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EFFECTS OF HABITAT SUCCESSION ON POPULATION AND REPRODUCTION OF THE ALLEGHENY MOUND ANT

By

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ABSTRACT

EFFECTS OF HABITAT SUCCESSION ON POPULATION AND REPRODUCTION OF THE ALLEGHENY MOUND ANT

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Reliance on transient habitats makes *Formica. exsectoides* Forel, the Allegheny mound ant, useful in the study of reproductive allocation as an adaptive strategy for coping with environmental change. The sex ratio of *F. exsectoides* was determined to be largely male-biased, yet variable among populations. Sex ratio estimates were influenced by sexual dimorphism in weight, but not energy content, and stage of pupal development. Population density and reproductive allocation were examined across a gradient of jack pine stand ages, grouped into young (2-16 y), mature (37-45 y) and old (66-77 y) classes. Forager abundance was greatest in mature stands, and declined in old stands. Composition of ground arthropod communities shifted as stands matured; the strongest change was an increase in predaceous beetles in old stands, possibly creating competition between beetles and *F. exsectoides* for protein resources. While this may cause decline of *F. exsectoides* in old stands, light availability may also play a role. Light environment was linked to mound reproductive status—mounds in low light produced only worker brood, mounds in intermediate light produced sexual-caste brood, and mounds in low light produced no brood. This suggests that low light makes older stands unsuitable for brood production, and that production of sexual forms may be a dispersal response to deteriorating conditions. Sex ratio was not associated with any measured variables related to habitat succession, but was related to population age.

This is dedicated to next year's brood, who will escape me.

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CHAPTER 1

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Introduction

Formica exsectoides Forel is a cosmopolitan and ecologically important ant species in the eastern United States. Ranging from Nova Scotia to northern Georgia and west to Ontario and Illinois, these ants are usually found in forest edges or openings (Creighton 1950). Often recognized by their conspicuous mound architecture, populations have been studied with respect to physical structure, placement, and orientation of mounds (McCook 1877, Andrews 1926, Cory & Haviland 1938, Dimmick 1951).

F. exsectoides is an aggressive predator (Ayre 1963) and has been investigated for its potential in biocontrol of forest pests (Allen *et. al.* 1970, Campbell 1991, Campbell *et. al.* 1991). *F. exsectoides* is also considered a pest species, mainly due to its practice of killing young trees in proximity to mounds (Andrews 1926, Wilson 1977, Patterson 1994), and its generally aggressive behavior (Levins and Traniello 1981, personal observation). Investigations into its control include Price (1945), Haviland (1947), and Weaver and Smith (1993). *F. exsectoides* has also been shown to affect the community structure of phloem-feeding insects through mutualistic associations with Homopterans (Andrews 1929, Bishop 1998). Despite this obvious interest in economic influences of *F. exsectoides*, knowledge of its basic biology is lacking.

In Michigan, *F. exsectoides* is commonly found in jack pine (*Pinus banksiana* Lamb.) ecosystems. *F. exsectoides* exhibits a patchy use of this habitat and can form large, locally dense populations. *F. exsectoides* colonies possess multiple queens (polygyny) (Bristow *et. al.* 1992) and may occupy multiple mounds (polydomy) (Nipson 1978, Oster and Wilson 1978). Colony expansion occurs by budding or production of sexual forms. Sexual forms are produced once a year in Michigan, with pupation occurring in late July/early August. No mating swarms have been observed—females exhibit calling by pheromone (Hölldobler and Wilson 1990), attracting males to mate on low shrubs and saplings. Mated queens may found new colonies by temporary social parasitism of *F. fusca* Linn., or may attempt to gain acceptance into an existing *F. exsectoides* mound (Wheeler 1933; Creighton 1950, Starr 1979).

The sex ratio of *F. exsectoides* is unknown. Knowledge of sex ratio is important for understanding the reproductive ecology of an organism. In ants, sex is determined by one or more loci, where heterozygotes are female and hemi- or homozygous individuals are male (Crozier 1971, Bull 1983). For simplicity, it can be generalized that females are diploid (arising from fertilized eggs) and males are haploid. This system of sex determination, coupled with caste differentiation displayed by highly eusocial Hymenoptera, places sex ratio on a behavioral, rather than evolutionary, time-scale of response. The lability of this trait makes it an important source of reproductive variation among species.

The primary objective of this study was to estimate the sex ratio of *F. exsectoides*. As well as providing long-overdue knowledge of the reproductive ecology of this species, results may contribute to the growing power of sex ratio meta-analyses to inform on

important evolutionary questions. Haplo-diploidy creates elevated relatedness between sisters, since sisters share an average of 75% of their genetic material, while parent and offspring share 50%, and sisters and brothers share (on average) only 25%.
Due to these relatedness asymmetries between siblings, studies of Hymenopteran sex ratios can provide valuable tests of kin selection theory, and insight into parent-offspring conflict, the mechanisms of social evolution and the maintenance of sex (Hamilton 1964, Trivers and Hare 1976, Charnov 1982, for review see Nonacs 1986, Pamilo and Crozier 1996). Individual studies are of limited utility in approaching such general questions. Syntheses of many sex ratio studies can be more informative, provided that the studies incorporated are compatible and relevant. Sex ratio data from *F. exsectoides* may also facilitate comparisons with *Formica exsecta* and the more extensively-studied European wood ants of the *Formica rufa* group (Pamilo and Rosengren 1983, Rosengren and Pamilo 1986).

A secondary objective of this study was to determine an appropriate method of estimating *F. exsectoides* sex ratio, by addressing questions of sexual dimorphism and intraspecific sex ratio variation. Measures of sex ratio generated by counting individuals of each sex have been displaced by often sophisticated attempts to quantify allocation of resources to each sex. The simplest of these is the use of weight ratios, correcting for the possibility of sexual size dimorphism. Boomsma (1989) stated that female investment had been "systematically overestimated", as male ants generally have higher respiration rates than females. His correction, supported by energetic data from *Lasius niger* (Boomsma 1989) and additional species (Boomsma *et. al.* 1995), is generally accepted, but should be used with the caveat that not all species will show the same extent of physiological dimorphism, and that metabolic activity may vary over development. Few studies (Boomsma and Isaaks 1985, MacKay 1985) incorporate the overall energy content of sexual forms. Energy content may be expected to differ between sexes, to an extent determined by dispersal patterns and founding biology. The investigation of these questions is vital to the future utility of this study to larger analyses, since combining sex ratio studies without reference to the method and organism of study can create misleading conclusions. While it is not essential that all authors estimate sex ratio by the same method, the products of differing methods must be biologically equivalent to facilitate meaningful comparisons.

Material and Methods

Site locations. Study sites were located in Oscoda, Crawford, and Roscommon counties, in the northern lower peninsula of Michigan (see Table 1 for details on site locations). All sites were noncontiguous and located at least 1 km apart, so populations were discrete. Vegetation consisted predominantly of jack pine (*Pinus banksiana*) with some interspersed red pine (*Pinus resinosa* Aiton), Northern pin oak (*Quercus ellipsoidalis* Hill), and cherry (*Prunus* spp.). Understory vegetation consisted primarily of blueberry (*Vaccinium angustifolium* Aiton), sand cherry (*Prunus pumila* L.), bracken fern (*Pteridum* spp.) and grasses (*Carex* spp.).

Brood collection and measurement. Partial excavations of 66 mounds from 6 sites were performed from July 18 to July 26, 1996. For each mound, a comparative index of mound surface area (longest slope * shortest slope) and the distance to the nearest neighboring mound were recorded. Presence or absence of reproductive caste brood was determined by excavating the mound to approximately 0.5 m below ground level (or until reproductive caste pupae were found). Reproductive caste pupae are easily distinguished from worker pupae by size and coloration.

Samples of 30-90 pupae were collected from mounds found to contain reproductive caste brood. Pupae were dried and weighed. Sex could usually be determined visually after removing the pupal case. Some pupae had not developed enough for sex to be distinguishable by morphology; these were scored as "undeveloped". Stage of pupal development was scored from 0 (undeveloped) to 3 (fully developed, pigmented, ready to

Table1. Pilot study site locations and descriptions

site	location	description
1	Crawford Co., T.25N, r.1W, s.23,24	open, grassy areas with scattered ~2m jack pine, blending back into 51 year-old mature jackpine
2	Crawford Co., T.25N, r.1W, s.10	66 year old jack pine stand surrounding Rock Cemetery; oak understory, grassy clearings created by treefall
3	Roscommon Co., T.24N, r.2W, s.17	mature jackpine with blueberry ground-cover, bordering clearcut, high recreation traffic
4	Crawford Co., T.25N, r.1W, s.26	recent jackpine clearcut. not yet re-planted
5	Oscoda Co., T.25N r.1E, s.19, 24	70 year old jack pine stand with a 1-acre grassy clearing created by wildfire
6	Oscoda Co., T.25N r.1E, s.10, 15	50-60 year old jack pine with large grassy clearings

eclose). Figure 1 presents representative photographs of pupal development stages. Stages presented are 1, 2, and 3, but half-stages were used where development was intermediate. Analysis of covariance was used to analyze pupal dry weights, with sex as the primary analysis variable and stage of development as the covariate (PROC ANCOVA, SAS Systems 1990).

Energy content. Caloric densities of pupae were measured using a semi-micro calorimeter (Parr Instrument Company, Moline, IL). Heat of combustion in calories/g (H_c) was calculated by the following equation:

$$H_c = ((B * \Delta T) - ((f_1 - f_2) * 1400)) / \text{sample weight}$$

where B is a caloric constant, calculated through calibration with benzoic acid, ΔT is the change in temperature occurring during combustion, and $(f_1-f_2)*1400$ is a correction factor accounting for the amount of fuse wire used to ignite the sample.

Since measurement accuracy declines sharply in samples below 0.01g weight (Terry Trier, personal communication), 3-5 individuals of each sex were measured together. At least three samples from each site were analyzed to determine energy content of males; it was often not possible to analyze three (or any) samples of females, as fewer females were available. The data were analyzed using a nested analysis of variance, in the following hierarchy: days (mounds (sex))). The variable "days" accounts for variation in calorimeter performance during the course of the analysis.

Calculation of sex ratio. Sex ratio was estimated numerically (number of males/total number of pupae) and by dry weight (dry weight of males/ dry weight of total sample). "Undeveloped" pupae were not included. As pupae within samples were usually of the

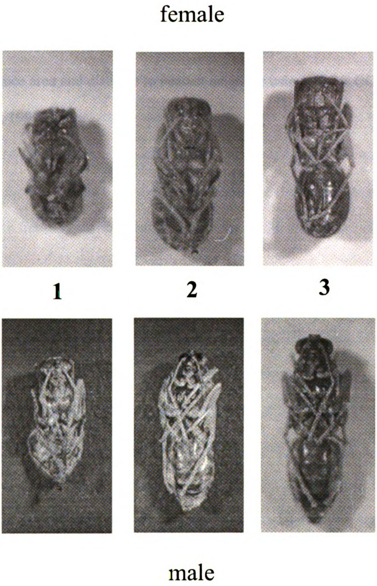

Figure 1. Representative stages of development of male and female pupae

same developmental stage, weights were not corrected for developmental stage. Numerical and weight-based sex ratio estimates were compared to each other using a paired t-test. Weight-based sex ratios were corrected using Boomsma's (1989) energetic cost ratio, and these corrected ratios were compared between mounds and between sites using a nested analysis of variance. Boomsma-corrected sex ratios were also correlated to mound surface area and distance to nearest neighbor using PROC CORR (SAS Systems, Inc., 1990).

Results

Mound measurements and collection of brood. Reproductive pupae were only found in 37 of 66 partially excavated mounds. Of these 37, only 33 mounds contained sufficient pupae (25-30) for sex ratio sampling. Table 2 provides a summary of excavations, sampling, and sex ratios for each site. Sites 4 and 6 contained relatively few mounds; all mounds within visual range were examined. A total of 1510 pupae were collected across the six sites. Of these, 134 had not developed sufficiently for sex to be morphologically distinguishable; these were classified as "undeveloped", and are subtracted from the total pupae sampled to provide the "# of pupae for ratios" shown in Table 2.

Mound surface area ranged from 0.9 m^2 to 12.5 m^2. Distance to nearest neighboring mound ranged from 1 m to 51 m. Only active/inhabited mounds were counted as "neighbors". Table 3 lists the mean and standard deviation of mound surface area indices and distance to nearest neighbor recorded at each site. The mean surface area index for mounds found to produce sexual brood was marginally greater than that for mounds that produced only worker brood ($p = 0.0560$, Table 4), but this relationship was variable among sites (Figure 3). Distance from nearest neighboring mound did not appear to be associated with production of sexual brood ($p = 0.8662$, Table 5; Figure 4).

Weight & Development. Females were heavier than males. The mean female weight (least squares mean, corrected for developmental stage using ANCOVA) across all sites

Table 2. Sampling data and sex ratios. by site. Boomsma cost ratio is generated by correcting dry weight ratios with Boomsma's (1989) energetic cost ratio: (weight males/weight total)$^{0.7}$

site	mounds excavated	mounds sampled	total pupae sampled	#pupae for ratios	numerical ratio	dry weight ratio	Boomsma cost ratio
1	10	7	347	301	0.791	0.768	0.831
2	13	8	389	319	0.902	0.882	0.916
3	14	7	382	348	0.820	0.781	0.841
4	5	2	66	61	0.453	0.384	0.512
5	20	6	232	222	0.658	0.605	0.704
6	5	3	114	96	0.959	0.952	0.966

Table 3. Site means and standard deviations of surface area and distance to nearest neighboring mound.

site	mound surface area (m^2)		distance to nearest neighbor (m)	
	mean	stdev	mean	stdev
1	5.119	2.02	11.103	10.09
2	4.715	1.97	11.834	7.54
3	5.337	3.81	14.537	13.85
4	4.001	1.35	19.507	10.16
5	5.402	3.64	12.763	13.67
6	3.639	1.75	25.298	16.51

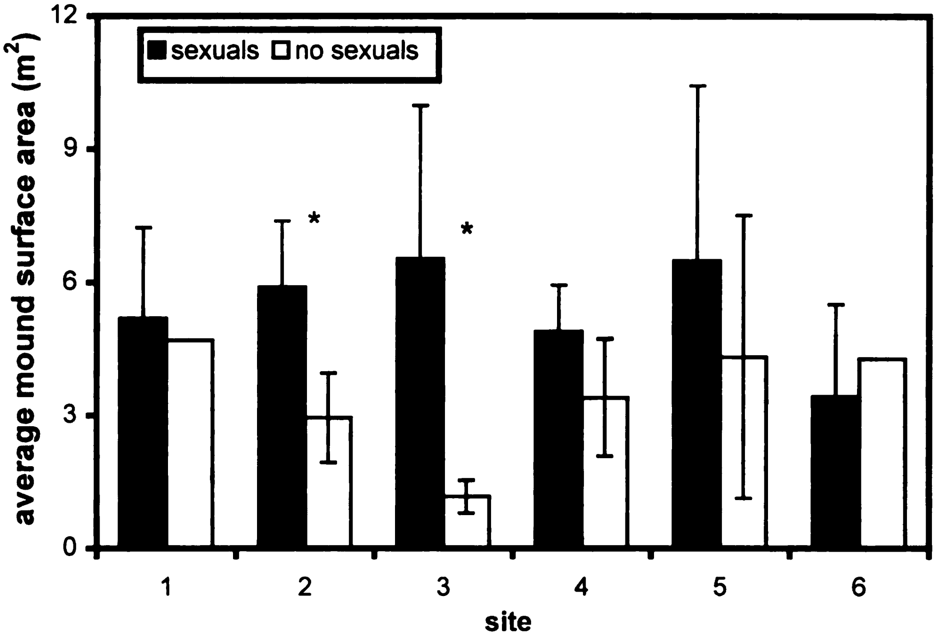

Figure 2. Mounds that produced sexual forms were larger in comparative surface dimensions than those that did not. Error bars represent one standard deviation.

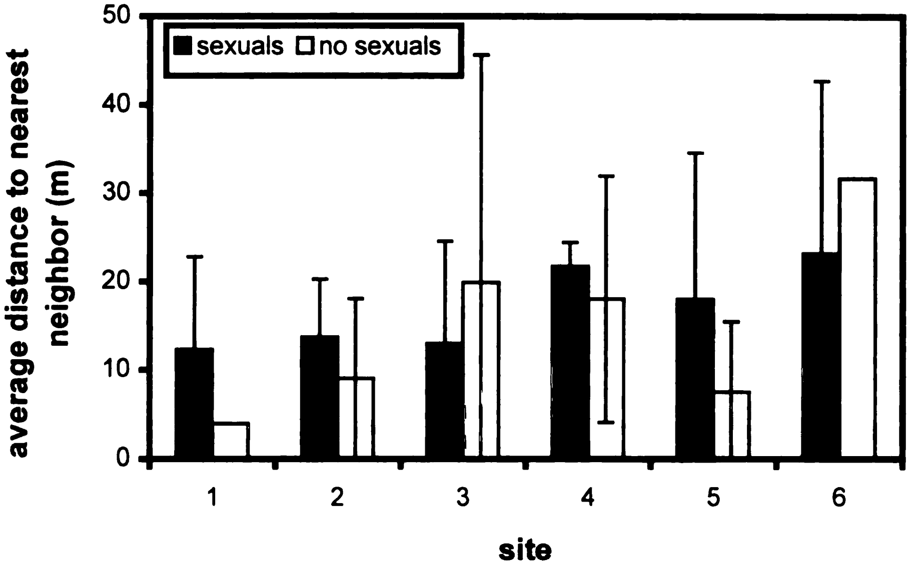

Figure 3. Distance from nearest neighboring mound is not related to production of sexual forms. Error bars represent one standard deviation.

Table 4. ANOVA test of H_o: Surface area indices of mounds producing sexual caste brood are equal to those of mounds producing worker brood only.

Source of variation	df	mean square	F	p
mound status	1	28.55	3.84	0.056
error	46	7.43		
total	47			

Table 5. ANOVA test of H_o: Distance to nearest neighbor is equal for mounds that produce sexual forms and mounds that produce workers only.

Source of variation	df	mean square	F	p
mound status	1	4.44	0.03	0.8662
error	46	154.64		
total	47			

was 0.0101 (stderr = 0.00007; n=276). The mean male weight across sites was 0.0081 (stderr = 0.00004; n=1100).

While significant differences in pupal weights of both sexes existed among sites, variation in weights was also found among mounds within sites. Male and female pupal dry weights with standard errors at each site are shown in Figure 4.

Some of this weight variation was linked to variation in stage of pupal development. Significant negative regression relationships existed between development stage and pupal dry weight for both sexes (Table 6, Figure 5). The regression slopes were similar between sexes, but development stage explained less variation in male weight ($r^2 = 0.1787$) than in females ($r^2 = 0.2934$), due to the presence of outliers in the male weight data. Mean stage of pupal development was not different among mounds or among sites (Figure 6).

Energy Content. There was no significant difference ($p = 0.5576$) between energy density (corrected Hc) of male and female pupae (Table 7 a,b; Figure 7). The least-squares means of energy density for males was 5526.2 cal/g, and for females 5507.9 cal/g. The overall nested ANOVA model: Hc = day (site (sex))) was not significant at $p = 0.5001$. As pupae could not be combusted individually, it was impossible to test for differences in caloric density among stages of development.

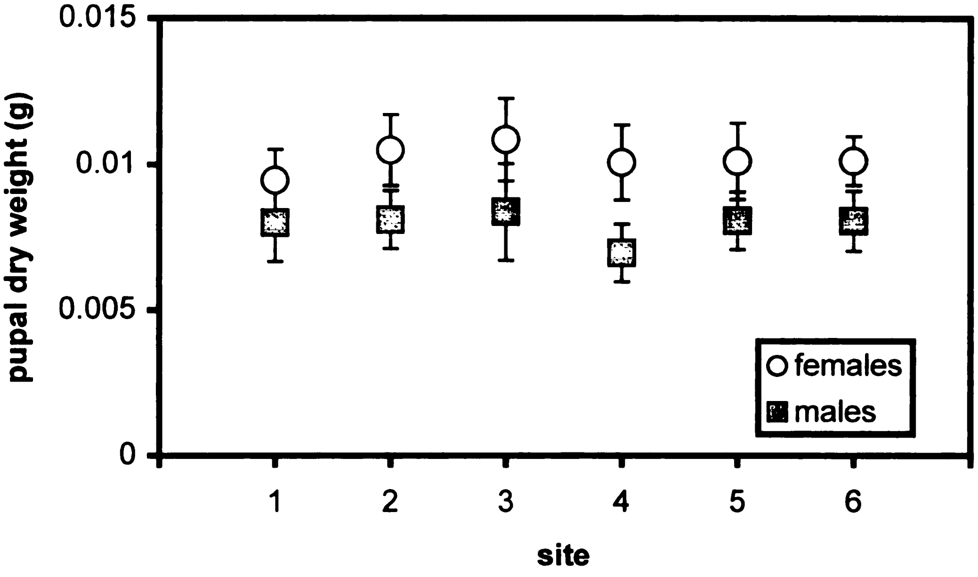

Figure 4. Females were heavier than males. Error bars represent one standard deviation.

Table 6. Analysis of covariance test of hypotheses H_0: Weight of male and female pupae are equal; H_0: There is no significant relationship between weight and developmental stage; and H_0: This relationship is the same for both sexes.

Source of variation	df	mean square	F	p
sex	1	0.0002445	160	0.0001
development	1	0.0003666	239.92	0.0001
sex*development	1	0.0000124	8.14	0.0044
error	1353	0.0000015		
total	1356			

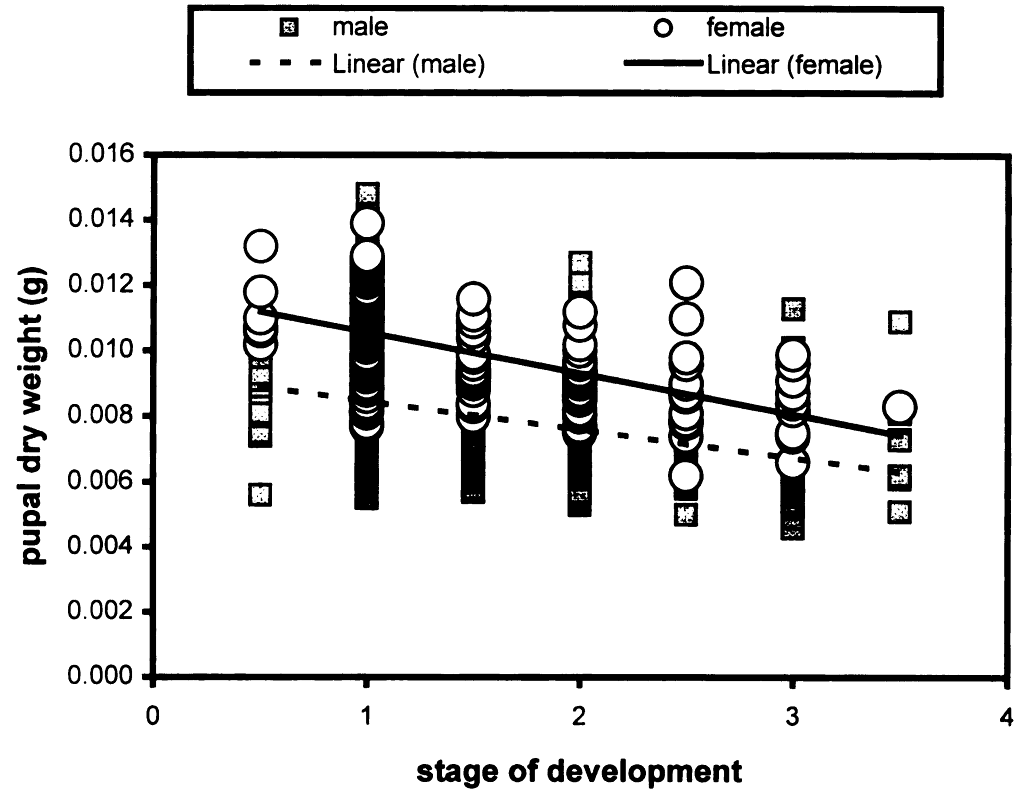

Figure 5. Pupal weight decreased over development. Regression slopes were significant at $p = 0.0001$; ANCOVA tests are provided in Table 6.

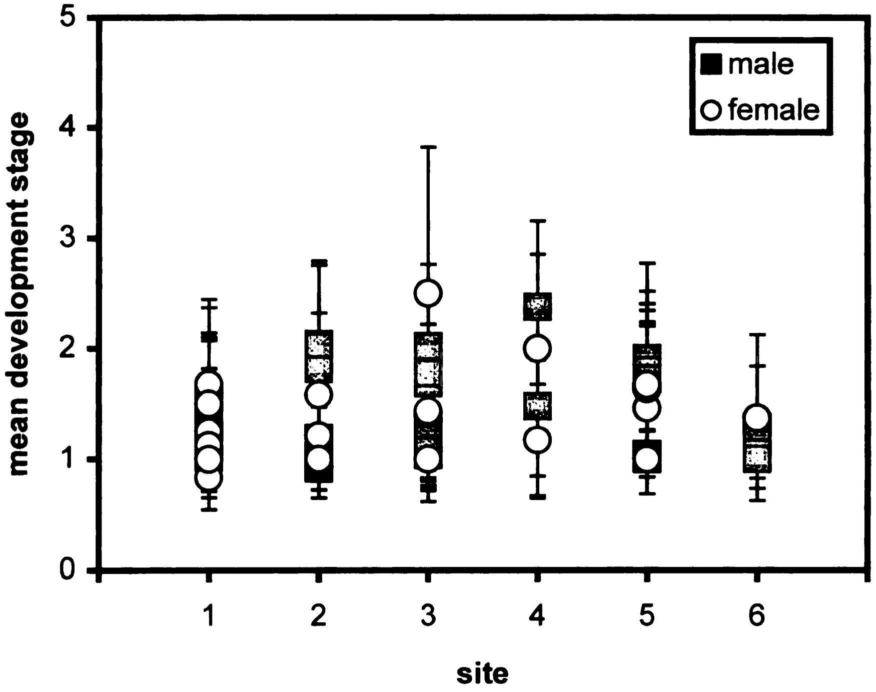

Figure 6. Average stage of pupal development did not differ significantly between sexes or among sites.

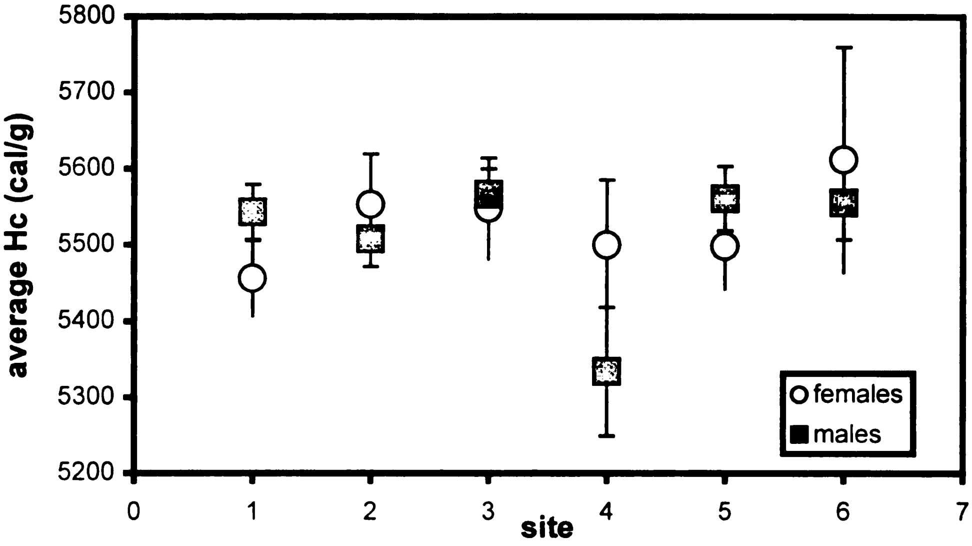

Figure 7. There was no significant difference in energy density (H_c) of reproductive caste pupae between sexes or among sites. ANOVA tests are provided in Table 7.

Table 7. Nested ANOVA performed on energy content data, testing H_0: H_c is equal for all sites and both sexes within sites. H_c = heat of combustion, in calories per gram.

Source of variation	df	mean square	F	p
model	6	15298	0.9	0.5002
sex	1	5904	0.35	0.5576
site	5	74435	0.88	0.5021
error	74	17013		
total	80			

Sex Ratios. As males and females had no consistent differences in energy content, caloric content data could not be used to estimate sex ratio. Numerical and dry-weight estimates of sex ratio were statistically different (t = 5.64997, df = 32, p = 0.0001). Due to the differences between male and female weights, using numbers of individuals to estimate sex ratio overestimated investment in males by an average of 2.8% compared to estimates based on weights. Whether this difference would be meaningful in a larger context depends largely on the precision of theoretical predictions tested. Applying Boomsma's (1989) correction increases the male bias to a greater extent than using the original numerical investment ratio. As sex ratio estimates based on dry weights seem more accurate in determining investment, and the Boomsma correction is accepted by convention, Boomsma-corrected weight ratios are discussed in the rest of this thesis.

Sex ratios were predominantly male-biased, but ranged from all-male (1.0) to almost all-female (0.08). Although considerable variation in sex ratio was present within sites, polydomy of *F. exsectoides* colonies makes mound to mound comparisons questionable without further genetic information. Thus sex ratio estimates from all mounds within a site are treated as samples of a single population, and only comparisons between population averages were performed. Population mean numerical and weight-based sex ratios are shown in Table 2.

Analysis of variance showed differences in sex ratio among sites (p = 0.0245, Table 8; Figure 8). Sites fell into two categories: those that were male-biased (sites 1, 2, 3, & 6), and sites with sex ratios not differing from 50/50 or equal investment (sites 4 & 5). Sites 4 and 5 still contained male-biased mounds, but these were balanced by female-biased mounds.

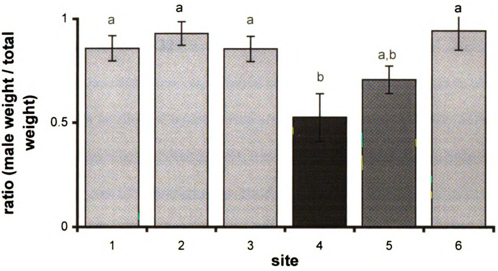

Figure 8. Sex ratio was predominantly male biased.

Table 8. ANOVA test of H_0: sex ratio is equal for all sites.

Source of variation	df	mean square	F	p
site	5	0.0806	3.10	0.0245
error	27	0.0230		
total	32			

Discussion

Mound measurements and collection of brood. The relatively low percentage of *F. exsectoides* mounds containing reproductive brood may be simply explained in the context of a polydomous colony—one or a few mounds within the colony may contain all of the colony's sexual offspring. While this may seem like "putting all of the pupae in one basket", several advantages are possible. The environment in some mounds might be better suited for production (or pupation) of sexual offspring. Some gain in efficiency of brood care might be attained by clustering sexual brood in a few mounds, rather than scattering it among many. Alternatively, a lack of production of sexual forms may reflect allocation to asexual colony expansion (budding). Budding should be a more successful method of expansion within habitats, since it does not depend on availability of *F. fusca* host nests. Alate sexual forms are usually vulnerable to predation during dispersal (Holldobler and Wilson 1990), and may be an unnecessary investment in stable habitats.

While mound surface area is not a good surrogate for direct measures of mound population (Cory and Haviland 1938), or even mound volume (Bristow *et. al.* 1992), the measurement has some value in comparisons between established mounds and newly-formed buds. As mound size increases over time (Andrews 1926, Haviland 1948), the tendency for mounds producing sexual brood to be larger in surface dimensions than mounds producing only workers may reflect differences in colony maturity (Figure 3). Measurements of distance to nearest neighboring mound may be expected to reflect crowding, local competition for resources, and frequency of budding, all factors that might influence reproductive allocation within a mound. Knowledge of relatedness and interaction behavior is necessary to understand which mounds are "neighbors" and which function as parts of the same mound. Current data do not show an effect of neighbor distance on reproduction.

Weight & Development. Differences in pupal development within and between mounds may arise from environmental factors (mound temperature, placement of pupae within mound), colony factors (time of egg-laying, maternal effects, nutrition), or genetic factors influencing development time. The data obtained in this study give no indication that males and females differ in development rates or emergence times (Figure 5, 6). While sex of "undeveloped" pupae cannot be determined (inviting conjecture that differences in development time may obscure the true sex ratio), relatively equal proportions of males and females at each developmental stage lends support to the contention that "undeveloped" pupae represent a random assortment of males and females (Figure 5).

Weight data indicated significant dimorphism between males and females, but this dimorphism was small relative to that shown by many ant species (Boomsma 1989, Crozier and Pamilo 1996). This is consistent with the founding biology of *F. exsectoides*—species with dependent (non-claustral) founding tactics, such as budding and social parasitism, may invest less in individual females. Founding biology also explains the lack of difference in male and female energy density, as females do not require large fat reserves for founding new colonies.

The sexual dimorphism in weight was partially masked by the presence of male weight outliers. These were usually males that were as heavy or heavier than females. These males were often also outliers with respect to appearance, possessing disproportionately large heads. The presence of these "heavy" males might indicate a dispersal polymorphism such as that described by Fortelius *et. al.* (1987) and Agosti and Hauschteck-Jungen (1987) in *Formica exsecta*, in which small males dispersed while larger males mated in the vicinity of the nest. The low frequency of these males among *F. exsectoides* sampled argues against that explanation. Alternatively, these males could be diploid. Nipson's (1978) study, as well as the colony structure and mating behavior of *F. exsectoides*, suggests that significant inbreeding may occur within populations. Inbreeding may lead to the production of diploid individuals homozygous at sex-determining loci—intended females who are morphologically male and reproductively dysfunctional (Crozier 1971, Pamilo *et.al.* 1993). The presence of diploid males as a significant fraction of the reproductive brood produced would lead an overestimation of male investment, and should be investigated in the future.

Sex Ratio. The strong male bias in sex ratio displayed by *F. exsectoides* is surprising, as relatedness asymmetries in eusocial Hymenoptera are expected to result in worker preference for a female bias (Trivers and Hare 1976). The male bias observed is consistent with observed sex ratios for other ant species that display colony expansion through budding (Bourke and Franks 1995, Pamilo and Rosengren 1983). Unfortunately, budding is associated with a suite of characteristics, such as polygyny and polydomy that may equally influence sex ratio (Boomsma 1993). The extent of any or all of the above traits may be related to genetic, social, or environmental factors.

Polygyny may lead to an increased worker preference for males, but the functional extent of polygyny in this species is incompletely known. *F. exsectoides* colonies are peculiar in that "foreign" queens are sometimes adopted into existing mounds (Creighton 1950, Nipson 1978). This apparently selfless act of adopting a stranger and raising her offspring might be explained if relatedness within populations is high (decreasing the genetic level of polygyny) or if inbreeding has depressed the population (by increasing diploid male production) to the extent that novel genetic input is desirable.

In highly polygynous colonies, relatedness between sisters will decrease (unless all queens are highly related!). Workers in these situations may attempt to produce their own (male) offspring. However, workers should prevent reproduction by their less-related sisters, leading reciprocal policing and a lack of worker-produced males. Additionally, workers of many ant species lack ovarian function, or are inhibited by the presence of a healthy queen. There are no firm reports of worker reproduction in queen-right colonies of *Formica* species (Crozier and Pamilo 1996). *F. exsectoides* has only been observed to lay trophic eggs in queenless laboratory colonies (C. Bristow, personal communication). All colonies found to contain sexual caste pupae also contained worker pupae, so it is unlikely that these mounds were queenless. While the majority of males in this study could be produced by workers, the production of such large numbers of worker-laid males in queenright colonies could indicate a decline in colony stability and a possible cause of colony failure (Starr 1977, Crozier and Pamilo 1996).

The difference in sex ratio between sites suggests an environmental influence. Two sites located within (site 4) or near (site 5) recently clear-cut forests had sex ratios not different from 50/50, while all sites located in mature forest had strongly male-biased sex ratios (Figure 8). If this difference arises from a true environmental influence (occurrence of aberrant sex ratios in these areas may be due to chance), two factors that might encourage increased female investment are food and availability of nest sites. Resource availability may contribute directly to sex ratio, either through developmental effects (lack of food results in fewer sexuals, more workers (Deslippe and Savolianen 1995, Herbers and Banschbach 1998)) or by influencing reproductive allocation. The theory of local resource competition (Clark 1978) states that, in a resource and dispersal-limited environment, male production will be favored since female offspring will compete directly with the mother for resources. While this theory is not consistent with observations in this study (clear-cut areas may be expected to have fewer resources than mature forest), it is certainly possible that differences in sex ratio among sites represent population-level strategies for coping with ecological conditions at each site. The effect of environment on sex ratio may also be indirect—Herbers (1986, 1993) shows the impact of ecological factors on queen number, which might be expected to affect sex ratio by altering the relatedness structure of the colony. More detailed comparisons between populations along a gradient of environmental conditions would be necessary to determine the existence of these strategies and the factors regulating them.

Factors that cause *Formica exsectoides* to present particular difficulties in sex ratio studies, such as polydomy, social parasitism, and specialization on transient habitats, increase the need to perform such studies, since understanding sex ratio and reproductive dynamics in only discrete, monogynous laboratory colonies gives an incomplete picture of the interaction of genetic and environmental factors in shaping reproductive strategy. While perhaps unfortunate from the standpoint of those hoping to integrate sex ratio studies in large-scale evolutionary analyses, this study indicates that intraspecific variation in sex ratio exists in *F. exsectoides*, whether due to environmental or genetic influences. As significant variation in sex ratio occurred between sites <u>within</u> a specific habitat (jack pine), whole species generalizations of sex ratio based on one or two studies will surely misrepresent true sex ratio dynamics of many ant species.

CHAPTER 2

Age of jack pine stands influences abundance of the Allegheny mound ant (Hymenoptera: Formicidae) and other terrestrial arthropods

Introduction

Formica exsectoides (Forel), the Allegheny mound ant, has been presented by Oster and Wilson (1978) as a primary example of a fully polygynous species. They propose that *F. exsectoides* fits an "adaptation syndrome" for habitats that are stable, long-lived, and patchy. These habitats are primarily persistant grassy clearings.

The syndrome includes traits displayed by *F. exsectoides* that allow it to rapidly expand to highly dense populations within a habitat. Such traits include polygyny (possessing multiple queens per colony) and polydomy (occupying more than one nest site per colony). Polygyny provides colonies with the potential for earlier production of sexual forms than single-queen colonies (Keller 1991). Polydomy and polygyny allow colonies to expand by mound budding or fission, rather than or in addition to the slower and riskier process of establishing new colonies through dispersal of alate sexual forms. *F. exsectoides* colonizes new habitats through temporary social parasitism of *Formica fusca* colonies, possibly an adaptation that allows *F. exsectoides* to skip the vulnerable early stages of claustral founding, creating lower rates of failure in young colonies and allowing rapid growth of the population (Bourke and Franks 1995).

In Michigan, *F. exsectoides* occurs in high abundance in jack pine (*Pinus banksiana* Lamb.) (Bristow *et. al.* 1992, Bishop 1998). An early successional tree adapted to cold climates and poor, sandy soils, jack pine is valued for rapid recolonization of burned areas and for covering land of poor soil quality. Jack pine management is usually focused on pulpwood production, but in Michigan, jack pine is managed extensively to create habitat for the Kirtland Warbler, an endangered species (Rudolph and Laidly 1990). Jack pine stands are generally maintained by wildfire. In the absence of fire, jack pine does not recruit well naturally—clear-cut stands are often replanted (Beland and Bergeron 1993, Duchesne and Sirois 1995). Jack pine stands may persist 80-100 years on poor soils, but it is a short-lived species and will eventually fall prey to disease or insect damage.

No habitat is truly stable—in fact, as an early successional community, jack pine stands may be considered quite transient. Little is known about causes of population decline and colony mortality in *F. exsectoides*, but habitat changes may play a role. Researchers studying *F. exsectoides* populations (Andrews 1926, 1929; Haviland 1948, Dimmick 1951) have reported observations suggesting that habitat suitability, particularly the light environment, may decline over as short a time span as 10 years. While distribution (Bristow *et. al.* 1992), mound placement (Andrews 1926) and population size (Cory and Haviland 1938) have been investigated, these studies focus on single populations of *F. exsectoides*, rather than presenting comparisons between populations in differing habitats.

I examine changes in *F. exsectoides* populations within various stages of this transient habitat. The high density and aggressive nature of *F. exsectoides* (Creighton

1950) gives it many important roles in jack pine communities. While the effects of *F. exsectoides* populations on arboreal insect communities have been examined by Bishop (1998), no data is available on the relationship between *F. exsectoides* and the ground-dwelling communities of jack pine. The majority of arthropod study in jack pine is based on the economic pests, such as *Choristoneura pinus* (jack pine bud worm) (Nealis and Lomic 1994, Volney and McCullough 1994). Studies of insect communities have generally focused on early stages of jack pine growth (Naylor and Bendell 1983, Spires and Bendell 1983, Beaudry *et. al.* 1997). Accordingly, this study also examines the ground arthropod community co-occurring with *F. exsectoides* and changes in abundance and composition of feeding guilds as jack pine ages.

Materials and Methods

Site selection & description. All sites described in this study are located in the Huron National Forest, Oscoda and Crawford Counties, Michigan (Figure 9a). Using maps showing stand composition and date of origin (date since last cut), the Big Creek (Oscoda Co. T25N, r1W) and Eldorado (Crawford Co. T25N, r1E) areas of the Huron National Forest were surveyed for presence of *F. exsectoides*. Of stands containing significant populations (>10 mounds), I selected five stands in each of three age classes for inclusion in the study. The three age categories were: young (2-16 years), mid-regrowth (37-44 years), and old (64-77 years). Stand codes, names, United States Forest Service designations, ages, and specific notes on sites are shown in Table 9.

Location and density of *F. exsectoides* mounds. Within each stand, I marked a 50m x 50m plot with wire stake flags. This plot was mapped using a grid of 5m^2 increments, compass, and surveyor's rope. Locations of all trees and all anthills were recorded. Longest slope and shortest slope were measured with a tape measure, and multiplied to yield a comparative measure of mound surface area in m^2. Species, distance to mound (m), and diameter at breast height (dbh) in cm were recorded for all trees greater than 1 cm in diameter within a 5m radius of each mound that showed activity (was not derelict).

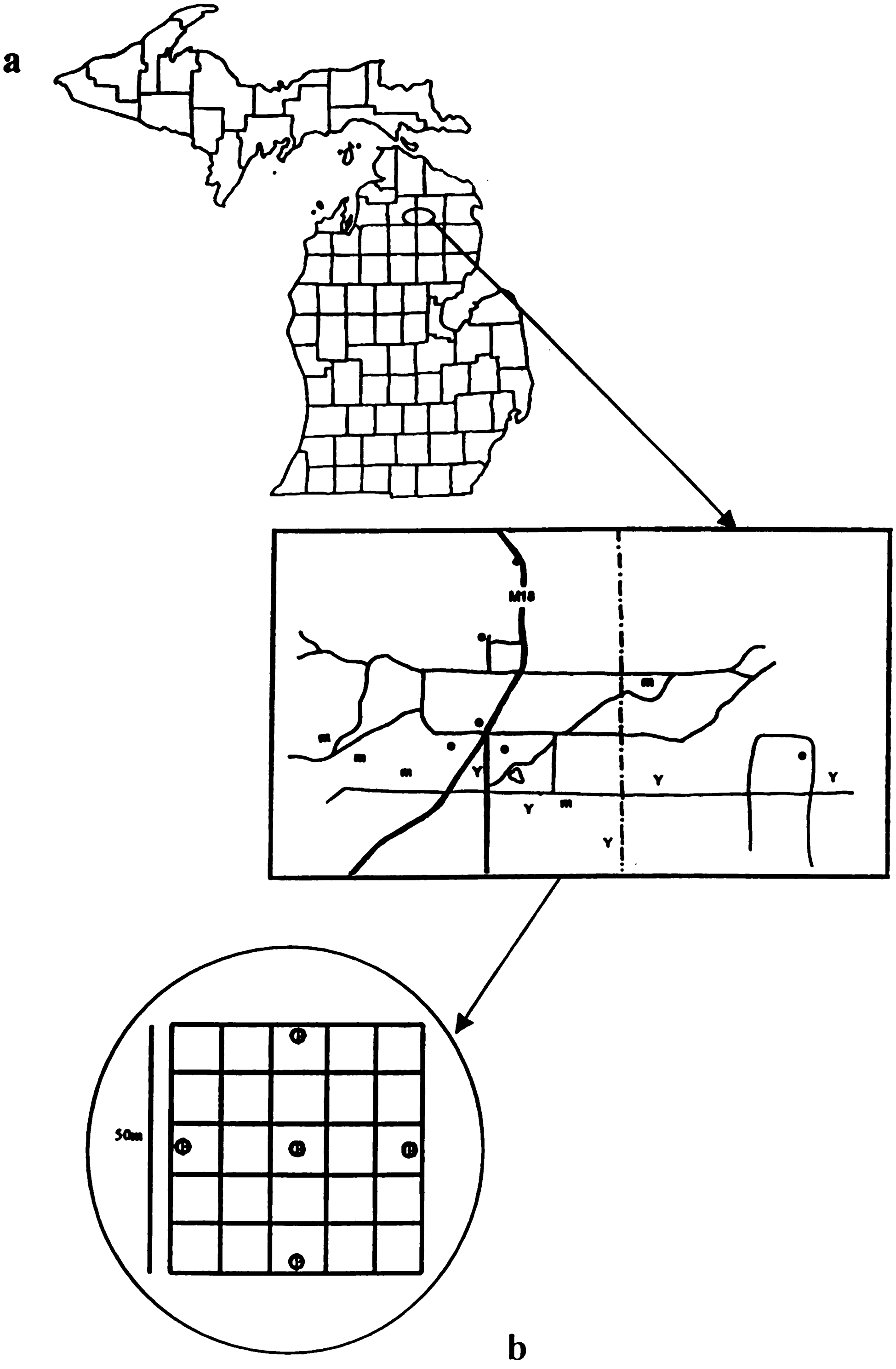

Figure 9. a) Stands were located in Crawford and Oscoda counties, Michigan. Letter indicates stand age class: Y = young, m = mature, o = old. b) Plot layout and pitfall trap locations.

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Table 9. Stand locations and descriptions for Chapters 2 & 3

Site Code	USFS compartment & stand #	Age Class	Stand Age	Notes
Y1	94/18	young	2	recent burn of 62 year-old redpine stand, debris still on ground; open/grassy
Y2	128/21	young	9	area cleared by a 3-acre fire within 61-yr old jack pine; sparse/grassy
Y3	78/1	young	16	naturally regenerating clearcut, very sparse
Y4	77/25	young	15	dense jack pine plantation, all trees 2-2.5m in height
Y5	75/7	young	8	dense jack pine plantation; all trees 1.5-2m in height
M1	77/1	mature	43	sparse stand, partially burned in 1966, many mature trees; grassy
M2	94/5	mature	44	mixed understory of grasses/moss; high recreation traffic
M3	56/19	mature	40	partially open, grassy
M4	49/35	mature	37	partially open, grassy
M5	54/8	mature	42	extremely dense stand, much undergrowth, oak understory
O1	72/50	old	64	much storm damage during 1997
O2	73/17	old	77	many openings due to tree fall, grassy areas
O3	55/12	old	74	very small stand, bordering on young jackpine plantation
O4	122/7	old	72	mature trees, primarily fern & moss understory
O5	75/6	old	72	mature trees, primarily fern & moss understory

Abundance of *F. exsectoides* and other ground-dwelling arthropods. To estimate the proportions and relative abundance of insect groups within these different jack pine stands, I placed five pitfall traps in each plot in the arrangement shown in Figure 9b. Pitfall traps consisted of two plastic cups (11 cm diameter, 7.5 cm in depth) nested inside each other and buried with the opening flush with the soil surface. Traps were filled with ethylene glycol to preserve samples until collection. Plastic lids held approximately 1cm above the soil surface by roofing nails helped to minimize debris and small mammals in traps. Trap lids were completely closed during non-collection periods. Traps were installed June 28-30, 1997, but left closed for one week to reduce captures of curious ants investigating the disturbance in the soil. Traps were opened for three one-week periods, with collections taking place on July 8-9, July 15-16, and August 3-4, 1997. Traps were emptied by removing the upper plastic cup and transferring the contents to a separate container filled with 95% EtOH, allowing replacement of the cup with minimal disturbance to the trap area.

Preserved pitfall samples were sorted and identified to either order or family level, with the exception of ants and Carabid beetles, which were identified to genus or species level. Ants were identified or confirmed by Gary Coovert, Dayton Museum of Discovery, Dayton, OH. F. Purrington, Ohio State University identified carabid beetles.

Data analysis.

Location and density of *F. exsectoides* mounds. To test whether mounds were clumped, randomly, or uniformly dispersed, I calculated Lloyd's indices of mean crowding (X^*) and patchiness (Pielou 1969), using a quadrat size of 25 m^2 (4 quadrats/plot). Since these indices are sensitive to quadrat size, I also performed a nearest neighbor analysis as described by Clark and Evans (1954) and Pielou (1969). Mound density was calculated as the sum of the total number of mounds divided by the total plot area. Tree density for each plot was calculated as the mean dbh of trees within the plot, multiplied by the number of trees per square meter, with the product variable in units of cm (tree diameter)/m^2 plot area.

Abundance of *F. exsectoides* and other ground-dwelling arthropods. Major taxonomic groups present in > 25% of samples and comprising > 0.1% of total invertebrates sampled were analyzed for differences among stand age classes using individual one-way analyses of variance (PROC GLM, SAS Systems, Inc.) A sequential Bonferroni correction was then used to assess the significance of the results (Rice 1990). Relative abundance of all major groups were also correlated to stand age, stand density, and to each other using a correlation analysis (PROC CORR, SAS Systems, Inc.), also corrected with a sequential Bonferroni procedure.

Composition of ground-dwelling insect communities. While families and genera within some orders (e.g. Orthoptera) are ecologically equivalent (due to similarities in feeding behavior), other higher taxonomic groups (such as Hemiptera and Coleoptera) are more diverse. The groups are therefore subdivided according to feeding behavior, using the categorizations of Herbivore, Predator, and Scavenger. Allocation of organisms to feeding category was based primarily on family-level feeding ecology described in Borror, Triplehorn, and Johnson (1989) and Arnett (1993). Ants, because they feed at multiple trophic levels and display complex behaviors such as homopteran tending, are

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considered separately from these designations as per the ecological guild divisions established by Moran and Southwood (1982). Parasitoids were noted but not included in the analysis.

Relative proportions of each feeding group examined were arcsine transformed and simultaneously compared within and among age classes using a multivariate analysis of variance (MANOVA) (PROC GLM, SAS Systems, Inc. 1990), with canonical correlation analyses performed to aid in interpretation of the analysis (Scheiner 1993). Similar MANOVA tests were performed to compare relative proportions of ant genera/species across stand ages. Relationships between feeding groups were examined by correlation analysis. Overall analyses were performed, then correlation analyses for each stand age class were performed separately to explore the interaction of age with feeding group correlations.

Stand ages, mound and tree densities, mound surface area indices, and relative densities of *F. exsectoides* (numbers of workers per trap) were correlated using PROC CORR (SAS Systems, Inc. 1990).

Results

Location and density of *F. exsectoides* mounds. Age-class means and standard errors for mound density and tree density are listed in Table 10. Average tree density increased with stand age (Figure 10), but stand age explained less than 50% of the observed variation in density ($R^2 = 0.4763$, $p = 0.0063$).

Mound density (mounds/m^2) was marginally correlated with stand density ($R^2 = 0.3603$, $p = 0.0232$) (Figure 11a). Further examination revealed an interaction of this relationship with stand age class—mound density had a strong negative correlation with stand density ($R^2 = 0.9948$, $p = 0.0026$), but only in young stands. Density of *F. exsectoides* mounds was greatest in young burned (Y1 and Y2) and mature jackpine stands (Figure 11a, b).

Nearest neighbor analyses showed that mounds were neither clumped nor overdispersed at the plot level; the mound dispersion pattern was not significantly different from the null expectation of randomness ($p>0.05$) (Table 11). Lloyd's Index of mean crowding showed an increase in number of mounds per 25 m^2 patch from young to mature stands, but a decrease from mature to old stands. Calculated values of Lloyd's patchiness index are similar across stand ages and approximately 1, indicating a random distribution of mounds in all three stand age classes (Table 12).

Mound surface area estimates ranged from 0.43 m^2 to 21.3 m^2. Mounds did not appear to be larger in older stands (Figure 12a), but site means for surface area estimates increased significantly with stand age ($R^2 = 0.3542$, $p = 0.001$) (Figure 12b).

Table 10. Stage age class means and standard deviations for mound and tree densities.

stand age class	mound density (mounds/m^2)	tree density (cm dbh/m^2 plot)
young	0.0042 (0.0012)	0.53 (0.33)
mature	0.0047 (0.0012)	1.05 (0.54)
old	0.0030 (0.0010)	1.37 (0.22)
overall	0.0039 (0.0013)	1.02 (0.50)

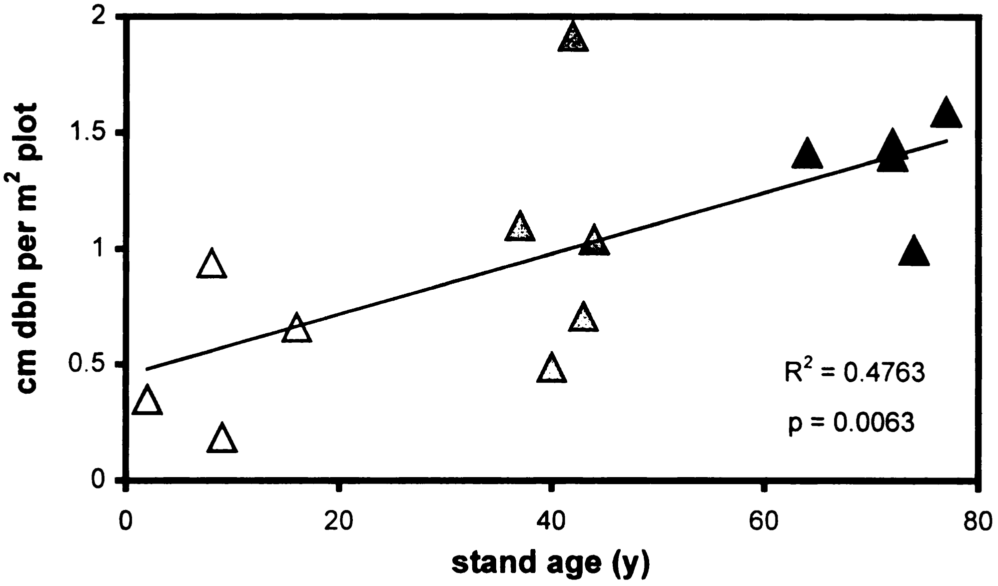


Figure 10. Stand density increased significantly with stand age. Density values represent both average diameter of trees in plot and density of individuals (stem density). No value is present for Y4, due to missing data on stem density for some areas of the plot.

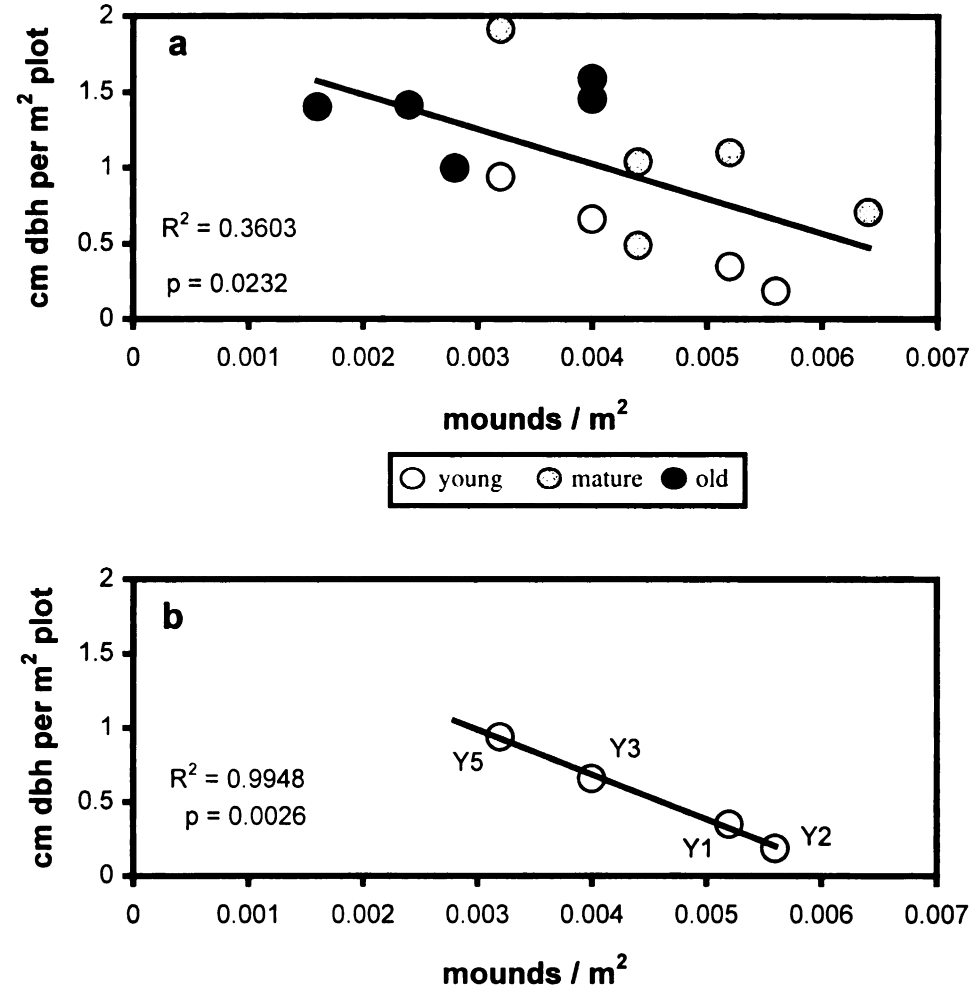


Figure 11. a. Mound density was negatively correlated with stand density. b. This correlation was strongest when only young stands were considered.

Table 11. Nearest neighbor analysis of mound dispersion (clark & Evans 1954), performed by calculating an index of Nonrandomness (R), by the formula $R = 2r(\rho)^{0.5}$. (Where r = distance to nearest neighbor, ρ = population density (mounds/m^2)) Analyses were performed on individual mounds and R tested against a null expectation of random dispersion (R = 1) using a t-test as prescribed in Pielou (1969).

	young	mature	old
n	52	59	37
mean r (m)	14.7	12.6	16.3
mean r	0.0042	0.0047	0.0030
mean R	1.30	1.14	1.10
non-random?	**no**	**no**	**no**

Table 12. Lloyd's indices of mean crowding (X*) and patchiness. X* = mean # of neighbors per individual within a sampling unit. Sampling units were 1250m2 (4 units/plot). The patchiness index indicates dispersion, with a null expectation of 1 indicating random dispersion.

	young	mature	old	overall
n	20	20	20	60
mean (mounds/25m^2)	2.5	3.0	1.9	2.5
var (mounds/25m^2)	3.11	3.05	2.16	2.86
X* (mean crowding)	2.74	3.02	2.06	2.64
patchiness	1.10	1.01	1.05	1.06
non-random?	**no**	**no**	**no**	**no**

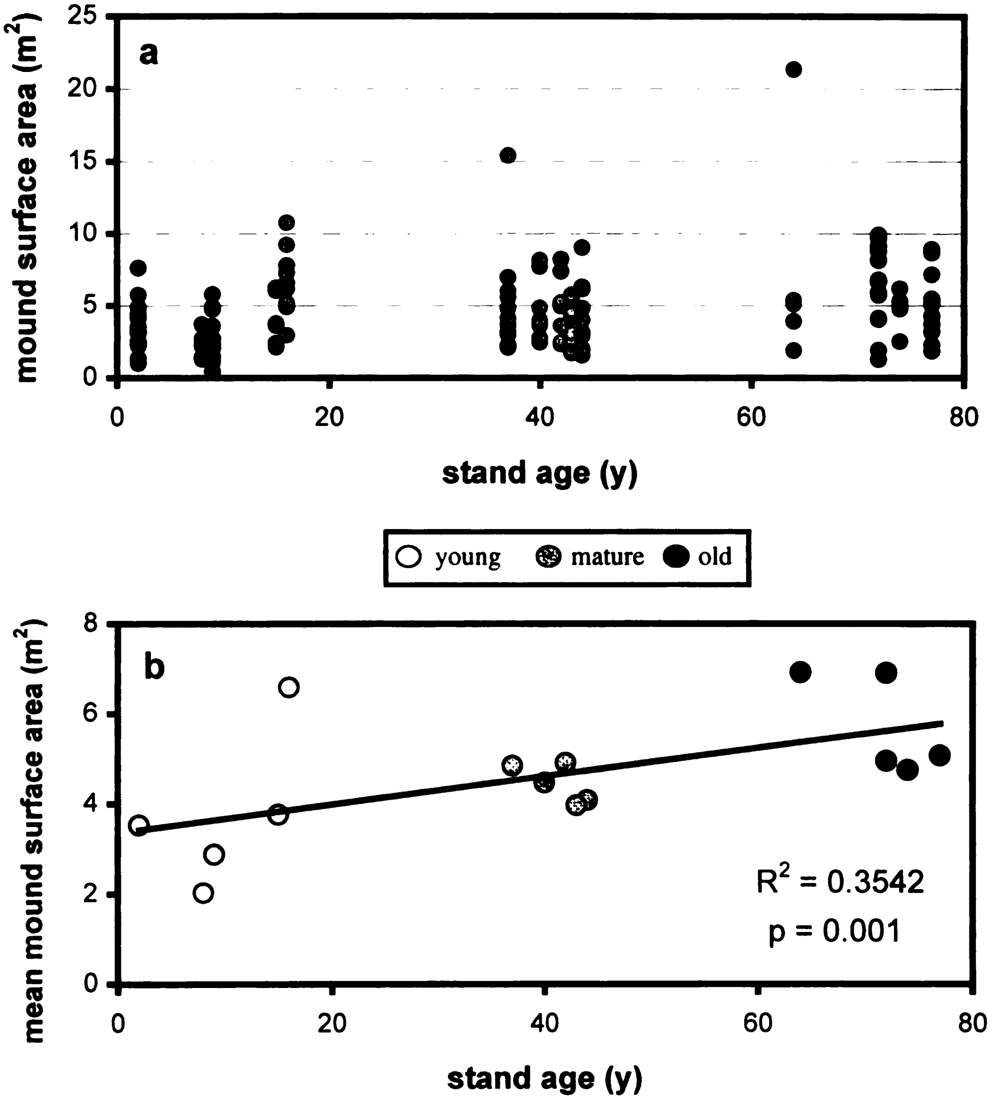


Figure 12. a. Mound surface area indices were not larger in older stands. b. Average mound surface area indices increased with stand age, possibly due to incomplete budding and a lack of small mounds.

Abundance of *F. exsectoides* and other ground-dwelling arthropods. A list of arthropods captured in jack pine is presented in Appendix A. Table 13 provides the mean number of individuals per trap and standard deviation of major invertebrate orders and families found in jack pine, as well as the percentage of all traps containing at least one individual from the group. Probability values reflect a test of the null hypothesis of no difference between stand age groups; 8 of 14 taxonomic groups tested were significantly different across stand age classes. Changes in total abundance of these same groups over collection dates and stand age are shown in Figure 13.

Arthropods were partitioned into the following groups: Ants, Herbivores, Predators, Scavengers, and Parasitoids. Organisms not included in these analyses due to low abundance or low frequency of occurrence included: gastropods (snails and slugs), oligochaetes (earthworms), and isopod crustaceans. Collembola were abundant in several samples but could not be counted accurately. Flying insects, particularly scavenging Diptera such as the Sepsidae and Lauxaniidae, were considered incidental. While these insects were probably attracted to the traps by odor cues, their representation in pitfall traps may not represent their normal range of ground activity.

Multivariate analysis of variance showed stand age class differences in proportions of feeding groups captured (Pillai's Trace: F = 5.5409, df = 10, p = 0.0001; Roy's Greatest Root: F = 7.4226, df = 5, p = 0.0001). Examination of standardized canonical coefficients revealed that changes in proportions of predators accounted for most of the variation between age classes, followed by scavengers, then ants (Table 14). Groups showing significant changes in univariate analyses were Predators (F = 14.33, p = 0.0001), and ants (F = 6.34, p = 0.0021). Changes in scavenger proportions across stand

Table 13. Invertebrates captured in pitfall traps. Values presented are: % of all traps containing at least one individual, and mean number of individuals per trap (standard deviation). P-values generated by one-way ANOVA on each taxonomic group. Entries in bold-face are significantly different after sequential Bonferroni adjustment for multiple comparisons.

	% traps	young n = 60	mature n = 71	old n = 67	p
Hymenoptera					
Formica exsectoides	**100**	**282 (303)**	**558 (924)**	**338 (992)**	**0.0001***
other Formicidae	**85**	**9.85 (11.02)**	**5.64 (8.24)**	**10.18 (8.94)**	**0.0068***
WASPS	**32**	**0.92 (1.62)**	**0.23 (0.51)**	**0.48 (0.70)**	**0.0008**
Orthoptera	**70**	**9.76 (9.16)**	**4.65 (5.36)**	**0.93 (1.58)**	**0.0001**
Coleoptera (predaceous)	**58**	**0.85 (1.52)**	**1.11 (1.64)**	**3.45 (4.31)**	**0.0001**
Coleoptera (other)	42	1.05 (1.65)	0.89 (1.65)	3.44 (4.31)	0.5311
Diptera	**86**	**5.58 (4.39)**	**3.87 (4.63)**	**1.22 (1.97)**	**0.0001**
Hemiptera					
Lygaeidae	84	18.63 (18.45)	22.70 (37.79)	14.22 (20.46)	0.1973
Thyreocoridae	**33**	**0.23 (0.56)**	**0.29 (0.76)**	**4.01 (6.03)**	**0.0001**
Homoptera	32	0.60 (1.03)	0.38 (0.88)	0.79 (1.38)	0.0982
Lepidoptera (adult)	28	0.50 (1.03)	0.77 (2.24)	0.43 (0.84)	
Arachnida					
Aranae	96	5.62 (3.38)	6.94 (4.56)	6.66 (4.83)	0.1962
Opiliones	**25**	**0.40 (1.01)**	**0.54 (1.71)**	**1.73 (3.29)**	**0.0011**
Diplopoda	**41**	**2.11 (3.84)**	**0.97 (1.47)**	**0.67 (1.63)**	**0.0032**

* p-value reflects analysis of log-transformed data. Means and standard deviations presented are untransformed data.

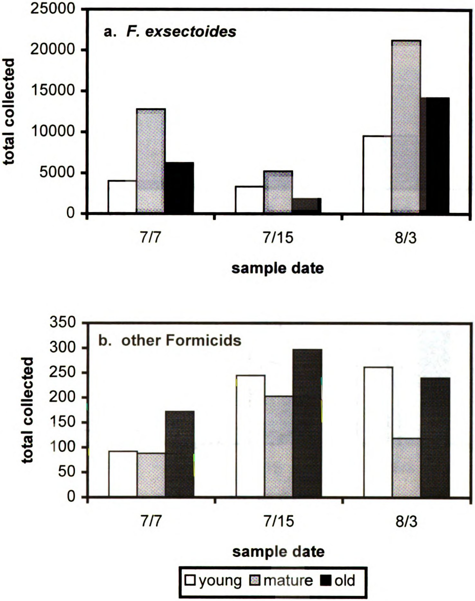


Figure 13. Total number of individuals captured per stand age class, sorted by taxon group and date. Taxon groups presented were present in over 25% of all traps and accounted for more than 1% of all invertebrates captured. These include: a) *F. exsectoides*, b) other Formicids, c) Orthoptera, d) Lygaeidae, e) Thyreocoridae, f) Aranae, g) predaceous Coleoptera, h) other Coleoptera, i) Opiliones, j) Diplopoda, k) Diptera.

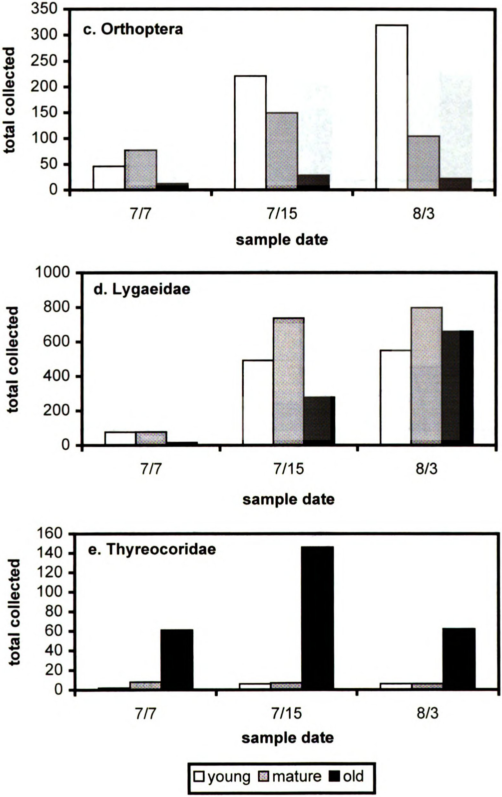
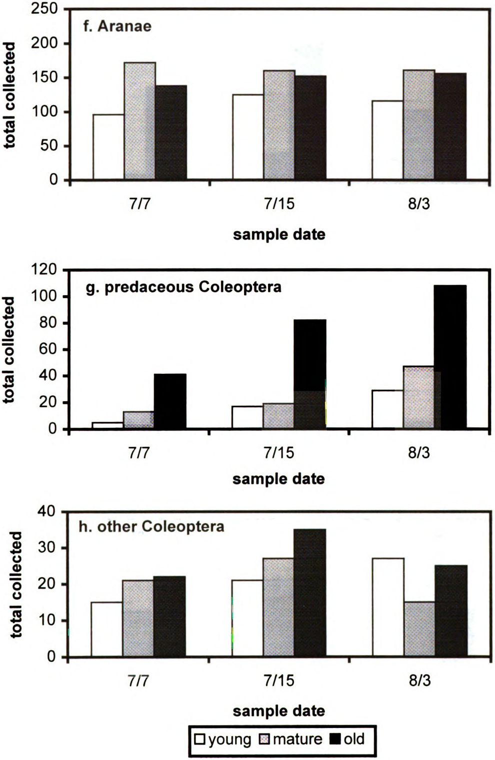


Figure 13. (CONTINUED)



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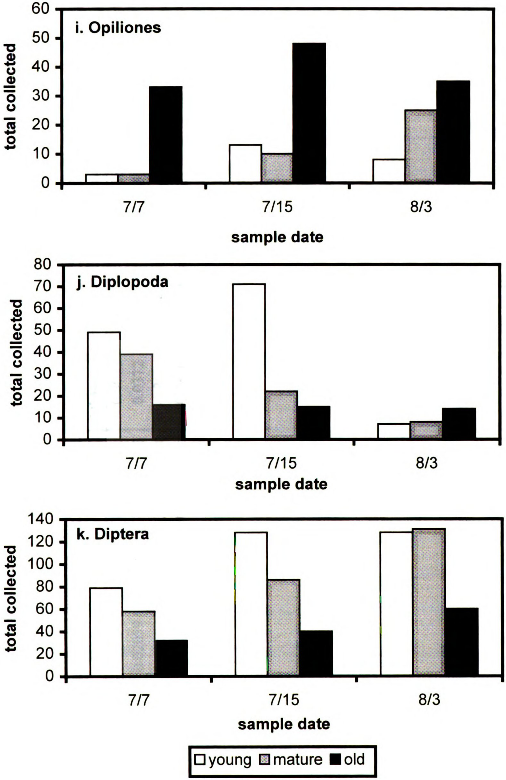

Figure 13. (CONTINUED)

Table 14. Univariate analyses testing H_0: feeding guild proportions are equal across stand age classes. Canonical coefficients were generated by MANOVA test; only those discussed in the text are shown.

	Univariate ANOVA				canonical coefficients			
	MSE	df_e	F	p	between (1)	between (2)	standardized (1)	standardized (2)
ANTS	0.09098	222	6.34	0.0021	0.4085	0.9128	-0.3178	2.2492
HERBIVORES	0.02059	222	1.39	0.2506	0.0372	-0.9993	-0.0651	1.3129
PREDATORS	0.00392	222	14.33	0.0001	-0.8979	-0.4403	-1.4467	0.7689
SCAVENGERS	0.00261	222	3.90	0.0216	0.0830	-0.9966	0.5281	0.1392

age classes were marginally significant (F = 3.90, p = 0.0216). Organisms assigned to each group and changes within feeding groups are presented below.

Ants. Ants were the most abundant group of organisms, comprising 87-93% of all invertebrates captured. Relative abundance of *F. exsectoides* foragers was not linearly correlated with stand age, mound surface area estimates, or tree density, but was significantly and positively correlated with mound density ($R^2 = 0.51$, p = 0.0027) (Figure 14). *F. exsectoides* foragers were most abundant in mature stands.

Only 2.2% of ants captured were species other than *F. exsectoides*. Ant genera/species captured in more than 3 traps are listed in Table 15, as well as the mean number of individuals per trap, standard deviation, and the percentage of all traps containing at least one individual of that genus/species. Probability values reflect a test of the null hypothesis of no difference between stand age groups; only *Formica fusca* (p = 0.0005) and *Aphaenogaster* spp. (p = 0.0001) were significantly different in average abundance per trap after correction for multiple comparisons.

MANOVA tests of stand age class differences in proportions of ant species were highly significant (Pillai's Trace: F = 5.2734, df = 8, p = 0.001; Roy's Greatest Root: F = 8.5993, df = 4, p = 0.0001). Standardized canonical coefficients indicated that these differences were largely due to shifts in abundance of *Aphaenogaster* spp. and *Dolichoderus plagiatus*, and that the correlation between groups shifted across age classes (Table 16).

Abundance of *F. exsectoides* was not correlated with abundance of other ants as a group ($R^2 = 0.0126$, p = 0.1387) (Figure 15). In individual species comparisons,

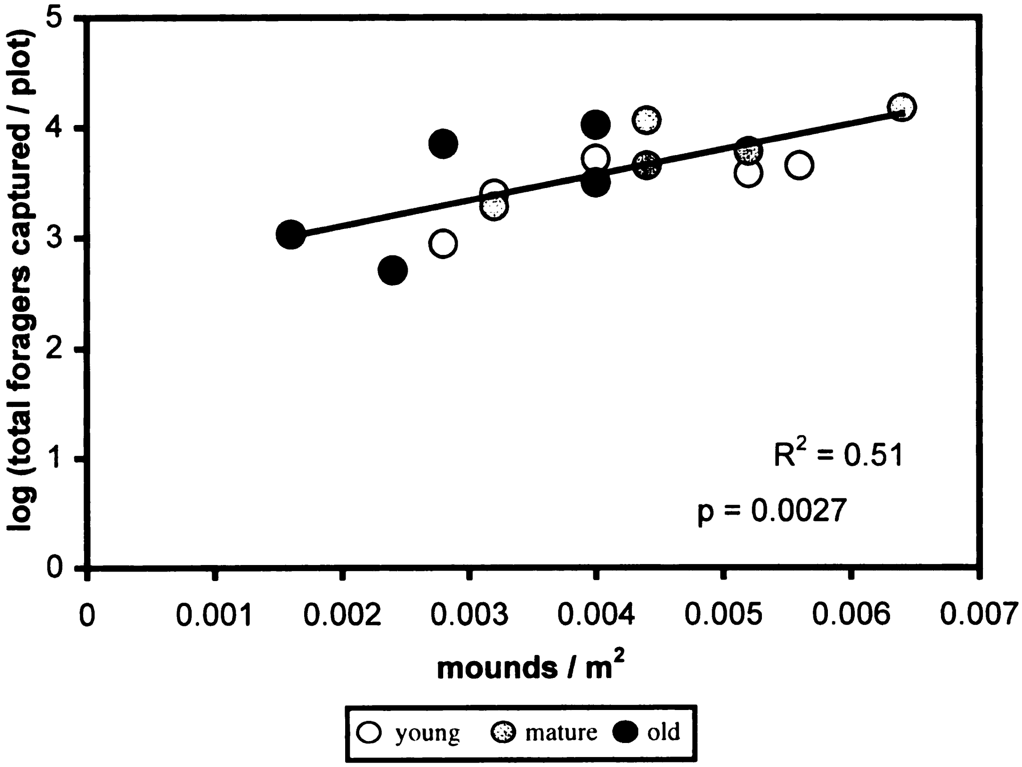

Figure 14. Mound density was positively correlated to log-transformed abundance of *F. exsectoides* foragers captured.

Table 15. Ant genera and species most commonly captured in jack pine. Values presented are: % of all traps containing at least one individual, and mean (stdev) for each age class. P-values were generated by one-way ANOVA for each species; bold species are significantly different after Bonferroni correction.

Genus/spp.	% traps	young	mature	old	p
Formica exsectoides	**100**	**281.81 (303)**	**558.27 (924)**	**337.58 (992)**	**0.0001***
***Aphaenogaster* spp.**	**73**	**3.12 (3.29)**	**2.59 (3.61)**	**8.05 (8.40)**	**0.0001**
Formica fusca	**39**	**4.30 (8.60)**	**1.22 (2.51)**	**1.00 (2.13)**	**0.0005**
Tapinoma sessile	30	0.83 (1.90)	0.51 (1.63)	0.62 (1.08)	0.4936
Dolichoderus plagiatus	19	0.48 (1.05)	0.26 (0.70)	0.20 (0.59)	0.1158
Lasius spp.	5	0.43 (1.87)	0.90 (7.22)	0.03 (0.17)	0.5268
Camponotus herculeanis	4	0.12 (0.37)	0.00 (0.00)	0.03 (0.17)	0.0143
Monomorium minimum	4	0.50 (1.97)	0.00 (0.00)	0.00 (0.00)	0.0146

* p-value reflects analysis of log-transformed data. Means and standard deviations are derived from untransformed data.

Table 16. Univariate analyses testing Ho: proportions of ant species are equal across stand age classes. Canonical correlations were generated by MANOVA test; only those discussed in the text are shown.

	Univariate ANOVA				canonical coefficients			
	MSE	df_e	F	p	between (1)	between (2)	standardized (1)	standardized (2)
F. exsectoides	0.100056	194	13.61	0.0001	0.9518	-0.3066	0.201	-1.815
Aphaenogaster	0.003833	194	0.73	0.4847	-0.9645	0.2642	-92.826	-159.524
F. fusca	0.002923	194	0.66	0.5163	-0.9614	0.2752	4.774	54.137
Dolichoderus	0.000030	194	0.67	0.5128	0.9604	-0.2788	-87.852	-105.263

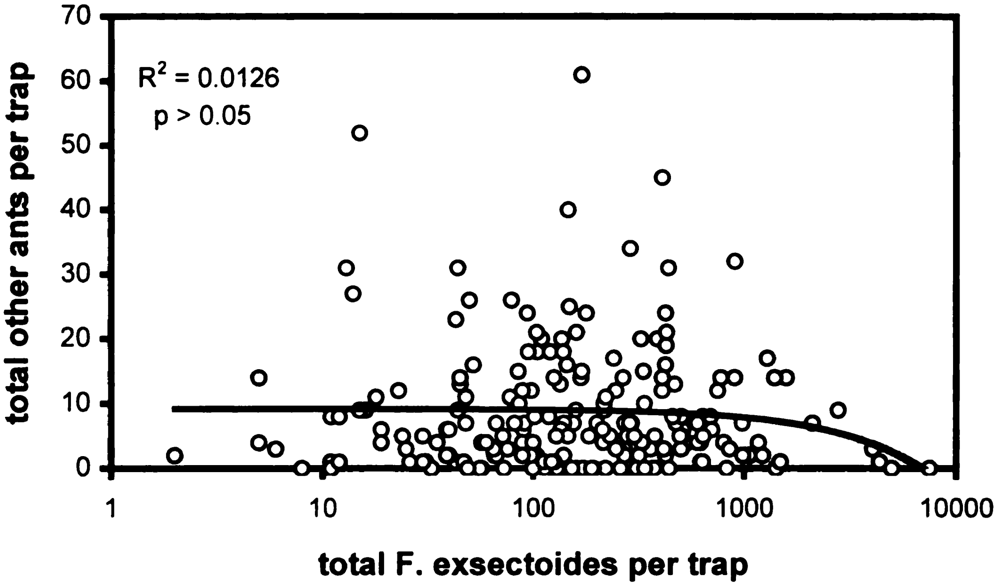

Figure 15. There was no significant relationship between abundance of *F. exsectoides* foragers per trap collection and abundance of other ants.

F. exsectoides forager abundance showed no significant relationship to any species but *C. herculeanis*. Abundance of *C. herculeanis* was significantly but weakly correlated to abundance of *F. exsectoides* ($R^2 = 0.0456$, $p = 0.0001$). Weak but significant relationships between relative abundance and stand age were found in *Aphaenogaster* spp. ($R^2 = 0.1211$, $p = 0.0001$), and *F. fusca* ($R^2 = 0.0445$, $p = 0.0011$). Additionally, abundance of *F. fusca* was strongly correlated to number of species captured ($R^2 = 0.7467$, $p = 0.0001$) (Figure 16a). While the large trap catches of *F. fusca* obtained in Y4 and Y5 (both dense jack pine plantations) lend those two data points strong influence, the relationship is still highly significant and reasonably strong if they are removed from the analysis ($R^2 = 0.4569$, $p = 0.0001$) (Figure 16b).

Herbivores. The Herbivore grouping contains taxa known to feed only on plant material, by chewing or sucking. These include Orthoptera, Hemiptera (Thyreocoridae, Lygaeidae), Homoptera, Lepidoptera, Coleoptera (Curculionidae). While overall proportions of herbivores did not vary significantly among stand age classes ($F = 1.39$, $df_e = 222$, $p = 0.2560$, Table 14), the composition of the herbivore group changed dramatically (Figure 17a,b). Abundance of Orthoptera declined from young to mature stands, then dropped sharply in old stands. Thyreocoridae were present in low numbers in young and mature stands, but increased from 1% of all herbivores in young and mature stands to 19% of the herbivores in old stands. Lygaeidae, the most abundant herbivores in all stand age classes, did not differ with stand age.

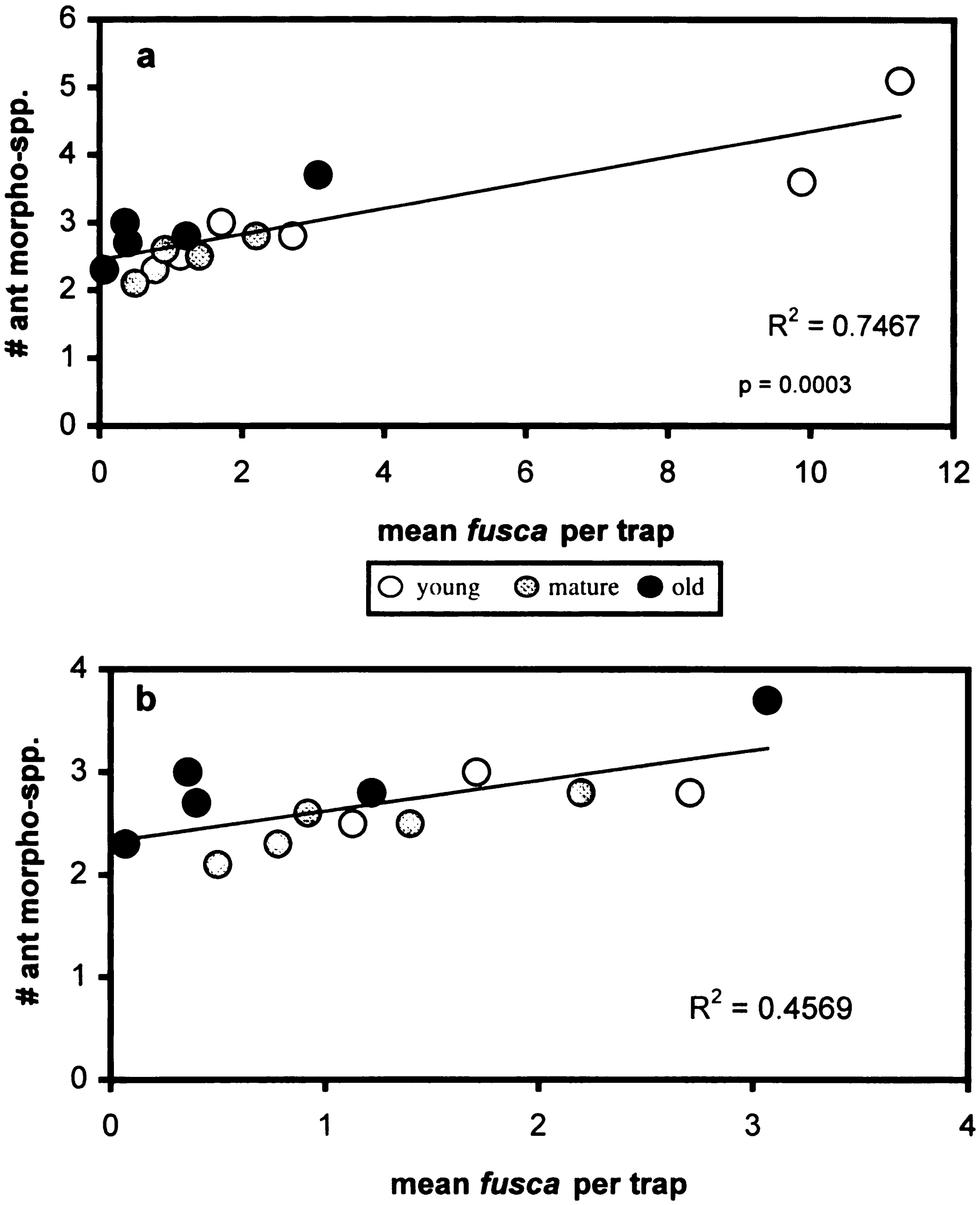

Figure 16. a) There was a significant positive relationship between mean number of *F. fusca* captured per trap and total number of ant morpho-species captured in all traps at a site. b) This relationship remained significant even when highly influential points (Y4 and Y5) were removed from the analysis.

Figure 17. Changes in composition of feeding guilds across stand age classes. a) herbivores b) herbivores (part 2) c) predators d) scavengers

Herbivores are presented in two graphs due to considerations of scale. Abbreviations of taxa are as follows: Or = Orthoptera, He(L) = Hemiptera (Lygaeidae), He(T) = Hemiptera (Thyreocoridae), Ho = Homoptera, La = soft-bodied Lepidopteran and Hymenopteran larvae, Le = Lepidoptera, Co = Coleoptera, Ar = Araneae, Chi = Chilopoda, Op = Opiliones, Di = Diptera, Dp = Diplopoda. Values shown are total pitfall trap captures, summed across traps, dates, and sites within stand age classes.

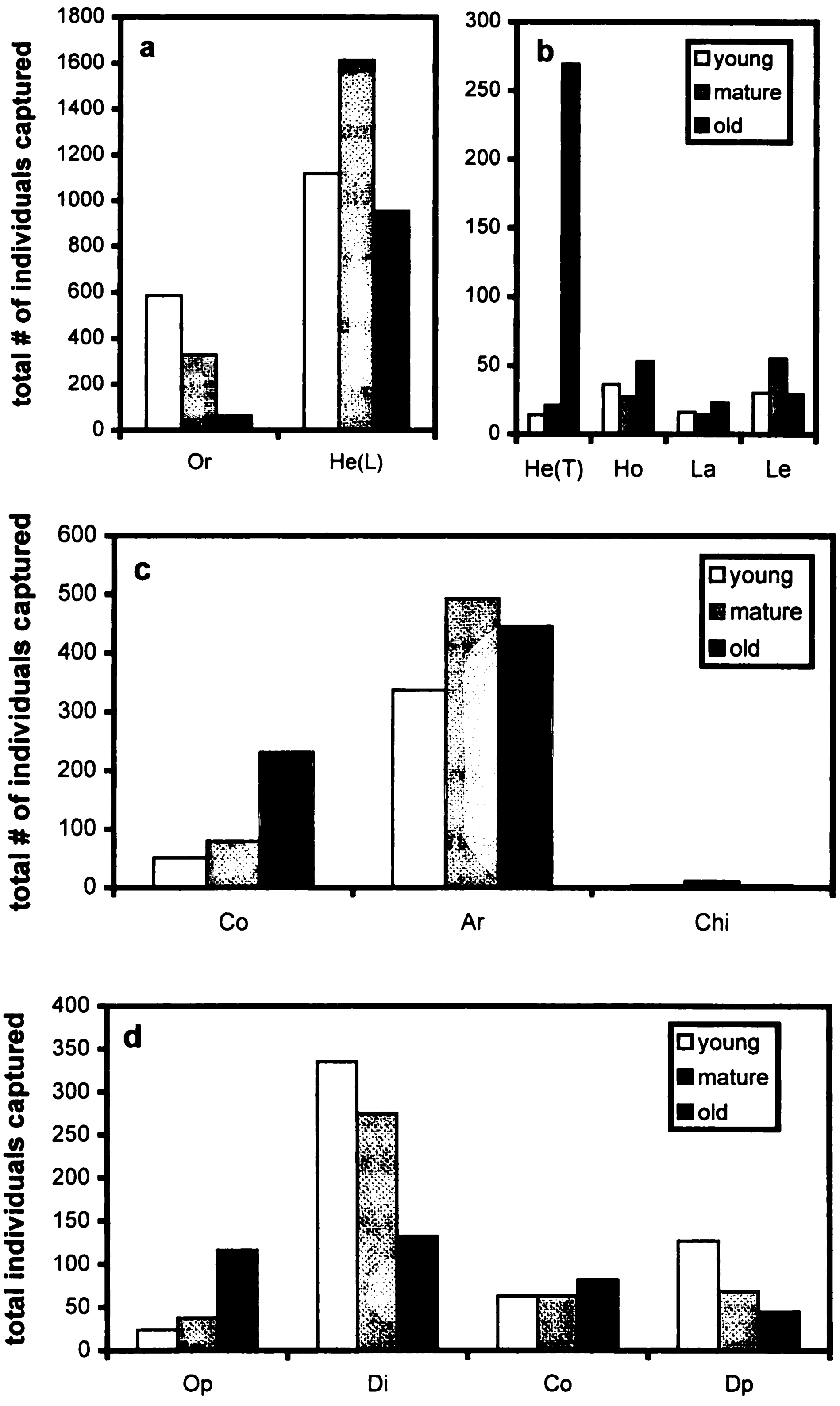

Figure 17. Changes in composition of feeding guilds across stand age classes. a) herbivores b) herbivores (part 2) c) predators d) scavengers

Predators. The primary group of non-ant predators captured was predaceous Coleoptera. These included individuals from the families Carabidae, Staphylinidae, Histeridae, Cicindellidae. Other predators were spiders (Aranae), centipedes (Chilopoda), and Hemiptera (Pentatomidae).

Abundance of predaceous Coleoptera increased dramatically in old stands (Figure 17c). The most commonly captured carabid beetles were *Myas cyanescens* Dejean and *Pterostichus pennsylvanicus* LeConte, both reported by Beaudry *et. al.* (1997) as having reduced abundance in recently clear cut areas.

Spiders as a group did not differ in relative abundance across stand ages; compositional shifts among spider families may have occurred but were not investigated. Centipedes comprised only 0.02% of arthropods sampled.

Scavengers. While many predators scavenge opportunistically, I have restricted the "scavenger" category to invertebrates feeding primarily on dead or decaying plant or vegetable matter. These include arachnids of the order Opiliones (daddy long-legs), Diptera (Mycetophilidae, Bibionidae, Lauxmonidae, Chironomidae, Muscidae, and Sepsidae), Coleoptera (primarily Scarabaeidae), and Diplopoda. Other scavengers present but not analyzed included collembolans and isopod crustaceans. Primary changes in scavenger abundance were due to an increase in Opiliones in old stands and decreases in Diptera and Diplopoda as stands aged (Figure 17d).

Relationships between feeding groups. Table 17 provides a summary of significant correlations between major arthropod orders and families of different feeding groups.

Table 17. Correlation between major invertebrate groups. "Correlation" = Pearson correlation coefficient, generated by PROC CORR. SAS Systems, Inc. Significance determined with a sequential Bonferroni adjustment of p-values. *Indicates values provides for only one stand age class.

group	correlation	R^2	overall p	significant?	age interaction?
F. exsectoides					
Orthoptera	0.279	0.0779	0.0001	yes	yes: R^2 increases to 0.1620 in young stands
Opiliones	-0.321	0.1037	0.0001	yes	yes: similar in old, young, disappears in mature
Coleoptera (P)	-0.243	0.0593	0.0006	yes	no
Opiliones					
Coleoptera (P)	0.484	0.2344	0.0001	yes	yes: similar in old, young, disappears in mature
Orthoptera	-0.188	0.0354	0.008	**marginal**	no
Lygaeidae	0.512*	0.2619*	0.0001*	yes*	*yes: only present in young stands
Coleoptera (P)					
Lygaeidae	0.161	0.0259	0.0234	no	yes: significant in young stands, marginal in old

Discussion

Location and density of *F. exsectoides* mounds. Stand age and tree density were correlated, but stand history and other stochastic affects may have more important impacts on vegetation than the passage of time alone (Figure 10). Much variation existed among young sites, due to differences in stand treatment/histories (Table 9). Y1 and Y2 were quite dense, due to recent reforestation by the United States Forest Service. Y3, a naturally regenerating clear-cut, was extremely sparse because jack pine is adapted for establishment after burning and may not regenerate well after harvest (Beland and Bergeron 1993, Zoladeski and Maycock 1991). Y4 and Y5 were of medium density, with high vegetational heterogeneity due to the patchiness of natural burn regeneration. Even within mature stands, much variation may exist due to small fires, tree mortality caused by disease or herbivory, storm damage, and thinning treatments. Old stands are increasingly vulnerable to damage and have greater probability of impact from random mortality factors simply by virtue of having been around longer.

F. exsectoides has been described as an opportunist species—although dispersal between patches may be risky and limited, once a suitable patch is colonized colonies quickly expand to control the entire territory (Oster and Wilson 1978). This dispersal limitation may be the key factor explaining high densities and smaller average mound surface area in young (especially clear-cut) stands (Figure 12b). In clear-cut stands, removal of trees and some ground vegetation by mechanical harvesting may be followed by several years of delay before replanting, potential cause for colony mortality *(personal observation*). Mechanical disturbance by machinery used to re-plant the stand may further diminish surviving populations, leaving open territories for new colonies. Recolonization of the site by *F. exsectoides* may be delayed by several factors. Colonists arriving at early stages of vegetation regeneration (before prey populations have established) may lack adequate resources for colony growth. Additionally, *F. exsectoides* queens found new colonies through temporary social parasitism of *Formica* of the *fusca* species group, and thus depend on the prior presence of a suitable host species to succeed in entering a 'new' habitat (Wheeler 1928, Wilson 1971).

The larger average mound size in old stands may be related to age of the population (older mounds may be larger) (Andrews 1926), but it also reflects the presence of fewer small mounds, possibly new mounds formed by budding or mound fission (Figure 12a,b). A large number of abandoned mounds and a lack of new mound establishment show the decline of F. exsectoides populations in old jack pine stands. Mound distribution was not different from a null expectation based on random dispersion. Isozyme studies performed by Nipson (1978) suggested that most *F.exsectoides* populations were the result of a single initial founding event, followed by budding of the primary colony and occasional acceptance of immigrant queens into these offshoot colonies. Given budding or colony fission as the usual method of within-site colony expansion, a null expectation of mound **clumping** seems reasonable. A lack of apparent clumping may therefore actually indicate some repellence between mounds, or specific nest sight requirements (such as microclimate effects) that may overrule any territory requirements for individual mounds.

Abundance of *F. exsectoides* and other ground-dwelling arthropods. Even at the coarse resolution of order-level comparisons, it is obvious that major shifts in ground arthropod communities occur as jack pine stands age. Significant changes in proportions of ants and predators occurred across stand age groups. This is inconsistent with the findings of Heatwole and Levins (1972), that trophic stucture remains constant on mangrove islands even while composition of trophic guilds varies. As this study only incorporates ground dwelling fauna, it may be that trophic guild stability is maintained when arboreal arthropods are considered. Proportions of herbivores and scavengers did not change, but alterations in the composition of these groups were evident.

Ants. The numerical dominance of *F.exsectoides* is in keeping with published accounts of their large population sizes and pugnacious natures (McCook 1877, Creighton 1950). Variation in numbers of *F. exsectoides* captured was substantial, and particularly large samples may have been caused by recruitment of foragers to trap areas. This variation may have concealed a correlation between stand age and *F. exsectoides* forager abundance, as differences were significant when stands were grouped as age classes (Table 15). Overall, *F. exsectoides* foragers were most abundant in mature stands, and least abundant in old stands.

The measurement of mound surface area provides only a comparative index of surface dimensions. As mound surface dimensions are only weakly correlated to overall nest volume (Bristow *et. al.* 1992), it is not surprising that this measurement was not correlated with forager abundance. Mound density, however, was correlated with forager abundance, lending support to the intuitive assumption that more mounds means more ants (Figure 14).

Levins and Traniello (1981) state that *F. exsectoides* is not a "truly territorial" ant (defending "territory" or potential foraging area), but defends only the nest and persistent food resources such as aphid colonies. Given the density of mounds and the extent of polydomy in *F. exsectoides* populations (Holldobler and Wilson 1990), their defense behavior seems functionally equivalent to that of a territorial species. As would be expected in that case, abundance of other ants was quite low. The absence of significant correlation between abundance of *F. exsectoides* and abundance of other ants (either as a group or as individual species) may arise from the general scarcity of non-*exsectoides*. Other species may be suppressed so effectively that responses to fluctuations in F. exsectoides abundance are not statistically detectable; a true test would require sampling in areas where *F. exsectoides* is absent.

The ants most commonly co-occuring with *F. exsectoides* were *Aphaenogaster* spp. and *F. fusca* (Table 15). Abundance of both showed weak but significant correlation to stand age, with *F. fusca* declining as stands matured and *Aphaenogaster* increasing. As the host species for *F. exsectoides* colony founding, establishment of *F. fusca* in a habitat must precede colonization by *F. exsectoides* (Wheeler 1928, Creighton 1950). *F. fusca* can then be expected to decline as *F. exsectoides* colonizes young habitats by parasitizing *fusca* nests. The relationship might be stronger if stands were sampled earlier in the colonization process. *Aphaenogaster* increases primarily in old stands; this may be due to a habitat preference or the decline of *F. exsectoides*.

F. fusca is described as "the submissive ant" (Savolianen 1990), or "singularly lacking in pugnacity" (Creighton 1950). This "docility", including a relative lack of even nest defense, may contribute to the large number of parasitic and slave-making species utilizing *F. fusca* as a host (Holldobler and Wilson 1990). Since Creighton (1950) also considers *F. fusca* is also considered to be "exceptionally adaptable in their environmental relationships", it seems more likely that their presence is limited by the presence of other species than by changes in habitat. The strong correlation between abundance of *F. fusca* and number of ant species found at each site may indicate that *F. fusca* is particularly amenable to coexistence, or that habitats where *F. fusca* has been suppressed are also unsuitable for more aggressive ant species.

It is interesting to note that, of eight ant species found to occur in jack pine, four are considered "pest" species (Patterson 1994). *F. exsectoides* alone is considered a pest within its natural setting, because of the damage it causes to young pine trees (Wilson 1977). *C. herculeanis*, *T. sessile*, and *M. minimum* are pests of human dwellings; *T. sessile* and *M. minimum* are particularly known for their cosmopolitan distribution and variety of habitats (Smith 1947). Factors contributing to this success may allow them to coexist with *F. exsectoides* in a habitat that is comparatively species-poor.

Herbivores. The overall decline in herbivores captured in pitfall traps is correlated to stand age. This may represent an overall shift in herbivore activity from ground vegetation to trees as trees mature and ground vegetation becomes less abundant due to shading or increased soil acidity caused by the decomposition of pine needles (Hendrix *et. al.* 1988). The sharp decline in numbers of orthopterans as stands mature is not quite matched by the sudden increase in Hemiptera in old stands. This change is almost entirely due to the increase of the small, well-armored Thyreocoridae (*Galgupha* spp.), generalist feeders on weedy plants. The presence of phloem-feeding insects, particularly homopterans, in pitfall traps is likely due the individuals being carried by ants.

Predators. Aside from *F. exsectoides*, the most numerically abundant predator in stands of all age classes were Aranae (spiders). The abundance of spiders was relatively constant across stand age classes (Table 13). Halaj *et. al.* (1997) found a negative impact of ants on hunting spiders in Douglas fir stands, but no relationship between ants and web-spinning spiders. A similar pattern may exist in this system, but spiders were not divided for analysis.

The primary source of variation in proportions of feeding groups between stand age classes was changing predator abundance; this change was largely caused by the sharp increase in predaceous Coleoptera in old stands. The three families of predaceous beetles found frequently in jack pine were Carabidae, Staphylinidae, and Histeridae. Carabidae and Staphylinidae are both very diverse families, occupying diverse habitats. It is surprising that these families are so poorly represented in young and mature stands. Beaudry *et. al.* (1997), in comparing carabid communities in burned and clear cut jack pine stands, found similar abundance and diversity of carabids in both stand types, but different species composition. The presence of *F. exsectoides* is therefore the more likely factor limiting carabid presence in young and mature stands, rather than habitat suitability. Hister beetles prey primarily on organisms attracted to decomposing plant or animal material (Borror, Triplehorn, and Johnson 1989), and thus may have been attracted to pitfall traps as good foraging locations.

Scavengers. Scavengers were especially likely to be attracted to pitfall traps by the odor of decomposition. Significant changes in scavenger abundance across stand age classes included a rise in numbers of Opiliones captured as stands aged and declines in Diptera and Diplopoda from young to old stands (Table 13, Figure 17d). Some Opiliones are predaceous. Moving Opiliones from the Scavenger to the Predator group creates a stronger downward trend in scavenger abundance as stands age and a stronger increase in predators in old stands. Scavengers, particularly those consuming decaying plant material, may follow the same trend as Herbivores—a decline in ground vegetation suggests a decline in rotting vegetation on the ground, and acidic conditions presented by decaying jack pine needles may discourage detritovores. This may explain a decline in Diplopoda, but explaining decline in the more diverse scavenging Diptera may require detailed analyses focused on that group.

Correlations between feeding groups. Of correlations significant after correction for multiple comparisons, several are explained by shared correlation to habitat: Opiliones and predaceous Coleoptera are both found primarily in old habitats, and are both negatively correlated to *F. exsectoides* abundance, which declined in old stands. Orthoptera were found primarily in young habitats, and were negatively correlated to Opiliones (Table 17). More interesting are the positive relationship between *F. exsectoides* and Orthoptera, and the negative relationships between *F. exsectoides* and predaceous Coleoptera and Opiliones. Although the correlations were weak and could be explained by habitat as above, all three of these correlations were stronger than the correlation between *F. exsectoides* and stand age.

F. exsectoides is an extremely generalist predator, feeding on anything that it can catch (Dimmick 1951, Ayre 1970). *F. exsectoides* have been observed feeding on Orthoptera, and I encountered Orthopteran fragments during nest excavations performed for sex ratio studies (see Ch.1, 3). Where Orthoptera are abundant, they could provide an abundant protein source for *F. exsectoides* brood development. Both predatory beetles and Opiliones may compete with *F. exsectoides* for living or dead protein resources. Alternatively, this too may be a habitat-associated correlation, where *F. exsectoides* and predaceous Coleoptera/Opiliones have opposing responses to an unmeasured environmental variable relating to stand age.

Conclusions. *F. exsectoides* was numerically dominant in all age classes of jack pine, but had a greater abundance of foragers and higher mound density in mature stands. Smaller *F. exsectoides* presence in young stands is probably due to the recent establishment of these populations, while the lower abundance and density in old stands is more likely a product of population decline. This suggests that *F. exsectoides* is limited by the "life span" of its habitat. Populations show the "dense, thorough occupation" of habitats described by Oster and Wilson (1978), but lower forager abundance and mound density in young stands indicates that *F. exsectoides* still requires substantial time investment to capitalize on a newly-colonized habitat. While the population decline seen in old stands may be exacerbated by competition from increasing numbers of predaceous arthropods, it is difficult to separate this influence from the impact of abiotic factors such as temperature and shading on brood development.

The ground arthropod community of jack pine changes significantly both in proportions and composition of feeding groups. Many of these changes seem likely to be driven by changes in ground vegetation as the trees mature and the canopy closes, while others may have a less direct relationship, being influenced by age-correlated changes in abundance of predators or competitors. The ant community exemplifies this pattern, as *F. exsectoides*, *F. fusca*, and *Aphaenogaster* spp. responded significantly to stand aging, *C. herculeanis* responded significantly to abundance of *F. exsectoides*, and the diversity of ants increased with the abundance of *F. fusca*.

Despite the well-described aggressiveness of *F. exsectoides*, this study provided no strong evidence that *F. exsectoides* excludes other ant species. Roberts and Gilliam (1995) review the proposed role of disturbance in maintaining species diversity by preventing competitive exclusion. If such an effect was present in this system, it might be revealed by more thorough examination of young jack pine stands (both burned and clear cut), before and after colonization by *F. exsectoides*. Additional data on colonization success, dispersal patterns, and habitat availability is necessary to comprehend the constraints and adaptations arising from specialization on transient environments.

CHAPTER 3

The impact of jack pine habitat succession on reproductive strategy of the Allegheny mound ant, *Formica exsectoides* Forel

Introduction

The study of allocation of reproductive efforts is essential to understanding the ecology and evolution of all organisms. Intrinsic to this study is the concept of the "trade-off": energy devoted to reproduction must be diverted from other tasks, such as growth, defense, or storage (Cody 1966, Lande 1982, Charnov 1993). Successful strategies are expected to possess features that will optimize allocation to reproduction versus other life functions.

Evaluating this trade-off for sexually reproducing organisms is more complicated. The cost of sexual reproduction (or "the cost of males" (Maynard Smith 1978, Williams 1975)) is assumed to be offset by an advantage gained through recombination, though the nature of that advantage (and its existence) remains in debate (Hurst and Peck 1996, Kondrashov 1993, Crow 1994). Organisms possessing the ability to reproduce by both sexual and asexual means are expected to reproduce sexually only when there is a benefit to doing so.

In addition to alternating between sexual and asexual strategies, reproductive strategies can include adjustment of the sex ratio, or allocation to males versus females. While it is intuitively obvious that the "cost of sex" may be minimized by producing fewer males, Fisher's (Fisher 1930, Trivers 1985) classic theory on the evolution of sex ratios proposes an explanation for the existence of equal investment sex ratios in so many diploid species. The basic argument is economic in nature—the supply of either sex will vary with the cost of production and the "demand" (a measure of mating success, dependant on availability of the opposite sex). When males and females are equal in cost, sex ratio should approach an equilibrium value of equal investment, since over-production of a rare sex will always be advantageous and overproduction of a common sex will be detrimental. This theory has been elaborated to include ecological and genetic factors leading to skewed sex ratios, such as Local Mate Competition (Hamilton 1967), Local Resource Competition (Clark 1978), differential mortality (usually of males), and inbreeding (Thornhill 1993). The theory of kin selection (Hamilton 1963, 1964, Trivers and Hare 1976) has led to an explosion of sex allocation studies of the eusocial Hymenoptera, particularly ants (for review see Nonacs 1986, Crozier and Pamilo 1996). The bulk of this study has been aimed at testing genetic relatedness hypotheses, with far fewer studies focused on the role of sex ratio as an environmentally variable reproductive strategy.

The genetic-relatedness hypothesis of sex ratio is based on the relatedness asymmetry between workers, males, and queens that is created by the haplo-diploid genetic structure of Hymenoptera (Figure 18). Elevated relatedness between female siblings provides not only an explanation for the evolution of eusociality (workers gain more by rearing their sisters than by producing their own offspring), but is expected to lead to female-biased sex ratios in colonies where workers control sex allocation (Trivers and Hare 1976). Elaboration of the genetic relatedness hypothesis to incorporate

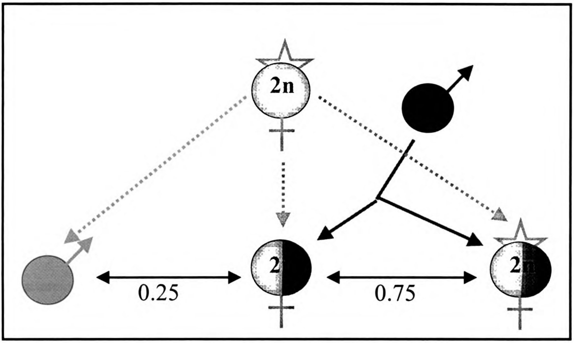

Figure 18. Creation of relatedness asymmetry by haplo-diploidy, as described by Trivers and Hare (1976).

polygyny, polyandry, and worker reproduction are reviewed in Bourke and Franks (1995) and Crozier and Pamilo (1996).

Theories regarding environmental influences on sex ratio are primarily based on resource-availability, although proposed mechanisms differ. The theory of local resource competition (Clark 1978) has been invoked as a possible explanation for male-biased sex ratios (Crozier and Pamilo 1996). In areas where resources are limited, female production would be expected to decrease if female offspring will compete directly with their mothers for resources. This theory is presented in a more general form by Cannings and Cruz Orive (1975), who predict a bias toward the more dispersive sex. Deslippe and Savolainen (1995) focused on food as the limited resource, but suggested a mechanism based on nutritional physiology (Wheeler 1994) rather than competition. They observed an increased allocation to female production in colonies of *Formica podzolica* that had been offered increased food and protein resources, and hypothesized that female allocation is often limited by colony nutrition (since underfed diploid offspring will develop as workers, while haploid offspring will always develop as males). Elmes and Wardlaw (1982a,b) hypothesize a relationship between female production and nest insolation, but present no specific data. Environmental factors may also have indirect effects on sex ratio, as Herbers (1993) describes evidence that ecological factors such as habitat and food availability may encourage polygyny, thereby altering the relatedness structure of colonies and potentially sex ratio.

Formica exsectoides Forel presents a particularly interesting challenge in the study of reproductive strategies in ants. While many features of its biology make the prospect of testing genetically-based models of sex ratio quite daunting (see Chapter 1: Discussion), it presents an excellent opportunity for investigating environmental impacts on reproductive strategy.

Reproduction in *F. exsectoides* can occur by colony fission/budding, or by production of alate sexual forms (Creighton 1950). Mated females attempt to re-enter established colonies, or found new colonies through temporary social parasitism of *Formica fusca* colonies (Wheeler 1928). *F. exsectoides* colonies are long-lived (Andrews 1929) and are not obligated to produce sexual forms for colony expansion within habitats.

F. exsectoides inhabits transient, early successional forest habitats. Colonies appear to decline as the forest ages (Andrews 1929, Chapter 2: Location and density of *F. exsectoides* mounds, Abundance of *F. exsectoides*). Several authors have hypothesized that insolation is an important factor (Cory and Haviland 1938; Wheeler 1928; Andrews

1926, 1929), but this has not been quantified. Numerous features of the habitat will be expected to vary along successional gradients, including abiotic factors (such as light availability) and biotic factors (such as composition of plant and animal communities). This variation follows predictable patterns, allowing adaptive changes in life history patterns to take place. Organisms inhabiting transient habitats should possess strategies to quickly exploit optimal habitats, and to alter or escape suboptimal habitats. Since colony growth may occur by sexual or asexual processes, and dispersal to new habitats is dependent on winged sexual forms, both allocation to sexual forms and variations in sex ratio represent potential strategic responses to environmental change.

The objective of this study is to determine whether differences in reproductive (brood production) status of *F. exsectoides* mounds and variation in sex ratio are consistently related to habitat age. In addition to stand age, I examined light availability which, while correlated to stand age, may be expected to vary within stands. Population-level variables, such as mound density and forager abundance are examined as potential links between changes in the forest environment and changes in reproductive allocation.

Methods

Site selection and description. Populations of *F. exsectoides* were studied in stands of jack pine (*Pinus banksiana* Lamb.) within the Huron National Forest, Michigan. Five sites in each of 3 jack pine age categories were selected to represent a gradient of jack pine succession. Further details on sites are presented in Chapter 2, with locations and descriptions of sites listed in Table 9.

Environmental variables. One 2500 m^2 plot was placed at random within each of the 15 study sites. These plots were mapped as a grid of 5m x 5m squares, using a compass, marker flags, and surveyor's rope. The location of all anthills was noted, even those that appeared to be newly-formed buds. A comparative index of mound surface area was calculated as longest slope x shortest slope in meters. Mounds were numbered and marked with wire-stake flags. Location of all trees > 1 cm diameter and overall stand density (cm dbh/m^2 plot) was recorded, as described in Chapter 2. Pitfall traps were used to measure relative abundance of *F. exsectoides* foragers and other ground-dwelling arthropods; trap placement and collection regimes are presented in Chapter 2.

Light availability. Light availability was measured using hemispherical photography. A Nikon camera mounted with an 8mm Sigma fish-eye lens was placed above each anthill and oriented north. Slides were taken above each anthill sampled for sex ratio, five additional anthills that were not sampled for sex ratio (if available), and above each pitfall trap. Pitfall traps were placed in fixed positions with respect to plot boundaries, therefore they were placed randomly with respect to anthills and provide a null expectation or random measurement of stand light availability. Slides were digitized and analyzed using the program GLI/C (Canham 1995), with defined latitude of 44.55°N and a growing season from 90 to 304 Julian days.

Mound reproductive status. Reproductive allocation was examined at the pupal stage, when the sex of individuals is identifiable by morphology but confusion caused by dispersal or loss of individuals to predation is not yet a factor. Whether mounds are producing sexual forms is also best determined at this stage, as pupation occurs in the upper portions of the mound. Sexual forms in the egg or larvae stage may never reach maturity, so observing these stages might provide a misleading picture of mound reproductive output. Mounds outside plot boundaries were periodically examined to check for pupation of reproductive forms, avoiding disturbance of study mounds before sampling. After pupation had occurred, all mounds within a plot were partially excavated to record the presence or absence of reproductive caste brood. Reproductive brood was considered absent if excavation of the mound to approximately 0.5m below the soil surface and sifting of the mound material through a large strainer revealed no reproductive caste pupae, prepupae, larvae, or alate adults. Mounds containing neither sexual nor worker brood were classified as "dead" even if adult workers were present on the mound surface. Mounds were excavated in an order based on computer-generated random number series to avoid biasing data by preferentially selecting large mounds for excavation.

Sex ratios. Samples of 30 reproductive-caste pupae were collected from the first 5 sexual-producing mounds excavated within each plot. Pupae were individually weighed and stored in a 10°C incubator for 10 days. This allowed development to continue, avoiding weight differences due to stage of development and problems with inability to determine sex of "undeveloped" pupae (see Chapter 1). The heaviest 5% of each sample was removed and frozen at -80°C for genetic analyses (see Appendix B: Attempted search for diploid males). These pupae were later returned to their original samples. Pupal cases were removed and the sex of each pupa recorded. Pupae were dried for 3 days at 70°C and re-weighed. A nested analysis of variance was used to analyze differences in pupal dry weights between sexes and among sites. Sex ratios were calculated using Boomsma's (1989) Energetic Cost Ratio (Bourke and Franks 1995), and sex ratios are expressed as proportion of male allocation.

Statistical analyses. An initial correlation analysis was performed to investigate the relationship between environmental variables. Mound density (mounds/m^2), stand density (cm dbh/m^2 plot), stand age (y), and light availability (% open sky) were analyzed using PROC CORR (SAS Systems 1990).

The percent of all living mounds within each stand that produced sexual caste pupae was correlated with the above variables. When significant correlation occurred between the proportion of sexual-producing mounds and two or more correlated variables, only comparisons with highest explanatory value are considered for discussion.

As 'young' stands were of heterogeneous origin (3 clear-cut, 2 burned (see Table 9)), analyses involving stand age were also performed using date since last cut (rather than date since major disturbance, i.e. cut **or** burn) for values of stand age. As *F. exsectoides* populations may be reduced or destroyed by clear-cutting but are in high abundance after fires (see Chapter 2 Discussion: Location and density of *F. exsectoides* mounds), date since last cut provides a more reasonable estimate of the age of the ant population, while date since major disturbance provides a better estimate of patch age.

Analysis of variance was used to analyze differences between light environments within stands. Specific comparisons were made between mounds producing sexual forms and mounds producing only workers, mounds producing brood and dead mounds, and mounds producing brood and random forest.

A nested analysis of variance was used to separately analyze differences in weights of each sex between sites and between mounds within sites. Binomial probability curves were generated to provide confidence curves for sex ratio estimates.

As comparisons were of greater interest than estimating actual values of sex ratio and mound-level sex ratio data were distributionally-challenged, a logistic regression (PROC CATMOD, SAS Systems) of raw count data was used to analyze the effect of mound-specific variables on sex ratio (Juliano 1993). The variables used in these analyses were light availability (% open sky) and mound surface area indices.

For analyses involving site-level variables, site averages for sex ratio were analyzed using simple regression. These analyses used stand age, % open sky, mound density, and *F. exsectoides* forager abundance as predictor variables. Additionally, sex ratio was regressed on abundance of *Formica fusca* L. As *F. fusca* is the host species for nest founding by *F. exsectoides* queens, female investment would be expected to increase in areas where *F. fusca* (and therefore nest sites) are abundant.

Results

Environmental and mound variables. Age-class means and standard deviations for mound density and tree density are presented in Chapter 2, Table 10. Age class means and standard deviations for abundance of ants are presented in Chapter 2, Table 15.

Light availability ranged from 11.8 to 82.4 percent open sky. While correlated with stand age, light environment was highly variable within stands ($R^2 = 0.4834$, $p = 0.0001$; Figure 19a).

The results of correlation analyses including mound density, stand density, stand age, light availability, and forager abundance values are presented in Table 18. Percent open sky is significantly and negatively correlated to both stand age and stand density. Relative abundance of *F. exsectoides* foragers was significantly correlated to mound density alone. Marginal correlations were present between mound surface area indices and stand age/open sky; all other correlations were not significant.

Mound reproductive status. Collections of reproductive caste pupae took place from July 19 to July 24, 1997. Of 241 mounds examined, 93 (38.5%) were dead and reproductive caste pupae were found in only 91 (38.1%). A few mounds in older stands (O4 and O1) contained only reproductive caste larvae at the time of initial excavation. Table 19 provides a summary of mound status for each site and each stand age class.

The percent of mounds examined that were classified as 'dead' increased significantly with stand age ($p = 0.0006$, Table 20), as did the proportion of mounds producing sexual forms ($p = 0.0107$, Table 21; Figure20). The percentage of dead

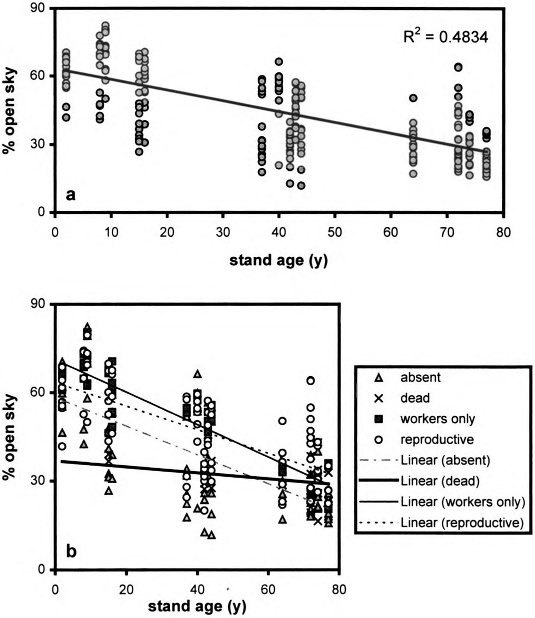

Figure 19. a) While correlated with stand age, light environment was highly variable within stands. b) Status groups differed in their relationship to the range of light available

Table 18. Correlation between environmental and F. exsectoides population characteristics. After correction for multiple comparisons, only correlations marked '***' ($p < 0.001$) can be considered significant. Correlations marked '**' ($p < 0.01$) or '*' ($p < 0.05$) were considered to be "marginal". Direction of correlation is listed below significance. "Mound size" refers to comparative surface area indices, NOT an absolute measure.

	stand age	stand density	% open sky	mound density	mound size
stand density	** (+)				
% open sky	*** (-)	*** (-)			
mound density	ns (-)	* (-)	ns (+)		
mound size	* (+)	ns (+)	* (-)	ns (-)	
forager abundance	ns (-)	ns (-)	ns (+)	** (+)	ns (-)

Table 19. Summary of mound status for each site and age class. Totals represent all mounds present in a 2500 m^2 plot. "Active" mounds contained brood, either a mixture of reproductive caste and worker brood or worker brood only. "Dead" mounds showed little or no ant activity and contained no brood.

	mounds mapped	active mounds		dead mounds
		reproductive	workers only	
young	59	26	26	7
Y1	14	6	7	1
Y2	17	7	7	3
Y3	11	4	6	1
Y4	9	5	2	2
Y5	8	4	4	0
mature	101	34	25	42
M1	20	5	11	4
M2	18	6	5	7
M3	13	9	2	2
M4	20	7	6	7
M5	30	7	1	22
old	81	31	6	44
O1	12	5	1	6
O2	26	6	4	16
O3	15	6	1	8
O4	15	10	0	5
O5	13	4	0	9

Table 20. ANOVA test of significance for the regression model: % dead mounds = stand age, when "stand age" = years since last major disturbance.

Source of variation	df	mean square	F	p
stand age	1	4621	19.88	0.0006
error	13	232		
total	14			

ANOVA test of significance for the regression model: % dead mounds = stand age, when "stand age" = years since last clear cut.

Source of variation	df	mean square	F	p
stand age (cut)	1	2539	6.47	0.0245
error	13	393		
total	14			

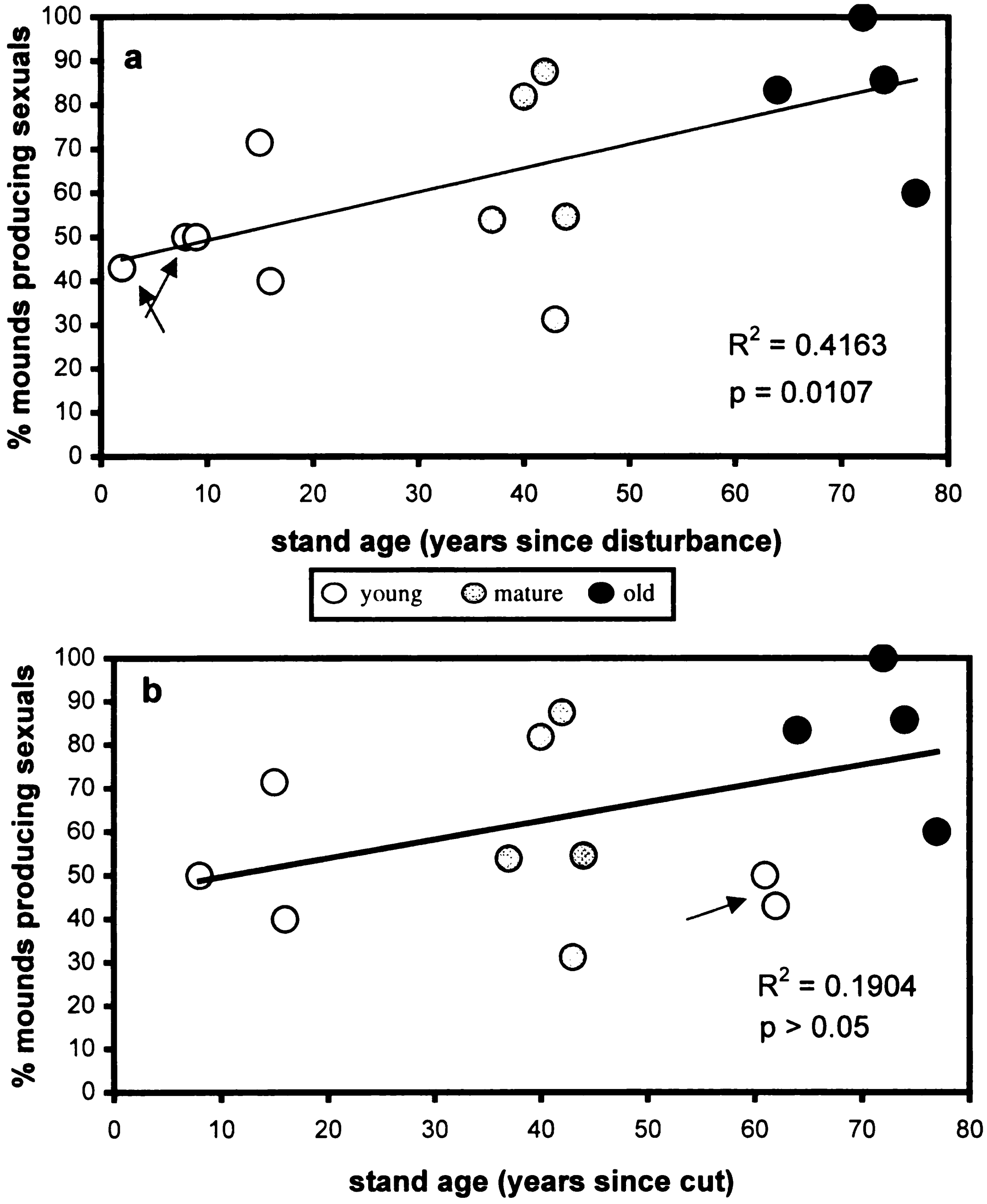

Figure 20. a) The percentage of mounds producing sexual caste brood increased with stand age, when "stand age" = date since major disturbance. b) This relationship was no longer significant when "years since clear-cut" is used as stand age. Arrows indicate burned stands.

Table 21. ANOVA test of significance for the regression model: % of mounds producing sexual forms = stand age, when "stand age" = years since last major disturbance.

Source of variation	df	mean square	F	p
stand age	1	2772	8.86	0.0107
error	13	313		
total	14			

mounds was, however, more strongly related to light availability (p = 0.0004, Figure21). The percent of mounds producing sexual forms was dependent on mound density in mature stands, but relatively independent of mound density in both young and old stands (Figure22 a, b).

Percent of mounds producing sexual forms was correlated to stand age, stand density, and light availability (Table 22), but stand age explained the highest proportion of variance.

Light environments differed significantly with mound status (Table 23, Figure23). There was no significant difference between light environment in locations where mounds were absent and locations where mounds were dead (p = 0.0672), but light environments of inhabited mounds had significantly greater percentages of open sky than either absent or dead (0.0001). Additionally, mounds containing worker brood only were found in locations with significantly greater percentages of open sky than mounds producing sexual forms (p = 0.0037).

Sex ratio. Pupal dry weights differed between sexes and between stand age classes (Table 24, p = 0.0001) (Figure 24). Dry weights of males and females were highly correlated to each other within stands. Dry weight was not related to sex ratio (Figure 25).

Sex ratios were male-biased at all sites, but sites differed in the composition of sex ratio (Figure26). Sites Y2, M3, M4, M5, and O2 display patterns indicative of "split" sex ratio (Boomsma and Grafen 1991; Godfray and Werren 1996), while other sites display a less-severe but more consistent male bias. While sites Y2 and M3 had sex ratios not

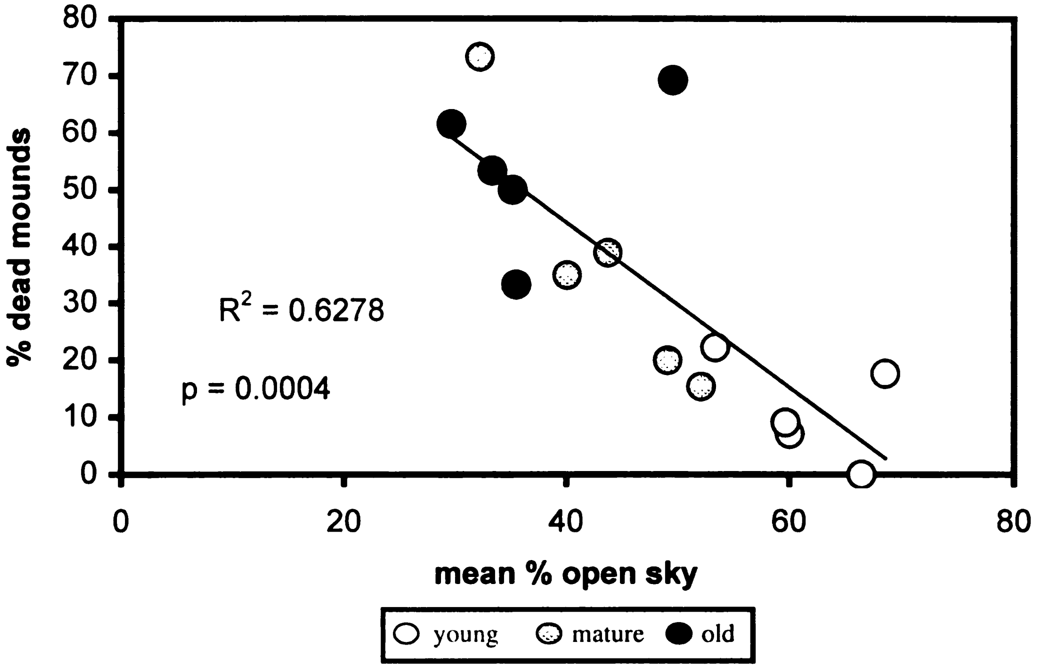

Figure 21. The percentage of mounds classified as "dead" (containing no brood, inactive) increased significantly as open sky decreased. Values of open sky are plot mean values.

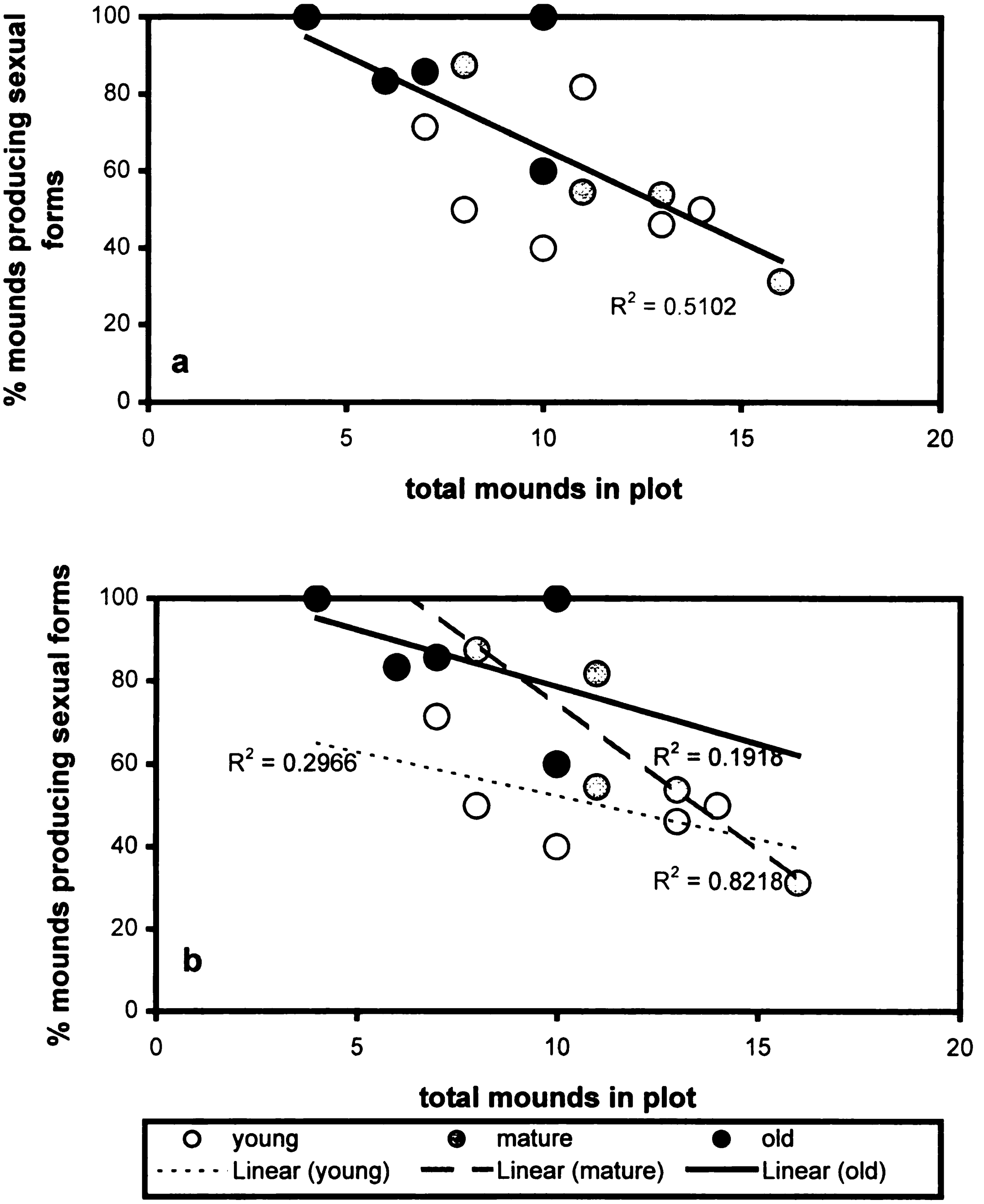

Figure 22. a) Sites with more mounds contained lower proportions of sexual-producing mounds. b) Plotting the interaction of this relationship with stand age reveals that this relationship is strongest in mature stands, while old and young stands are likely to produce sexual brood in a constant proportion of mounds.

Table 22. Correlation between percentages of dead and sexual-producing mounds and stand-specific variables. After correction for multiple comparisons, only correlations marked '***' (p < 0.001) and '**' (p < 0.01) can be considered significant. Correlations marked or '*' (p < 0.05) were considered to be "marginal". Direction of correlation is listed below significance. For the three correlated variables, stand age, stand density, and % open sky (see Table 18), shaded blocks indicate the correlation with greatest strength.

	stand age	stand density	open sky	mound density	forager abundance
% dead mounds	*** (+)	*** (+)	*** (-)	* (-)	ns (-)
% reproductive mounds	* (+)	* (+)	* (-)	** (-)	ns (-)

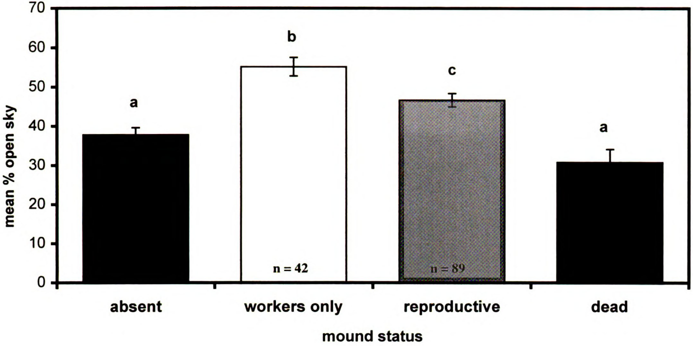

Figure 23. Light availability differed significantly among mounds of differing "status". "Absent" represents values of open sky measured where no *F. exsectoides* mounds were present. "Dead" represents values of light availability for mounds that were abandoned or producing no brood. "Workers only" and "reproductive" indicate absence or presence of reproductive caste pupae in active, brood-producing mounds. Values presented are least-squares means and standard errors. All differences were significant at $p < 0.001$

Table 23. ANOVA test of H_0: Light environment (% open sky) is equal for all mound "status" groups. Status groups were: absent = no mound present at measurement location; dead = inactive, not producing brood; workers only = producing only worker brood; reproductive = producing both sexual caste and worker brood.

Source of variation	df	mean square	F	p
status	3	4345.5	17.3	0.0001
error	228	251.2000		
total	231			

Table 24. ANOVA test of hypotheses H_0: Pupal dry weight is equal for males and females and H_0: Pupal dry weight does not differ among stand age classes.

Source of variation	df	mean square	F	p
model	3	0.00013049	115.01	0.0001
age	2	0.00008951	78.89	0.0001
sex	1	0.00022857	201.45	0.0001
error	1735	0.00000113		
total	1738			

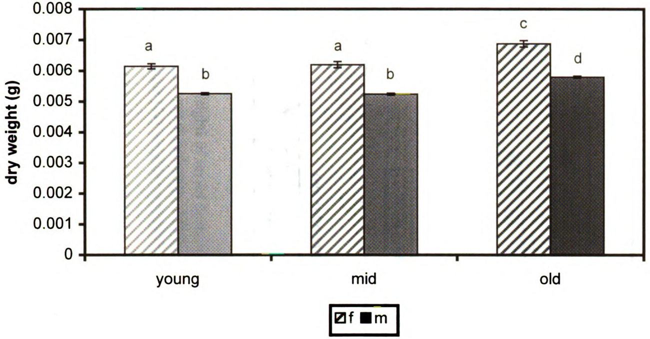

Figure 24. Females were significantly heavier than males. Both sexes were heavier in old stands than in young or mature stands. Values presented are least-squares means and standard errors. Significance tests are presented in Table 24.

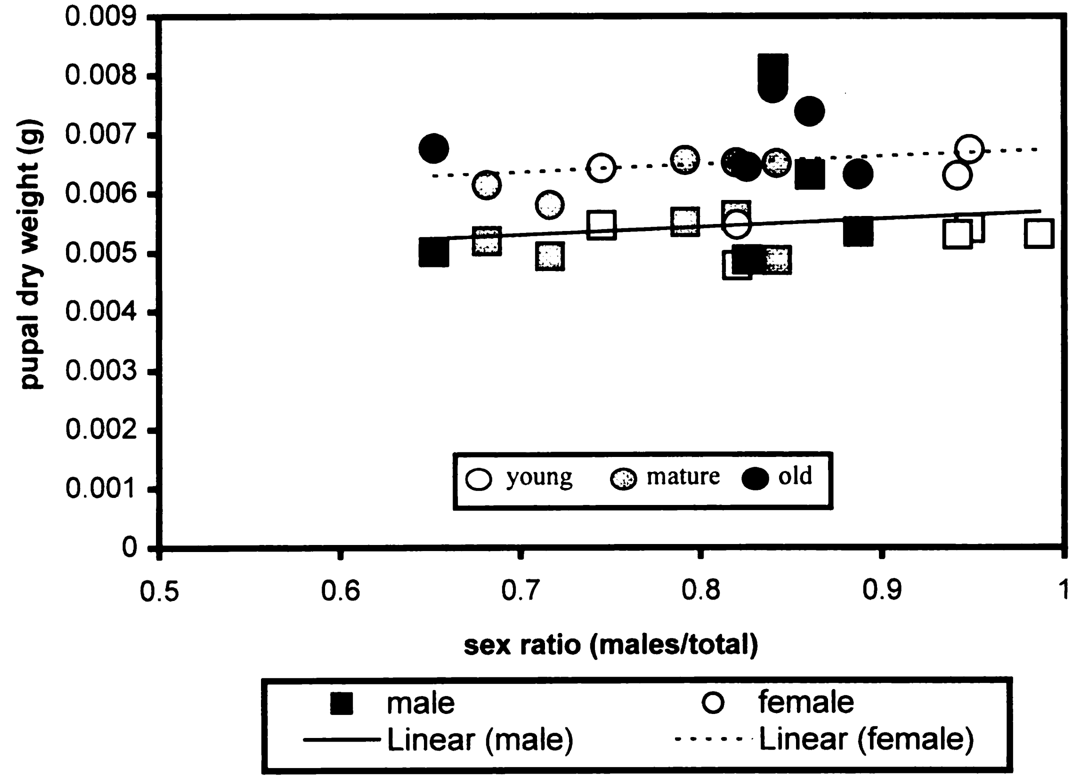

Figure 25. Pupal dry weights of sexual forms were not related to sex ratio. Regression slopes were not significantly different from zero for either sex. Each pair of male (❑) and female (❍) weights represents one stand.

Figure 26. Sex ratio of *F. exsectoides* populations in 15 jack pine stands.

Each donut represents one stand; each ring within a donut represents one mound. White represents allocation to females; grey represents allocation to males. Clear cut stands (Y3, Y4, and Y5) were significantly more male biased than burned (Y1, Y2), mature, and old stands, at p = 0.0245 (Table 25).

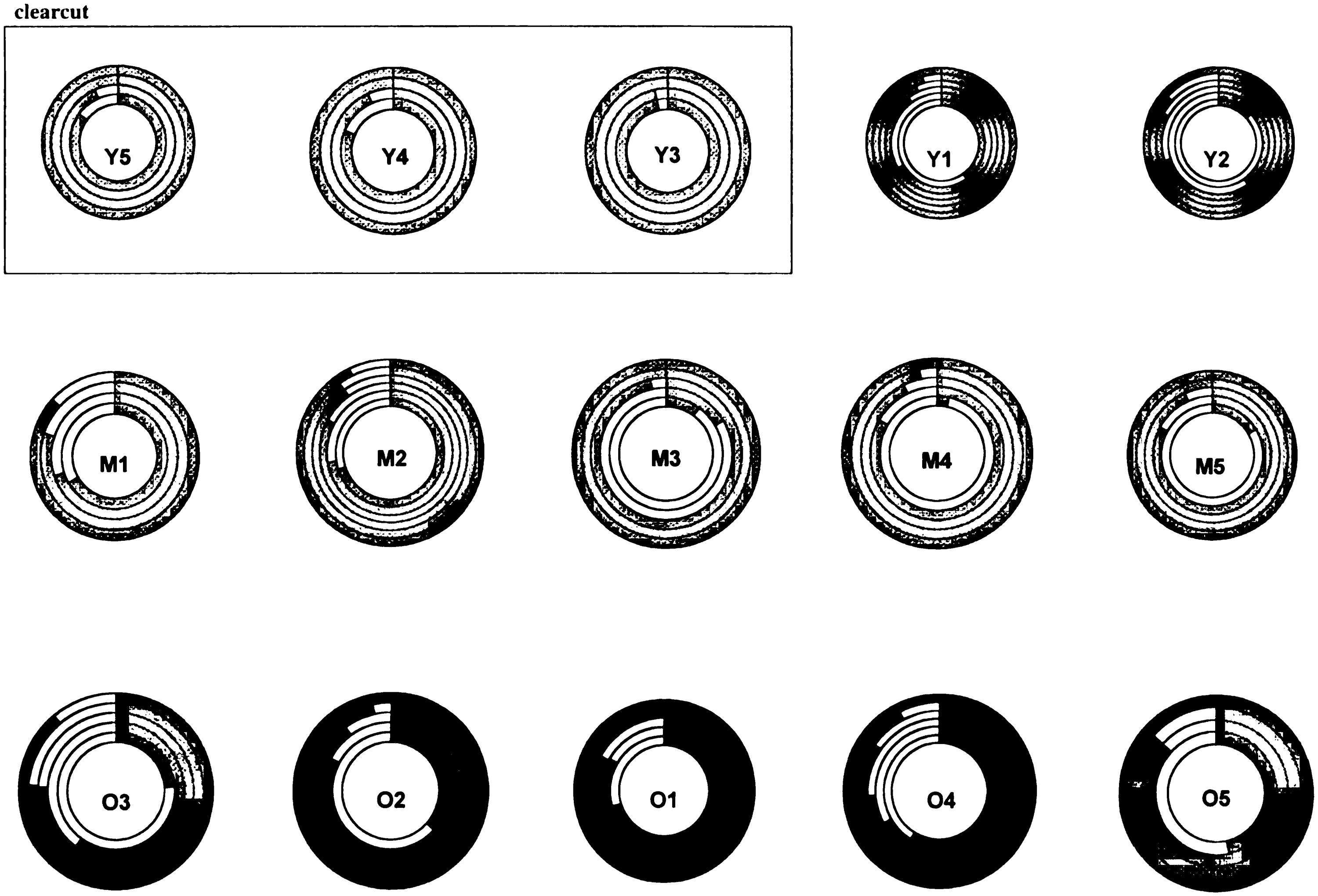

Figure 26. Sex ratio estimates for *F. exsectoides* populations in 15 jack pine stands.

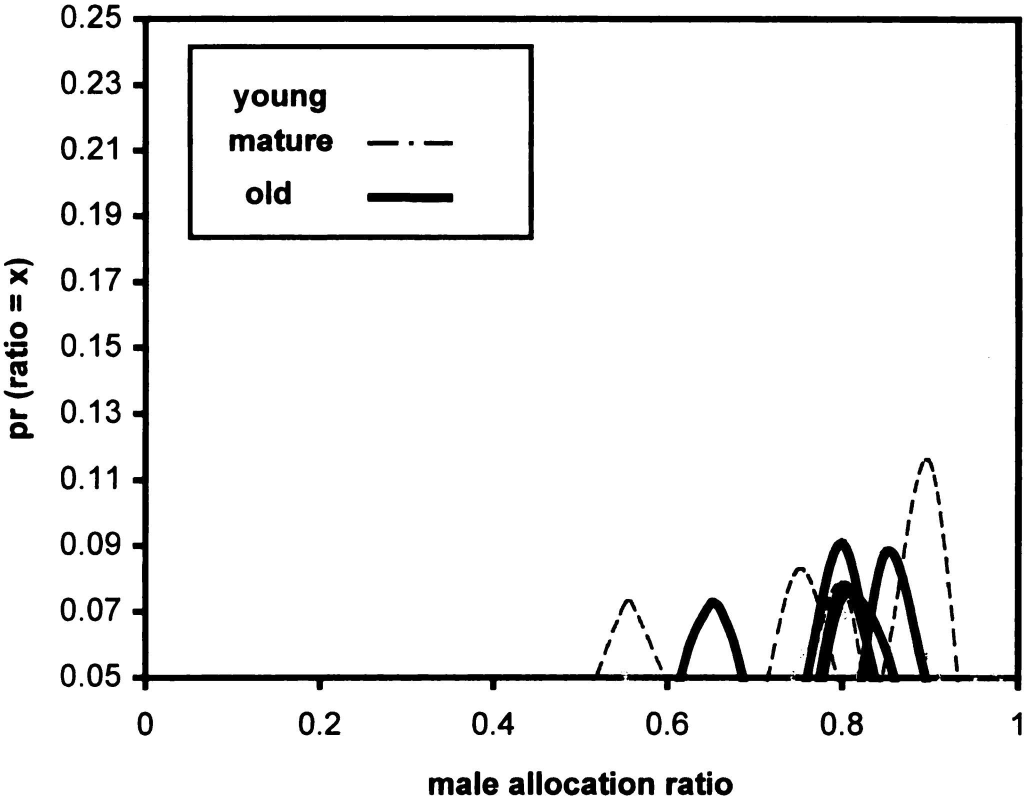

Figure 27. Confidence intervals for numerical sex ratio estimates, generated with binomial probability functions.

significantly different from 50-50 investment, and all other sites were male-biased, there were no significant differences in sex ratio among stand age classes (Figure 27). Sites Y3, Y4, and Y5 (three young clear-cut stands) were significantly more male-biased than any other stands (Table 25; Figure 26).

Logistic regression of raw counts of males and females on percent open sky for each mound yielded a cubic equation. Maximum likelihood tests indicated a probability of 0.0001 that the model produced an adequate description of the observed sex ratio data (Figure28). A similar analysis using mound surface area indices as the predictor variable yielded similar results. Analysis at the quadratic level yielded significant non-zero parameter estimates, but values generated by the equation did not fit the data ($p = 0.0001$, Figure29).

Sex ratio was not significantly related to mound density. Sex ratio was marginally related to abundance of *F. exsectoides* foragers (Figure 30a,b; Table 26), and also to relative abundance of *F. fusca* (Table 27, Figure31a). As the correlation with *F. fusca* appears to be based on two outlying data points, both clear-cut stands, and disappears when those stands are removed from the analysis (Figure31b). Sex ratio was not significantly related to stand age when stand age was determined by date since major disturbance (Table 32a, Figure 30a), but was related to stand ages determined by date since last cut (Table 28b, Figure 32b).

Table 25. ANOVA test of H_0: Sex ratio of young clear cut sites is equal to the sex ratio of all other sites.

Source of variation	df	mean square	F	p
stand	1	0.2914	6.08	0.0162
error	70	0.0480		
total	71			

Figure 28. Observed sex ratios were a poor fit to logistic regression models based on light availability.

The equation used to generate predicted values was:

sex ratio (males/total) = $1- (\exp(-9.5912 + 0.5570*sky + (-0.0116)*sky^2 + 0.000076*sky^3))$

Parameters were estimated using maximum likelihood functions (PROC CATMOD, SAS Systems, Inc., 1990), all parameters were significantly different from zero ($p = 0.0001$). The null hypothesis stating that this model adequately described the observed data was rejected at $p = 0.0001$. Linear, second-degree, and fourth-degree polynomial functions failed to provide parameters significantly different from zero or a better fit.

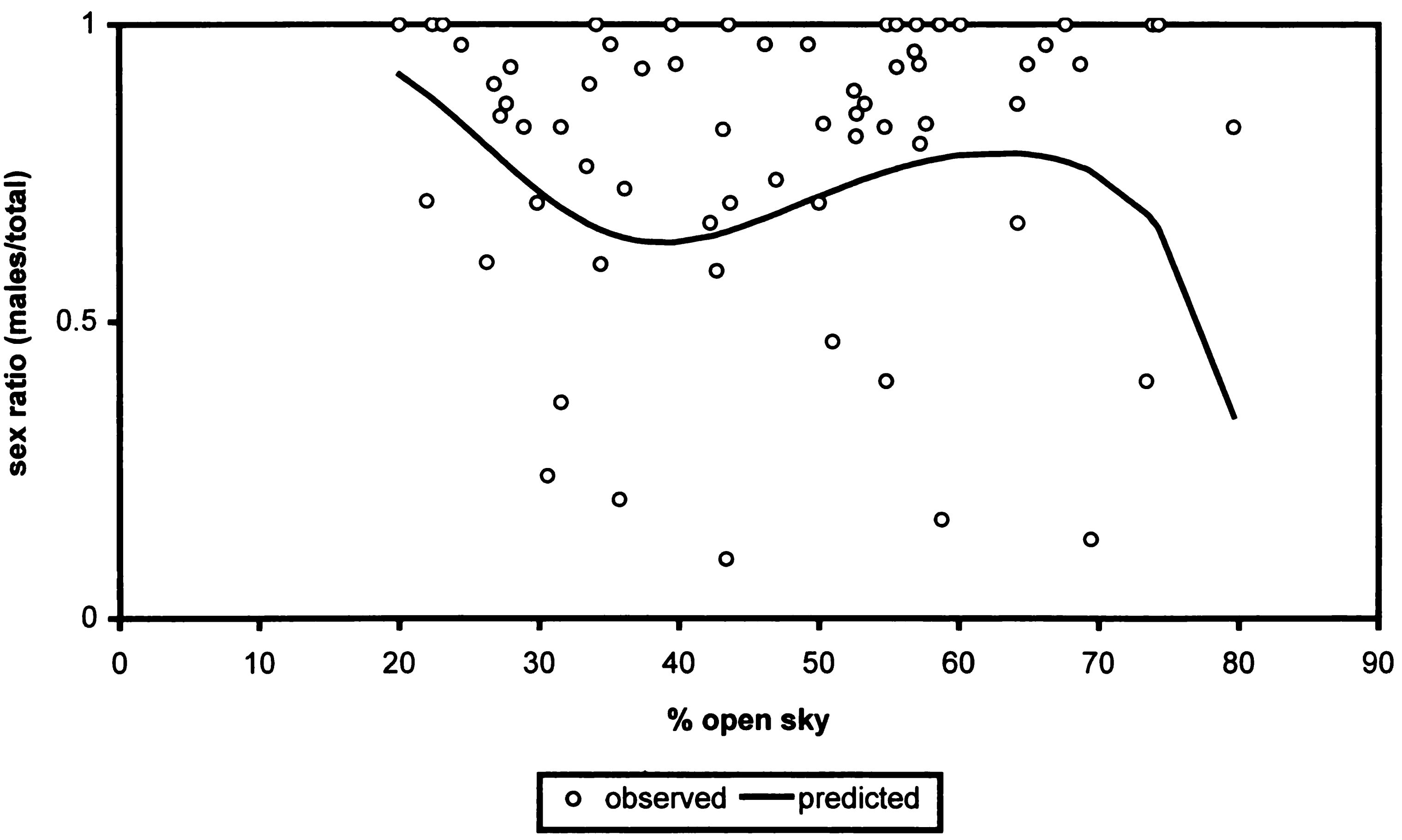

Figure 28. Observed sex ratios are a poor fit to logistic regression models based on light availability

Figure 29. Observed sex ratios were a poor fit to logistic regression models based on surface area indices.

Parameters were estimated using maximum likelihood functions (PROC CATMOD, SAS Systems, Inc., 1990), all parameters were significantly different from zero ($p = 0.0001$). The null hypothesis stating that this model adequately described the observed data was rejected at $p = 0.0001$. Linear and third-degree polynomial functions failed to provide parameters significantly different from zero or a better fit.

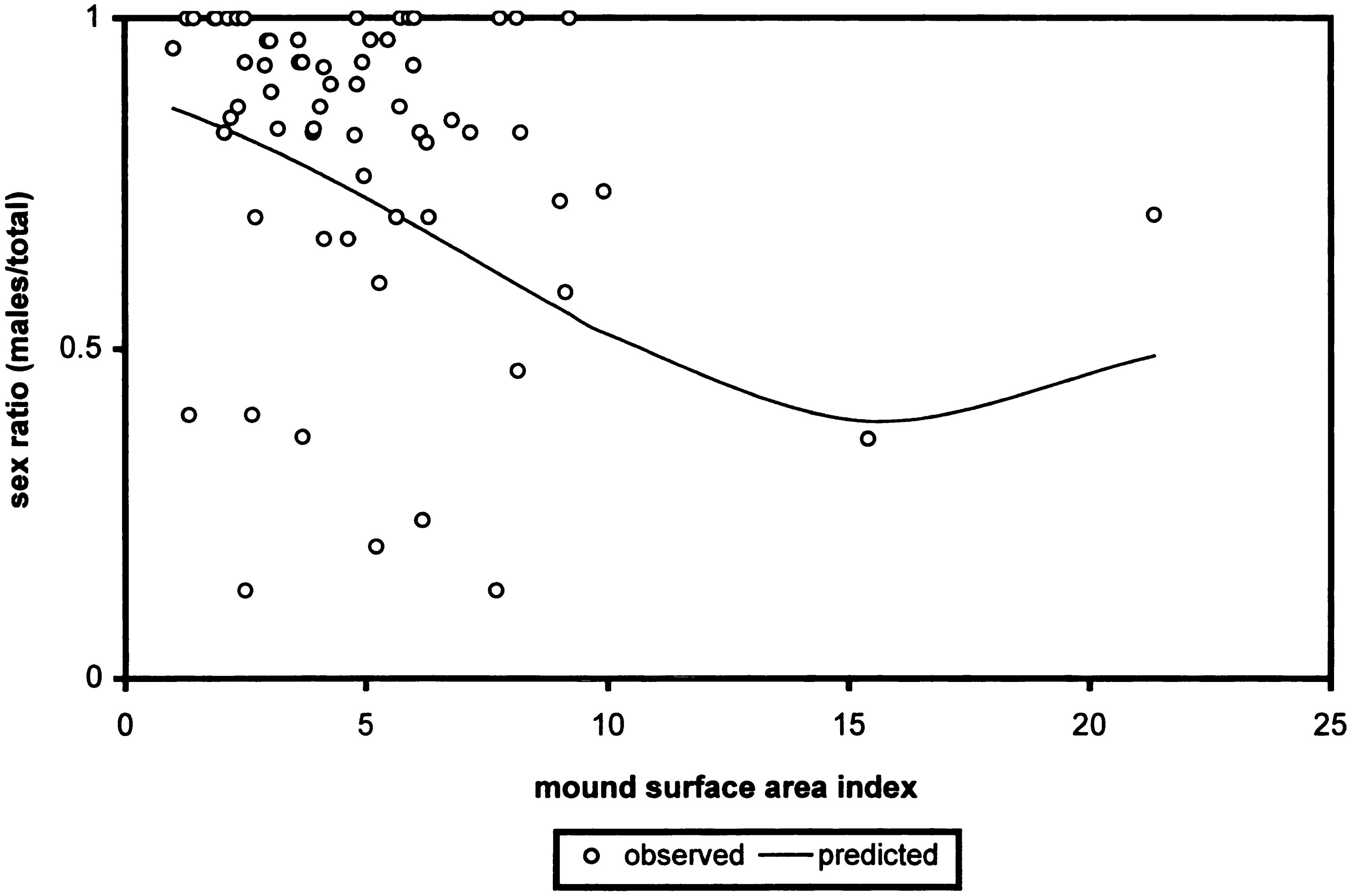

Figure 29. Observed sex ratios are a poor fit to logistic regression models based on surface area indices

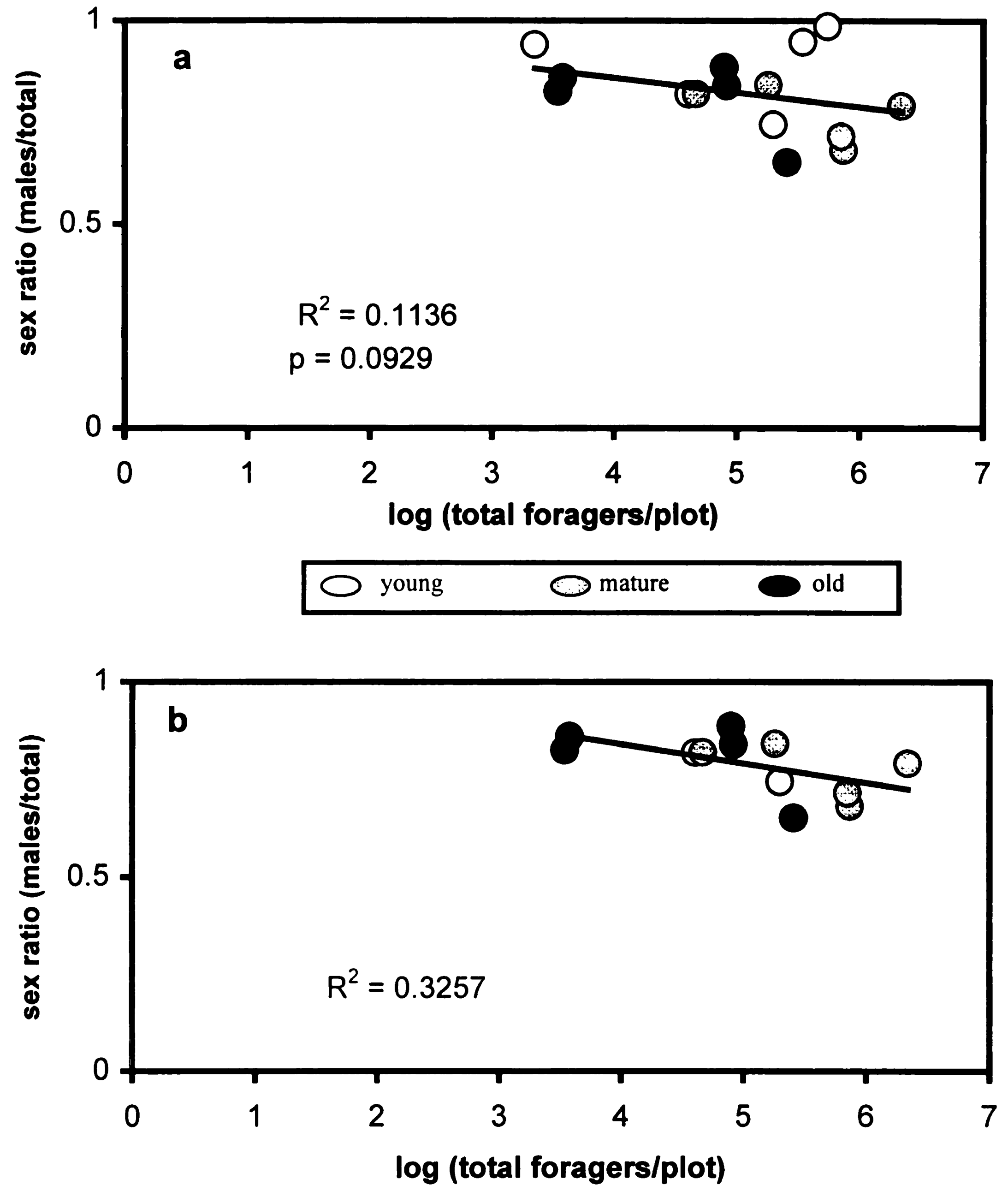

Figure 30. a) Sex ratio was not significantly related to abundance of *F. exsectoides* foragers captured in pitfall traps. b) This relarionship became significant when sex ratio outliers (young clear cut stands) are removed from the analysis.

Table 26. ANOVA test of significance for the regression model: sex ratio = log-transformed abundance of *F. exsectoides* foragers captured in pitfall traps.

Source of variation	df	mean square	F	p
forager abundance	1	0.0263	3.29	0.0929
error	13	0.0080		
total	14			

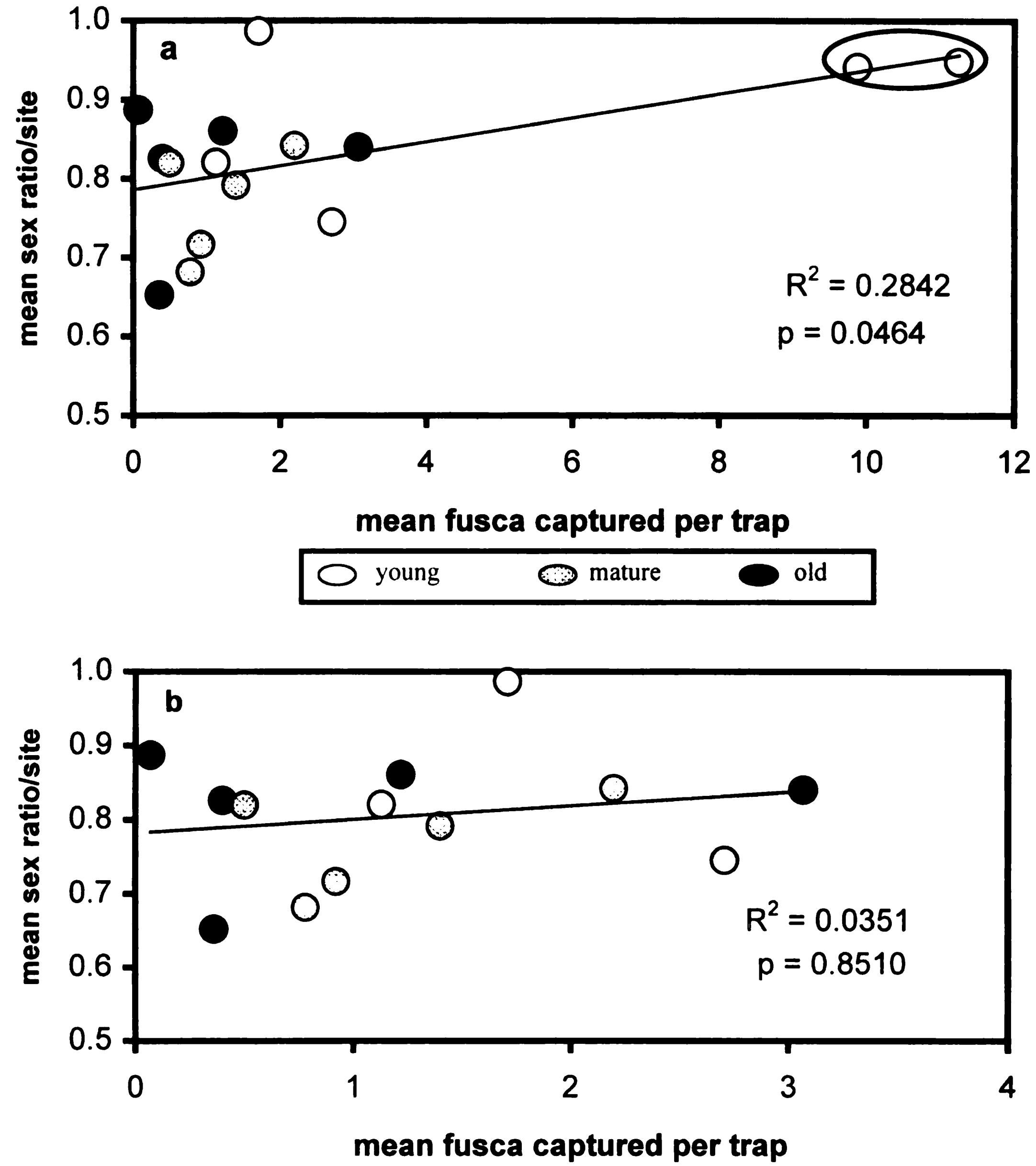

Figure 31. a) Sex ratio appeared to have a marginally significant negative relationship with relative abundance of *F. fusca*. b) This relationship disappeared when 2 young clear cut stands (outliers for both *fusca* abundance and sex ratio) are removed from the analysis. Values presented are mean *F. fusca* abundances for each stand, and total male allocation ratios for each stand. Significance tests for regression analyses are presented in Table 27a,b.

Table 27. ANOVA test of significance for the regression model: sex ratio = abundance of *F. fusca*. a) all sites b) excluding clear cut sites

a)

Source of variation	df	mean square	F	p
fusca abundance	1	0.0354	4.84	0.0464
error	13	0.0073		
total	14			

b)

Source of variation	df	mean square	F	p
fusca abundance	1	0.00023	0.04	0.851
error	10	0.00606		
total	11			

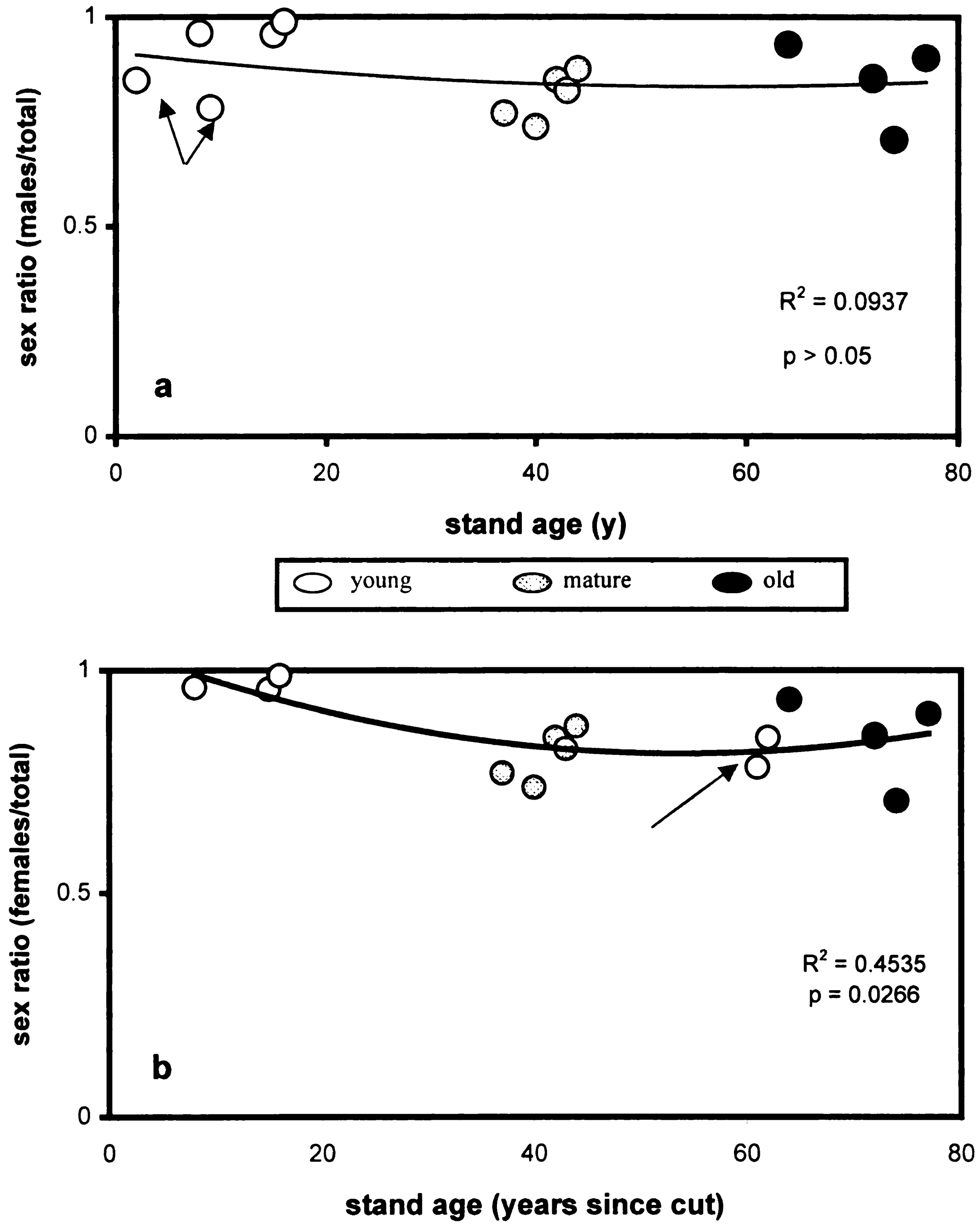

Figure 32. a) Sex ratio was not significantly related to stand age when "stand age" = years since major disturbance. b) Sex ratio was significantly related to stand age when "stand age" = years since clear cut. Curves in both a) and b) were fit using second-degree polynomial regression equations.

Table 28. a) ANOVA test of significance for the regression model: sex ratio = age + age^2, when "age" = years since last major disturbance of stand. b) ANOVA test of significance for the regression model: sex ratio = age + age^2, when "age" = years since last clear cut of stand.

Source of variation	df	mean square	F	p
model	2	0.0079	0.83	0.4593
age	1	0.00960	1.01	0.3355
age^2	1	0.00579	0.61	0.4509
error	12	0.00954		
total	14			

Source of variation	df	mean square	F	p
model	2	0.0295	4.98	0.0266
age (cut)	1	0.0435	7.34	0.0190
age $(cut)^2$	1	0.0312	5.26	0.0407
error	12	0.0059		
total	14			

Discussion

Environmental and mound variables. The biological concept of succession focuses on a progression of general changes that will take place as a community matures (Horn 1974). As forests grow and the canopy closes, light availability on the forest floor will decrease, causing composition changes in understory plant communities and related shifts in herbivore and predator communities and abundance. The responses of single species are more difficult to predict, especially given the amount of variability associated with any biological process, and the increasing likelihood of stochastic mischief with the passage of time. This is exemplified by the high variation in light environment within stands—older stands are especially vulnerable to mortality from insect attack, storm damage, or old age, so older stands possess greater variation in light environment (Figure 19).

While relative forager abundance was highest in mature stands, suggesting that mature stands represent a peak of *F. exsectoides* populations, this pattern was not significant. In fact, forager abundance was correlated significantly only to mound density (see Chapter 2). The stand variables in this study appear to have only an indirect effect on forager abundance, through their relationship with mound density (Table 18). Cory and Haviland (1938) failed to find a relationship between sunlight and ant activity, although this negative result may reflect a lack of quantitative data.

Mound status. The increase in the proportion of dead mounds in older stands is consistent with the observations of earlier authors that *F. exsectoides* populations occur in openings or forest edges, where sunlight is available (Haviland 1948). Andrews (1926) notes that mounds of *F. exsectoides* "arise and pass away in rhythms harmonious with phases of forestation", and offers two explanations for the apparent decline of *F. exsectoides* populations in mature forest. The first is that reduced light availability creates conditions unsuitable for brood production. The second is based on the argument that older trees will provide less food. Andrews speculates that young trees will sustain more aphids that will yield greater amounts of honeydew than old trees, and that ants will have further to travel to obtain the honeydew in old stands. While homopteran honeydew provides valuable carbohydrates to ant populations, it is not essential for ant survival (Bradley and Hinks 1968).

In a later study (Andrews 1929), Andrews examines the relationship of sunlight to mound temperatures. He concludes that mounds are heated by sunlight (rather than decay or ant activity), and that mound structure helps to retain heat. Andrews observed ants moving brood from areas of lower temperature to areas of higher temperature within the mound. These observations, combined with data from this study, lend support to Andrews' first explanation of *F. exsectoides* decline in old stands—prepupal stages of reproductive forms were found in shaded mounds in old stands (O1 and O4) as late as 22 July, when brood in mounds at stand edges or in young stands had already pupated. Additionally, old stands had higher pupal dry weights (Figure 24), which were explained by the inferior stage of pupal development in these samples, rather than greater investment per sexual (see Chapter 1: Weight and Developmental Stage). If lower light availability leads to delayed brood development, mounds in old stands may have insufficient time to complete their yearly cycle of brood production, and colonies would be expected to move to a more favorable location or become extinct.

The pattern displayed by mounds with respect to light availability (Figure 23) suggests that *F. exsectoides* prefers to construct mounds in areas with high insolation, and will expand the colony through budding while light conditions remain favorable. In studies of the related species *Formica ulkei* Emery, Scherba (1958) observed that new buds were formed primarily in non-shaded areas. In habitats with few suitable locations for budding, production of reproductive brood may be an adaptive response that will allow dispersal to younger habitats.

Elmes and Wardlaw (1982a,b), in population studies of *Myrmica sabuleti* and *Myrmica scabri*, stated that colonies producing sexual forms contain more workers than colonies that do not. They further cite the assertion of Brian and Brian (1951) that colonies of *Myrmica ruginodis* receiving high levels of insolation had higher worker numbers and larger worker size. Pamilo (1991) and Herbers and Stuart (1998) present the possibility that queen-worker conflict may occur over allocation to growth versus reproduction as well as sex ratio. If workers, having a shorter life expectancy, prefer greater allocation to reproduction, colonies with a larger workforce may be more likely to produce sexual forms.

Production of reproductive brood may also be a function of colony maturity. Old stands may contain higher proportions of mounds producing sexual forms because there are fewer young mounds in these stands. If budding represents an intermediate life history stage between ergonomic growth (worker production) and reproduction by sexual forms, the switch from mound fission to dispersal of sexual forms may represent age-specific reproductive response, rather than an adaptive response to the colony environment. This explanation seems less likely, since re-analysis of the data with burned stands aged by years from last cut (rather than years since fire event) yielded an inferior fit to the data (Figure 20a,b).

If colonies are polydomous and a fixed number of mounds in a population will produce sexual brood, the proportion of mounds producing reproductive forms might also be expected to increase in old stands, because there are fewer mounds. It is intriguing to observe that the relationship between the percentage of mounds producing sexual forms and the total number of mounds per plot is not consistent among stand ages (Figure 22). In both old and young stands, the proportion of mounds producing sexual brood declines only slightly with increasing mound density, while the relationship is more dramatic (and statistically significant) in mature stands. This implies that populations in young and old stands will increase output of reproductive forms as mound density increases, while a fixed number of mounds in mature stands will produce sexual brood without regard to increases in mound density.

Sex ratio. Sex ratio appeared more affected by factors intrinsic to the colony, such as age, size, and forager abundance, than by environmental influences. The extreme male bias of populations in clear cut stands may relate to resource shortages, although there is no indication that clear cut stands contain a lower abundance of invertebrate prey (Chapter 2). As previous populations are reduced or destroyed by cutting and replanting of stands, populations in clear-cut stands are more likely to represent recent colonization by *F. exsectoides*. Young colonies may specialize in the production of males.

The lack of a detectable influence of light environment on sex ratio may be due to a lack of response range—if there is a range of light environments within a given stand in which *F. exsectoides* mounds produce sexual forms (versus producing workers or moving out), this range might not be wide enough to allow a gradient of response in sex ratio. Or it may be that light environment is unimportant. Experimental manipulation of light environments would be useful, but logistical difficulties intervene.

Elmes and Wardlaw (1982a,b) draw the conclusion that, while production of males is dependent on the number of workers in the colony, production of females is dependent on the level of nest insolation. While the premise that separate factors may regulate allocation to each sex is intriguing, the fact that the authors have also linked colony size to level of insolation casts doubt on the independence of these factors. Elmes and Wardlaw were able to collect entire colonies to account for total production of each sex. Similar data for *F. exsectoides* colonies might suggest the presence (or absence) of such patterns, but collection of even a single colony of *F. exsectoides* in its entirety would require more resources than were available for this study.

It does appear, when age of population is taken into account (Figure 32b), that *F. exsectoides* increase allocation to females in mature stands. Mature stands present an environment where sexual production is less linked to mound density, indicating that production of sexual forms is not as strongly favored as in old stands. Since mature stands also contain the greatest abundance of foragers, more resources may be available per sexual form produced, encouraging increased investment in females. This explanation would be supported by higher pupal weights in mature stands, but this was not observed (Figure 24). Mature stands could represent an earlier phase of dispersal from the changing habitat (when the lifeboats are being provisioned and filled in an orderly fashion), while the increase in male allocation in old stands might represent a mass exodus (when the passengers are jumping onto anything that might float). Tests of this possibility would consider changes in overall output of sexual forms as a function of sex ratio.

Changes in sex ratio related to population (colony) age may reflect an adaptive life history progression. Oster and Wilson (1978) define the main stages of ant life history as foundation, ergonomic growth (production of only worker-caste brood), reproduction, and orphanage. Young colonies leaving the ergonomic stage may begin the reproductive stage by producing male-biased sex ratios, since males are usually the "cheaper" sex and tend to be overproduced by small colonies (Crozier and Pamilo 1996). Mature colonies may increase production of females at the "peak" of the reproductive stage, but may shift back toward male production as the colony senesces or habitat conditions become less favorable.

It is impossible to separate direct and indirect, genetic and environmental sources of the male bias found in *F. exsectoides* populations without detailed knowledge of population genetic structure. Given current knowledge of *F. exsectoides* biology, it is, however, possible to discuss some of the more likely explanations, and suggest direction (and caveat!) to future research.

Under the genetic relatedness hypothesis, increased male bias is expected under conditions of both polygyny and polyandry (both conditions which will decrease relatedness between female siblings) (Nonacs 1986). In inbred populations, however, multiple-mating and polygyny will be unlikely to decrease relatedness between sisters to the same extent as multiple mating to unrelated males or polygyny with unrelated queens. Further analysis of *F. exsectoides* population structure using hypervariable genetic markers is necessary, as allozyme studies indicate a lack of variation at many loci (Nipson 1971, Appendix B). This lack of variation may be indicative of high levels of inbreeding and high intercolony relatedness.

Local Mate Competition is popularly invoked as an explanation for female-biased sex ratios (Nonacs 1986, Crozier and Pamilo 1996), but it may as easily be invoked to explain male bias in *F. exsectoides*. If multiple mating is the norm for *F. exsectoides* queens, and males are the more dispersive sex (as is indicated by observed mating behavior, Bishop 1998), it is reasonable to expect a male bias in sex ratio. Each female produced will require several males to mate, and males will be more vulnerable to predation and random mortality factors during their dispersal. Given the lack of apparent advantage, for either workers or queens, to producing millions of males of which only a small fraction will mate successfully, female competition for sperm seems quite likely to be an important factor in this system.

While polygyny, polyandry, polydomy, poor survival in lab colonies, and unusual modes of colony founding and expansion may sound like a recipe for disaster in studies of reproductive allocation, these very traits and the apparent success of *F. exsectoides* as a eusocial species makes it important to understand how that success is shaped by reproductive strategies. Life history strategies of ants are, to date, poorly understood. Attempts to examine life history variation within species have rarely included age-specific variation within stages, although *F. exsectoides* variation in sex ratio within the reproductive stage indicates that this variation exists and must be taken into account in sex ratio studies. While sex ratio was not shown to respond to influences external to the colony, differential investment in sexual reproduction in response to environmental factors supports the possibility that such interactions may occur, and merit investigation in other species.

APPENDICES

APPENDIX A

APPENDIX A

Arthropods collected in jack pine (*Pinus banksiana* Lamb.) by pitfall trapping, July 7—August 4, 1997.

CLASS INSECTA

- Collembola

- Orthoptera:
  - Acrididae (Cyrtacanthacridinae, Oedipodinae)
  - Tettigoniidae
  - Gryllacrididae (Rhaphidophorinae)
  - Gryllidae

- Blattaria

- Hemiptera:
  - Lygaeidae
  - Pentatomidae
  - Thyreocoridae (*Galgupha* spp.)
  - Miridae
  - Tingidae

- Homoptera:
  - Aphididae
  - Membracidae
  - Cercopidae
  - Cicadellidae

- Neuroptera:
  - Myrmeleontidae

- Coleoptera:
  - Carabidae
    - *Notiophilus aenus* (Hbst.)
    - *Calosoma frigidum* Kirby
    - *Myas cyanescens* Dej.
    - *Pterostichus pennsylvanicus* LeC.
    - *Calathus gregarius* (Say)
    - *Synuchus impunctatus* (Say)
    - *Syntomus americanus* (Dej.)
  - Staphylinidae
  - Histeridae
  - Elateridae
  - Nitidulidae
  - Tenebrionidae
  - Mordellidae

- Bruchidae
- Curculionidae
- Scarabaeidae

- Diptera:
  - Mycetophilidae
  - Cecidomyiidae
  - Culicidae
  - Simuliidae
  - Therevidae
  - Dolichopodidae
  - Bombyliidae
  - Syrphidae
  - Pipunculidae
  - Sepsidae
  - Lauxaniidae
  - Muscoidea

- Lepidoptera

- Hymenoptera:
  - Braconidae
  - Ichneumonidae
  - Chrysididae
  - Apidae
  - Mutillidae
  - Pompilidae
  - Formicidae
    - *Formica exsectoides* Forel
    - *Formica fusca* Linn.
    - *Aphaenogaster* spp.
    - *Dolichoderus plagiatus*
    - *Camponotus herculeanis*
    - *Tapinoma sessile*
    - *Monomorium minimum*
    - *Lasius* spp.

ARACHNIDA

- Araneae
- Acari
- Opiliones

DIPLOPODA

CHILOPODA

ISOPODA

APPENDIX B

APPENDIX B

Attempted search for diploid males (or Things That Didn't Work)

Introduction

Inbreeding in ant populations is known to lead to the production of diploid males—although diploid eggs are generally female, homozygosity at sex determining loci can cause the egg to develop into an infertile male (Crozier 1971, Pamilo *et. al.* 1994). Founder events and habitat isolation increase the likelihood that queens and males will possess sufficient genetic similarity to cause a significant number of diploid males to be produced by the colony (Kukuk and May 1990, Ross and Fletcher 1986). Analysis of male weights in *F. exsectoides* showed a tendency toward dimorphism (possibly indicative of male diploidy) (Chapter 1). Fortelius *et al.* (1987), in a study of *Formica exsecta*, described a strong bimodal dimorphism of males, which they believed represented differing reproductive strategies: small males tended to disperse farther while large males mated in proximity of the nest. Since my data did not show such a strong bimodality, and the presence of a significant production of diploid males may distort the observed sex ratio, I felt that it was important to investigate this possibility. Diploid males are produced accidentally from eggs intended to be females; thus colonies producing a large percentage of diploid males could appear to exhibit a male bias when, functionally and intentionally, the bias may actually be toward females.

Methods

Diploid males can be detected with several techniques, including staining and counting of chromosomes, allozyme electrophoresis, and measuring total weight of DNA. I have chosen to use the former two options.

A. Chromosome staining

Brain tissue was dissected from reproductive caste pupae of *F. exsectoides*. Staining protocol followed Imai *et. al.* (1977). Tissues were placed in a hypotonic solution (1% sodium citrate) with 0.005% colchicine (measured in weight/volume) for 10-20 minutes, fixed with absolute ethyl alcohol and glacial acetic acid in the mixtures described by Imai et. al., and stained using a 10% solution of commercially prepared Giemsa stain (Sigma-Aldrich, St. Louis, MO).

B. Allozyme electrophoresis

Detection of diploid males through allozyme electrophoresis is dependent upon the presence of substantial levels of heterozygosity within populations. As homozygous diploid males will be indistinguishable from haploids, the extent of male diploidy among homozygotes must be estimated by multiplying the proportion of diploid individuals discovered among heterozygotes by the proportion of individuals homozygous at the loci under consideration (Owen and Packer 1994).

All allozyme assays were performed using cellulose-acetate electrophoresis, with agar overlay stains as outlined in Richardson *et. al.* (1986). Initial screenings for enzyme polymorphism were performed on fresh *F. exsectoides* workers from a queenless colony maintained in the Michigan State University Bug House. Abdomens were removed from workers to prevent disruption of buffer pH by formic acid (Nipson 1978). Approximately 10 workers were homogenized together in each lane of the cellulose acetate plate, allowing the screening of large numbers of workers for enzyme variation with lower investment in labor and materials. Approximately 30 workers were screened for variation at each of the following loci: PGM, PGD, PGK, MDH, ME, MPI, IDH, G6PD, LDH, ACO, ADH, GPI, GLDH, and HK. As cellulose acetate electrophoresis requires a very small amount of sample per plate, a single worker homogenate provided enough material for the assay of 5-8 enzymes. Plates were not multiply stained, but did have multiple origins in some cases.

Although homogenizing 10 individuals into a single lane removes the possibility of distinguishing between heterozygosity and within-species polymorphism, the only interest in this case was to locate loci with potentially informative variation. Enzyme loci showing uniform banding patterns across all workers screened were not considered for further investigation.

Using stains for the remaining enzymes, I then examined sexual caste pupae that had been collected from mounds outside of my study plots at sites Y3, Y2 and O3 (see Chapter 3: Methods). These pupae had been held in a 10°C incubator for 10 days, then frozen at -80°C. Two females and 3 males from each site were electrophoresed and stained for the following enzymes: ACO, GPI, HK, IDH, MDH, ME, PGM, and PGD.

Results

A. Chromosome staining

Although chromosomes could be clearly observed in some cells, no observed cells were in appropriate mitotic phases to determine chromosome number.

B. Allozyme electrophoresis

PGK, G6PD, and GLDH showed uniform single bands in all workers screened. MPI, ADH, and LDH presented technical difficulties in staining that would have been too time-consuming to resolve for this study.

Banding patterns produced for ACO, GPI, HK, IDH, MDH, ME, PGM, and PGD are presented in Table 29. Although electrophoresis of workers indicated some species-level variation at each of the loci tested, analysis of sexual forms from the three populations sampled revealed no within-population variation in banding patterns (except occasional distance variation caused by inadequately controlled running time).

Table 29. Allozyme banding patterns for *F. exsectoides*, values represent cm each band migrated from the origin. The column "workers" represents a summation of all bands in all samples (each sample contained multiple ants, see text). Columns Y1, Y4, and O4 contain consensus banding patterns from male and female sexual forms sampled in stands Y1, Y4, and O4—no within-stand variation in banding pattern occurred at any locus.

Enzyme	banding pattern (cm from origin)			
	workers	Y1	Y4	O4
ACO	0, 1.8	0, 1.8	0.3, 1.8	0, 1.8
GPI	0, 4	0, 3.5	0, 3.5	0, 4
HK	1.5, 2.1, 2.5	1.3, 2	1.3, 2	1.8
IDH	1.1, 1.3, 1.7	1	0.5-0.9	0.4-0.8
MDH	(-0.2), 2.5	0-0.5, 2.5-3.3	0-0.5, 2.5-3.4	0-0.5, 2.5-3.5
ME	2.5, 3.0, 3.5	2.2	2.2	2.2
PGM	0.9, 1.5	1.5	1.5	2.5
PGD	2, 3.1	2	2	2

Discussion

Although diploid males may be present in *F. exsectoides* populations, this study did not provide sufficient data to justify any conclusion on the matter. The lack of appropriate mitotic phases for karyotype determination is doubtlessly due only to the inappropriateness of the late pupal stage of development for this analysis (a lack of cell division), and could be remedied by substitution of a different life stage. As sex cannot be determined by morphology at the egg or larval stages, identification of diploid males by chromosome staining will require adults.

The uniformity of banding pattern within populations is more interesting, and may be consistent with inbreeding as suggested by Nipson (1978).

Four enzymes, ACO, GPI, HK, and PGM, showed population-level variation in banding pattern. In two of these four cases (GPI and PGM), O4 is the differing stand. O4 is more geographically distant from Y1 and Y4 than Y1 and Y4 are from each other, but a sample of three populations is not adequate for speculation about dispersal distances.

While a highly inbred population would seem to be a prime target for the investigation of the frequency of male diploidy and its effects on sex ratio, this same level of inbreeding leads to a reduction in genetic variation, making the identification of diploid males a more trying task. While the late pupal stage is more convenient for collection and determination of sex by morphology, it is inappropriate for karyotype analysis. Further attempts to investigate male diploidy in *F. exsectoides* must rely on hypervariable genetic markers, which can provide a wealth of information on the genetic structure of inbred populations and greater resolution than allozyme techniques.

APPENDIX C

APPENDIX C

Record of Deposition of Voucher Specimens*

The specimens listed on the following sheet(s) have been deposited in the named museum(s) as samples of those species or other taxa which were used in this research. Voucher recognition labels bearing the Voucher No. have been attached or included in fluid-preserved specimens.

Voucher No.: 1998-09

Title of thesis or dissertation (or other research projects):

Effects of habitat succession on population and reproduction of the Allegheny mound ant

Museum(s) where deposited and abbreviations for table on following sheets:

Entomology Museum, Michigan State University (MSU)

Other Museums:

Investigator's Name(s) (typed)
Heather C. Rowe

Date 12/09/1998

* Reference: Yoshimoto, C. M. 1978. Voucher Specimens for Entomology in North America. Bull. Entomol. Soc. Amer. 24:141-42.

Deposit as follows:

Original:	Include as Appendix C in ribbon copy of thesis or dissertation.
Copies:	Include as Appendix C in copies of thesis or dissertation. Museum(s) files. Research project files.

This form is available from and the Voucher No. is assigned by the Curator, Michigan State University Entomology Museum.

Voucher Specimen Data

Page 1 of 1 Pages

Species or other taxon	Label data for specimens collected or used and deposited	Number of:							
		Eggs	Larvae	Nymphs	Pupae	Adults ♀	Adults ♂	Other	Museum where deposited
Formica exsectoides Forel	Michigan: Oscoda County T25N, r1E, s.29 9 July 1997 H. Rowe				10	5	5	10 worker	MSU

(Use additional sheets if necessary)

Investigator's Name(s) (typed)

Heather Rowe

Date 12/09/98

Voucher No. 1998-09

Received the above listed specimens for deposit in the Michigan State University Entomology Museum.

Curator

9 Dec 1998

Date

REFERENCES CITED

REFERENCES CITED

Agosti, D.; Hauschteck-Jungen, E. 1987. Polymorphism of males in Formica exsecta Nyl. (Hymenoptera: Formicidae). **Insectes Sociaux**. 34(4): 280-290.

Allen, Douglas C.; Knight, Fred B.; Foltz, John L. 1970. Invertebrate predators of the Jack-Pine Budworm, *Choristoneura pinus*, in Michigan. **Annals of the Entomological Society of America**. 63(1): 59-64.

Andrews, E. A. 1926. Sequential distribution of *Formica exsectoides* Forel. **Psyche**. 33: 127-150.

Andrews, E. A. 1927. Ant mounds as to temperature and sunshine. **Journal of morphology and physiology**. 44(1): 1-20.

Andrews, E. A. 1929. The mound-building ant, *Formica exsectoides* F., associated with tree-hoppers. **Annals Entomological Society of America**. 22: 369-391.

Arnett, Ross H. 1993. **American insects: a handbook of the insects of America north of Mexico.** The Sandhill Crane Press, Inc.: Gainesville, FL.

Ayre, G. L. 1963. Laboratory studies of the feeding habits of seven species of ants (Hymenoptera: Formicidae) in Ontario. **The Canadian Entomologist**. 95: 712-715.

Beaudry, S.; Duchesne, L. C.; Cote, B. 1997. Short-term effects of three forestry practices on carabid assemblages in a jack pine forest. **Canadian Journal of Forest Research**. 27(12): 2065-2071.

Beland, Martin; Bergeron, Yves. 1993. Ecological factors affecting abundance of advanced growth in jack pine (*Pinus banksiana* Lamb.) stands of the boreal forest of northwestern Quebec. **The Forestry Chronicle**. 69 (5): 561-568.

Bishop, D. B. 1998. **Differential effects of the Allegheny mound ant, *Formica exsectoides*, Forel, on aphid, scale, and predator populations and their interactions in jack pine forests**. Ph.D. dissertation, Michigan State University.

Boomsma, J.J. 1989. Sex investment ratios in ants: has female bias been systematically overestimated? **American Naturalist**. 133: 517-532.

Boomsma, J.J. 1993. Sex ratio variation in polygynous ants. **Queen number and sociality in insects**. (ed. L. Keller) Oxford University Press: New York.

Boomsma, J.J.; Grafen, A. 1991. Colony-level sex ratio selection in the eusocial Hymenoptera. **Journal of Evolutionary Biology**. 3: 383-407.

Boomsma, J.J.; Isaaks, J. A. 1985. Energy investment and respiration in queens and males of *Lasius niger* (Hymenoptera: Formicidae). **Behavioral Ecology and Sociobiology**. 18: 19-27.

Boomsma, J.J.; Keller, L.; Nielsen, M. G. 1995. A comparative analysis of sex ratio investment parameters in ants. **Functional Ecology**. 9: 743-753.

Borror, Donald J.; Triplehorn, Charles; Johnson, Norman F. 1989. **An introduction to the study of insects** (6th edition). Saunders College Publishing: Forth Worth, TX.

Bourke, A. F. G.; Franks, N. 1995. **Social Evolution in Ants**. Princeton University Press: Princeton, NJ.

Bradley, G.A.; Hinks, J.D. 1968. Ants, aphids, and jack pine in Manitoba. **The Canadian Entomologist.** 100: 40-50.

Brian, M. V.; Brian, A. D. 1951. Insolation and ant population in the west of Scotland. **Transactions of the Royal Entomological Society of London**. 102: 303-330.

Bristow, C. M.; Cappaert, D.; Campbell, N. J.; Heise, A. 1992. Nest structure and colony cycle of the Allegheny mound ant, *Formica exsectoides* Forel (Hymenoptera: Formicidae). **Insectes Socieaux**. 39: 385-402.

Bull, J.J. 1983. **Evolution of sex determining mechanisms**. Benjamin Cummings, Menlo Park, CA.

Campbell, N. J. 1991. **An evaluation of *Formica exsectoides* Forel as a potential biological control agent of insect pests in pines**. Ph.D. dissertation, Michigan State University.

Campbell, N. J.; Bristow, C. M.; Ayers, G.; Simmons, G. 1991. Design and field test of portable colonies of the predaceous ant, *Formica exsectoides* (Hymenoptera: Formicidae). **Journal of the Kansas Entomological Society**. 64: 116-120.

Canham, Charles D. 1995. **GLI/C: Software for calculation of light transmission using color fisheye photography**. Institute for Ecosystem Studies: Millbrook, NY.

Cannings, C.; Cruz Orive, Luis, M. 1975. On the adjustment of the sex ratio and the gregarious behavior of animal populations. **Journal of Theoretical Biology**. 55: 115-136.

Charnov, Eric L. 1993. **Life history invariants: some explorations of symmetry in evolutionary ecology.** Oxford University Press, New York.

Charnov, Eric L. 1982. **The theory of sex allocation**. Princeton University Press, Princeton, NJ.

Clark, A. B. 1978. Sex ratio and local resource competition in a prosimian primate. **Science**. 201: 163-165.

Clark, P. J.; Evans, F. C. 1954. Distance to nearest neighbor as a measure of spatial relationships in populations. **Ecology**. 60: 316-317.

Cody, Martin L. 1966. A general theory of clutch size. **Evolution**. 20:174-184.

Cory, Ernest N.; Haviland, Elizabeth E. 1938. Population studies of *Formica exsectoides* Forel. **Annals Entomological Society of America**. 31: 50-56.

Creighton, W. S. 1950. The ants of North America. **Bulletin of the Museum of Comparative Zoology, Harvard University**. 104: 1-585.

Crow, J. F. 1994. Advantages of sexual reproduction. **Developmental Genetics**. 15(3): 205-213.

Crozier, R. H. 1971. Heterozygosity and sex determination in haplo-diploidy. **American Naturalist**. 105(945): 399-412.

Crozier, R. H.; Pamilo, P. 1996. **Evolution of Social Insect Colonies: Sex Allocation and Kin Selection.** Oxford University Press: New York.

Deslippe, Richard J.; Savolainen, Riita. 1995. Sex investment in a social insect: the proximate role of food. **Ecology**. 76(2): 375-382.

Dimmick, John F. 1951. An ecological study of *Formica Formica exsectoides*. **Illinois Academy of Science Transactions**. 44: 197-204.

Duchesne, S.; Sirois, L. 1995. The first stage of post-fire regeneration in subarctic coniferous populations. **Canadian Journal of Forest Research**. 25(2): 307-318.

Elmes, G.W.; Wardlaw, J.C. 1982. A population study of the ants *Myrmica sabuleti* and *Myrmica scabrinodis*, living at two sites in the south of England. I. A comparison of colony populations. **Journal of Animal Ecology**. 51: 651-664.

Fisher, R. A. 1930. **The genetical theory of natural selection**. Oxford University Press: Oxford, England.

Fortelius, W.; Pamilo, P.; Rosengren, R.; Sundström, L. 1987. Male size dimorphism and alternative reproductive tactics in *Formica exsecta* ants (Hymenoptera: Formicidae). **Ann. Zool. Fennici**. 24: 45-54.

Godfray, H.C.J.; Werren, J.H. 1996. Recent developments in sex ratio studies. **Trends in Ecology and Evolution.** 11(2): 59-63.

Halaj, J.; Ross, D. W.; Moldenke, A. R. 1997. Negative effects of ant foraging on spiders in Douglas-fir canopies. **Oecologia**. 109(2): 313-322.

Hamilton, W. D. 1963. The evolution of altruistic behavior. **American Naturalist**. 97: 354-356.

Hamilton, W.D. 1964. The genetical evolution of social behaviour. **Journal of Theoretical Biology.** 7: 1-32.

Hamilton, W.D. 1967. Extraordinary sex ratios. **Science**. 156: 477-488.

Haviland, E. E. 1947. The biology and control of the Allegheny mound ant. **Journal of Economic Entomology**. 40: 413-419.

Haviland, E.E. 1948. Mound changes after ten years in colonies of *Formica exsectoides* Forel. **Annals of the Entomological Society of America**. 41: 438.

Heatwole, Harold; Levins, Richard. 1972. Trophic structure stability and faunal change during recolonization. **Ecology**. 53(3): 531-534.

Hendrix, S.D.; Brown, V.K.; Dingle, H. 1988. Arthropod guild structure during early old field succession in a new and old world site. **Journal of Animal Ecology**. 57: 1053-1065.

Herbers, Joan M. 1986. Effects of ecological parameters on queen number in *Leptothorax longispinosis* (Hymenoptera: Formicidae). **Journal of the Kansas Entomological Society**. 59(4): 675-686.

Herbers, Joan M. 1993. Ecological determinants of queen number in ants. in **Queen Number and Sociality in Insects** (ed. L. Keller) Oxford University press: New York, p.262-293.

Herbers, Joan M.; Banschbach, Valerie. 1998. Food supply and reproductive allocation in forest ants: repeated experiments give different results. **Oikos**. 83: 145-151.

Herbers, Joan M.; Stuart, Robin J. 1998. Patterns of reproduction in slave-making ants. **Proceedings of the Royal Society of London, Series B**. 265: 875-887.

Hollbobler, Burt; Wilson, E. O. 1990. **The Ants**. Harvard University Press: Cambridge, MA.

Horn, H.S. 1974. The ecology of secondary succession. **Annual Review of Ecology and Systematics.** 5: 25-37.

Hurst, L. D.; Peck, J. R. 1996. Recent advances in understanding the evolution and maintenance of sex. **Trends in Ecology and Evolution**. 11(2): 46-52.

Imai, H. T.; Crozier, R. H.; Taylor, R. W. 1977. Karyotype evolution in Australian ants. **Chromosoma**. 59: 341-393.

Juliano, Steven A. 1993. Nonlinear curve fitting: predation and functional response curves. pp. 159-182 in **Design and analysis of ecological experiments**. (ed. Samuel M. Scheiner and Jessica Gurevitch) Chapman & Hall, New York.

Keller, L. 1991. Queen number, mode of colony founding, and queen reproductive success in ants (Hymenoptera: Formicidae). **Ethology, Ecology, and Evolution**. 3: 307-316.

Kondrashov, A. S. 1993. Classification of hypotheses on the advantage of amphimixis. **Journal of Heredity**. 84(5): 372-387.

Kukuk, P.; May, B. 1990. Diploid males in a primitively eusocial bee, *Lasioglossum* (*Dialictus*) *zephyrum* (Hymenoptera: Halictidae). **Evolution**. 44: 1522-1528.

Lande, R. 1982. A quantitative genetic theory of life history evolution. **Ecology**. 63: 607-615.

Levins, S.C.; Traniello, J.F.A. 1981. Territoriality, nest dispersion, and community structure in ants. **Psyche**. 88: 265-319.

MacKay, W. P. 1985. A comparison of the energy budgets of three species of *Pogonomyrmex* harvester ants (Hymenoptera: Formicidae). **Oecologia**. 66: 484-494.

Maynard Smith, J. 1978. **The Evolution of Sex**. Cambridge University Press: Cambridge, England.

McCook, H. C. 1877. Mound-making ants of the Alleghenies, their architecture and habits. **Transactions of the Entomological Society of America**. 6: 253-296.

Moran, V.C.; Southwood, T.R.E. 1982. The guild composition of arthropod communities in trees. **Journal of Animal Ecology**. 51: 289-306.

Naylor, B. J.; Bendell, J. F. 1983. Influence of habitat diversity on the abundance and diversity of small mammals in jack pine forests in Ontario. pp.295-307 in **Resources and Dynamics of the Boreal Zone**. (ed. Wein, Riewe, Methven) Association of Canadian Universities for Northern Studies.

Nealis, V.G.; Lomic, P.V. 1994. Host-plant influence on the population ecology of the jack pine budworm, *Choristoneura pinus* (Lepidoptera: Tortricidae). **Ecological Entomology**. 19: 367-373.

Nipson, H. E. 1978. **Inbreeding in the ant species *Formica exsectoides***. Ph.D. dissertation. Harvard University.

Nonacs, P. 1986. Ant reproductive strategies and sex allocation theory. **Quarterly Review of Biology**. 61: 1-21.

Oster, George F.; Wilson, Edward O. 1978. **Caste and ecology in the social insects**. Princeton University Press, Princeton, NJ.

Owen, R. E.; Packer, L. 1994. Estimation of the proportion of diploid males in populations of Hymenoptera. **Heredity**. 72: 219-227.

Pamilo, P.; Rosengren, R. 1983. Sex ratio strategies in *Formica* ants. **Oikos**. 40: 24-35.

Pamilo, P.; Sundström, L.; Fortelius, W.; Rosengren, R. 1994. Diploid males and colony-level selection in *Formica* ants. **Ethology, Ecology, and Evolution**. 6: 221-235.

Patterson, R.S. 1994. Biological control of introduced ant species. in **Exotic ants: biology, impact, and control of introduced species**. (ed. D. F. Williams) Westview Press: Boulder, CO, p.293-307.

Pielou, E. C. 1969. **Mathematical Ecology**. John Wiley: New York.

Price, W. A. 1945. The Allegheny mound ant and its control. **Journal of Economic Entomology**. 38(6): 706.

Rice, W. R. 1990. A consensus combined p-value test and the family-wide significance of component tests. **Biometrics**. 46: 303-308.

Richardson, B. J.; Baverstock, P. R.; Adams, M. 1986. **Allozyme Electrophoresis: a handbook for animal systematics and population studies**. Academic Press.

Roberts, Mark R.; Gilliam, Frank S. Patterns and mechanisms of plant diversity in forested ecosystems: implications for forest management. **Ecological Applications**. 5(4): 969-977.

Rosengren, R.; Pamilo, P. 1986. Sex ratio strategy as related to queen number, dispersal behavior and habitat quality in *Formica* ants (Hymenoptera: Formicidae). **Entomol. General**. 11: 139-151.

Ross, K. G.; Fletcher, D. J. C. 1986. Diploid male production—a significant colony mortality factor in the fire ant, *Solenopsis invicta* (Hymenoptera: Formicidae). **Behavioral Ecology and Sociobiology**. 19: 283-291.

Rudolph, T. D.; Laidly, P. R. 1990. *Pinus banksiana* Lamb (Jack Pine). pp.280-293 in **Silvics of North America: 1. Conifers**. (ed. Burns, Russell M.; Honkala, Barbara H.) Agriculture Handbook 654. U.S. Department of Agriculture, Forest Service, Washington, D.C.

Savolainen, Riita. 1990. Colony success of the submissive ant *Formica fusca* within territories of the dominant *Formica polyctena*. **Ecological Entomology**. 15: 79-85.

Scheiner, S. M. 1993. MANOVA: Multiple response variables and multispecies interactions. pp. 94-112 in **Design and Analysis of Ecological Experiments**. (ed. Samuel M. Scheiner, Jessica Gurevitch) Chapman & Hall: New York.

Scherba, Gerald. 1958. Reproduction, nest orientation and population structure of an aggregation of mound nests of *Formica ulkei* Emery (Formicidae). **Insectes Sociaux**. 5(2): 201-213.

Spires, S.; Bendell, J.F. 1983. Early postfire effects on some invertebrates, small mammals and birds in north-central Ontario. pp.308-318 in **Resources and Dynamics of the Boreal Zone: Proceedings of a conference held at Thunder Bay, Ontario, August 1982**. Association of Canadian Universities for Northern Studies.

Smith, M. R. 1947. A generic and subgeneric synopsis of the United States ants, based on workers. **American Midland Naturalist**. 37: 521-647.

Starr, C. K. 1979. Origin and evolution of insect sociality: a review of modern theory. in **Social Insects, vol.1** (ed. Hermann, H. R.). Academic Press: New York, p.35-80.

Thornhill, Nancy Wilmsen.(ed.) 1993. **The natural history of inbreeding and outbreeding**. University of Chicago Press, Chicago, IL.

Trivers, R. L. 1985. **Social Evolution**. Benjamin Cummings: Menlo Park, CA.

Trivers, R. L.; Hare, H. 1976. Haplodiploidy and the evolution of the social insects. **Science**. 191: 249-263.

Volney, W.J.A.; McCullough, D.G. 1994. Jack pine budworm population behavior in northwestern Wisconsin. **Canadian Journal of Forest Research**. 24: 502-510.

Weaver, J.E.; Smith, B.D. 1993. Allegheny mound ant control, Grant Co, WV, 1992. **Insecticide and Acaracide Tests**. 18: 337.

Wheeler, Diana E. 1994. Nourishment in ants: patterns in individuals and societies. pp.245-277 in **Nourishment and Evolution in Insect Societies**. (ed. James H. Hunt and Christine A. Nalepa) Westview Press: Boulder, CO.

Wheeler, W.M. 1928. **The social insects: their origin and evolution**. Harcourt, Brace and Company, New York.

Wheeler, W.M. 1933. **Colony-founding among ants**. Harvard University Press: Cambridge, MA.

Williams, G. C. 1975. **Sex and Evolution**. Princeton University Press: Princeton, NJ.

Wilson, E. O. 1971. **The Insect Societies**. Harvard University Press: Cambridge, MA.

Wilson, Louis F. 1977. **A guide to insect injury of conifers in the Lake States**. USDA Forest Service, Agriculture Handbook No. 501: Washington, D.C.

Zoladeski, C. A.; Maycock, P. F. 1990. Dynamics of the boreal forest in northwest Ontario. **American Midland Naturalist**. 124(2): 289-300.