

**Current Biology, Volume 23**

**Supplemental Information**

**Ecology, Not the Genetics**

**of Sex Determination, Determines**

**Who Helps in Eusocial Populations**

**Laura Ross, Andy Gardner, Nate Hardy, and Stuart A. West**

## Supplemental Data:

Figure S1: Relates to figure 3 and 4. Phylogenetic relationships among social lineages. Branch lengths are proportional to time.

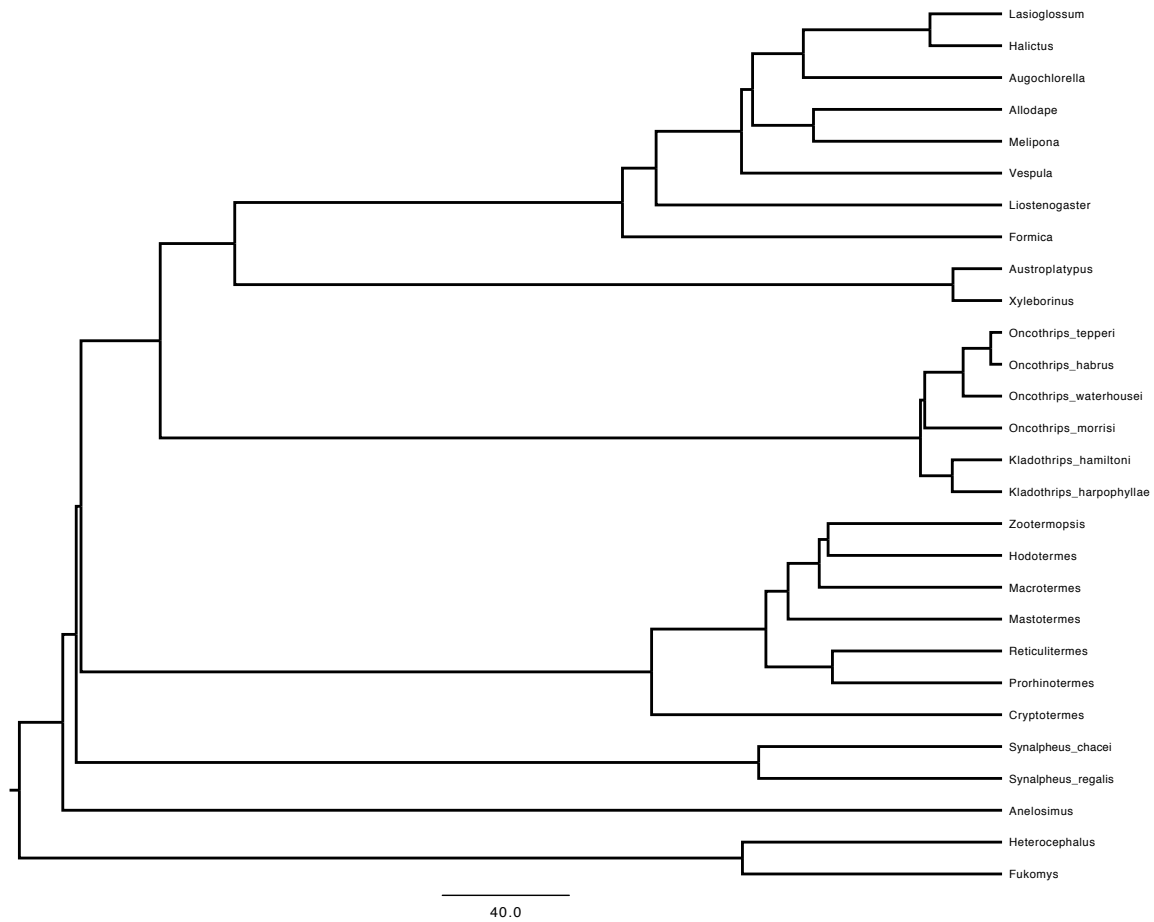


Figure S2: Relates to figure 4 and table S2. Proportion of eusocial origins with female/unbiased helpers (based on table S2). Data uncorrected for phylogeny.

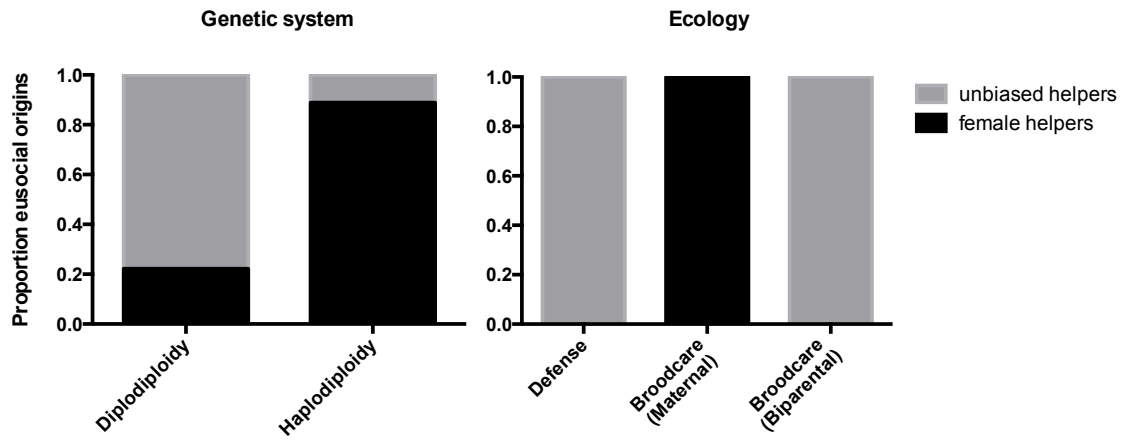


Figure S3: Relates to figure 2. Illustration of life-for-life relatedness for male or female helpers toward either offspring or siblings for haplodiploid and diploid taxa. The pink ant represents a female while the blue ant represents a male.

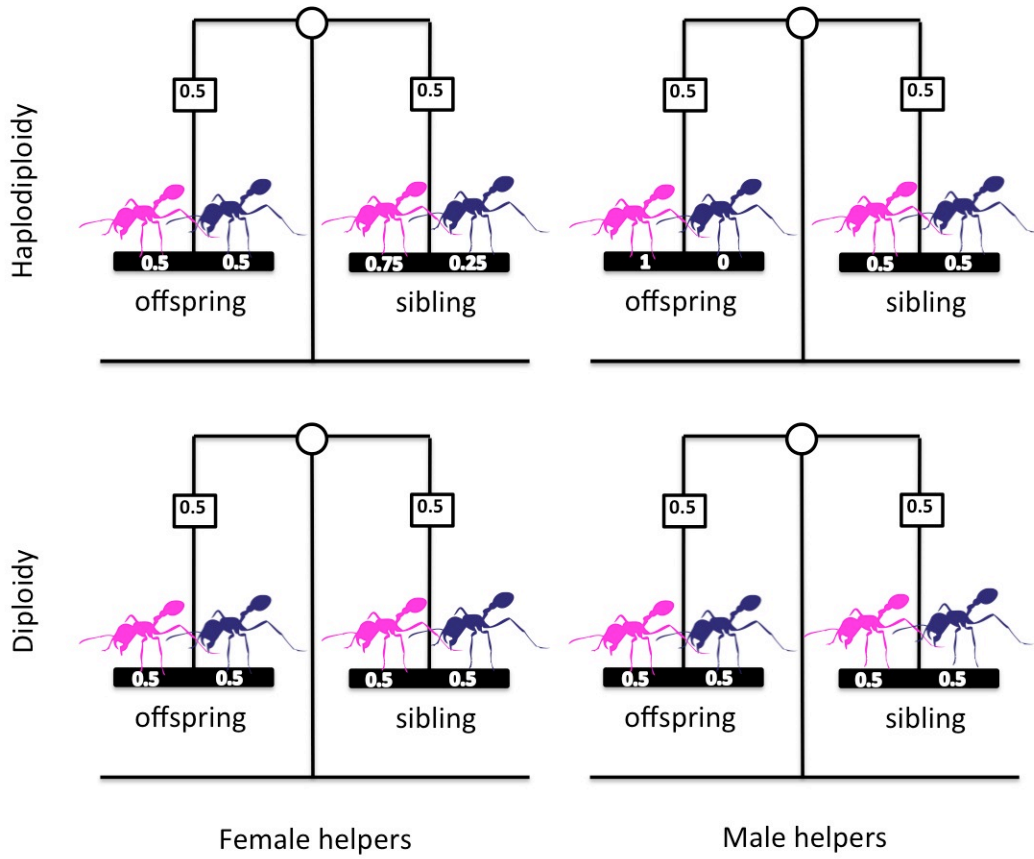


Table S1: Relates to figure 3 and 4. Genbank Accession numbers for DNA sequences used in phylogenetic analysis.

Taxon	Ef1a	16S	Cytb	COI	LWO	28S	18S	Wingless	12S
<b>Allodape</b>	NA	NA	JN564695.1	JN564689.1	NA	NA	NA	NA	NA
<b>Anelosimus</b>	NA	JX977513.1	NA	JX977273.1	NA	JX977391.1	EF050203.1	NA	NA
<b>Augochlorella</b>	NA	NA	NA	JX546143.1	AY227935.1	JQ279154.1	AY995647.1	AY222572.1	NA
<b>Austroplatypus</b>	NA	NA	NA	FJ867832.1	NA	JN544546.1	NA	NA	NA
<b>Cryptotermes</b>	NA	HQ215842.1	AF189122.1	EU253840.1	NA	FJ806528.1	FJ806329.1	FJ706793.1	FJ805969.1
<b>Formica</b>	JQ519673.1	FR865871.1	JX170891.1	JX829423.1	FJ824464.1	FJ407396.1	FJ824197.1	AY233599.1	NA
<b>Fukomys</b>	NA	NA	AY425867.1	NA	NA	NA	NA	NA	AF012213.1
<b>Halictus</b>	EF601166.1	NA	NA	JX833115.1	JQ861245.1	JQ279228.1	AY995651.1	JQ266713.1	NA
<b>Heterocephalus</b>	NA	NC_015112.1	NC_015112.1	NA	NA	JH165919.1	NA	NA	NC_015112.1
<b>Hodotermes</b>	NA	NA	EU253913.1	JN615371.1	NA	JN615328.1	FJ806328.1	NA	FJ805968.1
<b>Kladothrips hamiltoni</b>	NA	AF386659.1	NA	U93515.1	NA	NA	NA	AF386728.1	NA
<b>Kladothrips harpophyllae</b>	NA	AF386664.1	NA	U93514.1	NA	NA	NA	NA	NA
<b>Lasioglossum</b>	NA	NA	NA	JX831587.1	JQ342159.1	JQ279184.1	GU320090.1	JQ266745.1	NA
<b>Liostenogaster</b>	NA	AF066939.1	NA	GU596840.1	NA	GU596730.1	EF190726.1	NA	GU596568.1
<b>Macrotermes</b>	NA	JX050221.1	JX501185.1	JF923342.1	NA	JQ957912.1	FJ806334.1	NA	JX050221.1
<b>Mastotermes</b>	EU414693.2	NC_018120.1	NC_018120.1	JN615367.1	NA	DQ441950.1	FJ806331.1	FJ802921.1	NC_018120.1
<b>Melipona</b>	EU163257.1	NC_004529.1	NC_004529.1	JN315139.1	AF344607.1	EU049745.1	HQ651726.1	GU245576.1	NC_004529.1
<b>Oncothrips habrus</b>	NA	DQ246465.1	NA	AY920992.1	NA	NA	NA	DQ246503.1	NA
<b>Oncothrips morrisi</b>	NA	AF386660.1	NA	U93532.1	NA	NA	NA	AF386729.1	NA
<b>Oncothrips tepperi</b>	NA	U93545.1	NA	U93527.1	NA	NA	NA	AF386731.1	NA
<b>Oncothrips waterhousei</b>	NA	AF386673.1	NA	AY920991.1	NA	NA	NA	NA	NA
<b>Prorhinotermes</b>	NA	EU253752.1	EU253925.1	EU253849.1	NA	EU253672.1	EU253791.1	NA	EU253713.1
<b>Reticulitermes</b>	AB602838.1	GU070788.1	JX501149.1	JQ431093.1	NA	JQ431133.1	JQ972694.1	NA	FJ805972.1
<b>Synalpheus chacei</b>	NA	HQ435440.1	NA	HQ435381.1	NA	NA	NA	NA	NA
<b>Synalpheus regalis</b>	NA	HQ435474.1	NA	HQ435406.1	NA	NA	NA	NA	NA
<b>Vespa</b>	NA	AY206804.2	NA	JX832523.1	NA	AF142527.1	AF142515.1	DQ353143.1	GU596560.1
<b>Xyleborinus</b>	AF259876.1	NA	NA	AB588938.1	NA	HM099727.1	AJ850038.1	NA	AF259835.1
<b>Zootermopsis</b>	NA	NC_018123.1	NC_018123.1	JN615368.1	NA	JN615325.1	EU253799.1	NA	NC_018123.1

Table S2: Relates to figure 3 and 4. Data included in the comparative analysis (including references). We were forced to exclude one of the Hymenopteran origins (*Microstigma*) because of a lack of sequence data. The representative taxon is the one for which we collected sequence data, life history data is extrapolated across larger taxonomic groups. Further justification for the assignments for some taxonomic groups is provided below. Taxa with an asterisk behind their name were excluded from the second analyses that followed the stricter “Crespi-Boomsma” definition of eusociality.

Order	Origin	Representative taxon	Genetic system	Helper sex	Helper task	Sister group	Ancestral care
<b>Hymenoptera</b>	1 [S1, S2]	<i>Formica fusca</i>	haplodiploidy [S1, S3]	female [S3]	broodcare [S3-5]	Solitary wasps [S4-7]	maternal [S2, S6, S7]
<b>Hymenoptera</b>	2 [S1, S2]	<i>Vespula germanica</i>	haplodiploidy [S1, S6]	female [S6]	broodcare [S4-6]	Solitary wasps [S4-7]	maternal [S2, S6, S7]
<b>Hymenoptera</b>	3 [S1, S2]	<i>Liostenogaster flavolineata</i>	haplodiploidy [S1, S8]	female [S8, S9]	broodcare [S4, S5, S9]	Solitary wasps [S4-7]	maternal [S2, S6, S7]
<b>Hymenoptera</b>	4 [S1, S2]	<i>Melipona subnitida</i>	haplodiploidy [S1, S9]	female [S9]	broodcare [S4, S5, S9]	Solitary bees [S4, S5, S9]	maternal [S2, S9]
<b>Hymenoptera</b>	5 [S1, S2]	<i>Allodape punctata</i>	haplodiploidy [S1, S9]	female [S9]	broodcare [S4, S5, S9]	Solitary bees [S4, S5, S9]	maternal [S2, S9]
<b>Hymenoptera</b>	6 [S1, S2]	<i>Lasioglossum rowheri</i>	haplodiploidy [S1, S9]	female [S9]	broodcare [S4, S5, S9]	Solitary bees [S4, S5, S9]	maternal [S2, S9]
<b>Hymenoptera</b>	7 [S1, S2]	<i>Halictus marginatus</i>	haplodiploidy [S1, S9]	female [S9]	broodcare [S4, S5, S9]	Solitary bees [S4, S5, S9]	maternal [S2, S9]
<b>Hymenoptera</b>	8 [S1, S2]	<i>Augochlorella striata</i>	haplodiploidy [S1, S9]	female [S9]	broodcare [S4, S5, S9]	Solitary bees [S4, S5, S9]	maternal [S9, S10]
<b>Rodentia</b>	1	<i>Heterocephalus glaber</i> *	diplodiploidy [S9, S10]	both [S9]	broodcare [S9, S10]	Bathyergidae [S10, S11]	biparental [S10, S11]
<b>Rodentia</b>	2	<i>Fukomys damarensis</i> *	diplodiploidy [S9, S10]	both [S9]	broodcare [S9, S10]	Bathyergidae [S10, S11]	biparental [S11, S12]
<b>Thysanoptera</b>	1	<i>Kladothrips hamiltoni</i>	haplodiploidy [S12, S13]	both [S12, S13]	defense [S13, S14]	NA	NA
<b>Coleoptera</b>	1	<i>Austroplatypus incompertus</i>	diplodiploidy [S14-16]	female [S15, S16]	broodcare [S15, S16]	Platypodinae [S16]	maternal [S16, S17]
<b>Coleoptera</b>	2	<i>Xyleborinus saxesnii</i> *	haplodiploidy [S17]	female [S17]	broodcare [S17, S18]	Xyleborini [S16, S18]	maternal [S16, S19]
<b>Decapoda</b>	1 [S19, S20]	<i>Synalpheus chacei</i> *	diplodiploidy [S20]	both [S20]	defense [S19, S20]	NA	NA
<b>Decapoda</b>	2 [S19, S20]	<i>Synalpheus regalis</i> *	diplodiploidy [S20]	both [S20]	defense [S20, S21]	NA	NA
<b>Araneae</b>	1	<i>Anelosimus eximius</i>	diplodiploidy [S21]	female [S21]	broodcare [S21, S22]	subsocial <i>Anelosimus</i> [S16, S22]	maternal [S16, S22-S24]
<b>Isoptera</b>	1 [S23-25]	<i>Macrotermes subhyalinus</i>	diplodiploidy [S25]	both [S25, S26]	broodcare [S24, S27]	<i>Cryptocercidae</i> [S28, S29]	biparental [S23, S24, S28]
<b>Isoptera</b>	2 [S23-25]	<i>Mastotermes darwiniensis</i>	diplodiploidy [S25]	both [S25, S26]	broodcare [S24, S27]	<i>Cryptocercidae</i> [S28, S29]	biparental [S23, S24, S28]
<b>Isoptera</b>	3 [S23-25]	<i>Cryptotermes secundus</i>	diplodiploidy [S25]	both [S25, S26]	defense [S24, S27]	<i>Cryptocercidae</i> [S28, S29]	biparental [S23, S24, S28]
<b>Isoptera</b>	4 [S23-25]	<i>Prorhinotermes simplex</i>	diplodiploidy [S25]	both [S25, S26]	defense [S24, S27]	<i>Cryptocercidae</i> [S28, S29]	biparental [S23, S24, S28]
<b>Isoptera</b>	5 [S23-25]	<i>Reticulitermes flavipes</i>	diplodiploidy [S25]	both [S25, S26]	broodcare [S24, S27]	<i>Cryptocercidae</i> [S28, S29]	biparental [S23, S24, S28]
<b>Isoptera</b>	6 [S23-25]	<i>Zootermopsis nevadensis</i>	diplodiploidy [S25]	both [S25, S26]	defense [S24, S27]	<i>Cryptocercidae</i> [S28, S29]	biparental [S23, S24, S28]
<b>Isoptera</b>	7 [S23-25]	<i>Hodotermes mossambicus</i>	diplodiploidy [S25]	both [S25, S26]	broodcare [S24, S27]	<i>Cryptocercidae</i> [S28, S29]	biparental [S13, S28, S30]

Table S3: Relates to figure 3 and 4. All available estimates of helper sex ratios in eusocial societies, including information on helper task.

Order	family	species	Colony sex ratio	Worker sex ration	Male helping (1= defense, 2= alloparental care)	Alloparental care (1=provisioning, 2=egg care, 3= transport)	Further comments
<b>Araneae</b>	Theridiidae	<i>Anelosimus eximius</i>	0.08-0.15 [S31]	-	none [S31]	present (1) [S25, S31]	
<b>Coleoptera</b>	Curculionidae	<i>Austroplatypus incompertus</i>	0.5 [S15, S16]	0 (female only) [S15]	none [S15]	present (1,2) [S15, S17]	
<b>Coleoptera</b>	Curculionidae	<i>Xyleborinus saxesnii</i>	0.05-0.12 [S17]	-	none [S17, S32]	present (1,2)	males occasionally allogroom, but this is considered courtship behaviour
<b>Decapoda</b>	Alpheidae	<i>Synalpheus chacei</i>	0.5 [S20, S32]	-	1 [S20]	absent [S20, S32]	
<b>Decapoda</b>	Alpheidae	<i>Synalpheus regalis</i>	0.5 [S20, S32]	-	1 [S20]	absent [S20, S31]	
<b>Hymenoptera</b>	-	All eusocial species	variable	0 (female only)	none (see comment)	in all species studied [S1,S2,S3]	Only a single species shows male cooperative behaviour; the ant <i>Cardiocondyla</i> where males were observed to carry larva
<b>Isoptera</b>	Hodotermitidae	<i>Anacanthotermes ahngerianus</i>		male biased (soldiers+workers) [S33]	yes	present [S24, S27]	
<b>Isoptera</b>	Hodotermitidae	<i>Hodotermes mossambicus</i>		male biased (soldiers) [S33]	yes	present [S24, S27]	
<b>Isoptera</b>	Kalotermitidae	<i>Cryptotermes domesticus</i>	0.5	0.5 (soldiers)	1	absent [S24, S27]	
<b>Isoptera</b>	Kalotermitidae	<i>Cryptotermes secundus</i>	0.5	0.2 (soldiers)	1	absent [S24, S27]	
<b>Isoptera</b>	Kalotermitidae	<i>Incisitermes schwarzi</i>	-	0.55 (0.61) [S33]	1	absent [S24, S27]	
<b>Isoptera</b>	Kalotermitidae	<i>Neotermes connexus</i>	fem biased	fem biased (soldiers) [S33]	1	absent [S24, S27]	
<b>Isoptera</b>	Rhinotermitidae	<i>Coptotermes lacteus</i>		fem biased (soldiers) [S33]	1	absent [S24, S27]	
<b>Isoptera</b>	Rhinotermitidae	<i>Prorhinotermes simplex</i>	0.5 [S25, S34]	0.5 (soldier) [S34, S35]	1+2	absent [S24, S27]	
<b>Isoptera</b>	Rhinotermitidae	<i>Reticulitermes flavipes</i>	≈ 0.5 [S35, S36]	≈ 0.5 [S35, S36]	1+2	present [S24, S27]	
<b>Isoptera</b>	Rhinotermitidae	<i>Reticulitermes kanmonensis</i>		≈ 0.4 [S25, S36]	1+2	present [S24, S27]	
<b>Isoptera</b>	Rhinotermitidae	<i>Reticulitermes speratus</i>		0.5 [S36]	1+2	present [S24, S27]	
<b>Isoptera</b>	Rhinotermitidae	<i>Reticulitermes virginicus</i>		female only [S36]	1+2	present [S24, S27]	
<b>Isoptera</b>	Rhinotermitidae	<i>Schedorhinotermes lamanianus</i>		female biased (soldiers+pseudoworkers) [S33]	1+2	present [S24, S27]	
<b>Isoptera</b>	Termitidae	<i>Amitermes evuncifer</i>		unbiased (soldiers) [S33]	1+2	present [S24, S27]	
<b>Isoptera</b>	Termitidae	<i>Constrictotermes cyphergaster</i>		male biased (soldiers+workers) [S37]	1+2	present [S24, S27]	
<b>Isoptera</b>	Termitidae	<i>Cornitermes walkeri</i>		male biased (soldiers+workers) [S33]	1+2	present [S24, S27]	
<b>Isoptera</b>	Termitidae	Macrotermitinae		fem biased (soldiers) [S33]	1+2	present [S24, S27]	
<b>Isoptera</b>	Termitidae	<i>Mastotermes darwiniensis</i>	≈ 0.5 [S25, S34, S38]		1+2	present [S24, S27]	
<b>Isoptera</b>	Termitidae	<i>Microcerotermes fuscotibialis</i>		fem biased (soldiers) [S33]	1+2	present [S24, S27]	
<b>Isoptera</b>	Termitidae	<i>Nasutitermes arborum</i>		male biased (soldiers) [S33]	1+2	present [S24, S27]	
<b>Isoptera</b>	Termitidae	<i>Rhynchotermes perarmatus</i>		male biased (workers) + fem biased (soldiers) [S33]	1+2	present (1,2,3)	
<b>Isoptera</b>	Termitidae	<i>Sphaerotermes</i>		male biased (soldiers) [S33]	1+2	present (1,2,3)	

		<i>sphaerotherax</i>						
<b>Isoptera</b>	Termitidae	<i>Termes hospes</i>		fem biased (soldiers) [S33]	1+2	present (1,2,3)		
<b>Isoptera</b>	Termitidae	<i>Trinervitermes spp.</i>		fem biased (workers) + male biased (soldiers) [S33]	yes	present (1,2,3)		
<b>Isoptera</b>	Termopsidae,	<i>sp.</i>		unbiased (soldiers) [S33]				
<b>Rodentia</b>	Bathyergidae	<i>Fukomys damarensis</i>	0.59 [S39, S40], 0.22-0.29 [S39, S41]	-	1+2 [S30, S41]	present (3)		
<b>Rodentia</b>	Bathyergidae	<i>Heterocephalus glaber</i>	slight male bias [S42]	-	1+2 [S40, S42]	present (1,3)		
<b>Thysanoptera</b>	Phlaeothripidae	<i>Kladothrips hamiltoni</i>	0.94 (dispersers) [S30]	0.45 (soldiers) [S30, S43]	1 [S15, S16, S43]	absent		
<b>Thysanoptera</b>	Phlaeothripidae	<i>Oncothrips habrus</i>	0.49 (dispersers) [S30]	0.89 (soldiers) [S30, S43]	1 [S30, S43]	absent		High % soldier reproduction, biased soldier SR due to LMC
<b>Thysanoptera</b>	Phlaeothripidae	<i>Oncothrips morrissi</i>	0.69 (dispersers) [S30]	0.69 (soldiers) [S30]				High % soldier reproduction, biased soldier SR due to LMC
<b>Thysanoptera</b>	Phlaeothripidae	<i>Oncothrips tepperi</i>	0.89 (dispersers) [S30]	0.89 (soldiers) [S30, S43]	1 [S30, S43]	absent		High % soldier reproduction, biased soldier SR due to LMC
<b>Thysanoptera</b>	Phlaeothripidae	<i>Oncothrips waterhousei</i>	0.49 (dispersers) [S30]	0.88 (soldiers) [S30, S43]	1 [S30, S43]	absent		High % soldier reproduction, biased soldier SR due to LMC



Table S4: Relates to figure 4. Full summary table comparative analysis. The intercepts shows the estimate for diploid taxa with helpers that preform broodcare and a ancestral biparental care system. All other fixed effects are displayed as the difference from the intercept. We also estimated the phylogenetic signal (the binary equivalent to Pagel's lamda), which was 0.11 (CI95%: 0.002-0.47).

	Posterior mean	95% Credibility interval	Effective samples	pMCMC
<b>FIXED EFFECTS</b>				
<b>Intercept (broodcare.biparental +diploidy)</b>	-5.32	-10.87 - -1.08	1201	0.004**
<b>broodcare.maternal</b>	9.51	2.04 - 10.2	1000	0.001**
<b>Defense</b>	-3.50	-11.21 - 3.58	1000	0.382
<b>haplodiploidy</b>	2.63	-3.41 - 9.77	1392	0.416
<b>RANDOM EFFECTS</b>				
<b>phylogeny</b>	1.29	0.01 - 3.87	1000	

Table S5: Relates to figure 4. Full summary table comparative analysis for the restricted model that excludes those taxa considered “cooperative breeders” by Crespi and Yanega [S44, S45] (indicated with an asterisks in table S2). The intercepts shows the estimate for diploid taxa with helpers that preform broodcare and a ancestral biparental care system. All other fixed effects are displayed as the difference from the intercept. We also estimated the phylogenetic signal (the binary equivalent to Pagel’s lamda), which was 0.14 (CI95%: 0.003-0.67).

	Posterior mean	95% Credibility interval	Effective samples	pMCMC
<b>FIXED EFFECTS</b>				
<b>Intercept (broodcare.biparental +diploidy)</b>	-4.97	-10.40 – -0.046	1049	0.032*
<b>broodcare.maternal</b>	9.24	2.88 – 16.13	1000	<0.001**
<b>Defense</b>	-3.41	-11.14 – 4.36	1000	0.430
<b>haplodiploidy</b>	2.21	-4.11 – 9.04	1000	0.504
<b>RANDOM EFFECTS</b>				
<b>phylogeny</b>	2.15	0.011 – 8.79	1000	

Table S6: Relates to figure 2. Consanguinities of family members under diploidy and haplodiploidy, assuming outbreeding.

<b>Actor</b>	<b>Consanguinity</b>				
	Self	Daughter	Son	Sister	Brother
<i>Diploidy</i>					
Female	1/2	1/4	1/4	1/4	1/4
Male	1/2	1/4	1/4	1/4	1/4
<i>Haplodiploidy</i>					
Female	1/2	1/4	1/2	3/8	1/4
Male	1	1/2	0	1/4	1/2

## Supplemental Experimental Procedures:

### Assignment of helper sex ratio and helper task

#### *Helper sex ratio*

In general, we consider the helper sex ratio female only if more than 90% of helpers are female (sex ratio of 0.1) or if males have never been observed performing cooperative behaviours. In some species, there are only colony-level estimates of the sex ratio, in which case we use these estimates. In some taxa, both worker and reproductives sex ratios are highly skewed, in which case we assign female-biased helping only when the sex ratio of the helpers is more female-biased than that of the reproductives. The only exception to these rules are for determining the assignments for the thrips and the termites. Soldier sex ratio in both groups appears to be variable, yet in both cases this variation has appeared after the origin of eusociality. In thrips male soldier are present in all species studied but the sex ratio of soldiers is biased (in comparison to the reproductives) in a few of the more derived species, this bias is absent however in the most ancestral eusocial taxa and can be understood in terms of worker reproduction and local mate competition [S13, S30, S33, S46]. Likewise although several termites have a biased soldier caste this again appears to be a derived state [S33, S42, S46] not present either at the origin of eusociality or at the origins of a true worker caste. Therefore for both thrips and the primitive and advanced termites we have assigned the helper sex ratio as unbiased.

#### *Helper task*

Helpers in eusocial societies often engage in both broodcare and defense. However the importance of these two types of behaviours at the origin of eusociality differ (referred to as fortress defense vs. life insurance, Crespi, Queller). For example soldiers appeared at the origin of eusociality in thrips and termites, and primitive termites and thrips do not display alloparental broodcare, while all eusocial Hymenoptera provide some form of alloparental care. We expect alloparental care to be sex-linked if there is a pre-existing bias toward maternal care, while we do not expect such bias for defensive tasks. We have therefore divided the eusocial taxa in those that 1) *never* provide broodcare and those that 2) *do* provide some form of broodcare (evolving either from maternal or paternal care). We recognize three different behaviors as alloparental care: (1) direct provisioning of the young; (2) care for the eggs; and (3) transport of eggs and young. We indicate the presence of these behaviours in table S3

## R Code and prior used for the analysis in the MCMCglmm package [S44]

```
library(MCMCglmm)

eus<-read.table("/Users/lauraross/eusoc_analysis_dataframe.txt",header=T)
tree1300.eus <- read.tree("/Users/lauraross/eusocial_1300.trees")

INtree.eus<- inverseA(tree1300.eus[[1]], nodes="TIPS")

gelmanprior<-list(B=list(mu=c(0,0,0,0),V=gelman.prior(~ecology+genetic.sys, data=eus,
scale=1+1+pi^2/3)), R=list(V=1,fix=1),G=list(G1=list(V=diag(1)*0.1, nu=1)))

mphyLMCMC.start<-MCMCglmm(helper.sex~ecology+genetic.sys, random=~animal, data=eus,
ginverse=list(animal=INtree.eus$Ainv), family="categorical", prior=gelmanprior,
pl=TRUE,slice=TRUE, nitt=1000, thin=1, burnin=0)

mphyLMCMC<-mphyLMCMC.start

for(i in 1:1300){

INtree.eus<- inverseA(tree1300.eus[[i]],nodes="TIPS")

start<-list(Liab=mphyLMCMC$Liab[1,], R=1, G=list(G1=mphyLMCMC$VCV[1,1]))

mphyLMCMC<-MCMCglmm(helper.sex~ecology+genetic.sys, random=~animal, data=eus,
family="categorical", prior=gelmanprior,
ginverse=list(animal=INtree.eus$Ainv), pl=TRUE,slice=TRUE, nitt=1000, thin=1,
burnin=999, start=start, verbose=FALSE)

if(i>300){
mphyLMCMC.start$VCV[i-300,]<-mphyLMCMC$VCV[1,]
mphyLMCMC.start$Sol[i-300,]<-mphyLMCMC$Sol[1,]
mphyLMCMC.start$Liab[i-300,]<-mphyLMCMC$Liab[1,]
}

print(i)
}

summary(mphyLMCMC.start)
hist(mphyLMCMC.start$Liab)
hist(mphyLMCMC.start$VCV[,1]/(rowSums(mphyLMCMC.start$VCV)+pi^2/3))
mean(mphyLMCMC.start$VCV[,1]/(rowSums(mphyLMCMC.start$VCV)+pi^2/3))
HPDinterval(mphyLMCMC.start$VCV[,1]/(rowSums(mphyLMCMC.start$VCV)+pi^2/3))

write.table(predict(mphyLMCMC.start, interval = "confidence"),"/Users/lauraross/
eusoc_analysis_predictions.csv",sep = ",")
```

**Analysis notes:** We marginalized over the posterior distribution of trees by sampling a tree from the posterior at iteration  $t$ , running the MCMC comparative analysis for 1000 iterations and saving

the last MCMC sample. The values of the latent variables and variance components were passed as starting values to the analysis at iteration  $t+1$  for which a new tree from the posterior sample was taken. The process was repeated for 1300 iterations (i.e.  $t=1300$ ) disposing the first 300 iterations as burnin. If the posterior distribution of the tree topology is variable then it is not clear how the latent variables at ancestral nodes from tree  $t$  should map onto the latent variables at ancestral nodes from tree  $t+1$  when defining the starting value.

Given that the introduction of latent variables at ancestral nodes was a computational trick in order to exploit algorithms for solving sparse linear systems rather than a necessity for valid inference, we ran models in which the parameter space was not augmented with the ancestral states and the only latent variables were associated with extant taxa which could be defined consistently between trees. For large phylogenies this may prove to be too slow, in which case 1000 independent MCMC analyses (one for each tree) could be performed, although the number of iterations per tree would probably have to be increased with a suitable burnin period to ensure that the posterior distribution of the comparative analysis parameters is adequately sampled.

## Theory: Why doesn't the genetics of sex determination matter?

Inclusive fitness is gained by sending copies of one's genes into future generations. Hence, an actor values a recipient according to how well the recipient transmits copies of the actor's genes to future generations [S47]. This is the product of two quantities: their consanguinity ( $p$ ), i.e. the extent to which the actor and recipient share genes in common; and the recipient's reproductive value ( $v$ ), i.e. the extent to which they transmit their own genes to future generations [S48-50]. The consanguinities for diploid and haplodiploid family members are summarized in Table S6. The reproductive values are  $v_{\text{♀}} = 1/(1-z)$  for females and  $v_{\text{♂}} = 1/z$  for males under diploidy, and  $v_{\text{♀}} = 2/(1-z)$  for females and  $v_{\text{♂}} = 1/z$  for males under haplodiploidy, where  $z$  is the population sex ratio (proportion male).

Thus, in a diploid population, a female who raises  $b$  extra siblings at a cost of  $c$  to her own reproduction increases her inclusive fitness by an amount  $b \times (z \times p_{\text{Bro}|♀} \times v_{\text{♂}} + (1-z) \times p_{\text{Sis}|♀} \times v_{\text{♀}}) - c \times (z \times p_{\text{Son}|♀} \times v_{\text{♂}} + (1-z) \times p_{\text{Dau}|♀} \times v_{\text{♀}})$ , which is positive if  $c/b < 1/2$ . For a male, the calculation is  $b \times (z \times p_{\text{Bro}|♂} \times v_{\text{♂}} + (1-z) \times p_{\text{Sis}|♂} \times v_{\text{♀}}) - c \times (z \times p_{\text{Son}|♂} \times v_{\text{♂}} + (1-z) \times p_{\text{Dau}|♂} \times v_{\text{♀}})$ , which is also positive if  $c/b < 1/2$ . In a haplodiploid population, the calculation for a female is  $b \times (z \times p_{\text{Bro}|♀} \times v_{\text{♂}} + (1-z) \times p_{\text{Sis}|♀} \times v_{\text{♀}}) - c \times (z \times p_{\text{Son}|♀} \times v_{\text{♂}} + (1-z) \times p_{\text{Dau}|♀} \times v_{\text{♀}})$ , yielding exactly the same condition  $c/b < 1/2$ . And for a male the calculation is  $b \times (z \times p_{\text{Bro}|♂} \times v_{\text{♂}} + (1-z) \times p_{\text{Sis}|♂} \times v_{\text{♀}}) - c \times (z \times p_{\text{Son}|♂} \times v_{\text{♂}} + (1-z) \times p_{\text{Dau}|♂} \times v_{\text{♀}})$ , which also yields the condition  $c/b < 1/2$ . Hence, the genetics of sex determination neither predisposes haplodiploid versus diploids to eusociality nor predisposes females versus males to altruistic helping in haplodiploid societies [S1, S51-55].

The above argument has been framed in terms of consanguinity and reproductive value. However, the concept of life-for-life relatedness, which combines these quantities into a single measure of value, is more commonly encountered in the social insect literature. Life-for-life relatednesses are recovered by multiplying the consanguinity of actor and recipient by the recipient's reproductive value, and dividing by the product of the actor's consanguinity to self and the actor's own reproductive value. For example, this recovers the familiar  $3/4$  relatedness between haplodiploid sisters:  $R_{\text{Sis}|♀} = (p_{\text{Sis}|♀} \times v_{\text{♀}})/(p_{\text{Self}|♀} \times v_{\text{♀}}) = 3/4$ . Assuming an even sex ratio, the other consanguinities are:  $R_{\text{Bro}|♀} = 1/4$ ,  $R_{\text{Dau}|♀} = 1/2$ ,  $R_{\text{Son}|♀} = 1/2$ ,  $R_{\text{Sis}|♂} = 1/2$ ,  $R_{\text{Bro}|♂} = 1/2$ ,  $R_{\text{Dau}|♂} = 1$ ,  $R_{\text{Son}|♂} = 0$  under haplodiploidy; and  $R_{\text{Sis}|♀} = R_{\text{Bro}|♀} = R_{\text{Dau}|♀} = R_{\text{Son}|♀} = R_{\text{Sis}|♂} = R_{\text{Bro}|♂} = R_{\text{Dau}|♂} = R_{\text{Son}|♂} = 1/2$  under diploidy. The consequent equal valuation of siblings and offspring under both genetic modes is illustrated in Figure S3, and the above argument framed in terms of consanguinity and reproductive value reveals that this result continues to hold even when sex ratios are biased.

## Supplemental References:

- S1. Trivers, R. L., and Hare, H. (1976). Haplodiploidy and the evolution of the social insects. *Science* 191, 249–263.
- S2. Hughes, W. O. H., Oldroyd, B. P., Beekman, M., and Ratnieks, F. L. W. (2008). Ancestral Monogamy Shows Kin Selection Is Key to the Evolution of Eusociality. *Science* 320, 1213–1216.
- S3. Hölldobler, B., and Wilson, E. O. (1990). *The ants* (Cambridge, Massachusetts: Belknap Press).
- S4. Toth, A. L., Varala, K., Newman, T. C., Miguez, F. E., Hutchison, S. K., Willoughby, D. A., Simons, J. F., Egholm, M., Hunt, J. H., Hudson, M. E., et al. (2007). Wasp Gene Expression Supports an Evolutionary Link Between Maternal Behavior and Eusociality. *Science* 318, 441–444.
- S5. Amdam, G. V., Csondes, A., Fondrk, M. K., and Page, R. E. (2006). Complex social behaviour derived from maternal reproductive traits. *Nature* 439, 76–78.
- S6. Jeanne, R. L. (1980). Evolution of social behavior in the Vespidae. *Annu. Rev. Entomol.* 25, 371–396.
- S7. Reeve, H. K. (1993). Haplodiploidy, Eusociality and Absence of Male Parental and Alloparental Care in Hymenoptera: A Unifying Genetic Hypothesis Distinct from Kin Selection Theory. *Philos T Roy Soc B* 342, 335–352.
- S8. Strassmann, J. E., Hughes, C. R., Turillazzi, S., Solís, C. R., and Queller, D. C. (1994). Genetic relatedness and incipient eusociality in stenogastrine wasps. *Animal Behaviour* 48, 813–821.
- S9. Michener, C. D. (1974). *The social behavior of the bees: a comparative study* (Cambridge, Massachusetts: Belknap Press).
- S10. Bennett, N. C., and Faulkes, C. G. (2000). *African Mole-Rats* (Cambridge University Press).
- S11. Burda, H. (1999). Syndrome of eusociality in African subterranean mole-rats (Bathyergidae, Rodentia), its diagnosis and aetiology (New York City: Springer).
- S12. Crespi, B. J. (1992). Eusociality in Australian gall thrips. *Nature* 359, 724–726.
- S13. Kranz, B. D., Schwarz, M. P., Mound, L. A., and Crespi, B. J. (2001). Social biology and sex ratios of the eusocial gall-inducing thrips *Kladothrips hamiltoni*. *Ecol Entomol* 24, 432–442.
- S14. Smith, S., Beattie, A., Kent, D., and Stow, A. (2009). Ploidy of the eusocial beetle



*Austroplatypus incompertus* (Schedl)(Coleoptera, Curculionidae) and implications for the evolution of eusociality. *Insectes Sociaux* 56, 285–288.

- S15. Kent, D. S., and Simpson, J. A. (1992). Eusociality in the beetle *Austroplatypus incompertus* (Coleoptera: Curculionidae). *Naturwissenschaften* 79, 86–87.
- S16. Choe, J. C., and Crespi, B. J. (1997). *The Evolution of Social Behaviour in Insects and Arachnids* (Cambridge: Cambridge University Press).
- S17. Biedermann, P. H. W., and Taborsky, M. (2011). Larval helpers and age polyethism in ambrosia beetles. *Proceedings of the National Academy of Sciences* 108, 17064–17069.
- S18. Normark, B. B., Jordal, B., and Farrell, B. (1999). Origin of a haplodiploid beetle lineage. *P Roy Soc Lond B Bio* 266, 2253–2259.
- S19. Duffy, J. E., Morrison, C. L., and Ríos, R. N. (2000). Multiple origins of eusociality among sponge-dwelling shrimps (*Synalpheus*). *Evolution* 54, 503–516.
- S20. Duffy, J. E. (1996). Eusociality in a coral-reef shrimp. *Nature* 381, 512–514.
- S21. Rypstra, A. L. (1993). Prey size, social competition, and the development of reproductive division of labor in social spider groups. *Am Nat* 142, 868–880.
- S22. Samuk, K. M., LeDue, E. E., and Aviles, L. (2011). Sister clade comparisons reveal reduced maternal care behavior in social cobweb spiders. *Behav Ecol* 23, 35–43.
- S23. Legendre, F., Whiting, M. F., Bordereau, C., Canello, E. M., Evans, T. A., and Grandcolas, P. (2008). The phylogeny of termites (Dictyoptera: Isoptera) based on mitochondrial and nuclear markers: implications for the evolution of the worker and pseudergate castes, and foraging behaviors. *Molecular Phylogenetics and Evolution* 48, 615–627.
- S24. Inward, D. J., Vogler, A. P., and Eggleton, P. (2007). A comprehensive phylogenetic analysis of termites (Isoptera) illuminates key aspects of their evolutionary biology. *Molecular Phylogenetics and Evolution* 44, 953–967.
- S25. Thorne, B. L. (1997). Evolution of eusociality in termites. *Annu Rev Ecol Syst* 28, 27–54.
- S26. Korb, J., Buschmann, M., Schafberg, S., Liebig, J., and Bagnères, A. G. (2012). Brood care and social evolution in termites. *P R Soc B* 279, 2662–2671.
- S27. Korb, J., Buschmann, M., Schafberg, S., Liebig, J., and Bagnères, A.-G. (2012). Brood care and social evolution in termites. *P R Soc B* 279, 2662–2671.
- S28. Rosengaus, R. B., and Traniello, J. F. A. (1991). Biparental care in incipient colonies of the dampwood termite *Zootermopsis angusticollis* Hagen (Isoptera: Termopsidae). *J*

*Insect Behav* 4, 633–647.

- S29. Inward, D., Beccaloni, G., and Eggleton, P. (2007). Death of an order: a comprehensive molecular phylogenetic study confirms that termites are eusocial cockroaches. *Biology Letters* 3, 331–335.
- S30. Chapman, T. W. (2002). The evolution of soldier reproduction in social thrips. *Behav Ecol* 13, 519–525.
- S31. Vollrath, F. (1986). Eusociality and extraordinary sex ratios in the spider *Anelosimus eximius* (Araneae: Theridiidae). *Behav Ecol Sociobiol* 18, 283–287.
- S32. Tóth, E., and Bauer, R. T. Gonopore sexing technique allows determination of sex ratios and helper composition in eusocial shrimps. *Marine Biology* 151, 1875–1886.
- S33. Roisin, Y. (2001). Caste sex ratios, sex linkage, and reproductive strategies in termites. *Insectes Sociaux* 48, 224–230.
- S34. Roisin, Y. Morphology, development and evolutionary significance of the working stages in the caste system of *Prorhinotermes* (Insecta, Isoptera). *Z. Morph. u. Okol. Tiere* 107, 339–347.
- S35. Dean, S. R., and Gold, R. E. (2004). Sex Ratios and Development of the Reproductive System in Castes of *Reticulitermes flavipes* (Kollar) (Isoptera: Rhinotermitidae). *Ann Entomol Soc Am* 97, 147–152.
- S36. Matsuura, K. (2006). A novel hypothesis for the origin of the sexual division of labor in termites: which sex should be soldiers? *Evol Ecol* 20, 565–574.
- S37. Moura, F. M. S., Vasconcellos, A., Silva, N. B., and Bandeira, A. G. (2010). Caste development systems of the Neotropical termite *Constrictotermes cyphergaster* (Isoptera, Termitidae). *Insectes Sociaux* 58, 169–175.
- S38. Nalepa, C. A., Miller, L. R., and Lenz, M. Flight characteristics of *Mastotermes darwiniensis* (Isoptera, Mastotermitidae). *Insectes Sociaux* 48, 144–148.
- S39. Jacobs, D. S., Bennett, N. C., Jarvis, J. U. M., and Crowe, T. M. (1991). The colony structure and dominance hierarchy of the Damaraland mole-rat, *Cryptomys damarensis* (Rodentia: Bathyergidae), from Namibia. *J Zoology* 224, 553–576.
- S40. Hazell, R. W. A., Bennett, N. C., Jarvis, J. U. M., and Griffin, M. (2000). Adult dispersal in the co-operatively breeding Damaraland mole-rat (*Cryptomys damarensis*): a case study from the Waterberg region of Namibia. *J Zoology* 252, 19–25.
- S41. Bennett, N. C., and Jarvis, J. U. (1988). The social structure and reproductive biology of colonies of the mole-rat, *Cryptomys damarensis* (Rodentia, Bathyergidae). *Journal of Mammalogy*, 293–302.

- S42. Sherman, P. W., Jennifer U. M.) Jarvis, J. U. M., and Alexander, R. D. (1991). *The Biology of the Naked Mole-Rat*.
- S43. Crespi, B. J., and Mound, L. A. (1997). Ecology and evolution of social behavior among Australian gall thrips and their allies. *The evolution of social behavior in insects and arachnids*, 166–180.
- S44. Hadfield, J. D. (2010). MCMC Methods for Multi-Response Generalized Linear Mixed Models: The MCMCglmm R Package. *Journal of Statistical Software* 33, 1–22.
- S45. Crespi, B. J., and Yanega, D. (1995). The definition of eusociality. *Behav Ecol* 6, 109–115.
- S46. Abe, T., Bigness, D. E., and Higashi, M. (2000). *Termites: evolution, sociality, symbioses, ecology* (New York City: Springer).
- S47. Hamilton, W. (1964). The genetical evolution of social behaviour. I. *Journal of Theoretical Biology*.
- S48. Fisher, R. A. (1930). *The Genetical Theory of Natural Selection* (Oxford: Clarendon Press).
- S49. Bulmer, M. (1994). *Theoretical Evolutionary Ecology* (Sunderland, Massachusetts: Sinauer).
- S50. Hamilton, W. D. (1972). Altruism and related phenomena, mainly in social insects. *Annu Rev Ecol Syst*, 193–232.
- S51. Charlesworth, B. (1978). Some models of the evolution of altruistic behaviour between siblings. *Journal of Theoretical Biology* 72, 297–319.
- S52. Charnov, E. L. (1978). Evolution of eusocial behavior: offspring choice or parental parasitism? *Journal of Theoretical Biology* 75, 451–465.
- S53. Craig, R. (1982). Evolution of male workers in the Hymenoptera. *Journal of Theoretical Biology* 94, 95–105.
- S54. Pamilo, P. (1991). Evolution of the sterile caste. *Journal of Theoretical Biology* 149, 75–95.
- S55. Bourke, A. F. G., and Franks, N. R. (1995). *Social Evolution in Ants* (Princeton, New Jersey: Princeton University Press).