

Electronic Supplementary Material for: Division of labour promotes the spread of information in colony emigrations by the ant *Temnothorax rugatulus*

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Supplementary methods

Data collection

Experiments were video recorded using three cameras with 1k resolution. One camera gave a bird's-eye view of the whole arena; the other two were positioned above each candidate nest and captured the nest interior at sufficiently high resolution to make the ants' paint marks identifiable. We manually reviewed these recordings to compile a complete record of each ant's arrivals, departures, and recruitment acts (see Table S1 for a complete list of recorded actions). For each action, we noted the time of occurrence, the identity of the ant and the nest at which the action occurred. We also recorded the origin and destination of every tandem run and transport, as determined from the recording of the whole arena.

Table S1 Ethogram of individual behaviours observed in video-recorded emigrations. Each row shows the symbol and definition of a particular action, along with the set of identifiers used to specify where it occurred.

Symbol	Definition	Location
<i>E</i>	Enter a candidate nest	{ <i>good, mediocre</i> }
<i>L</i>	Leave a candidate nest	{ <i>good, mediocre</i> }
<i>LB</i>	Begin leading a tandem run	{ <i>old</i> → <i>good</i> , <i>old</i> → <i>mediocre</i> , <i>mediocre</i> → <i>good</i> }
<i>FB</i>	Begin following a tandem run	{ <i>old</i> → <i>good</i> , <i>old</i> → <i>mediocre</i> , <i>mediocre</i> → <i>good</i> }
<i>TB</i>	Transport a brood item (<i>i.e.</i> , egg, larva, pupa) into a candidate nest	{ <i>good, mediocre</i> }
<i>TU</i>	Transport an unknown object (<i>i.e.</i> , detritus for use as building material) into a candidate nest	{ <i>good, mediocre</i> }
<i>TA</i>	Transport an adult ant into a candidate nest	{ <i>good, mediocre</i> }
<i>BC</i>	Be carried into a candidate nest	{ <i>good, mediocre</i> }
<i>TBO</i>	Transport a brood item out of a candidate nest	{ <i>good, mediocre</i> }
<i>TAO</i>	Transport an adult ant out of a candidate nest	{ <i>good, mediocre</i> }
<i>BCO</i>	Be carried out of a candidate nest	{ <i>good, mediocre</i> }

Behavioural features of individual ants

Table S2 List of measured features, their definitions, domain and whether they are defined for all ants or only for those ants involved in transport activity.

Feature	Definition	Domain	Always defined
Visits	Average number of visits to any potential new nest (<i>i.e.</i> , good or mediocre nest)	$[0, \infty)$	✓
Visit duration	Average duration of visits to any potential new nest	$[0, \infty)$	✓
Visits both	Proportion of emigrations in which the ant visits both the good and the mediocre nest before transporting an item or be carried	$[0,1]$	✓
Total visits duration	Cumulative duration of visits to any potential new nest before transporting an item	$[0, \infty)$	✗
Time of 1 st visit	Average proportion of the emigration at which the ant visits a potential new nest for the first time	$[0,1]$	✓
Tandem runs led	Average number of tandem runs led including only runs from the old nest to either of the two potential nests and from the mediocre nest to the good nests	$[0, \infty)$	✓
Tandem runs followed	Average number of tandem runs followed including only runs from the old nest to either of the two potential nests and from the mediocre nest to the good nests	$[0, \infty)$	✓
Reverse tandem runs led	Average number of reverse tandem runs led including only runs from the either of the two potential nests to the old nest	$[0, \infty)$	✓
Reverse tandem runs followed	Average number of reverse tandem runs followed including only runs from the either of the two potential nests to the old nest	$[0, \infty)$	✓
Items transported	Average number of items (<i>i.e.</i> , adult ant, brood, dirt or unknown object) transported towards any nest	$[0, \infty)$	✓
Transport duration	Average duration of a round-trip transport of any item	$[0, \infty)$	✗
Time of 1 st transport	Average proportion of the emigration at which the ant transports her first item	$[0,1]$	✗
Be carried	Averaged number of times an ant is carried towards any nest	$[0, \infty)$	✓

Measures of heterogeneity

To quantify the degree of heterogeneity in the distribution of a behavioural feature, we used both Lorenz curves and Gini coefficients. A Lorenz curve is a probability plot that assesses how much the distribution of a feature across individuals varies from a hypothetical uniform distribution. In particular, it plots the cumulative portion of the total amount of a certain feature (*e.g.*, total number of tandem runs) against the cumulative portion of the population of ants, with ants ordered by increasing values of the feature. When a feature is equally distributed across the population, the corresponding Lorenz curve is a line with slope one (line of uniformity); the higher the degree of nonuniformity of the distribution, the larger the area between the line of uniformity and the Lorenz curve. The Gini coefficient is the ratio of this area to

the total area under the line of uniformity. A Gini coefficient of one indicates maximal heterogeneity (e.g., one individual performs all of the actions measured by the feature) while a value of zero means that all individuals contribute equally to the feature).

Measures of division of labour

We used an information-theoretic framework that measures both the degree of specialization of each *ant* and the degree at which *tasks* are performed by the same individuals. Specialization of individual workers, *i.e.*, their tendency to perform certain tasks over others, can be quantified by the index $DOL_{indiv} = I(ant; task)/H(task)$, while the degree to which tasks are performed by the same individuals is defined as $DOL_{task} = I(ant; task)/H(ant)$. In these equations, function I is the mutual information between two variables and function H is the Shannon entropy of one variable. $DOL_{indiv} = 1$ indicates that given the identity of an ant we have complete knowledge of the task performed by that ant, whereas $DOL_{indiv} = 0$ indicates that tasks are performed randomly by ants and no association is present. $DOL_{task} = 1$ indicates that given a task we have complete knowledge of the identity of the ant performing that task whereas $DOL_{task} = 0$ indicates that ants randomly perform all tasks. An aggregate measure of division of labour is their geometric mean $DOL = \sqrt{DOL_{indiv} DOL_{task}}$. Values of DOL close to 1 correspond to a high degree of division of labour while values close to 0 represent instead the absence of any predictive relationship between a task to be performed and the ants that perform it. To compute these measures for each colony, we considered six tasks: leading a forward tandem run, following a forward tandem run, leading a reverse tandem run, following a reverse tandem run, transporting an item to a nest, and being carried to a nest. We then counted how many times each ant performed each task over the course of all emigrations. Using this data, we computed DOL measures both among the individuals of the colony (*i.e.*, variable *ant* represents the unique id of each ant) and among behavioural castes (*i.e.*, variable *ant* represents the role assigned to each ant during the clustering analysis).

Supplementary results

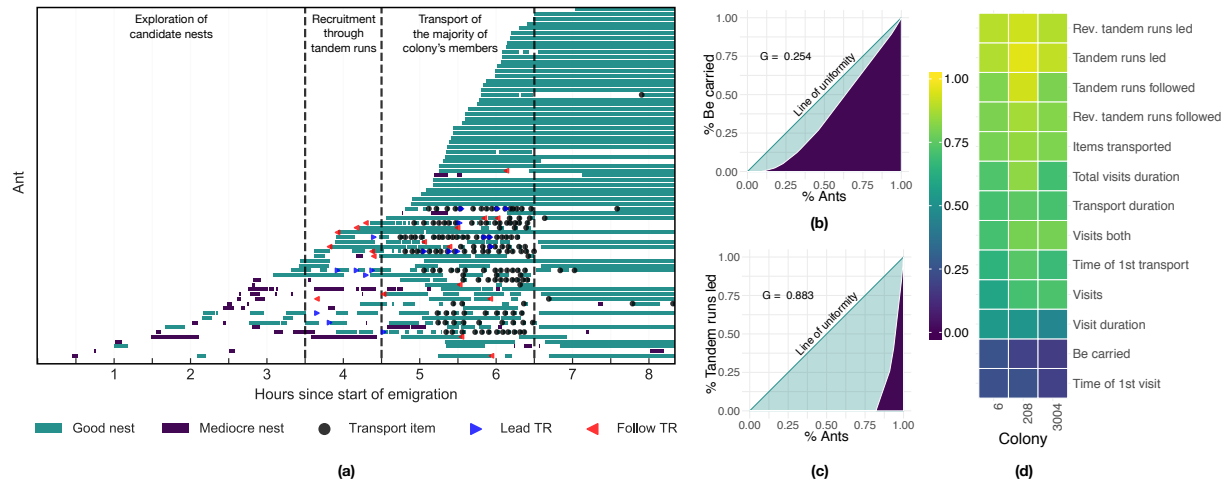


Figure S1. Panel (a) shows the individual behaviour over time of each ant in colony 6 during the first emigration. Horizontal bars denote time spent inside a candidate nest; symbols denote the occurrence of transport and tandem running behaviours; dashed vertical lines denote, from left to right, the exploration phase, the initial recruitment through tandem running, and the transport phase. Panel (b) and panel (c) show the Lorenz curves of the distribution of behaviours be-carried and leading tandem run across the members of colony 6 in all emigrations. Panel (d) provides a heatmap representation of the Gini coefficients computed for a number of different behavioural features for each considered colony (higher values correspond to more unequal distributions).

Behavioural diversity of workers

The contributions of individual workers in an emigration are far from equal. The majority of colony members experience the decision-making process only passively, when carried from one nest to another by a nestmate (Figure 1a, upper portion). A smaller portion of the workforce instead contributes actively to finding a new home and moving there (Figure 1a, lower portion). Initially, a few ants discover and perform short visits to the candidate nests (0–3.5 hours). Some of these ants later recruit other colony members through a number of tandem runs (3.5–4.5 hours). Finally, the bulk of the colony is carried to the chosen nest by this hard-working minority (4.5–6.5 hours).

For each colony, we measured the distribution of several behavioural features across colony members. Figure 1d provides a compact representation of how this distribution varied among behavioural features. Some behaviours were performed at similar frequency by all colony members. Being carried to a nest, for example, happens to most ants one time (Figure 1b), and the time of first visit was similar for most ants. Other behaviours, such as leading a tandem run, following a tandem run, or transporting a nestmate, had a much more unequal distribution, with only few workers performing them (Figure 1c). The remaining behavioural features, mostly concerned with visits to candidate nests, showed an intermediate level of diversity.

High level clustering

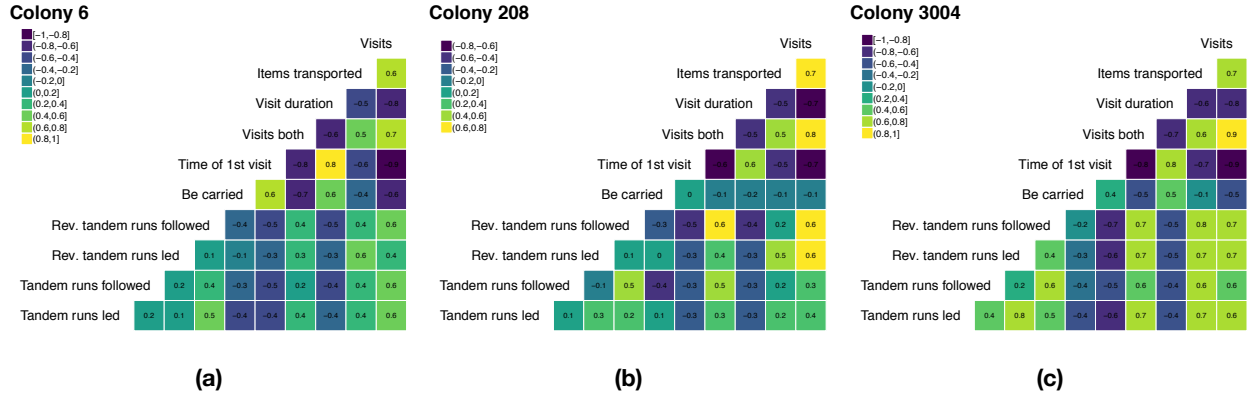


Figure S2 Matrix of Pearson correlation coefficients between all behavioural features considered at the higher level of the hierarchy for (a) colony 6, (b) colony 208, and (c) colony 3004.

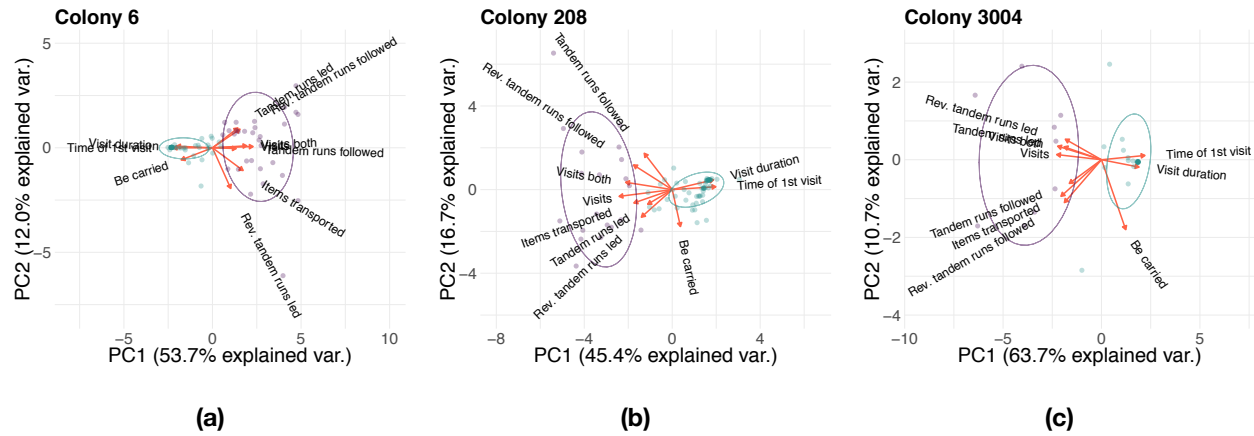


Figure S3 PCA representation over the first two components of the loadings for the behavioural features considered at the higher level of the hierarchy for (a) colony 6, (b) colony 208, and (c) colony 3004. Coloured points represent clustered workers (purple colour for active workers, green colour for inactive workers).

Low level clustering

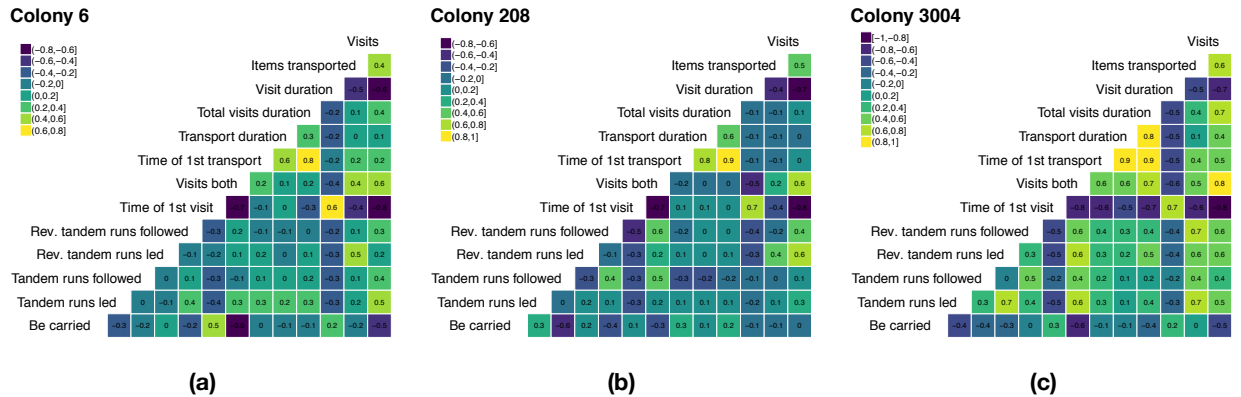


Figure S4 Matrix of Pearson correlation coefficients between all behavioural features considered at the lower level of the hierarchy for active workers of (a) colony 6, (b) colony 208, and (c) colony 3004.

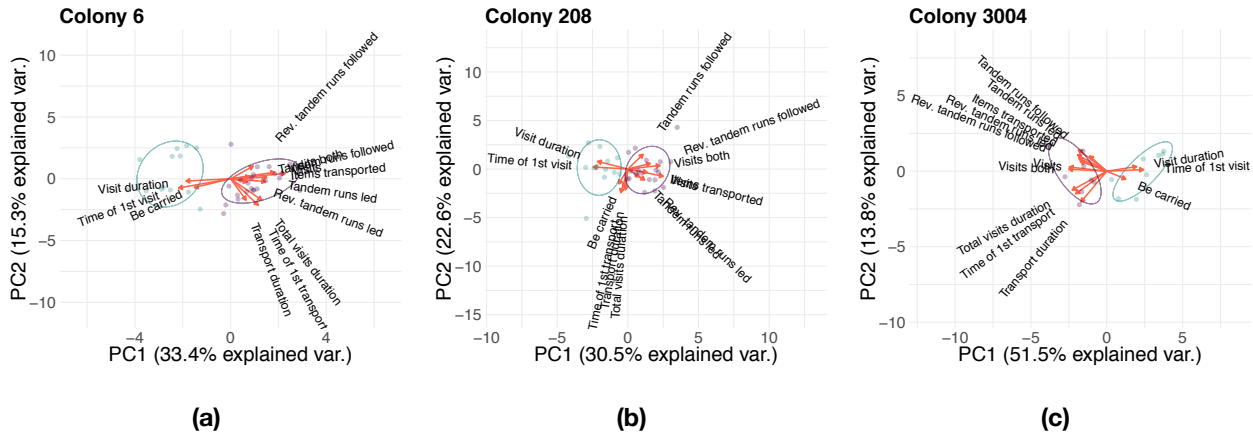


Figure S5 PCA representation over the first two components of the loadings for the behavioural features considered at the lower level of the hierarchy for active workers of (a) colony 6, (b) colony 208, and (c) colony 3004. Coloured points represent clustered workers (purple colour for primary workers, green colour for secondary workers).

In the case of active workers, we considered all behavioural features included at the higher level of the hierarchy plus three additional features related to transport: the average duration of a transport event, the time of first transport, and the cumulative visit duration before transporting an item. Behavioural features generally had a low level of pairwise correlation with the exception of two sets of features with high intra-set correlation (Pearson correlation coefficients in Figure S3). One set included features related to transport events, and the other included features related to the timing of visits. As the PCA (see Figure S4) showed two separate groups of workers, we further divided active workers in two groups: *primary* workers and *secondary* workers.

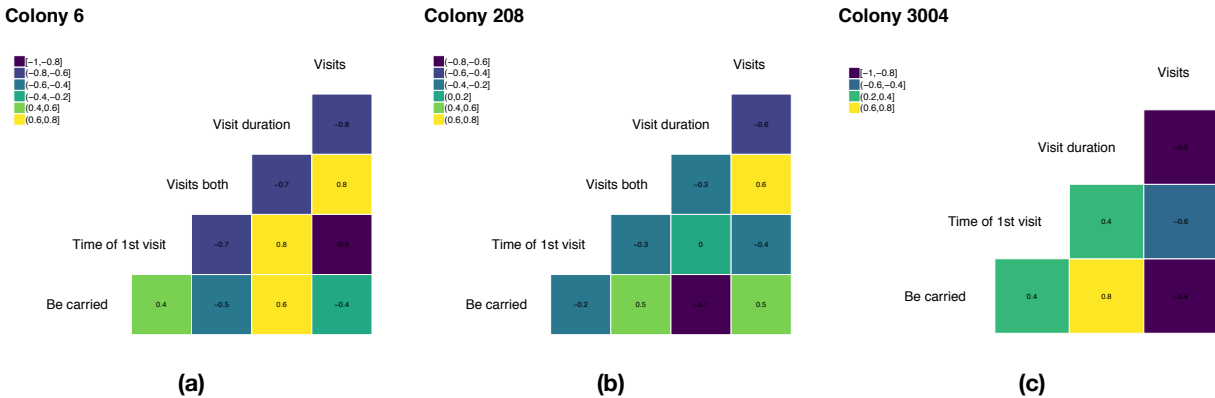


Figure S6 Matrix of Pearson correlation coefficients between all behavioural features considered at the lower level of the hierarchy for inactive workers of (a) colony 6, (b) colony 208, and (c) colony 3004.

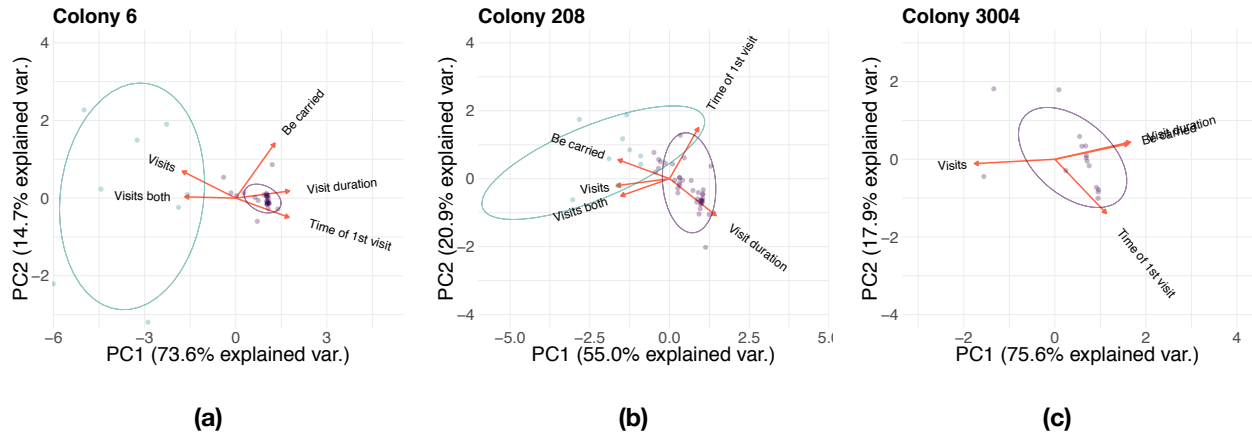


Figure S7 PCA representation over the first two components of the loadings for the behavioural features considered at the lower level of the hierarchy for inactive workers of (a) colony 6, (b) colony 208, and (c) colony 3004. Coloured points represent clustered workers (purple colour for passive workers, green colour for wandering workers).

In the case of inactive workers, we considered only a few behavioural features: the number of times they have been carried to a nest, the number of visits, their average duration, time of the first visit, and whether they visited both nests (this feature was not considered for colony 3004 as inactive workers never visited both nests). The number of visits to a candidate nest is positively correlated to the likelihood of visiting both nests whereas the mean visit duration is positively correlated with the number of times a worker is carried to a nest (Figure S5). From the PCA (Figure S6), we observe that loadings are still divided in two separate sets pointing in opposite directions. We therefore clustered inactive workers in two groups: *passive* workers and *wandering* workers.

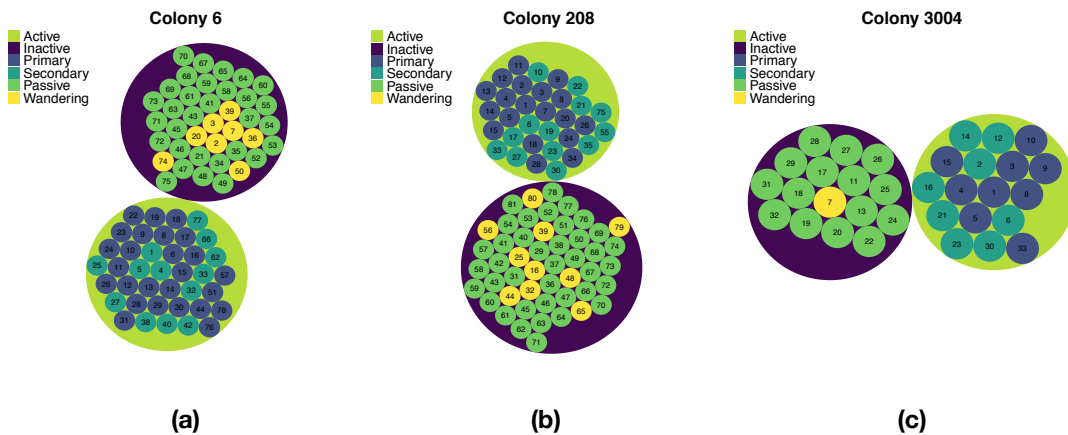


Figure S8 Circle plots representing the overall results of the clustering of ants for (a) colony 6, (b) colony 208, and (c) colony 3004.

Network analysis

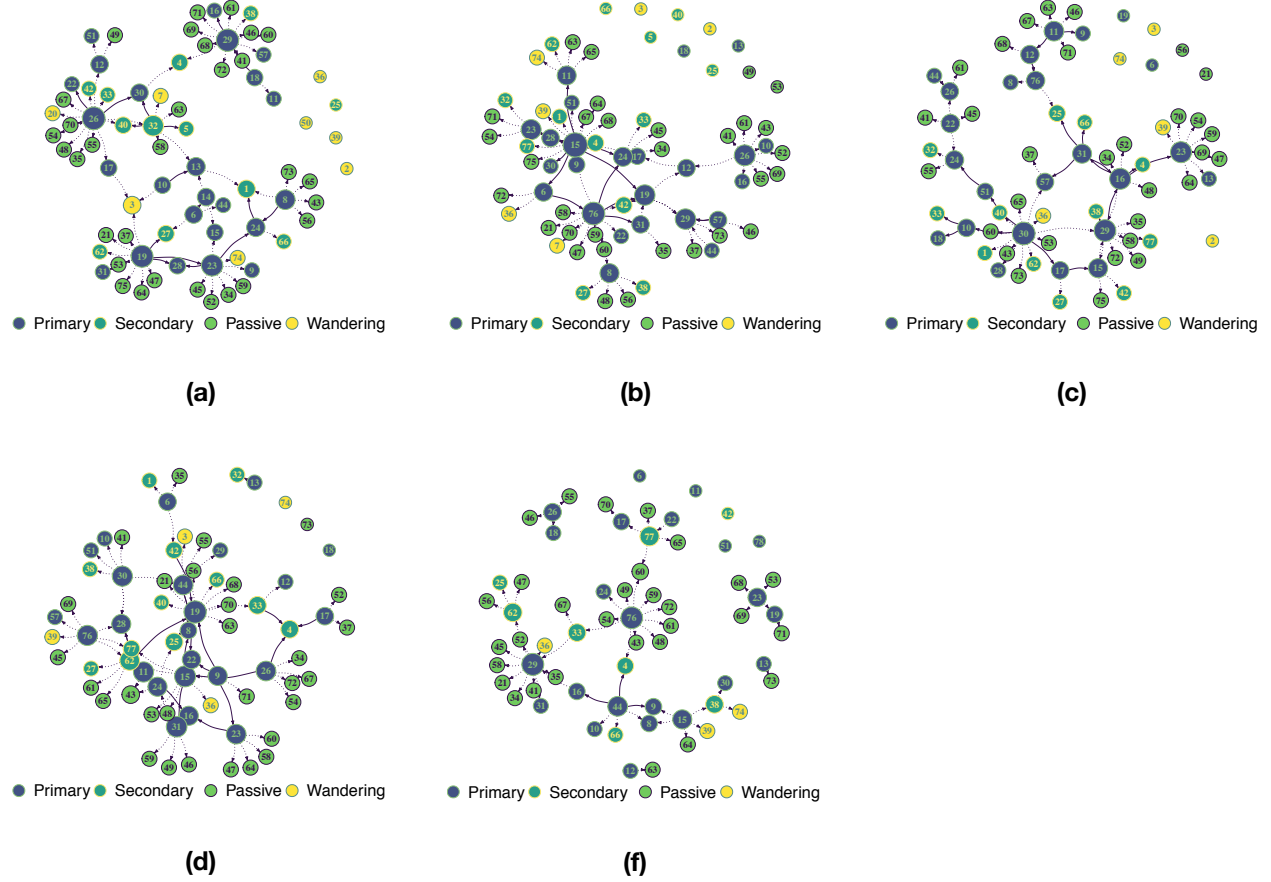


Figure S9 Illustration of recruitment networks for treatment 1–5 of colony 6. Solid arrows represent tandem runs (both forward and reverse), dotted arrows represent transport events.

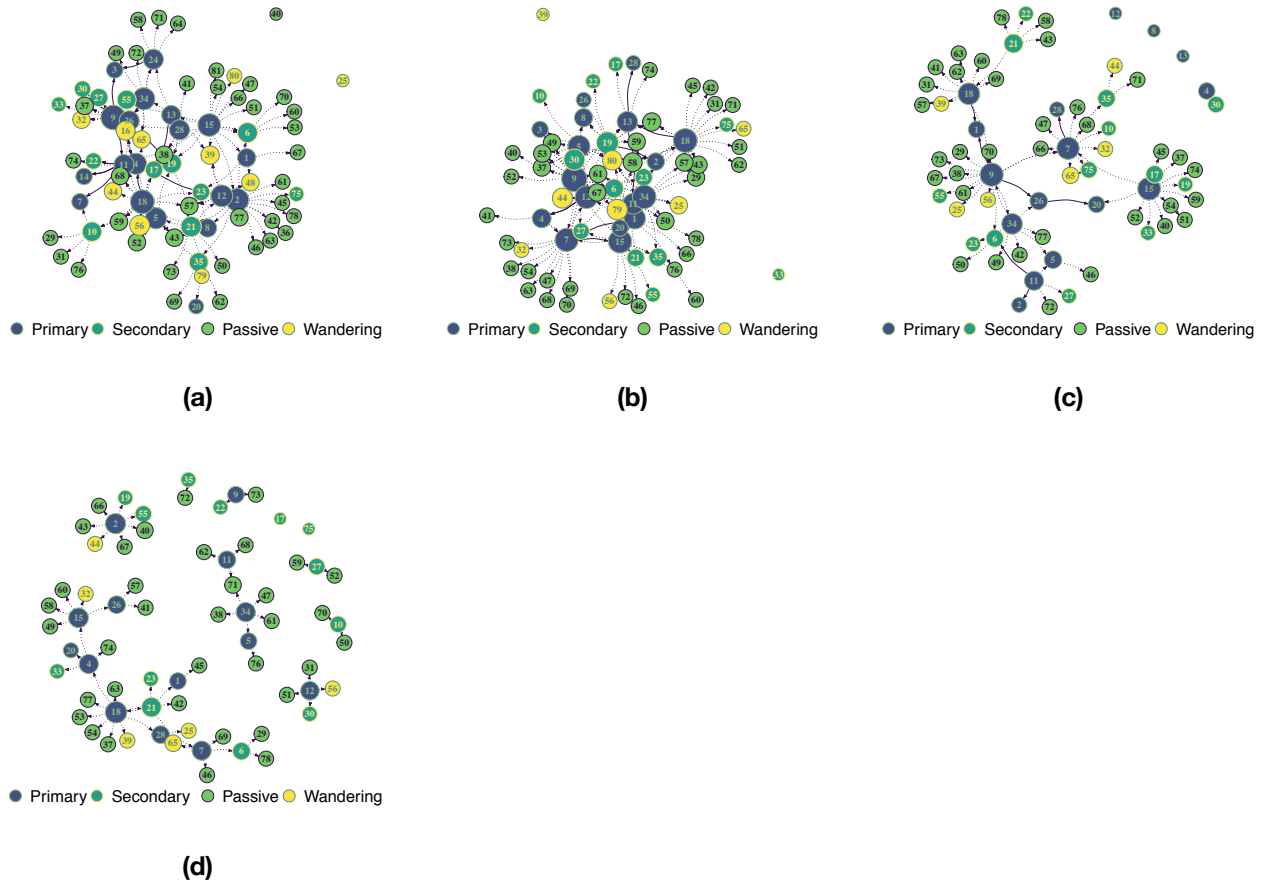


Figure S10 Illustration of recruitment networks for treatment 2–5 of colony 208. Solid arrows represent tandem runs (both forward and reverse), dotted arrows represent transport events.

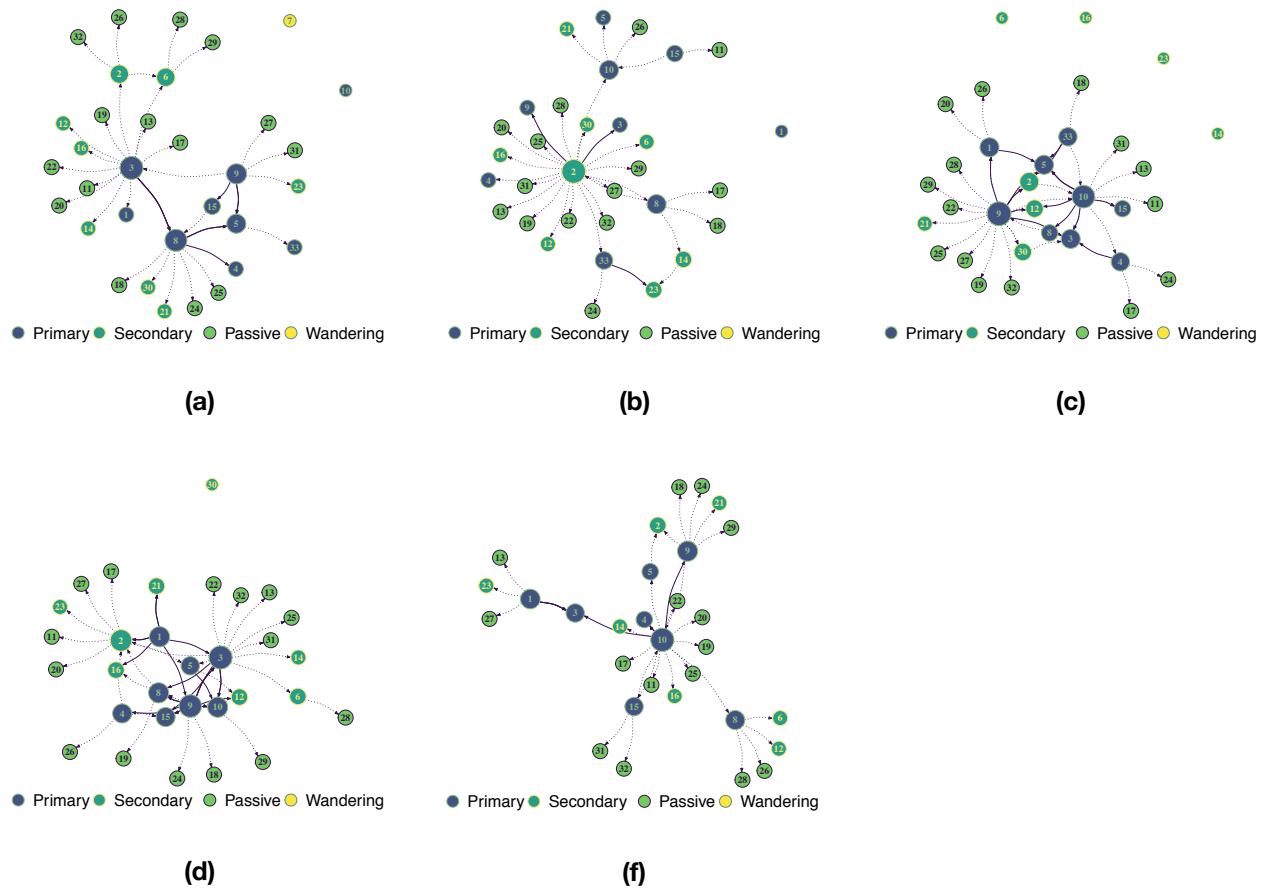


Figure S11 Illustration of recruitment networks for treatment 1–5 of colony 3004. Solid arrows represent tandem runs (both forward and reverse), dotted arrows represent transport events.

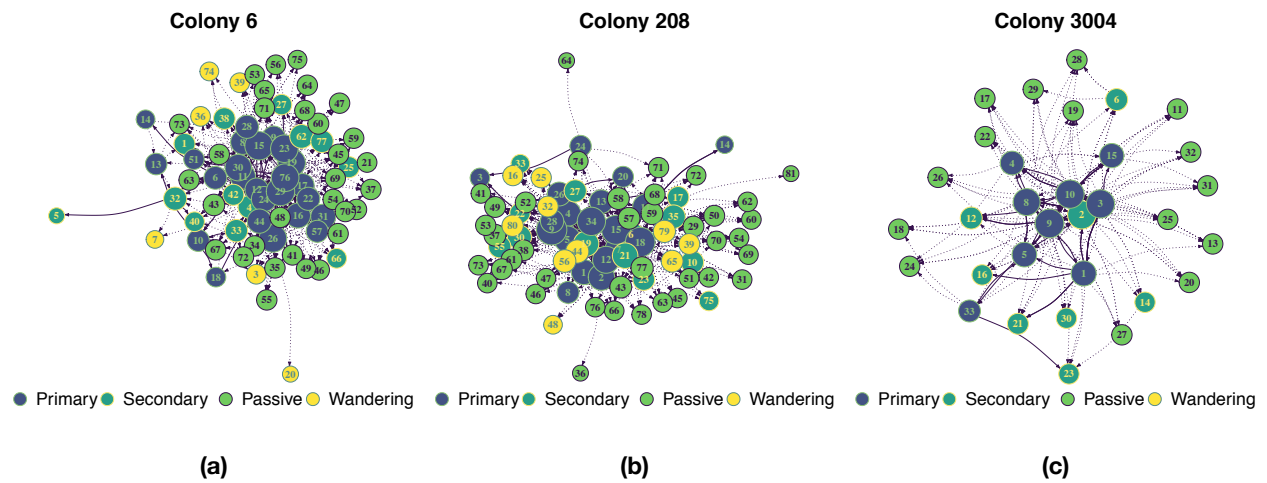


Figure S12 Aggregate recruitment network for (a) colony 6, (b) colony 208, and (c) colony 3004. Solid arrows represent tandem runs (both forward and reverse), dotted arrows represent transport events. Isolated nodes are not shown.

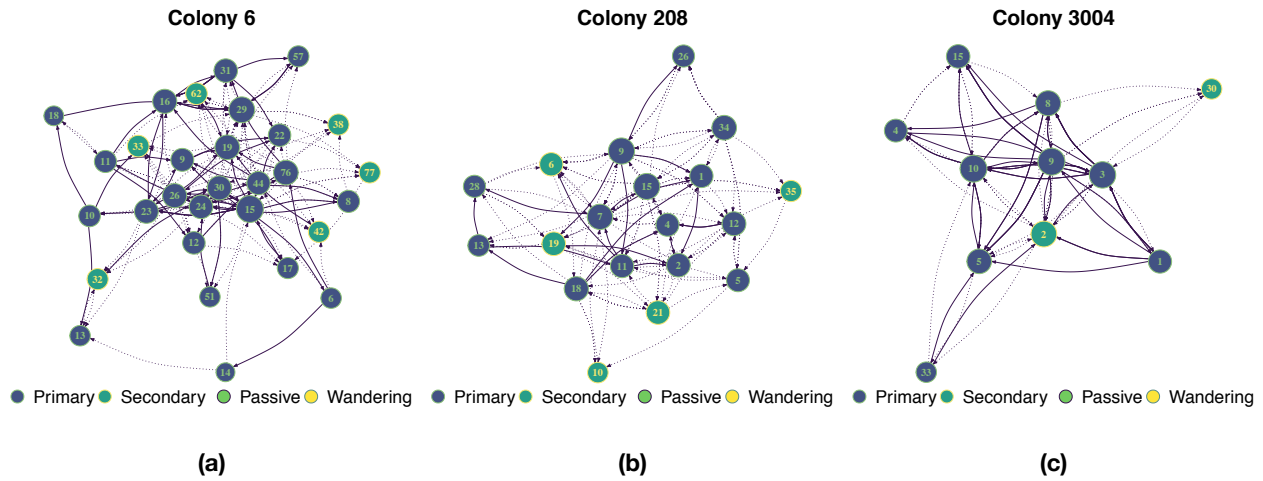


Figure S13 Core of the aggregate recruitment network for (a) colony 6, (b) colony 208, and (c) colony 3004. Solid arrows represent tandem runs (both direct and reverse), dotted arrows represent transport events. Isolated nodes not shown.

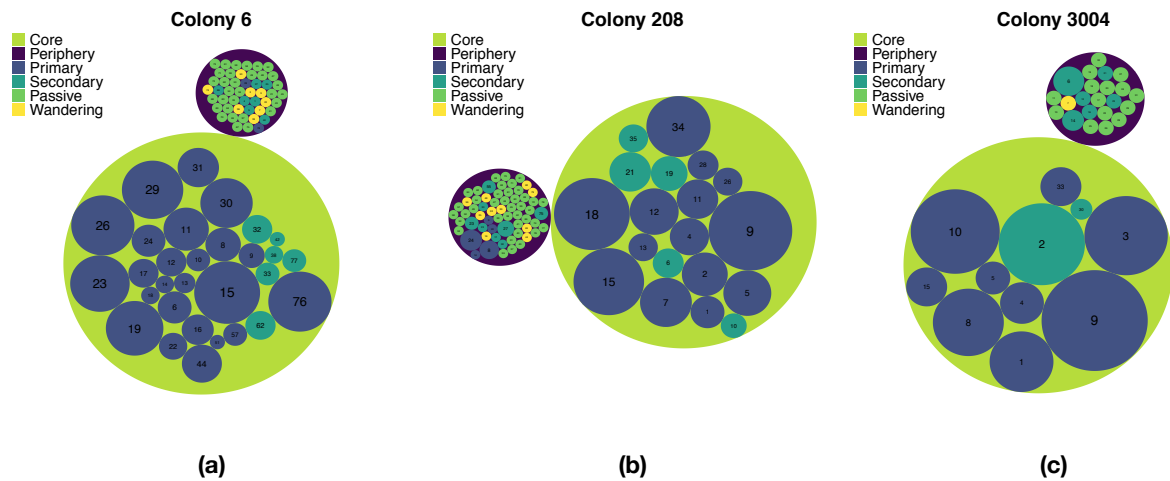


Figure S14 Illustration of the relations between the role covered by each ant as defined from the clustering analysis and their location in the aggregate network (i.e., core versus periphery), respectively, (a) for colony 6, (b) for colony 208, and (c) for colony 3004.

Division of labour

Table S3 Measures of division of labour for each colony and their mean and standard deviation divided, respectively, in ant versus task and role versus task.

Colony	(ant, task)			(role, task)		
	DOI	DOI _{indiv}	DOI _{task}	DOI	DOI _{indiv}	DOI _{task}
6	0.23	0.43	0.12	0.33	0.28	0.39
208	0.24	0.48	0.12	0.33	0.34	0.32
3004	0.23	0.34	0.15	0.26	0.21	0.33
mean ± sd	0.23 ± 0.01	0.42 ± 0.07	0.13 ± 0.02	0.31 ± 0.04	0.28 ± 0.07	0.35 ± 0.04

Spread of information

Table S4 Number and proportion of recruitment events that turned the recruited worker into a recruiter herself. Results divided for forward tandem runs and transport events.

<i>Colony</i>	<i>Forward tandem runs</i>	<i>Transports of adults</i>
6	13/23 = 0.57	19/257 = 0.07
208	3/9 = 0.33	31/240 = 0.13
3004	8/15 = 0.53	14/121 = 0.12