeny)? 4) How does it work (mechanism/causation)? The emergence of modern  
93 neuroscience and neurogenetics followed in the footsteps of ethology by providing a  
94 mechanistic framework as a powerful approach to the design of behavioural studies and their  
95 interpretation in the context of brain functions (Tinbergen question #4).

96 However, modern neuroscience research is now largely framed in the context of causation and  
97 mechanism (Tinbergen question #4). Consequently, the diversity of animal species used for  
98 basic neurogenetic research has, historically, been reduced to just a few genetically tractable  
99 species that have sequenced genomes, including the roundworm *Caenorhabditis elegans*  
100 (Bargmann, 1998), the fruit fly *Drosophila melanogaster* (Bellen et al., 2010), the zebrafish  
101 *Danio rerio* (Stewart et al., 2014), and the laboratory mouse *Mus musculus* (Lehner, 2013).

102 While there is no doubt that these models have been instrumental in the phenomenal progress  
103 made in cellular and molecular neuroscience over the past three decades, the decline in model  
104 diversity is clearly an unfortunate side effect, and has negatively impacted our ability to  
105 understand behaviour in ecologically and evolutionary relevant contexts (Fitzpatrick et al.,  
106 2005; Walton et al., 2020).

107 Yet, recent advances in genetic and genomic approaches have enabled investigators to adopt  
108 modern molecular approaches for understanding the behaviour of a wider assortment of  
109 animal species in the lab and in their natural environments. Some of the first non-drosophilid

110 insects for which genomic and molecular tools for studying behaviour have made a  
111 tremendous impact are the social insects, particularly in the several independent social clades  
112 within the order Hymenoptera. One of the best established model species for sociogenomic  
113 and genetic studies is the European honey bee *Apis mellifera*. As the most dominant  
114 domesticated insect, the long tradition of beekeeping has provided investigators with a rich  
115 knowledge of its behaviour and physiology, including well-established techniques for  
116 manipulating colony demography and colony genetics via artificial inseminations (Page et al.,  
117 2002; Vanengelsdorp & Meixner, 2010). Not surprisingly, the economic importance of the  
118 honey bee has led to the development of diverse molecular and genetic resources, including a  
119 relatively well-annotated genome, high-resolution population genetics, and successful genome  
120 transformations (Ament et al., 2010; Ben-Shahar, 2014; Ben-Shahar et al., 2004; Chen et al.,  
121 2021; Foret et al., 2009; Sinha et al., 2006; Y. Wang et al., 2006; Weinstock et al., 2006;  
122 Whitfield et al., 2006; Whitfield et al., 2003). Subsequently, the progress in analysing honey  
123 bee behaviour with molecular and genetic tools has paved the way for similar breakthroughs  
124 in many additional social insect species (Karen M. Kapheim et al., 2020; Kapheim et al.,  
125 2015; R. Libbrecht et al., 2013; Sadd et al., 2015; A. L. Toth et al., 2010). Nevertheless, the  
126 genetic mechanisms by which specific genes and neuronal signalling pathways regulate the  
127 complex behaviour of social insects had remained mostly a mystery until the discovery of the  
128 gene *foraging* (*for*), a cGMP-dependent protein kinase (PKG), and its role in the regulation of  
129 behavioural plasticity in colonies of social insects (Figure 1D). Subsequent studies of this  
130 gene of in the context of division of labour have provided some important mechanistic  
131 insights into animal sociality.

### 132 **The role of *foraging* in regulating division of labour in social insect colonies**

133 The *foraging* (*for*) gene encodes a cGMP-dependent protein kinase (K. Osborne et al., 1997),  
134 a highly conserved enzyme present in all animal genomes sequenced to date (Anreiter &

135 Sokolowski, 2019; Lucas, Kornfein, et al., 2010). The *for* gene was first identified as a  
136 naturally polymorphic locus that regulates larval foraging behaviour in *Drosophila* (J.S. de  
137 Belle & Sokolowski, 1987; Sokolowski, 1980), and was later identified as a gene that encodes  
138 one of the two cGMP-dependent protein kinases (PKG) encoded by the *Drosophila* genome  
139 (K. Osborne et al., 1997). The highly conserved PKG family is broadly expressed in both  
140 neuronal and non-neuronal tissues, and has been implicated in numerous cellular and  
141 physiological functions via its kinase activity (Dason et al., 2019; Douglas et al., 2005;  
142 Francis et al., 2010; Lohmann et al., 1997; X. Wang & Robinson, 1997). In *Drosophila*, *for* is  
143 expressed in diverse tissues, including the peripheral (PNS) and central (CNS) nervous  
144 systems of both larvae and adults (Anreiter et al., 2017). Several genetic studies have revealed  
145 that the natural polymorphism in *for* contributes to diverse sensory functions, including the  
146 visual, olfaction, and gustatory pathways (Lin et al., 1992). In the CNS, *for* is expressed in the  
147 Kenyon cells of the mushroom bodies, the central complex, and the antennal lobes. It has  
148 been implicated in regulating neuronal and behavioural plasticity in diverse cognitive and  
149 motor functions such as neuronal sensitization and habituation, learning and memory, and  
150 sleep (Eddison et al., 2012; Engel et al., 2000; Scheiner et al., 2004). In addition to regulating  
151 behaviour via neuronal functions, *for* also indirectly contributes to feeding and foraging  
152 behaviours via its role in metabolic plasticity and the response to stress (Dason et al., 2020;  
153 Kaun et al., 2008; Kaun & Sokolowski, 2009; Kent et al., 2009).

154 The role of the cGMP-PKG signalling axis in regulating feeding behaviours is conserved in  
155 other animal species. For example, cGMP-signalling has been shown to regulate the response  
156 threshold of insects to pheromones (Boekhoff et al., 1993; Ziegelberger et al., 1990), and to  
157 influence olfactory communication in rats (Kroner et al., 1996; Moon et al., 1998). In the  
158 worm *Caenorhabditis elegans*, the PKG-ortholog *egl-4* plays a role in foraging, olfaction,  
159 sleep, body size and lifespan (Fujiwara et al., 2002; L'Etoile et al., 2002; Raizen et al., 2008).



160 Yet, in spite of its broad role in regulating different forms of behavioural plasticity in diverse  
161 animal species, the identities of the specific neuronal substrates modulated by *for* in the  
162 context of foraging behaviour and social insect division of labour remain mostly unknown.  
163 However, studies in genetically tractable species provide some mechanistic insights into how  
164 PKG signalling might be regulating neuronal processes associated with division labour. For  
165 example, studies in the mouse, indicate that PKG activity can have broad effects on long-term  
166 neuronal processes and behavioural plasticity via diverse downstream effectors. Neuronal  
167 PKG activity can modulate long-term potentiation (LTP) in the mammalian brain via a direct  
168 modulation of cAMP-phosphodiesterases, which regulate cAMP levels and synaptic plasticity  
169 (Bollen et al., 2014). In addition, studies of *Egl-4*, the ortholog of *for* in the worm *C. elegans*,  
170 has shown that sensory-induced neuronal plasticity within the chemosensory system depends  
171 on the transition of activated PKG to the nucleus, which leads to changes in neuronal gene  
172 expression via the modulation of heterochromatin binding proteins (Lee et al., 2010).  
173 Together, these examples illustrate the broad role *for* and related kinase genes play in  
174 regulating neuronal plasticity and organismal behaviour via diverse molecular and cellular  
175 mechanisms in the peripheral and central nervous systems of invertebrates and vertebrates.

### 176 **The *foraging* gene in social bees**

177 The realization that *for* plays a key role in regulating feeding-related behavioural  
178 plasticity in *Drosophila* suggested that the phylogenetically conserved PKG signalling pathway  
179 is also likely to be driving similar foraging-related decisions in other insect species, including  
180 in the regulation of foraging vs. brood care behaviour in social insect division of labour.  
181 Recently, *for* has also been implicated in the regulation of social networks in *Drosophila*  
182 (Alwash et al., 2021). By using transcriptomics and gene candidate approaches (Fitzpatrick et  
183 al., 2005), multiple studies have identified several key genes that play a role in regulating  
184 division of labour in honey bees including the acetylcholine esterase (*AchE*) gene (Shapira et

185 al., 2001), the *foraging* (*for*) gene (Ben-Shahar et al., 2003; Ben-Shahar et al., 2002), and the  
186 manganese transporter *malvolio* (Ben-Shahar et al., 2004). In terms of how *for* exerts its action  
187 in honey bees, follow-up studies have identified an increase in brain expression levels of *for*  
188 just prior to the initiation of foraging, which suggests that its activity may serve as a trigger for  
189 the transition from in-hive behaviours to foraging outside (Heylen et al., 2008). Furthermore,  
190 QTL analysis of foraging-related traits in honey bee populations has identified an association  
191 with QTLs in the *for* locus, suggesting that in addition to its role at the developmental and  
192 physiological timescales, *for* may play a role in shaping division of labour at the evolutionary  
193 timescale as well (Ruppell et al., 2004).

194         The discovery of *for* as a putative key regulator of division of labour in the honey bee  
195 was followed by studies investigating its role in regulating division of labour across the social  
196 hymenopteran phylogeny. It was shown that changes in *for* expression are also associated  
197 with division of labour in other species of social bees, including the Asian honey bee *A.*  
198 *cerana* (Ma et al., 2018), as well as different bumblebee species (Kodaira et al., 2009;  
199 Tobback et al., 2011). Interestingly, in contrast to the reported increased *for* expression in the  
200 brains of honey bee foragers, the expression of *for* in association with the size-dependent  
201 regulation of division of labour in bumblebees was variable, indicating higher *for* expression  
202 in *Bombus terrestris* foragers but lower in brains of *B. ignitus* foragers (Couvillon et al., 2010;  
203 Kodaira et al., 2009; Shpigler et al., 2013). Together, these studies suggest that the functional  
204 relationship between *for* activity in the brain and the behavioural states that define the  
205 probability of performing specific tasks by individual bees is complex and likely involve both  
206 central and peripheral neuronal pathways.

## 207 **The *foraging* gene in ants**

208         In ants, *for* has been shown to be involved in age-related division of labour, as in  
209 honey bees (Krista K Ingram et al., 2016; K. K. Ingram et al., 2005; Lucas et al., 2015;  
210 Manfredini et al., 2014; Oettler et al., 2015). However, in contrast to social bees, which  
211 primarily regulate division of labour on a physiological timescale, studies of *for* in ants  
212 revealed its role in regulating polyethisms in association with both age-dependent and  
213 developmentally-regulated morphological castes. For example, colonies of the ant *Pheidole*  
214 *pallidula* have two morphologically-distinct workers (Passera, 1985), each associated with  
215 specific behavioural repertoires. The majors have a large head with large mandibles and are  
216 mainly involved in the defence of the colony and processing of large food items, while the  
217 minors are smaller and are mainly involved in brood care and foraging. This morphological  
218 and behavioural division of labour was associated with changes in brain activity; minors had  
219 lower brain PKG activity than majors (Lucas & Sokolowski, 2009). In addition, although  
220 minors and majors typically perform distinct roles in the colony, when confronted with  
221 changing needs of the colony, majors may assist minors in foraging activities and minors may  
222 help with defence tasks (Aarab, 1991). Accordingly, experimental manipulations showed that  
223 majors that respond to foraging stimuli show minor-like, low brain PKG activity. Similarly,  
224 when participating in colony defence, minors showed a significant increase in brain PKG  
225 activity to major-like levels (Lucas & Sokolowski, 2009). Moreover, pharmacological  
226 treatment of minors with the PKG activator 8-Br-cGMP induced brain PKG activity, and led  
227 to an increase in defence and decrease in foraging behaviours (Lucas & Sokolowski, 2009),  
228 thus establishing a causal association between PKG activity and worker behavioural  
229 plasticity. The *foraging* gene in *P. pallidula* therefore appears to modulate both foraging  
230 behaviour and defence behaviour (Anreiter & Sokolowski, 2019; Lucas, Hughson, et al.,  
231 2010; Lucas & Sokolowski, 2009) and, under conditions that require plastic task reallocation,  
232 can drive neuronal plasticity even in species with a robust morphological division of labour

233 system (Anreiter & Sokolowski, 2019; Lucas, Hughson, et al., 2010). These findings suggest  
234 that *for* exerts at least two levels of control over worker behaviour in ants: (1) Differences in  
235 baseline brain PKG activity are associated with caste polyethism at the developmental  
236 timescale; and (2) Plasticity in PKG activity allows colony-level responses to environmental  
237 changes at the physiological timescale. This link between defence and PKG activity has also  
238 been demonstrated in a field study, where feeding the ant *Allomerus octoarticulatus* with a  
239 PKG activator, increased defence behaviour of the ant-plant *Cordia nodosa* against  
240 herbivores, suggesting a molecular basis for ant-plant mutualism (Malé et al., 2017).

241 In another ant species, *Solenopsis invicta*, *foraging* gene expression in workers was  
242 influenced by colony demography, in particular whether the colony had one queen  
243 (monogynous) or multiple queens (polygynous) (Lucas et al., 2015). Workers from  
244 monogynous colonies of *S. invicta*, which are very territorial and aggressive (Chirino et al.,  
245 2012; Tschinkel, 2006), had higher brain expression levels of *for* relative to workers from the  
246 more docile polygynous colonies (Lucas et al., 2015). Altogether, these results suggest that  
247 the *for* gene might be involved in regulating differential levels of aggression associated with  
248 territorial foraging or caste polyethisms. Recently, *for* has been shown to regulate aggression  
249 in flies as well, providing further support to the idea that the role of *for* in regulating  
250 behavioural plasticity across distant species is conserved not only for foraging behaviour but  
251 may be also for defence behaviour (S. Wang & Sokolowski, 2017).

252 Moreover in the ant *S. invicta*, *for* expression level was shown to be associated with  
253 reproductive state (future queen vs. workers) and the number of future queens (monogynous  
254 vs. polygynous) (Lucas et al., 2015), suggesting that in addition to its role in regulating  
255 behavioural plasticity in workers, *for* also contributes to reproductive division of labour in *S.*  
256 *invicta*. Interestingly, variations in colony demography (monogynous vs. polygynous) exist in

257 several ant species, and are thought to confer colony-level benefits by increasing genetic  
258 diversity, and therefore, amplifying worker behavioural differences that underlie division of  
259 labour (Cronin et al., 2013; Krista K Ingram, 2002; Manfredini et al., 2014; Pearcy et al.,  
260 2004; Ross & Keller, 1995).

## 261 **The neuroethological context of the regulation of division of labour by *for***

262         Based on theoretical and empirical studies, one of the primary models for self-  
263 organized division of labour in social insect colonies is based on the idea that animals at  
264 different behavioural states exhibit different behavioural response thresholds to task-specific  
265 stimuli (Beshers & Fewell, 2001; Duarte et al., 2012). Consequently, although workers are  
266 exposed to many or all task-stimuli in the colony, the probability that an individual worker  
267 will respond to the stimulus for a particular task varies (Bonabeau et al., 1998; Detrain et al.,  
268 1999; Duarte et al., 2012; Leitner & Dornhaus, 2019). Subsequently, molecular, genetic, and  
269 neuroethological studies have suggested that the PKG activity of *for* regulates division of  
270 labour, at least in part, via the modulation of task-specific sensory thresholds (Ben-Shahar et  
271 al., 2003; George et al., 2020; Heylen et al., 2008; Thamm & Scheiner, 2014).

272         As a kinase, *for* is likely to have numerous protein targets in the insect brain (X. Wang  
273 & Robinson, 1997). To date, the specific protein targets that mediate its role in behavioural  
274 plasticity in general, and division of labour in particular, remain mostly elusive, however,  
275 studies in various species suggest that *for* may have targets within the sensory system. Studies  
276 in *Drosophila*, for example, have indicated that *for* contributes to sucrose responsiveness in  
277 adults (Scheiner et al., 2004) and the larval nociception threshold (Dason et al., 2020). Similar  
278 effects on sensory thresholds have also been identified in *C. elegans* and rodent models,  
279 further indicating a conserved role for cGMP-dependent signalling in modulating behavioural  
280 states via sensory functions (Gangadharan et al., 2017; Jaumann et al., 2012; Levy &  
281 Bargmann, 2020). Subsequently, several studies in the honey bee have indicated that *for*

282 activity plays a role in regulating the response threshold to light, indicating that increased  
283 PKG activity drives positive phototaxis in foragers (Ben-Shahar, 2005; Ben-Shahar et al.,  
284 2003). In addition, *for* has also been implicated in honey bee sucrose responsiveness and the  
285 response threshold to queen pheromones (Fussnecker et al., 2011; Thamm & Scheiner, 2014).  
286 Together, these studies suggest that *for* is likely to regulate division of labour via multimodal  
287 modulation of sensory functions.

288         In addition to its role in regulating sensory functions, *for* has been shown to contribute  
289 to both simple forms of neuronal plasticity, such as the habituation in the giant fiber escape  
290 circuit responsiveness to sugar (Engel et al., 2000; Scheiner et al., 2004), as well as more  
291 complex processes such as visual memories (Kuntz et al., 2017; Z. Wang et al., 2008) and  
292 associative learning (Kaun et al., 2007; Mery et al., 2007) in flies. Because division of labour  
293 is often associated with differential learning and memory capacities (Ben-Shahar & Robinson,  
294 2001; Ben-Shahar et al., 2000; Roussel et al., 2009), it has been hypothesized that *for* is also  
295 likely to also modulate division of labour via its action in central neuronal circuits associated  
296 with higher level cognitive functions (Ben-Shahar, 2005; Menzel et al., 2006). Interestingly,  
297 in *Pheidole* ants, the expression pattern of *for* in the brains of majors is specific to a subregion  
298 of the mushroom bodies, while both minor and major workers share its expression in several  
299 brain regions including the lobula, which plays a role in the integration of visual cues (Lucas  
300 & Sokolowski, 2009). Although it is still unknown how the action of *for* in this specific brain  
301 region is contributing to the behaviours exhibited by minors and majors, the well-established  
302 anatomical and physiological role of the mushroom bodies in regulating division of labour in  
303 diverse social insects suggest that *for*-dependent signalling in this brain region is important for  
304 cognitive processes associated with division of labour. This is further supported by the recent  
305 findings that *for* activity is associated with interindividual differences in the dance intensity of  
306 returning foragers (George et al., 2020), and the impact of cGMP pharmacological treatment

307 on spatial learning in honey bee foragers (Tsvetkov et al., 2019).

### 308 **The *foraging* gene in other social contexts**

309       The complex role of *for* in regulating division of labour in bee and ant species  
310 indicates that this gene is highly pleiotropic, regulating not only foraging but also many other  
311 behavioural aspects of division of labour and other sociality-related traits. The study of *for* in  
312 other social species, such as wasps and termites, will yield important discoveries for  
313 understanding how *for* regulates social behaviours across diverse insect taxa. For example, a  
314 study on the comparisons of brain gene expression between the social wasp *Polistes metricus*  
315 and the honey bee *Apis mellifera* allowed to identify common molecular roots for division of  
316 labour, demonstrating the importance of *for* across lineages (Amy L Toth et al., 2010). In the  
317 termite *Reticulitermes flavipes*, a recent study showed differences in mRNA expression  
318 depending on caste, temperature and photoperiod (Merchant et al., 2019), suggesting the  
319 possibility that the differential expression of *for* may very well be important for this social  
320 insect group as well. Future comparisons between termites and social hymenopteran will  
321 likely shed even more light on how *for* regulates division of labour across phylogenetically  
322 distant social systems (Hymenoptera/Isoptera) with distinct developmental processes (e.g.,  
323 holometabolism vs. hemimetabolism).

324       Although the precise origin of insect eusociality remains mostly unknown at the  
325 molecular level, it is clear that some key neuronal signalling pathways, such as *for*, contribute  
326 to social interactions between conspecifics in non-social insects as well. For example, flies  
327 with low *for* expression are more sensitive to social context in learning paradigms than those  
328 with high expression levels (Kohn et al., 2013). In addition, *for* has been shown to be  
329 associated with population density-dependent regulation of polyphenisms in some aphid  
330 species (Tarès et al., 2013), and between solitary and gregarious forms in the locust

331 *Schistocerca gregaria* (Lucas, Kornfein, et al., 2010; Rogers et al., 2003; Tobback et al.,  
332 2013).

### 333 **Concluding remarks**

334         The association between the PKG activity of the *foraging* gene and behavioural  
335 plasticity in response to changes in the social and biotic environments, especially in the  
336 context of feeding and foraging behaviours, is conserved across a broad range of animal  
337 species, from insects to humans (Anreiter & Sokolowski, 2019; Armstrong et al., 2010;  
338 Chardonnet et al., 2014; Kohlmeier et al., 2019; Lucas, Kornfein, et al., 2010; Malé et al.,  
339 2017; Merchant et al., 2019; Robertson & Sillar, 2009; Struk et al., 2019; Tarès et al., 2013;  
340 Tobback et al., 2013). Additionally, it remains unknown whether all or only some of the  
341 alternative transcripts transcribed by *for* in at least some social insect species play a role in  
342 regulating neuronal plasticity and behaviour in social insects (Lucas & Sokolowski, 2009;  
343 Merchant et al., 2019; NCBI, 2021). Furthermore, the specific molecular targets and neuronal  
344 signalling pathways modulated by *for* activity across the evolutionary, developmental, and  
345 physiological timescales remain a mystery. Nevertheless, recent developments in transposon-  
346 mediated transgenesis and *Cas9*/CRISPR-dependent genome editing in several social insect  
347 species (Ben-Shahar, 2014; Chen et al., 2021; Kohno et al., 2016; McAfee et al., 2019;  
348 Schulte et al., 2014; Tribble et al., 2017) will likely yield new insights into the molecular and  
349 cellular pathways that underlie the role of *for*, and other important signalling pathways, in the  
350 “social” brain.

### 351 **Acknowledgements**

352         We thank Jeffrey Dason, Ina Anreiter, and Chun-Fang Wu for their kind invitation to  
353 contribute to this special issue; Joël Meunier and Romain Libbrecht for the pictures of the  
354 ants; and Nicole Leitner for helpful comments on previous drafts of this manuscript.

355



356 **Disclosure statement**

357           The authors report no conflict of interests.

358 **Funding**

359           This work was supported by the European Union's Horizon 2020 research and  
360 innovation program under the grant agreement 773324, and APR-IR 2017-00117111 from  
361 Région Centre-Val de Loire to CL; and grants R21NS089834 from the NIH, and 1754264 and  
362 1707221 from the NSF to YB.

363

364 **ORCID**

365 Christophe Lucas [0000-0003-2839-7583](https://orcid.org/0000-0003-2839-7583)

366 Yehuda Ben-Shahar [0000-0002-2956-2926](https://orcid.org/0000-0002-2956-2926)

367

368 **References**

- 369 Aarab, A. (1991). *Polyéthisme, régulation sociale et éthogénèse chez les deux sous-castes*  
370 *morphologiques de la fourmi Pheidole pallidula*, Université Paris-Nord].
- 371 Alwash, N., Allen, A.M., Sokolowski, M.B, Levine, J. (2021) The *Drosophila melanogaster*  
372 *foraging* gene affects social networks. *Journal of Neurogenetics*.
- 373 Ament, S. A., Wang, Y., & Robinson, G. E. (2010). Nutritional regulation of division of labor  
374 in honey bees: toward a systems biology perspective. *Wiley Interdiscip Rev Syst Biol*  
375 *Med*, 2(5), 566-576. <https://doi.org/10.1002/wsbm.73>
- 376 Anreiter, I., Kramer, J. M., & Sokolowski, M. B. (2017). Epigenetic mechanisms modulate  
377 differences in *Drosophila* foraging behavior. *Proceedings of the National Academy of*  
378 *Sciences*, 114(47), 12518-12523.
- 379 Anreiter, I., & Sokolowski, M. B. (2019). The foraging gene and its behavioral effects:  
380 Pleiotropy and plasticity. *Annual Review of Genetics*, 53.
- 381 Armstrong, G. A., López-Guerrero, J. J., Dawson-Scully, K., Peña, F., & Robertson, R. M.  
382 (2010). Inhibition of protein kinase G activity protects neonatal mouse respiratory  
383 network from hyperthermic and hypoxic stress. *Brain Research*, 1311, 64-72.
- 384 Bargmann, C. I. (1998). Neurobiology of the *Caenorhabditis elegans* genome. *Science*,  
385 282(5396), 2028-2033. <http://www.ncbi.nlm.nih.gov/pubmed/9851919>
- 386 Bellen, H. J., Tong, C., & Tsuda, H. (2010). 100 years of *Drosophila* research and its impact  
387 on vertebrate neuroscience: a history lesson for the future [10.1038/nrn2839]. *Nat Rev*  
388 *Neurosci*, 11(7), 514-522. <http://dx.doi.org/10.1038/nrn2839>
- 389 Ben-Shahar, Y. (2005). The foraging gene, behavioral plasticity, and honeybee division of  
390 labor. *J Comp Physiol A Neuroethol Sens Neural Behav Physiol*, 191(11), 987-994.  
391 <https://doi.org/10.1007/s00359-005-0025-1>
- 392 Ben-Shahar, Y. (2014). A piggyBac route to transgenic honeybees. *Proc Natl Acad Sci U S A*,  
393 111(24), 8708-8709. <https://doi.org/10.1073/pnas.1407876111>
- 394 Ben-Shahar, Y., Dudek, N. L., & Robinson, G. E. (2004). Phenotypic deconstruction reveals  
395 involvement of manganese transporter malvolio in honey bee division of labor. *J Exp*  
396 *Biol*, 207(Pt 19), 3281-3288. <https://doi.org/10.1242/jeb.01151>
- 397 Ben-Shahar, Y., Leung, H. T., Pak, W. L., Sokolowski, M. B., & Robinson, G. E. (2003).  
398 cGMP-dependent changes in phototaxis: a possible role for the foraging gene in honey  
399 bee division of labor. *J Exp Biol*, 206(Pt 14), 2507-2515.  
400 <http://www.ncbi.nlm.nih.gov/pubmed/12796464>

401 Ben-Shahar, Y., Robichon, A., Sokolowski, M. B., & Robinson, G. E. (2002). Influence of  
402 gene action across different time scales on behavior. *Science*, 296(5568), 741-744.  
403 <https://doi.org/10.1126/science.1069911>

404 Ben-Shahar, Y., & Robinson, G. E. (2001). Satiation differentially affects performance in a  
405 learning assay by nurse and forager honey bees. *J Comp Physiol A Neuroethol Sens*  
406 *Neural Behav Physiol*, 187(11), 891-899.  
407 <http://www.ncbi.nlm.nih.gov/pubmed/11866187>

408 Ben-Shahar, Y., Thompson, C. K., Hartz, S. M., Smith, B. H., & Robinson, G. E. (2000).  
409 Differences in performance on a reversal learning test and division of labor in honey bee  
410 colonies. *Animal Cognition*, 3(3), 119-125. <https://doi.org/10.1007/s100710000068>

411 Beshers, S. N., & Fewell, J. H. (2001). Models of division of labor in social insects. *Annual*  
412 *Review of Entomology*, 46, 413-440. <Go to ISI>://000167877300015

413 Boekhoff, I., Seifert, E., Goggerle, S., Lindemann, M., Kruger, B. W., & Breer, H. (1993).  
414 Pheromone-induced second-messenger signalling in insect antennae. *Insect. Biochem.*  
415 *Molec. Biol.*, 23, 757-762.

416 Bollen, E., Puzzo, D., Rutten, K., Privitera, L., De Vry, J., Vanmierlo, T., Kenis, G., Palmeri,  
417 A., D'Hooge, R., Balschun, D., Steinbusch, H. M., Blokland, A., & Prickaerts, J. (2014).  
418 Improved long-term memory via enhancing cGMP-PKG signaling requires cAMP-PKA  
419 signaling. *Neuropsychopharmacology*, 39(11), 2497-2505.  
420 <https://doi.org/10.1038/npp.2014.106>

421 Bonabeau, E., Theraulaz, G., & Deneubourg, J.-L. (1998). Fixed response thresholds and the  
422 regulation of division of labor in insect societies. *Bulletin of Mathematical Biology*,  
423 60(4), 753-807. <https://doi.org/10.1006/bulm.1998.0041>

424 Burkhardt, R. W. (2014). Tribute to Tinbergen: Putting Niko Tinbergen's 'Four Questions' in  
425 Historical Context. *Ethology*, 120(3), 215-223. <https://doi.org/10.1111/eth.12200>

426 Buttstedt, A., Ihling, C. H., Pietzsch, M., & Moritz, R. F. (2016). Royalactin is not a royal  
427 making of a queen. *Nature*, 537(7621), E10.

428 Cash, A. C., Whitfield, C. W., Ismail, N., & Robinson, G. E. (2005). Behavior and the limits  
429 of genomic plasticity: power and replicability in microarray analysis of honeybee  
430 brains. *Genes Brain Behav*, 4(4), 267-271. [https://doi.org/10.1111/j.1601-](https://doi.org/10.1111/j.1601-183X.2005.00131.x)  
431 [183X.2005.00131.x](https://doi.org/10.1111/j.1601-183X.2005.00131.x)

432 Chardonnet, F., Capdevielle-Dulac, C., Chouquet, B., Joly, N., Harry, M., Le Ru, B., Silvain,  
433 J.-F., & Kaiser, L. (2014). Food searching behaviour of a Lepidoptera pest species is

434 modulated by the foraging gene polymorphism. *Journal of Experimental Biology*,  
435 217(19), 3465-3473.

436 Chen, Z., Traniello, I. M., Rana, S., Cash-Ahmed, A. C., Sankey, A. L., Yang, C., &  
437 Robinson, G. E. (2021). Neurodevelopmental and transcriptomic effects of  
438 CRISPR/Cas9-induced somatic orco mutation in honey bees. *J Neurogenet*, 1-13.  
439 <https://doi.org/10.1080/01677063.2021.1887173>

440 Chirino, M. G., Gilbert, L. E., & Folgarait, P. J. (2012). Behavioral discrimination between  
441 monogyne and polygyne red fire ants (Hymenoptera: Formicidae) in their native range.  
442 *Annals of the Entomological Society of America*, 105(5), 740-745.

443 Corona, M., Libbrecht, R., & Wheeler, D. E. (2016). Molecular mechanisms of phenotypic  
444 plasticity in social insects. *Current Opinion in Insect Science*, 13, 55-60.  
445 <https://doi.org/https://doi.org/10.1016/j.cois.2015.12.003>

446 Couvillon, M. J., Jandt, J. M., Duong, N., & Dornhaus, A. (2010). Ontogeny of worker body  
447 size distribution in bumble bee (*Bombus impatiens*) colonies. *Ecol Entomol*, 35(4), 424-  
448 435. <https://doi.org/10.1111/j.1365-2311.2010.01198.x>

449 Cronin, A. L., Molet, M., Doums, C., Monnin, T., & Peeters, C. (2013). Recurrent evolution  
450 of dependent colony foundation across eusocial insects. *Annual Review of Entomology*,  
451 58, 37-55.

452 Dason, J. S., Allen, A. M., Vasquez, O. E., & Sokolowski, M. B. (2019). Distinct functions of  
453 a cGMP-dependent protein kinase in nerve terminal growth and synaptic vesicle  
454 cycling. *Journal of Cell Science*, 132(7), 1-12.

455 Dason, J. S., Cheung, A., Anreiter, I., Montemurri, V. A., Allen, A. M., & Sokolowski, M. B.  
456 (2020). *Drosophila melanogaster* foraging regulates a nociceptive-like escape behavior  
457 through a developmentally plastic sensory circuit. *Proceedings of the National Academy*  
458 *of Sciences*, 117(38), 23286-23291.

459 de Belle, J. S., Hilliker, A. J., & Sokolowski, M. B. (1989). Genetic localization of foraging  
460 (for): a major gene for larval behavior in *Drosophila melanogaster*. *Genetics*, 123(1),  
461 157-163.  
462 [http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Cita](http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=2509284)  
463 [tion&list\\_uids=2509284](http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=2509284)

464 de Belle, J. S., & Sokolowski, M. B. (1987). Heredity of rover/sitter: alternative foraging  
465 strategies of *Drosophila melanogaster* larvae. *Heredity*, 59, 73-83.

466 Detrain, C., Deneubourg, J. L., & Pasteels, J. M. (1999). *Information processing in social*  
467 *insects*. Berlin: Birkhäuser Verlag.

468 Douglas, S. J., Dawson-Scully, K., & Sokolowski, M. B. (2005). The neurogenetics and  
469 evolution of food-related behaviour. *Trends in Neurosciences*, 28(12), 644-652.

470 Duarte, A., Pen, I., Keller, L., & Weissing, F. J. (2012). Evolution of self-organized division  
471 of labor in a response threshold model. *Behavioral Ecology and Sociobiology*, 66(6),  
472 947-957. <https://doi.org/10.1007/s00265-012-1343-2>

473 Eddison, M., Belay, A. T., Sokolowski, M. B., & Heberlein, U. (2012). A genetic screen for  
474 olfactory habituation mutations in *Drosophila*: analysis of novel foraging alleles and an  
475 underlying neural circuit. *PLoS ONE*, 7(12), e51684.  
476 <https://doi.org/10.1371/journal.pone.0051684>

477 Engel, J. E., Xie, X. J., Sokolowski, M. B., & Wu, C. F. (2000). A cGMP-dependent protein  
478 kinase gene, foraging, modifies habituation-like response decrement of the giant fiber  
479 escape circuit in *Drosophila*. *Learn Mem*, 7(5), 341-352.  
480 [http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Cita](http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=11040266)  
481 [tion&list\\_uids=11040266](http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=11040266)

482 Fitzpatrick, M. J., Ben-Shahar, Y., Smid, H. M., Vet, L. E., Robinson, G. E., & Sokolowski,  
483 M. B. (2005). Candidate genes for behavioural ecology. *Trends Ecol Evol*, 20(2), 96-  
484 104. <https://doi.org/10.1016/j.tree.2004.11.017>

485 Foret, S., Kucharski, R., Pittelkow, Y., Lockett, G. A., & Maleszka, R. (2009). Epigenetic  
486 regulation of the honey bee transcriptome: unravelling the nature of methylated genes.  
487 *BMC Genomics*, 10, 472. <https://doi.org/1471-2164-10-472> [pii]  
488 10.1186/1471-2164-10-472

489 Francis, S. H., Busch, J. L., Corbin, J. D., & Sibley, D. (2010). cGMP-dependent protein  
490 kinases and cGMP phosphodiesterases in nitric oxide and cGMP action. *Pharmacol*  
491 *Rev*, 62(3), 525-563. <https://doi.org/10.1124/pr.110.002907>

492 Fujiwara, M., Sengupta, P., & McIntire, S. L. (2002). Regulation of body size and behavioral  
493 state of *C. elegans* by sensory perception and the EGL-4 cGMP-dependent protein  
494 kinase. *Neuron*, 36(6), 1091-1102. <Go to ISI>://000180011600012

495 Fussnecker, B. L., McKenzie, A. M., & Grozinger, C. M. (2011). cGMP modulates responses  
496 to queen mandibular pheromone in worker honey bees. *J Comp Physiol A Neuroethol*  
497 *Sens Neural Behav Physiol*, 197(9), 939-948. [https://doi.org/10.1007/s00359-011-0654-](https://doi.org/10.1007/s00359-011-0654-5)  
498 [5](https://doi.org/10.1007/s00359-011-0654-5)

499 Gangadharan, V., Wang, X., & Luo, C. (2017). Cyclic GMP-dependent protein kinase-I  
500 localized in nociceptors modulates nociceptive cortical neuronal activity and pain

501 hypersensitivity. *Mol Pain*, 13, 1744806917701743.  
502 <https://doi.org/10.1177/1744806917701743>

503 George, E. A., Bröger, A.-K., Thamm, M., Brockmann, A., & Scheiner, R. (2020). Inter-  
504 individual variation in honey bee dance intensity correlates with expression of the  
505 foraging gene. *Genes, Brain and Behavior*, 19(1), e12592.  
506 <https://doi.org/https://doi.org/10.1111/gbb.12592>

507 Gordon, D. M. (2016). From division of labor to the collective behavior of social insects.  
508 *Behav Ecol Sociobiol*, 70, 1101-1108. <https://doi.org/10.1007/s00265-015-2045-3>

509 Greenberg, J. K., Xia, J., Zhou, X., Thatcher, S. R., Gu, X., Ament, S. A., Newman, T. C.,  
510 Green, P. J., Zhang, W., Robinson, G. E., & Ben-Shahar, Y. (2012). Behavioral  
511 plasticity in honey bees is associated with differences in brain microRNA transcriptome.  
512 *Genes Brain Behav*, 11(6), 660-670. <https://doi.org/10.1111/j.1601-183X.2012.00782.x>

513 Haydak, M. H. (1970). Honey bee nutrition. *Annual Review of Entomology*, 15(1), 143-156.

514 Heylen, K., Gobin, B., Billen, J., Hu, T. T., Arckens, L., & Huybrechts, R. (2008). Amfor  
515 expression in the honeybee brain: a trigger mechanism for nurse-forager transition. *J*  
516 *Insect Physiol*, 54(10-11), 1400-1403. [https://doi.org/S0022-1910\(08\)00154-6](https://doi.org/S0022-1910(08)00154-6) [pii]  
517 10.1016/j.jinsphys.2008.07.015

518 Hölldobler, B., & Wilson, E. O. (2009). *The superorganism : the beauty, elegance, and*  
519 *strangeness of insect societies*. W.W. Norton & Company.

520 Ingram, K. K. (2002). Plasticity in queen number and social structure in the invasive  
521 Argentine ant (*Linepithema humile*). *Evolution*, 56(10), 2008-2016.

522 Ingram, K. K., Gordon, D. M., Friedman, D. A., Greene, M., Kahler, J., & Peteru, S. (2016).  
523 Context-dependent expression of the foraging gene in field colonies of ants: the  
524 interacting roles of age, environment and task. *Proceedings of the Royal Society B:*  
525 *Biological Sciences*, 283(1837), 20160841.

526 Ingram, K. K., Oefner, P., & Gordon, D. M. (2005). Task-specific expression of the *foraging*  
527 gene in harvester ants. *Molecular Ecology*, 14(3), 813-818. <Go to  
528 [ISI>://000227132300013](https://doi.org/10.1111/j.1365-3113.2004.03001.x)

529 Jaumann, M., Dettling, J., Gubelt, M., Zimmermann, U., Gerling, A., Paquet-Durand, F., Feil,  
530 S., Wolpert, S., Franz, C., Varakina, K., Xiong, H., Brandt, N., Kuhn, S., Geisler, H. S.,  
531 Rohbock, K., Ruth, P., Schlossmann, J., Hutter, J., Sandner, P., Feil, R., Engel, J.,  
532 Knipper, M., & Ruttiger, L. (2012). cGMP-Prkg1 signaling and Pde5 inhibition shelter  
533 cochlear hair cells and hearing function. *Nat Med*, 18(2), 252-259.  
534 <https://doi.org/10.1038/nm.2634>

535 Kamakura, M. (2011). Royalactin induces queen differentiation in honeybees. *Nature*,  
536 473(7348), 478-483.

537 Kapheim, K. M., Jones, B. M., Sovik, E., Stolle, E., Waterhouse, R. M., Bloch, G., & Ben-  
538 Shahr, Y. (2020). Brain microRNAs among social and solitary bees. *R Soc Open Sci*,  
539 7(7), 200517. <https://doi.org/10.1098/rsos.200517>

540 Kapheim, K. M., Pan, H., Li, C., Salzberg, S. L., Puiu, D., Magoc, T., Robertson, H. M.,  
541 Hudson, M. E., Venkat, A., Fischman, B. J., Hernandez, A., Yandell, M., Ence, D.,  
542 Holt, C., Yocum, G. D., Kemp, W. P., Bosch, J., Waterhouse, R. M., Zdobnov, E. M.,  
543 Stolle, E., Kraus, F. B., Helbing, S., Moritz, R. F., Glastad, K. M., Hunt, B. G.,  
544 Goodisman, M. A., Hauser, F., Grimmelikhuijzen, C. J., Pinheiro, D. G., Nunes, F. M.,  
545 Soares, M. P., Tanaka, E. D., Simoes, Z. L., Hartfelder, K., Evans, J. D., Barribeau, S.  
546 M., Johnson, R. M., Massey, J. H., Southey, B. R., Hasselmann, M., Hamacher, D.,  
547 Biewer, M., Kent, C. F., Zayed, A., Blatti, C., 3rd, Sinha, S., Johnston, J. S., Hanrahan,  
548 S. J., Kocher, S. D., Wang, J., Robinson, G. E., & Zhang, G. (2015). Social evolution.  
549 Genomic signatures of evolutionary transitions from solitary to group living. *Science*,  
550 348(6239), 1139-1143. <https://doi.org/10.1126/science.aaa4788>

551 Kaun, K. R., Chakaborty-Chatterjee, M., & Sokolowski, M. B. (2008). Natural variation in  
552 plasticity of glucose homeostasis and food intake. *J Exp Biol*, 211(Pt 19), 3160-3166.  
553 <https://doi.org/211/19/3160> [pii]  
554 10.1242/jeb.010124

555 Kaun, K. R., Hendel, T., Gerber, B., & Sokolowski, M. B. (2007). Natural variation in  
556 *Drosophila* larval reward learning and memory due to a cGMP-dependent protein  
557 kinase. *Learn Mem*, 14(5), 342-349. <https://doi.org/14/5/342> [pii]  
558 10.1101/lm.505807

559 Kaun, K. R., & Sokolowski, M. B. (2009). cGMP-dependent protein kinase: linking foraging  
560 to energy homeostasis [Research Support, N.I.H., Extramural  
561 Review]. *Genome*, 52(1), 1-7. <https://doi.org/10.1139/G08-090>

562 Kent, C. F., Daskalchuk, T., Cook, L., Sokolowski, M. B., & Greenspan, R. J. (2009). The  
563 *Drosophila foraging* gene mediates adult plasticity and gene-environment interactions  
564 in behaviour, metabolites and gene expression in response to food deprivation. *PLoS*  
565 *Genet.*, 5(8), e1000609. <https://doi.org/10.1371/journal.pgen.1000609>

566 Kodaira, Y., Ohtsuki, H., Yokoyama, J., & Kawata, M. (2009). Size-dependent foraging gene  
567 expression and behavioral caste differentiation in *Bombus ignitus*. *BMC Res Notes*, 2,  
568 184. <https://doi.org/10.1186/1756-0500-2-184>



569 Kohlmeier, P., Alleman, A. R., Libbrecht, R., Foitzik, S., & Feldmeyer, B. (2019). Gene  
570 expression is more strongly associated with behavioural specialization than with age or  
571 fertility in ant workers. *Molecular Ecology*, 28(3), 658-670.

572 Kohn, N. R., Reaume, C. J., Moreno, C., Burns, J. G., Sokolowski, M. B., & Mery, F. (2013).  
573 Social environment influences performance in a cognitive task in natural variants of the  
574 *foraging* gene. *PLoS ONE*, 8(12), e81272.

575 Kohno, H., Suenami, S., Takeuchi, H., Sasaki, T., & Kubo, T. (2016). Production of  
576 Knockout Mutants by CRISPR/Cas9 in the European Honeybee, *Apis mellifera* L.  
577 *Zoolog Sci*, 33(5), 505-512. <https://doi.org/10.2108/zs160043>

578 Kroner, C., Boekhoff, I., Lohmann, S. M., Genieser, H. G., & Breer, H. (1996). Regulation of  
579 olfactory signalling via cGMP-dependant protein kinases. *Trends Biochem. Sci.*, 22(8),  
580 307-312.

581 Kuntz, S., Poeck, B., & Strauss, R. (2017). Visual Working Memory Requires Permissive and  
582 Instructive NO/cGMP Signaling at Presynapses in the Drosophila Central Brain. *Curr*  
583 *Biol*, 27(5), 613-623. <https://doi.org/10.1016/j.cub.2016.12.056>

584 L'Etoile, N. D., Coburn, C. M., Eastham, J., Kistler, A., Gallegos, G., & Bargmann, C. I.  
585 (2002). The cyclic GMP-dependent protein kinase EGL-4 regulates olfactory adaptation  
586 in *C. elegans*. *Neuron*, 36(6), 1079-1089. <https://doi.org/S0896627302010668> [pii]

587 Lattorff, H. M., & Moritz, R. F. (2013). Genetic underpinnings of division of labor in the  
588 honeybee (*Apis mellifera*). *Trends Genet*, 29(11), 641-648.  
589 <https://doi.org/10.1016/j.tig.2013.08.002>

590 Lee, J. I., O'Halloran, D. M., Eastham-Anderson, J., Juang, B. T., Kaye, J. A., Scott Hamilton,  
591 O., Lesch, B., Goga, A., & L'Etoile, N. D. (2010). Nuclear entry of a cGMP-dependent  
592 kinase converts transient into long-lasting olfactory adaptation. *Proc Natl Acad Sci U S*  
593 *A*, 107(13), 6016-6021. <https://doi.org/10.1073/pnas.1000866107>

594 Lehner, B. (2013). Genotype to phenotype: lessons from model organisms for human  
595 genetics. *Nat Rev Genet*, 14(3), 168-178. <https://doi.org/10.1038/nrg3404>

596 Leitner, N., & Dornhaus, A. (2019). Dynamic task allocation: how and why do social insect  
597 workers take on new tasks? *Animal Behaviour*, 158, 47-63.  
598 <https://doi.org/https://doi.org/10.1016/j.anbehav.2019.09.021>

599 Levy, S., & Bargmann, C. I. (2020). An Adaptive-Threshold Mechanism for Odor Sensation  
600 and Animal Navigation. *Neuron*, 105(3), 534-548 e513.  
601 <https://doi.org/10.1016/j.neuron.2019.10.034>



602 Libbrecht, R., & Keller, L. (2013). Genetic compatibility affects division of labor in the  
603 Argentine ant *Linepithema humile*. *Evolution: International Journal of Organic*  
604 *Evolution*, 67(2), 517-524.

605 Libbrecht, R., Oxley, P. R., Kronauer, D. J., & Keller, L. (2013). Ant genomics sheds light on  
606 the molecular regulation of social organization. *Genome Biol*, 14(7), 212.  
607 <https://doi.org/10.1186/gb-2013-14-7-212>

608 Lin, M., Nairn, A. C., & Guggino, S. E. (1992). cGMP-dependant protein kinase regulation of  
609 a chloride channel in T84 cells. *Am. J. Physiol.*, 262(5 Pt 1), 1304-1312.

610 Lohmann, S. M., Vaandrager, A. B., Smolenski, A., Walter, U., & De Jonge, H. R. (1997).  
611 Distinct and specific functions of cGMP-dependent protein kinases. *Trends Biochem*  
612 *Sci*, 22(8), 307-312. [https://doi.org/10.1016/s0968-0004\(97\)01086-4](https://doi.org/10.1016/s0968-0004(97)01086-4)

613 Lucas, C., Hughson, B. N., & Sokolowski, M. B. (2010). Job switching in ants: role of a  
614 kinase. *Communicative and Integrative Biology*, 3(1), 6-8.  
615 <https://doi.org/10.4161/cib.3.1.9723>

616 Lucas, C., Kornfein, R., Chakaborty-Chatterjee, M., Schonfeld, J., Geva, N., Sokolowski, M.  
617 B., & Ayali, A. (2010). The locust *foraging* gene. *Archives of Insect Biochemistry and*  
618 *Physiology*, 74(1), 52-66. <https://doi.org/10.1002/arch.20363>

619 Lucas, C., Nicolas, M., & Keller, L. (2015). Expression of *foraging* and *Gp-9* are associated  
620 with social organization in the fire ant *Solenopsis invicta*. *Insect Molecular Biology*,  
621 24(1), 93-104.

622 Lucas, C., & Sokolowski, M. B. (2009). Molecular basis for changes in behavioral state in ant  
623 social behaviors. *Proceedings of the National Academy of Sciences of the United States*  
624 *of America*, 106(15), 6351-6356. <https://doi.org/doi:10.1073/pnas.0809463106>

625 Ma, W., Jiang, Y., Meng, J., Zhao, H., Song, H., & Shen, J. (2018). Expression  
626 Characterization and Localization of the foraging Gene in the Chinese Bee, *Apis cerana*  
627 *cerana* (Hymenoptera: Apidae). *J Insect Sci*, 18(2): 1-5.  
628 <https://doi.org/10.1093/jisesa/iey034>

629 Malé, P.-J. G., Turner, K. M., Doha, M., Anreiter, I., Allen, A. M., Sokolowski, M. B., &  
630 Frederickson, M. E. (2017). An ant–plant mutualism through the lens of cGMP-  
631 dependent kinase genes. *Proceedings of the Royal Society B: Biological Sciences*,  
632 284(1862), 20170896.

633 Manfredini, F., Lucas, C., Nicolas, M., Keller, L., Shoemaker, D., & Grozinger, C. M. (2014).  
634 Molecular and social regulation of worker division of labour in fire ants. *Molecular*  
635 *Ecology*, 23(3), 660-672.

636 McAfee, A., Pettis, J. S., Tarpy, D. R., & Foster, L. J. (2019). Feminizer and doublesex  
637 knock-outs cause honey bees to switch sexes. *PLoS Biol*, *17*(5), e3000256.  
638 <https://doi.org/10.1371/journal.pbio.3000256>

639 Menzel, R., Leboulle, G., & Eisenhardt, D. (2006). Small brains, bright minds. *Cell*, *124*(2),  
640 237-239. <https://doi.org/Doi> 10.1016/J.Cell.2006.01.011

641 Merchant, A., Song, D., Yang, X., Li, X., & Zhou, X. J. (2019). Candidate foraging gene  
642 orthologs in a lower termite, *Reticulitermes flavipes*. *Journal of Experimental Zoology*  
643 *Part B: Molecular and Developmental Evolution*, *334*(3), 168-177.

644 Mery, F., Belay, A. T., So, A. K., Sokolowski, M. B., & Kawecki, T. J. (2007). Natural  
645 polymorphism affecting learning and memory in *Drosophila*. *Proc Natl Acad Sci U S A*,  
646 *104*(32), 13051-13055. <https://doi.org/0702923104> [pii]  
647 10.1073/pnas.0702923104

648 Moon, C., Jaber, P., Otto-Bruc, A., Baehr, W., Palczewski, K., & Ronnet, G. V. (1998).  
649 Calcium-sensitive particulate guanylyl cyclase as a Modulator of cAMP in olfactory  
650 Receptor Neurons. *The J. of Neurosc.*, *18*(9), 3195-3205.

651 NCBI. Retrieved 9th April 2021 from [https://www.ncbi.nlm.nih.gov/protein/?term=cGMP-](https://www.ncbi.nlm.nih.gov/protein/?term=cGMP-dependent+protein+kinase%5BAll+Fields%5D+AND+(ant+%5BAll+Fields%5D+or+bee%5BAll+Fields%5D+or+termite%5BAll+Fields%5D+or+wasp%5BAll+Fields%5D+not+Nasonia+not+Amantichitinum+not+fungi+not+chordates+not+arachnids+not+blatella))  
652 [dependent+protein+kinase%5BAll+Fields%5D+AND+\(ant+%5BAll+Fields%5D+or+bee%5BAll+Fields%5D+or+termite%5BAll+Fields%5D+or+wasp%5BAll+Fields%5D+](https://www.ncbi.nlm.nih.gov/protein/?term=cGMP-dependent+protein+kinase%5BAll+Fields%5D+AND+(ant+%5BAll+Fields%5D+or+bee%5BAll+Fields%5D+or+termite%5BAll+Fields%5D+or+wasp%5BAll+Fields%5D+not+Nasonia+not+Amantichitinum+not+fungi+not+chordates+not+arachnids+not+blatella))  
653 [not+Nasonia+not+Amantichitinum+not+fungi+not+chordates+not+arachnids+not+blat](https://www.ncbi.nlm.nih.gov/protein/?term=cGMP-dependent+protein+kinase%5BAll+Fields%5D+AND+(ant+%5BAll+Fields%5D+or+bee%5BAll+Fields%5D+or+termite%5BAll+Fields%5D+or+wasp%5BAll+Fields%5D+not+Nasonia+not+Amantichitinum+not+fungi+not+chordates+not+arachnids+not+blatella))  
654 [ella\)](https://www.ncbi.nlm.nih.gov/protein/?term=cGMP-dependent+protein+kinase%5BAll+Fields%5D+AND+(ant+%5BAll+Fields%5D+or+bee%5BAll+Fields%5D+or+termite%5BAll+Fields%5D+or+wasp%5BAll+Fields%5D+not+Nasonia+not+Amantichitinum+not+fungi+not+chordates+not+arachnids+not+blatella)  
655

656 Nygaard, S., Zhang, G., Schjøtt, M., Li, C., Wurm, Y., Hu, H., Zhou, J., Ji, L., Qiu, F., &  
657 Rasmussen, M. (2011). The genome of the leaf-cutting ant *Acromyrmex echinatior*  
658 suggests key adaptations to advanced social life and fungus farming. *Genome Research*,  
659 *21*(8), 1339-1348.

660 Oettler, J., Nachtigal, A.-L., & Schrader, L. (2015). Expression of the foraging gene is  
661 associated with age polyethism, not task preference, in the ant *Cardiocondyla obscurior*.  
662 *PLoS ONE*, *10*(12), e0144699.

663 Osborne, K., Robichon, A., Burgess, E., Butland, S., Shaw, R., Coulthard, A., Pereira, H.,  
664 Greenspan, R., & Sokolowski, M. (1997). Natural behavior polymorphism due to a  
665 cGMP-dependent protein kinase of *Drosophila*. *Science*, *277*(5327), 834-836.

666 Oster, G. F., & Wilson, E. O. (1978). *Caste and ecology in the social insects*. Princeton, New  
667 Jersey: Princeton University Press.

668 Page, R. E., Jr., Gadau, J., & Beye, M. (2002). The emergence of hymenopteran genetics.  
669 *Genetics*, *160*(2), 375-379. <https://www.ncbi.nlm.nih.gov/pubmed/11861545>

670 Passera, L. (1985). Soldier determination in ants of the genus *Pheidole*. In J. A. L. Watson, B.  
671 M. Okot-Kotber, & C. Noirot (Eds.), *Caste differentiation in social insects* (pp. 331-  
672 346). Oxford: Pergamon Press.

673 Patel, N., Haydak, M., & Gochnauer, T. (1960). Electrophoretic components of the proteins in  
674 honeybee larval food. *Nature*, 186(4725), 633-634.

675 Percy, M., Aron, S., Doums, C., & Keller, L. (2004). Conditional use of sex and  
676 parthenogenesis for worker and queen production in ants. *Science*, 306(5702), 1780-  
677 1783.

678 Raizen, D. M., Zimmerman, J. E., Maycock, M. H., Ta, U. D., You, Y.-j., Sundaram, M. V.,  
679 & Pack, A. I. (2008). Lethargus is a *Caenorhabditis elegans* sleep-like state. *Nature*,  
680 451(7178), 569-572.

681 Robertson, R. M., & Sillar, K. T. (2009). The nitric oxide/cGMP pathway tunes the  
682 thermosensitivity of swimming motor patterns in *Xenopus laevis* tadpoles. *Journal of*  
683 *Neuroscience*, 29(44), 13945-13951.

684 Robinson, G. E., Grozinger, C. M., & Whitfield, C. W. (2005). Sociogenomics: social life in  
685 molecular terms. *Nat Rev Genet*, 6(4), 257-270. <https://doi.org/10.1038/nrg1575>

686 Rogers, S. M., Matheson, T., Despland, E., Dodgson, T., Burrows, M., & Simpson, S. J.  
687 (2003). Mechanosensory-induced behavioural gregarization in the desert locust  
688 *Schistocerca gregaria*. *Journal of Experimental Biology*, 206(Pt 22), 3991-4002.  
689 [http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Cita](http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=14555739)  
690 [tion&list\\_uids=14555739](http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=14555739)

691 Ross, K. G., & Keller, L. (1995). Joint influence of gene flow and selection on a  
692 reproductively important genetic polymorphism in the fire ant *Solenopsis invicta*. *The*  
693 *American Naturalist*, 146, 325-348.  
694 [http://my.unil.ch/serval/document/BIB\\_6181B2F1C88C.pdf](http://my.unil.ch/serval/document/BIB_6181B2F1C88C.pdf) or available through:  
695 Universite de Lausanne website [http://www.unil.ch/dee/page6998\\_en.html](http://www.unil.ch/dee/page6998_en.html)

696 Roussel, E., Carcaud, J., Sandoz, J.-C., & Giurfa, M. (2009). Reappraising Social Insect  
697 Behavior through Aversive Responsiveness and Learning. *PLoS ONE*, 4(1), e4197.  
698 <https://doi.org/10.1371/journal.pone.0004197>

699 Rueffler, C., Hermisson, J., & Wagner, G. P. (2012). Evolution of functional specialization  
700 and division of labor. *Proc Natl Acad Sci U S A*, 109(6), E326-335.  
701 <https://doi.org/10.1073/pnas.1110521109>

702 Ruppell, O., Pankiw, T., & Page, R. E., Jr. (2004). Pleiotropy, epistasis and new QTL: the  
703 genetic architecture of honey bee foraging behavior. *J Hered*, 95(6), 481-491.  
704 <https://doi.org/10.1093/jhered/esh072>

705 Sadd, B. M., Barribeau, S. M., Bloch, G., de Graaf, D. C., Dearden, P., Elsik, C. G., Gadau,  
706 J., Grimmelikhuijzen, C. J., Hasselmann, M., Lozier, J. D., Robertson, H. M., Smagghe,  
707 G., Stolle, E., Van Vaerenbergh, M., Waterhouse, R. M., Bornberg-Bauer, E., Klasberg,  
708 S., Bennett, A. K., Camara, F., Guigo, R., Hoff, K., Mariotti, M., Munoz-Torres, M.,  
709 Murphy, T., Santessmasses, D., Amdam, G. V., Beckers, M., Beye, M., Biewer, M.,  
710 Bitondi, M. M., Blaxter, M. L., Bourke, A. F., Brown, M. J., Buechel, S. D., Cameron,  
711 R., Cappelle, K., Carolan, J. C., Christiaens, O., Ciborowski, K. L., Clarke, D. F.,  
712 Colgan, T. J., Collins, D. H., Cridge, A. G., Dalmay, T., Dreier, S., du Plessis, L.,  
713 Duncan, E., Erler, S., Evans, J., Falcon, T., Flores, K., Freitas, F. C., Fuchikawa, T.,  
714 Gempe, T., Hartfelder, K., Hauser, F., Helbing, S., Humann, F. C., Irvine, F., Jermin, L.  
715 S., Johnson, C. E., Johnson, R. M., Jones, A. K., Kadowaki, T., Kidner, J. H., Koch, V.,  
716 Kohler, A., Kraus, F. B., Lattorff, H. M., Leask, M., Lockett, G. A., Mallon, E. B.,  
717 Antonio, D. S., Marxer, M., Meeus, I., Moritz, R. F., Nair, A., Napflin, K., Nissen, I.,  
718 Niu, J., Nunes, F. M., Oakeshott, J. G., Osborne, A., Otte, M., Pinheiro, D. G., Rossie,  
719 N., Rueppell, O., Santos, C. G., Schmid-Hempel, R., Schmitt, B. D., Schulte, C.,  
720 Simoes, Z. L., Soares, M. P., Swevers, L., Winnebeck, E. C., Wolschin, F., Yu, N.,  
721 Zdobnov, E. M., Aqrawi, P. K., Blankenburg, K. P., Coyle, M., Francisco, L.,  
722 Hernandez, A. G., Holder, M., Hudson, M. E., Jackson, L., Jayaseelan, J., Joshi, V.,  
723 Kovar, C., Lee, S. L., Mata, R., Mathew, T., Newsham, I. F., Ngo, R., Okwuonu, G.,  
724 Pham, C., Pu, L. L., Saada, N., Santibanez, J., Simmons, D., Thornton, R., Venkat, A.,  
725 Walden, K. K., Wu, Y. Q., Debyser, G., Devreese, B., Asher, C., Blommaert, J.,  
726 Chipman, A. D., Chittka, L., Fouks, B., Liu, J., O'Neill, M. P., Sumner, S., Puiu, D., Qu,  
727 J., Salzberg, S. L., Scherer, S. E., Muzny, D. M., Richards, S., Robinson, G. E., Gibbs,  
728 R. A., Schmid-Hempel, P., & Worley, K. C. (2015). The genomes of two key  
729 bumblebee species with primitive eusocial organization. *Genome Biol*, 16, 76.  
730 <https://doi.org/10.1186/s13059-015-0623-3>

731 Scheiner, R., Sokolowski, M. B., & Erber, J. (2004). Activity of cGMP-dependent protein  
732 kinase (PKG) affects sucrose responsiveness and habituation in *Drosophila*  
733 *melanogaster*. *Learn Mem*, 11(3), 303-311. <https://doi.org/10.1101/lm.71604>

734 11/3/303 [pii]

735 Schulte, C., Theilenberg, E., Muller-Borg, M., Gempe, T., & Beye, M. (2014). Highly  
736 efficient integration and expression of piggyBac-derived cassettes in the honeybee (*Apis*  
737 *mellifera*). *Proc Natl Acad Sci U S A*, *111*(24), 9003-9008.  
738 <https://doi.org/10.1073/pnas.1402341111>

739 Schwander, T., & Keller, L. (2008). Genetic compatibility affects queen and worker caste  
740 determination. *Science*, *322*(5901), 552-552.

741 Schwander, T., Lo, N., Beekman, M., Oldroyd, B. P., & Keller, L. (2010). Nature versus  
742 nurture in social insect caste differentiation. *Trends in Ecology & Evolution*, *25*(5), 275-  
743 282.

744 Shapira, M., Thompson, C. K., Soreq, H., & Robinson, G. E. (2001). Changes in neuronal  
745 acetylcholinesterase gene expression and division of labor in honey bee colonies. *J Mol*  
746 *Neurosci*, *17*(1), 1-12.  
747 [http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Cita](http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=11665858)  
748 [tion&list\\_uids=11665858](http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=11665858)

749 Shpigler, H., Tamarkin, M., Gruber, Y., Poleg, M., Siegel, A. J., & Bloch, G. (2013). Social  
750 influences on body size and developmental time in the bumblebee *Bombus terrestris*.  
751 *Behavioral Ecology and Sociobiology*, *67*(10), 1601-1612.  
752 <https://doi.org/10.1007/s00265-013-1571-0>

753 Simola, D. F., Graham, R. J., Brady, C. M., Enzmann, B. L., Desplan, C., Ray, A., Zwiebel,  
754 L. J., Bonasio, R., Reinberg, D., Liebig, J., & Berger, S. L. (2016). Epigenetic  
755 (re)programming of caste-specific behavior in the ant *Camponotus floridanus*. *Science*,  
756 *351*(6268), aac6633. <https://doi.org/10.1126/science.aac6633>

757 Sinha, S., Ling, X., Whitfield, C. W., Zhai, C., & Robinson, G. E. (2006). Genome scan for  
758 cis-regulatory DNA motifs associated with social behavior in honey bees. *Proceedings*  
759 *of the National Academy of Sciences*, *103*(44), 16352-16357.  
760 <https://doi.org/10.1073/pnas.0607448103>

761 Smith, C. R., Toth, A. L., Suarez, A. V., & Robinson, G. E. (2008). Genetic and genomic  
762 analyses of the division of labour in insect societies. *Nature Reviews Genetics*, *9*(10),  
763 735-748. [https://doi.org/Doi 10.1038/Nrg2429](https://doi.org/10.1038/Nrg2429)

764 Sokolowski, M. B. (1980). Foraging strategies of *Drosophila melanogaster*: a chromosomal  
765 analysis. *Behavior Genetics*, *10*(3), 291-302.

766 Stewart, A. M., Braubach, O., Spitsbergen, J., Gerlai, R., & Kalueff, A. V. (2014). Zebrafish  
767 models for translational neuroscience research: from tank to bedside. *Trends in*

768 *Neurosciences*, 37(5), 264-278.  
769 <https://doi.org/http://dx.doi.org/10.1016/j.tins.2014.02.011>

770 Struk, A. A., Mugon, J., Huston, A., Scholer, A. A., Stadler, G., Higgins, E. T., Sokolowski,  
771 M. B., & Danckert, J. (2019). Self-regulation and the *foraging* gene (PRKG1) in  
772 humans. *Proceedings of the National Academy of Sciences of the United States of*  
773 *America*, 116(10), 4434-4439.

774 Tarès, S., Arthaud, L., Amichot, M., & Robichon, A. (2013). Environment exploration and  
775 colonization behavior of the pea aphid associated with the expression of the *foraging*  
776 gene. *PLoS ONE*, 8(5), e65104.

777 Thamm, M., & Scheiner, R. (2014). PKG in honey bees: spatial expression, Amfor gene  
778 expression, sucrose responsiveness, and division of labor. *J Comp Neurol*, 522(8), 1786-  
779 1799. <https://doi.org/10.1002/cne.23500>

780 Tinbergen, N. (1963). On aims and methods of Ethology. *Zeitschrift für Tierpsychologie*,  
781 20(4), 410-433. <https://doi.org/10.1111/j.1439-0310.1963.tb01161.x>

782 Tobback, J., Mommaerts, V., Vandersmissen, H. P., Smagghe, G., & Huybrechts, R. (2011).  
783 Age- and task-dependent foraging gene expression in the bumblebee *Bombus terrestris*.  
784 *Archives of Insect Biochemistry and Physiology*, 76(1), 30-42.  
785 <https://doi.org/10.1002/arch.20401>

786 Tobback, J., Verlinden, H., Vuerinckx, K., Vleugels, R., Vanden Broeck, J., & Huybrechts, R.  
787 (2013). Developmental-and food-dependent foraging transcript levels in the desert  
788 locust. *Insect Science*, 20(6), 679-688.

789 Toth, A. L., & Robinson, G. E. (2007). Evo-devo and the evolution of social behavior. *Trends*  
790 *Genet*, 23(7), 334-341. [https://doi.org/S0168-9525\(07\)00155-2](https://doi.org/S0168-9525(07)00155-2) [pii]  
791 10.1016/j.tig.2007.05.001

792 Toth, A. L., Varala, K., Henshaw, M. T., Rodriguez-Zas, S. L., Hudson, M. E., & Robinson,  
793 G. E. (2010). Brain transcriptomic analysis in paper wasps identifies genes associated  
794 with behaviour across social insect lineages. *Proceedings of the Royal Society B:*  
795 *Biological Sciences*, 277(1691), 2139-2148.

796 Toth, A. L., Varala, K., Henshaw, M. T., Rodriguez-Zas, S. L., Hudson, M. E., & Robinson,  
797 G. E. (2010). Brain transcriptomic analysis in paper wasps identifies genes associated  
798 with behaviour across social insect lineages. *Proc Biol Sci*, 277(1691), 2139-2148.  
799 <https://doi.org/rspb.2010.0090> [pii]  
800 10.1098/rspb.2010.0090



801 Toth, A. L., Varala, K., Newman, T. C., Miguez, F. E., Hutchison, S. K., Willoughby, D. A.,  
802 Simons, J. F., Egholm, M., Hunt, J. H., Hudson, M. E., & Robinson, G. E. (2007). Wasp  
803 gene expression supports an evolutionary link between maternal behavior and  
804 eusociality. *Science*, *318*(5849), 441-444. <https://doi.org/10.1126/science.1146647>

805 Tribble, W., Olivos-Cisneros, L., McKenzie, S. K., Saragosti, J., Chang, N. C., Matthews, B.  
806 J., Oxley, P. R., & Kronauer, D. J. C. (2017). orco Mutagenesis Causes Loss of  
807 Antennal Lobe Glomeruli and Impaired Social Behavior in Ants. *Cell*, *170*(4), 727-735  
808 e710. <https://doi.org/10.1016/j.cell.2017.07.001>

809 Tschinkel, W. R. (2006). *The fire ants*. Cambridge: Harvard University Press.

810 Tsvetkov, N., Madani, B., Krimus, L., MacDonald, S. E., & Zayed, A. (2019). A new protocol  
811 for measuring spatial learning and memory in the honey bee *Apis mellifera*: effects of  
812 behavioural state and cGMP. *Insectes Sociaux*, *66*(1), 65-71.  
813 <https://doi.org/10.1007/s00040-018-0641-8>

814 Vanengelsdorp, D., & Meixner, M. D. (2010). A historical review of managed honey bee  
815 populations in Europe and the United States and the factors that may affect them. *J*  
816 *Invertebr Pathol*, *103 Suppl 1*, S80-95. <https://doi.org/10.1016/j.jip.2009.06.011>

817 Walton, A., Sheehan, M. J., & Toth, A. L. (2020). Going wild for functional genomics: RNA  
818 interference as a tool to study gene-behavior associations in diverse species and  
819 ecological contexts. *Hormones and Behavior*, *124*, 104774.

820 Wang, S., & Sokolowski, M. B. (2017). Aggressive behaviours, food deprivation and the  
821 *foraging* gene. *Royal Society Open Science*, *4*(4), 170042.

822 Wang, X., & Robinson, P. J. (1997). Cyclic GMP-dependent protein kinase and cellular  
823 signaling in the nervous system. *Journal of Neurochemistry*, *68*(2), 443-456.

824 Wang, Y., Jorda, M., Jones, P. L., Maleszka, R., Ling, X., Robertson, H. M., Mizzen, C. A.,  
825 Peinado, M. A., & Robinson, G. E. (2006). Functional CpG methylation system in a  
826 social insect. *Science*, *314*(5799), 645-647. <https://doi.org/314/5799/645> [pii]  
827 10.1126/science.1135213

828 Wang, Z., Pan, Y., Li, W., Jiang, H., Chatzimanolis, L., Chang, J., Gong, Z., & Liu, L.  
829 (2008). Visual pattern memory requires foraging function in the central complex of  
830 *Drosophila*. *Learn Mem*, *15*(3), 133-142. <https://doi.org/10.1101/lm.873008>

831 Weinstock, G. M., Robinson, G. E., Gibbs, R. A., Worley, K. C., Evans, J. D., Maleszka, R.,  
832 Robertson, H. M., Weaver, D. B., Beye, M., Bork, P., Elsik, C. G., Hartfelder, K., Hunt,  
833 G. J., Zdobnov, E. M., Amdam, G. V., Bitondi, M. M. G., Collins, A. M., Cristino, A.  
834 S., Lattorff, H. M. G., Lobo, C. H., Moritz, R. F. A., Nunes, F. M. F., Page, R. E.,

835 Simoes, Z. L. P., Wheeler, D., Carninci, P., Fukuda, S., Hayashizaki, Y., Kai, C.,  
836 Kawai, J., Sakazume, N., Sasaki, D., Tagami, M., Albert, S., Baggerman, G., Beggs, K.  
837 T., Bloch, G., Cazzamali, G., Cohen, M., Drapeau, M. D., Eisenhardt, D., Emore, C.,  
838 Ewing, M. A., Fahrbach, S. E., Foret, S., Grimmelikhuijzen, C. J. P., Hauser, F.,  
839 Hummon, A. B., Huybrechts, J., Jones, A. K., Kadowaki, T., Kaplan, N., Kucharski, R.,  
840 Leboulle, G., Linial, M., Littleton, J. T., Mercer, A. R., Richmond, T. A., Rodriguez-  
841 Zas, S. L., Rubin, E. B., Sattelle, D. B., Schlipalius, D., Schoofs, L., Shemesh, Y.,  
842 Sweedler, J. V., Velarde, R., Verleyen, P., Vierstraete, E., Williamson, M. R., Ament, S.  
843 A., Brown, S. J., Corona, M., Dearden, P. K., Dunn, W. A., Elekonich, M. M., Fujiyuki,  
844 T., Gattermeier, I., Gempe, T., Hasselmann, M., Kadowaki, T., Kage, E., Kamikouchi,  
845 A., Kubo, T., Kucharski, R., Kunieda, T., Lorenzen, M. D., Milshina, N. V., Morioka,  
846 M., Ohashi, K., Overbeek, R., Ross, C. A., Schioett, M., Shippy, T., Takeuchi, H., Toth,  
847 A. L., Willis, J. H., Wilson, M. J., Gordon, K. H. J., Letunic, I., Hackett, K., Peterson,  
848 J., Felsenfeld, A., Guyer, M., Solignac, M., Agarwala, R., Cornuet, J. M., Monnerot, M.,  
849 Mougel, F., Reese, J. T., Vautrin, D., Gillespie, J. J., Cannone, J. J., Gutell, R. R.,  
850 Johnston, J. S., Eisen, M. B., Iyer, V. N., Iyer, V., Kosarev, P., Mackey, A. J., Solovyev,  
851 V., Souvorov, A., Aronstein, K. A., Bilikova, K., Chen, Y. P., Clark, A. G., Decanini, L.  
852 I., Gelbart, W. M., Hetru, C., Hultmark, D., Imler, J. L., Jiang, H. B., Kanost, M.,  
853 Kimura, K., Lazzaro, B. P., Lopez, D. L., Simuth, J., Thompson, G. J., Zou, Z., De  
854 Jong, P., Sodergren, E., Csuros, M., Milosavljevic, A., Osoegawa, K., Richards, S., Shu,  
855 C. L., Duret, L., Elhaik, E., Graur, D., Anzola, J. M., Campbell, K. S., Childs, K. L.,  
856 Collinge, D., Crosby, M. A., Dickens, C. M., Grametes, L. S., Grozinger, C. M., Jones,  
857 P. L., Jorda, M., Ling, X., Matthews, B. B., Miller, J., Mizzen, C., Peinado, M. A., Reid,  
858 J. G., Russo, S. M., Schroeder, A. J., St Pierre, S. E., Wang, Y., Zhou, P. L., Jiang, H.  
859 Y., Kitts, P., Ruef, B., Venkatraman, A., Zhang, L., Aquino-Perez, G., Whitfield, C. W.,  
860 Behura, S. K., Berlocher, S. H., Sheppard, W. S., Smith, D. R., Suarez, A. V., Tsutsui,  
861 N. D., Wei, X. H., Wheeler, D., Havlak, P., Li, B. S., Liu, Y., Sodergren, E., Jolivet, A.,  
862 Lee, S., Nazareth, L. V., Pu, L. L., Thorn, R., Stolc, V., Newman, T., Samanta, M.,  
863 Tongprasit, W. A., Claudianos, C., Berenbaum, M. R., Biswas, S., de Graaf, D. C.,  
864 Feyereisen, R., Johnson, R. M., Oakeshott, J. G., Ranson, H., Schuler, M. A., Muzny,  
865 D., Chacko, J., Davis, C., Dinh, H., Gill, R., Hernandez, J., Hines, S., Hume, J.,  
866 Jackson, L., Kovar, C., Lewis, L., Miner, G., Morgan, M., Nguyen, N., Okwuonu, G.,  
867 Paul, H., Santibanez, J., Savery, G., Svatek, A., Villasana, D., Wright, R., & Consort, H.



868 G. S. (2006). Insights into social insects from the genome of the honeybee *Apis*  
869 *mellifera*. *Nature*, 443(7114), 931-949. <https://doi.org/Doi> 10.1038/Nature05260

870 Wheeler, D. E. (1991). The developmental basis of worker caste polymorphism in ants. *The*  
871 *American Naturalist*, 138(5), 1218-1238.

872 Whitfield, C. W., Ben-Shahar, Y., Brillet, C., Leoncini, I., Crauser, D., Leconte, Y.,  
873 Rodriguez-Zas, S., & Robinson, G. E. (2006). Genomic dissection of behavioral  
874 maturation in the honey bee. *Proc Natl Acad Sci U S A*, 103(44), 16068-16075.  
875 <https://doi.org/10.1073/pnas.0606909103>

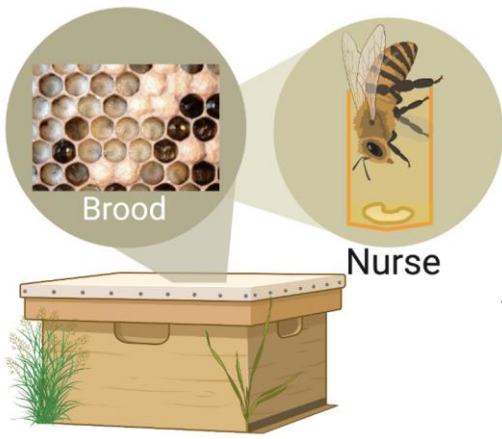
876 Whitfield, C. W., Cziko, A. M., & Robinson, G. E. (2003). Gene expression profiles in the  
877 brain predict behavior in individual honey bees. *Science*, 302(5643), 296-299.  
878 [http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Cita](http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=14551438)  
879 [tion&list\\_uids=14551438](http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=14551438)

880 Wilson, E. O. (1987). Causes of ecological success: the case of the ants. *The Journal of*  
881 *Animal Ecology*, 1-9.

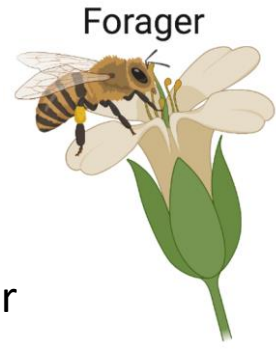
882 Ziegelberger, G., van den Berg, M. J., Kaissling, K. E., Klumppt, S., & Schultz, J. E. (1990).  
883 Cyclic GMP levels and guanylate cyclase activity in pheromone-sensitive antennae of  
884 slimoths *Antherae polyphemus* and *Bombyx mori*. *J. Neurosci.*, 10, 217-225.  
885  
886

887 **Figure 1. cGMP-dependent protein kinase is a key node in the gene network that**  
888 **regulates behavioral states associated with division of labour.** (A) Division of labour in the  
889 honey bee *Apis mellifera* is regulated solely by age-dependent processes. Young bees start  
890 their lives as nurses and typically start foraging when they are three weeks old. (B) Division  
891 of labour in the ant *Pheidole pallidula*. Tasks are regulated by developmental caste  
892 determination of “majors” and “minors”, as well by age-dependent regulation of behaviours  
893 such as nursing and foraging. Division of labour is also associated with task-specific sensory  
894 thresholds (A-B); and changes in neuronal anatomy and physiology (C). (D) Current models  
895 stipulate that *for* is a conserved regulatory node in the gene networks that regulate behavioral  
896 states across diverse social insect species. Therefore, changes in PKG activity can lead to  
897 shifts in the activity of peripheral and central neuronal populations that drive task-specific  
898 behavioral repertoires.

A



Age-dependent  
Division of labour



**Nursing state:**

1. Negative phototaxis
2. Low response threshold to brood pheromone
3. High response threshold to sugar

**Foraging state:**

1. Positive phototaxis
2. High response threshold to brood pheromone
3. Low response threshold to sugar
4. Attraction to floral odours

B



Major



Developmental  
caste-dependent  
Division of labour



Minor

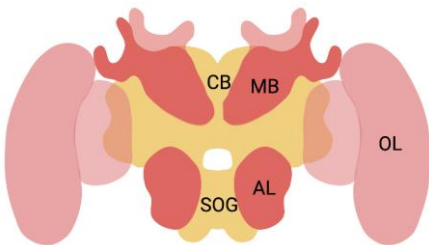
**Soldier state:**

1. Guarding
2. Low response threshold to intruders
3. High response threshold to food
4. Process large food items

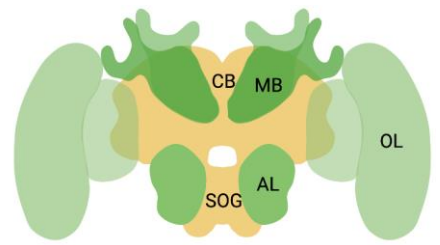
**Forager state:**

1. Food search
2. High response threshold to intruders
3. Low response threshold to food
4. Recruitment to food sources/intruders

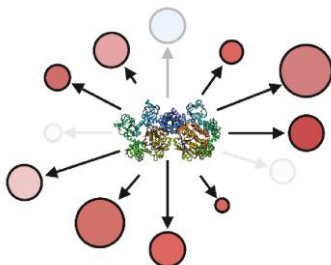
C



Brain anatomy  
and physiology



D



PKG-associated  
gene network

