tions of Tween 20 undergo autoxidation[9]. Ascorbic acid itself can autoxidize and generate H_2O_2 at an obviously slow rate[10], but in the presence of other autoxidizable substances ascorbic acid can stimulate both O_2 consumption and H_2O_2 production[11]. Ascorbic acid can react with superoxide anion to generate H_2O_2[12]. In the presence of traces of metal ions, superoxide anion and H_2O_2 might generate the highly reactive hydroxy radical. If catalase is inhibited by azide, the membrane lipid peroxidation[13,14] may progress due to the increased generation of H_2O_2 and other oxidants. The depletion of cellular reduced glutathione and the oxidation of protein thiol groups may cause cell lysis. We have a hypothesis that the oxidative damage of membrane ATPase is crucial for this, since according to our unpublished result, the potassium loss preceded the cell lysis, and the inactivation of ATPase occurring during the treatment could be prevented by the addition of DTT. Vitamin E is a radical scavenger[15,16] and may play a structural role in protecting membrane lipid-protein complexes against oxidative damage.

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A queen bee extract (*Apis mellifera* L.) (Hymenoptera) reduces the fecundity of *Tenebrio molitor* L. (Coleoptera)

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Summary. 3 different doses of crude queen bee extract were administered to pairs of adult *Tenebrio molitor* L.; the extract exerted an inhibitory action on fecundity, but did not induce complete sterility of the females.

The queen of *Apis mellifera* is known to hinder the development of the worker bees' ovaries by means of a pheromone[1-3] which has been isolated and identified as trans-9-keto-2-decenoic acid[4,5]. Both the pure substance (natural or synthetic) and crude extracts of queen bee are active in other insects as well: in *Kalotermes flavicollis* (Isoptera) they influence caste differentiation[6-8], in *Aedes aegypti* (Diptera) they cause death and delay moulting and metamorphosis[9,10]; but more frequently they exert an inhibitory action on fecundity, as they do in bees. This is so in *Formica fusca* (Hymenoptera)[11], *Musca domestica* (Diptera)[12] and *K. flavicollis*[8], and even in a crustacean, *Leander serratus*[11].

The experiments reported here show that crude extracts of queen bee, administered orally, also reduce fecundity in *Tenebrio molitor* L. (Coleoptera).

Queen bees at least 1-year-old were homogenized in alcohol. The lipoproteins were precipitated from the solute with acetone[13]; the solute was then concentrated in a vacuum at 30 °C, dissolved in alcohol, and spread on wheat wafers which were fed to pairs of *Tenebrio molitor* as their only food. 3 doses were used: 1:100, 2:100 and 4:100 (ratios by weight of crude extract to wafer at room conditions). The controls were fed wafers treated with alcohol alone. Every 4 days the pairs were given drinking water on dampened filter paper.

Single pairs of *T. molitor* which had moulted less than 24 h earlier were placed in plastic boxes with fine wood sawdust, where they laid their eggs. On the 5th, 9th, 13th and 17th days after metamorphosis each pair was moved to another box and the eggs counted. Thus 4 lots of eggs, laid successively, were counted per couple.

Since the treatment with the 3 different doses of extract was done at different times a separate series of control pairs was set up in each case.

Table 1 shows the average number of eggs per lot laid by

Table 1. Average number of eggs (by lot) laid by treated and control pairs

	Treated insects			Controls			
Dose	No. pairs	No. lots	No. eggs per lot	No. pairs	No. lots	No. eggs per lot	t
1:100	27	108	25.56 ± 1.32	20	80	27.46 ± 1.73	0.89
2:100	23	92	23.64 ± 1.57	23	92	28.68 ± 1.64	2.22*
4:100	24	96	22.08 ± 1.52	22	88	28.72 ± 1.67	2.94*
Total	74	296	23.82 ± 0.85	65	260	28.32 ± 1.00	3.51*

* $p < 0.05$.

Table 2. Variation of the average fecundity of treated and control couples in the period following metamorphosis

Days	Treated insects		Controls		t
	No. females	No. eggs	No. females	No. eggs	
1–5	74	12.82 ± 1.21	65	16.95 ± 1.59	2.09*
6–9	74	34.03 ± 1.74	65	40.54 ± 1.99	2.47*
10–13	74	28.42 ± 1.31	65	30.00 ± 1.29	0.85
14–17	74	20.07 ± 1.33	65	25.78 ± 1.55	2.81*

* $p < 0.05$.

treated and control females. No females died during the study and all laid eggs.

The pairs treated with the 1:100 dose laid the same number of eggs on the average as the controls. Those treated with the 2 larger doses, instead, laid fewer. The variance, according to Bartlett's test, was found to be homogeneous and therefore not altered by the treatment.

Table 2 shows the average number of eggs laid in the 4 consecutive time intervals by all treated and control pairs. The treated pairs always laid fewer eggs than the controls. The effect of the extract on their fecundity was almost immediate; their 1st lot of eggs was already smaller on the average than those of the controls. The reduction of fecundity remained nearly constant throughout the period of extract administration and there was no accentuation of the effect. 3 treated females (out of 74) and 7 controls (out of 65) only began laying eggs after the 5th day; the treatment thus did not delay the onset of oviposition but only reduced the number of eggs laid.

Some eggs laid either by the control pairs (11.8%) or by the treated ones (12,6%) dried up, remaining whitish; no significant differences were found.

On the whole the queen bee extract given by mouth to pairs of *T. molitor* acted very quickly in reducing the average fecundity of females, but did not induce complete sterility nor produce cumulative effects on fecundity.

Some researches have shown that the 'royal substance' of *Apis mellifera* hinders the enlargment of the corpora allata in orphan worker bees[14] and in orphan pseudergates of *K. flavicollis*[15]. It may act in the same way in other insects as well: reducing, that is, the fecundity of females by making the corpora allata wholly or partly inactive.

It is even more interesting, however, that the 'royal substance' of *Apis mellifera* has been shown to be active in an ever wider number of species, and thus does not have a strictly specific action.

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Measuring nanogram amounts of DNA by photographing fluorescence in thin layers of agarose

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Summary. A method is described which permits the measurement of DNA amounts of as low as 0.5 ng by photographing the fluorescence of ethidium bromide after complete removal of unbound dye. The DNA is immobilized in thin layers of agarose.

A reliable and easily-reproducible assay for the measurement of minute quantities of DNA is desirable when the results depend very much on the precise determination of these values. Normally the evaluation has to be made with a sample that is taken from a small aliquot with limited amounts of DNA. Typical applications of such assays are during DNA-protein binding studies and for the determination of specific radioactivities where only the removal of amounts of DNA in the ng range is acceptable. Previous attempts to solve this problem have been published[3–6] and the assay which is described relies on information conveyed in all of them. The given method may be used to estimate DNA amounts of as low as 0.5 ng.

The following DNAs were used: calf thymus and plasmid col. E1 (Boehringer, Mannheim); phage lambda (a gift of H. Bujard, Heidelberg); lambda rif^{d18} (a gift of P. Venetianer, Szeged); pBR 322 and several DNA inserts in this plasmid; *E. coli* chromosomal, and several chloroplast DNAs[7]. The latter DNAs were prepared as described before[7] and all were stored sterile at 4 °C in 0.5 mM EDTA in concentrations of approximately 0.5 mg/ml. DNA concentrations were determined photometrically on the basis of a molar extinction coefficient of 6800 M^{-1} cm^{-1} at 260 nm in 1 × SSC, pH 6.8.

For the assay, 4 ml of 1% high-temperature gelling agarose (MCI, Rockville) in 10 mM NaCl were allowed to solidify