ly, elicit stable changes in the activities of various enzymes associated with both favourable and unfavourable consequences. In our experiments with hypercholesterolic SWR/y mice, postnatal administration of IPA resulted in a long-term induction of the activities of microsomal enzymes and 7-cholesterol hydroxylase which caused a stable decrease in blood cholesterol level[26]. Postnatal treatment of galactosemic rats with galactose elevated the activity of G-6-PD, enhanced galactose oxidation and considerably alleviated the symptoms of galactesemia such as cataracts, hepatomegaly and splenomegaly[27]. By contrast, when administered to rats, cortisol suppressed metabolic responses to the hormone in adults and weakened their protective reactions to stressing agents[28]. The data obtained indicate that postnatal induction may serve as an efficient means for long-term control of gene expression providing obvious beneficial effects. However, uncontrolled exposure to some genetic inducers during early postnatal life may be deleterious.

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Hybridization and inbreeding effects on genome coadaptation in a haplo-diploid hymenoptera: *Cothonaspis boulardi* (Eucoilidae)

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Summary. Hybridization and inbreeding effects were tested in 2 populations of a haplo-diploid parasitic wasp, *Cothonaspis boulardi.* No inbreeding depression was observed. This suggests adaptation of the genome to total homozygosity and the absence of new gene rearrangements which could be lethal after hybridization.

It has been postulated that in parthenogenetic species with a haplo-diploid system of reproduction, genomes are coadapted under conditions of hemizygosity[2] and thus indirectly under homozygosity[3]. Since disadvantageous genes are immediately revealed in haploid males, they should not be able to accumulate in such haplo-diploid systems[4,5]. With this haplo-diploid system, therefore, no inbreeding effects are expected. Nevertheless, some theoretical considerations predict that variability due to deleterious alleles is only reduced compared with that in diploid species and not completely eliminated[6]. Experimental investigations have demonstrated inbreeding effects for morphological traits, oviposition rate and honey yield of bees[7–10] and fecundity, fertility[11] and egg hatchability[12] of predaceous Acarina. However, in the latter group, arrhenotoky is now contested[13].

Though the sex locus problem has not always been excluded in these studies, the results have been interpreted as suggesting that a) lethality exists on male haploid loci in systems of balanced polymorphism[6], b) heterotic mechanisms play a role in the diploid part of the population[5], c) inbreeding apparently brings about the loss of internal genetic balance and causes low viability possibly through disruption of polygenic complexes[11,14].

These hypotheses were tested by hybridization studies. Hybridization may disturb the presumed coadapted genetic systems and destroy the favorable gene combinations exist-

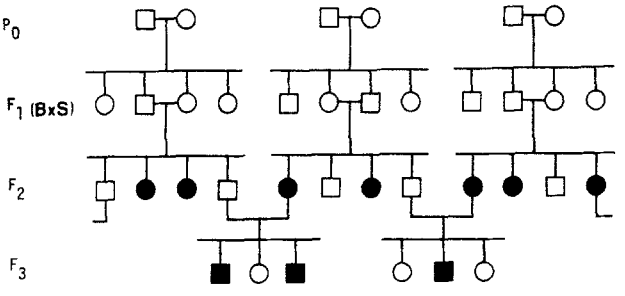

Fig. 1. Diagram of crosses. ■, inbred males and ●, inbred females. Open squares and circles are non-inbred individuals.

ing in parents. The fitness effects of such perturbations were observed in an inbreeding experiment.

Materials and methods. *Cothonaspis boulardi*, a haplo-diploid hymenoptera (family Eucoilidae) was reared in its host, larvae of *Drosophila melanogaster*. Virgin adults were obtained after dissection of parasitized *Drosophila* pupae. Couples were immediately formed, placed in 100-ml plastic boxes,fed with honey and kept at 22°C. Every 2nd day each box received 0–48-h-old eggs or larvae of *Drosophila* in sufficient numbers to minimize superparasitism. Productivity (number of descendants) and sex-ratio (percent males) were measured during a 10-day period in which more than 90% of the total offspring was produced.

2 stocks of parasites, recently caught in 2 geographically distant areas, France (Malaucène) and Guadeloupe (Petit Bourg), and their hybrids were studied. In each stock and their hybrids, unrelated individuals were randomly crossed. They constituted the parental P_0 generation (figure 1). Among the resulting progeny, individuals were crossed by brother-sister matings. The F_2 offspring of these F_1 sib couples consisted of normal males and inbred females. These F_2 males and females were then randomly crossed (non-inbred matings) leading to F_3 male offspring of inbred origin and to non inbred F_3 female offspring. Thus, at each generation inbreeding concerns one sex only. Differential mortality between males and females should then result and lead both to a reduction in total offspring production and a variation in sex ratio. In this context, a high percentage of males in the offspring of F_1 and of females in the offspring of F_2 is to be expected.

Results and discussion. Distributions of sex ratio obtained in various generations are shown in figure 2. No modification of sex ratio appeared in subsequent generations of the France stock ($F=0.35$; $p > 0.50$) nor of the hybrid involving France females ($F=1.4$; $p > 0.20$). On the other hand, Guadeloupe stock and hybrids involving Guadeloupe females showed a significant increase in the percentage of males among the offspring of F_1 (stock: $F=13.2$; $p < 0.001$; hybrids: $F=4.6$; $0.01 < p < 0.05$). No effect was observed among the offspring of F_2. These sex ratio variations are not correlated with a decrease in productivity[15]. Instead, an unexplained slight increase in offspring production by F_1 was observed ($F=2.7$ and $F=4.7$; $0.01 < p < 0.05$ for the 2 cases). This sex ratio divergence cannot, therefore, be explained by differential mortality between inbred males and females. It can be suggested that inbreeding may interfere with a sex determination mechanism, as in other species of hymenoptera. In these species, such as *Habrobracon*[16] or bees[7,17] sex determination is under the control of a sex locus with many alleles[18]. Males develop normally from haploid eggs but they can also develop from diploid eggs if the alleles on the sex locus are homozygous[19]. In this case, inbreeding increases the probability of a diploid egg being homozygous in sex alleles and consequently developing into a diploid male. Thus the percentage of males in the offspring of F_1 is increased without any modification of total productivity. F_2 couples then involve either haploid males, for which no effect appears in the progeny, or diploid males. In this latter case, triploid females[20] appear in the offspring of F_2; nevertheless, the sex ratio comes back to the normal value.

How can the absence of an inbreeding effect in the France stock be explained? It may be supposed that this stock either has no sex locus, or carries a neutral allele which even in a homozygous condition allows diploid eggs to develop into females. This neutral allele hypothesis implies that development of an egg towards the female sex is determined by the diploidy of the genome rather than heterozygosity. Therefore, active alleles, when they exist, would act as recessive modifiers which when homozygous would lead diploid eggs to develop into males. Such a neutral allele could also exist in the Guadeloupe population, but at low frequency. The differences between the 2 stocks France and Guadeloupe could thus be explained by differences in allele frequency.

Another explanation of our results could be that the sex ratio deviation in inbred offspring is due to factors affecting the proportion of eggs fertilized. But physiology of the F_1 females is not involved since these females are not inbred. Gametic incompatibility could therefore exist, resulting in the failure of egg fertilization by sperm genetically too closely related. Sib matings lead to such gametic incompatibility in the American oyster[21].

In conclusion, it apprears that apart from interference with sex determination, inbreeding has no effect on fitness in this species. As is the case with many parasites and some social hymenoptera[22], this absence of inbreeding depression is consistent with the idea that their gene pool is adapted to hemizygosity and thus to complete homozygosity. Selection acting mainly on males may eliminate deleterious combinations of genes so that even hybridization between different

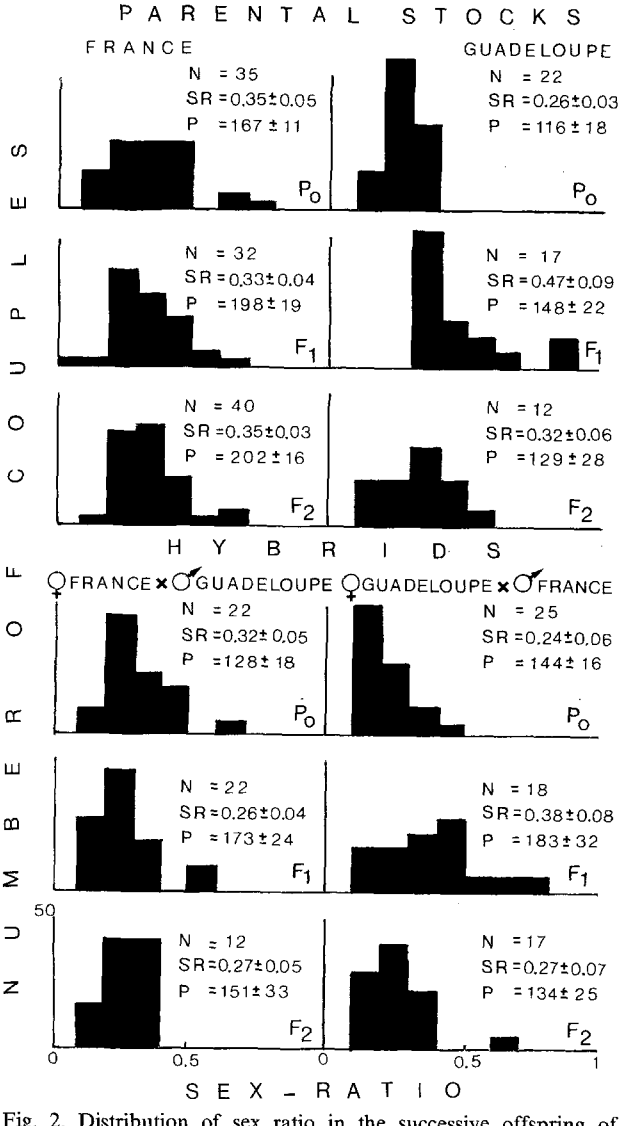

Fig. 2. Distribution of sex ratio in the successive offspring of generations P_o, F_1 and F_2, for the 2 populations and their hybrids. For rapid graphical comparison, the distributions are based on 100 couples. N, total number of couples studied; SR, sex ratio (percent males); P, productivity (number of descendants).

stocks does not lead to unfavorable genetic interactions. It is interesting to note in this context, that in the X-chromosomal haplo-diploid situation in *Drosophila* synthetic, lethal combinations do not occur[23].

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Giemsa centromere staining of *Humulus lupulus* L. metaphase chromosomes

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Summary. Giemsa staining of hop (*Humulus lupulus* L.) chromosomes at metaphase revealed kinetochore-like structures in the centromeric region.

Pardue and Gall[2] were the first to demonstrate the differential staining of mouse chromosomes with Giemsa stain. Since then, numerous workers have demonstrated the differential staining of animal chromosomes with Giemsa; the types of bands obtained have been designated as G-bands, C-bands, R-bands etc. These techniques have made it possible to identify different chromosomes and to detail their morphological features. The different Giemsa banding techniques were initially applied to animal chromosomes only, as plant chromosomes posed some difficulties. By now many workers have perfected these techniques on plant chromosomes also[3–5]. In plants C-banding has been demonstrated clearly, but there is no report of G-banding yet[6]. C-banding reveals the centromeric type of constitutive heterochromatin. In this paper an attempt was made to demonstrate the centromeric heterochromatin in *Humulus lupulus* L. metaphase chromosomes.

Materials and methods. Root tips were collected from the field during morning hours, thoroughly washed with distilled water and pre-treated in 0.05% aqueous colchicine for 4–5 h. The fixation was carried out in 1:3 acetic acid-alcohol for 24 h, followed by storage in 70% alcohol. The tips were hydrolyzed in 0.2 N HCl for 10 min at 60 °C and then washed thoroughly with distilled water. Squashing was performed in 45% glacial acetic acid. The cover slips were removed in 95% alcohol and air dried. The air-dried cover slips were kept for 2 days as such and then were placed in 0.007 N NaOH for 2 min at room temperature. After removal from NaOH, they were washed for about 10 min with distilled water and air dried. The air-dried cover slips were incubated in 2×SSC (0.3 M NaCl+0.03 M trisodiumcitrate, pH 7.0) at 60 °C for 45 min, then washed with distilled water for about 15 min, air dried and kept overnight. The staining was performed in Giemsa (E. Merck) diluted 50 times with M/15 Sörensen's phosphate buffer (pH 6.8) at room temperature. The optimum staining takes place in about 30 min. Then the cover slips were washed with distilled water, air dried and mounted in Euparal. All the solutions were prepared freshly.

Results and discussion. The diploid chromosome number of *H. lupulus* L. is 2 n = 20. In conventional staining techniques like Feulgen and aceto-orcein, all the 20 chromosomes show an unstained gap in the centromeric region whereas the Giemsa technique produced 2 darkly stained dots (figure). Giemsa Cd-banding technique of Eiberg[7] has revealed 2 dots in the region of the centromere in human chromosomes. Evans and Ross[8] have concluded that the Cd-bands are essentially the kinetochores. It has now been shown by electron microscopic studies that the centromere consists of 2 disc- or ball-shaped structures which are designated as kinetochores[9]. Stack[10], studying *Allium cepa, Ornithogalum virens, Tradescantia edwardsiana* and *Rhoeo discolor,* reports specific staining of the centromeric regions with Giemsa as paired dots and denotes them as kineto-