

approximate specific activity of 0.3 mCi/ μ g. The minimal detectable quantity of α -interferon was 0.05 ng/ml. The specificity of the radioimmunoassay was confirmed; a) the assay did not cross-react with β -interferon, γ -interferon, or ACTH; b) the recombinant α -interferon Ro 22-8181 inhibited the binding of 125 I- α -interferon to a level comparable with inhibition by BALL-1 cell α -interferon⁷. A linear correlation existed between the radioimmunoassay (y) and the virus inhibition assay (x), with a regression line of y on x as $y = 0.659x + 245$ (u) ($p < 0.01$). Circulating α -interferon was extracted and concentrated from plasma either by silicic acid or antibody immunoadsorption, and the dilution curves of plasma and extracted samples of plasma were completely parallel to the standard curve⁷. Each assay was standardized by introducing appropriate concentrations of internal standard α -interferon, and the inter-assay variation in our hands was 4.12%⁷. The values below the sensitivity limit of the assay were calculated as 0.05 ng/ml, according to the previous results of extraction studies⁷. Peripheral blood was drawn heparinized, and plasma was immediately separated and stored at -20°C until assayed.

Results and discussions

Circulating α -interferon in the plasma of healthy individuals was low in children and reached the highest level in the young adult, subsequently declining gradually with age (fig.). Circulating α -interferon was highest at ages 30–39 years; 0.201 ± 0.059 ng/ml in males ($n = 19$) and 0.184 ± 0.076 ng/ml in females ($n = 14$). These results seem to reflect a gradual change in immune surveillance of the host during aging.

Previous studies have shown that, although circulating α -interferon did not change significantly after measles virus infection⁸, circulating α -interferon did increase af-

ter hepatitis A virus infection (Shiozawa K. et al., submitted). Circulating α -interferon was, however, significantly low (below 0.05 ng/ml, the sensitivity limit in this assay) in certain diseases such as rheumatoid arthritis⁷, and in some patients with adult T cell leukemia, for whom α -interferon therapy was highly effective⁹. This is in contrast with the present finding that circulating α -interferon is maintained up to a certain level even in very elderly individuals, unless they suffer from certain diseases. These findings appear to suggest that circulating α -interferon, the concentration of which may reflect that of endogenous α -interferon, is kept to a certain level, and to maintain the level of endogenous α -interferon would often be mandatory for the host's intact immune protection. There is evidence that endogenous α -interferon is indeed required for protection against tumor growth in vivo¹⁰ and replication of retroviruses in vitro^{11,12}.

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Genetic structure and division of labor in honeybee societies

N. W. Calderone*, G. E. Robinson and R. E. Page Jr

Department of Entomology, The Ohio State University, Columbus (Ohio 43210, USA)

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Summary. Recent studies have demonstrated a genotypic component to the division of labor among worker honeybees. However, these studies used artificially-selected strains of bees or colonies derived from queens that were instrumentally inseminated with the semen from very few males. We present evidence for genotypic variability among groups of workers performing tasks in colonies with naturally-mated queens. These results demonstrate that genetic structure is a level of social organization in honeybees.

Key words. Social behavior; honeybee; division of labor; behavior genetics.

Division of labor is fundamental to the complex organization of insect societies^{1,2}. The prevailing model of division of labor explains behavioral differentiation among

individual workers solely on the basis of age and environmental factors^{1–3}. However, nestmates may differ from one another genetically as a consequence of polyandry,

polygyny, and recombination. Recent studies suggest that genotypic variability plays a significant and previously unrecognized role in division of labor.

Calderone and Page⁴ demonstrated that workers of the same age from two selected strains of honeybees differed in their likelihood of collecting pollen and differed in the age at which they made the transition from nest to foraging activities. Robinson and Page⁵ found differences among workers in their tendencies to guard the nest entrance and remove corpses, using honeybee colonies derived from instrumentally-inseminated queens with three electrophoretically-distinct subfamilies (in the polyandrous honeybee, workers with the same father comprise a subfamily). Frumhoff and Baker⁶ studied colonies composed of two visually-distinguishable subfamilies and demonstrated a genetic component to variability in the frequency of grooming behavior.

Selected strains and colonies comprised of only a few, genetically-marked subfamilies are powerful techniques in genetic analyses of colony social organization. However, results that demonstrate a genetic basis for division of labor using these methods may not necessarily be generalizable to colonies under more natural conditions. Artificial selection may result in much larger behavioral differences than those found among related nestmates in wild-type colonies. Similarly, results based on colonies with only two or three subfamilies may be an artifact of the family structures compared to those found in wild-type colonies with queens that normally mate with about 17 males⁷. We compared the behavior of workers from different subfamilies in unselected colonies with naturally-mated queens. Allelic variants of the enzyme malate dehydrogenase (*Mdh*) were used as phenotypic markers.

Materials and methods

Techniques for studying the *Mdh* locus in honeybees are well established^{5,8}, including their use as subfamily markers. Using the 'slow' (S), 'medium' (M), and 'fast' (F) electromorphs of *Mdh*, we identified two naturally-mated queens that were homozygous for the S allele and that were both producing three biochemically-distinguishable groups of worker offspring (SS, SM, SF). Due to polyandry, each of these three allozyme-phenotype groups included individuals from one-or-more subfamilies.

Workers that were either guarding the entrance, located in the honey-storage area, foraging for pollen, or foraging for nectar were collected (n = 200/colony for each group) and electrophoretically assayed to establish their allozyme phenotype. Individuals were identified as guards using unambiguous behavioral criteria⁹. Bees from the honey-storage area were collected immediately after collecting the guards. The number of guards for each of the three allozyme phenotypes was compared with the corresponding numbers of honey-storage-area

bees because these groups of workers are thought to be similar in age^{3,10}.

Foragers were sampled by collecting workers that alighted on a screen that was temporarily placed over the entrance of each colony. Pollen foragers were identified by the presence of their conspicuous pollen loads. Alighting workers that did not carry pollen were classified on the basis of their crop contents^{11,12}. All but six were found to have at least 20 µl of nectar with a sugar concentration of at least 30% and were classified as nectar foragers. Outgoing foragers, workers on orientation flights, and water collectors were thus not included in the sample. The distributions of pollen foragers and nectar foragers were compared directly because these tasks are normally performed by bees of the same age¹³⁻¹⁶.

Guards and honey-storage-area bees were not compared with pollen foragers or nectar foragers because these tasks are performed at different ages and the relative frequencies of the subfamilies comprising the three allozyme-phenotype groups may have changed over time. This could occur if sperm contained in the queen's spermatheca were incompletely mixed.

In each comparison (guards versus honey-storage-area bees and pollen foragers versus nectar foragers), the two phenotypic distributions of workers are expected to be the same if variability among workers of the same age is solely environmental, because workers within a colony share a common environment. Differences in the distributions of workers among the three allozyme-phenotype groups greater than that expected on the basis of sampling error alone demonstrates genotypic variability.

Results and discussion

Phenotypic representation of pollen foragers and nectar foragers among the three allozyme-phenotype groups differed significantly in both colonies, while guards and honey-storage-area bees differed in one colony (table). These results are consistent with results of recent studies that used selected strains⁴ or colonies with small num-

Frequencies of guards, honey-storage-area bees, pollen foragers, and nectar foragers in each of three, allozyme-phenotype groups (SS, SM, SF) of worker honeybees (results of G-test¹⁷: ns = not significant; *p < 0.05; **p < 0.01; ***p < 0.001). Both Colony A and Colony B were derived from naturally-mated queens.

	Colony A					Colony B				
	SS	SM	SF	df	G	SS	SM	SF	df	G
Guards	163	12	25	2	5.2 ^{ns}	180	20	0	1	4.2*
Honey-storage-area bees	175	4	20 ¹			166	34	0		
Pollen foragers	192	4	4	2	25.8***	182	17	1	2	9.3**
Nectar foragers	161	13	26			162	31	6 ^a		

^an = 199 because the phenotype of one individual could not be determined.

bers of genetically-marked subfamilies^{5,6}. Together, these results demonstrate genetic contributions to variability in worker behavior. These results were obtained even though the assay we used is relatively insensitive compared to one in which each allozyme-phenotype group corresponds to a single subfamily.

The observed differences in allozyme-phenotype frequencies between guards and honey-storage-area bees may be a consequence of changing patterns of sperm use over time if guards and honey-storage-area bees have different age distributions. Although workers from these groups are likely to be very similar in age, this relationship is not firmly established^{3,10}. This argument is less tenable as an explanation for the differences between pollen foragers and nectar foragers. Workers performing these tasks have not been shown to represent different age groups, therefore, we suggest that the observed differences in this case are primarily a consequence of genotypic effects on worker behavior. These results demonstrate that genetic structure is an important component of the social organization in unselected honeybee colonies with naturally-mated queens.

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- * Present address: U.S.D.A., Beneficial Insects Laboratory, BARC-EAST, Bldg 476, Beltsville, MD 20705, USA.
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Cadmium effects and biochemical status in hamsters following acute exposure in late gestation

D. P. Hanlon and V. H. Ferm

Department of Anatomy, Dartmouth Medical School, Hanover (New Hampshire 03756, USA)

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Summary. A normally teratogenic dose of cadmium ions administered to hamsters late in gestation does not cross the placental barrier nor does it result in placental pathology. Our finding, which differs from data for other rodents, may be due to differences in the chemical status of cadmium in the placental cytosols.

Key words. Placenta; cadmium ion; biochemical status; hamster.

The cadmium ion (Cd^{++}) is a potent teratogen in mammals and other vertebrates². The deleterious effect of Cd^{++} on in utero development is not restricted to the production of fetal abnormalities but also produces profound toxic responses in both placentas and fetuses of certain rodents during late gestation³⁻⁷. This paper reports our findings for the fetotoxic and placental-toxic effects of Cd^{++} administered to hamster dams late in gestation. Our study includes an appraisal of the distribution and chemical status of Cd in the maternofetal system.

Materials and methods

Pregnant hamsters (LKV strain) were obtained from the Charles River Breeding Labs on day 5 of gestation. The dams were held individually in cages and fed rat chow and water ad libitum. Individuals were injected i.p. with 7.80 μmoles of Cd^{++} (as the sulfate salt) per kg on the morning of day 14 of gestation (about 36 h before birth). Carrier-free $^{109}\text{Cd}^{++}$ (New England Nuclear) served as the radiotracer (5–10 $\mu\text{C}/\mu\text{M}$ Cd). Animals were sacrificed 24 h post-injection using CO_2 inhalation. Maternal