Phagostimulatory Responses of Male and Female *Sitophilus granarius* L. to Newly Harvested and Stored Wheat Grains

H.Z. Levinson and K.R. Kanaujia

Max-Planck-Institut für Verhaltensphysiologie, D-8131 Seewiesen

The granary weevil *Sitophilus granarius* L. is known to develop mainly in stored cereal seeds [1], but does not usually grow in immature kernels. Shifts in the chemical composition during storage including reduction in water and other volatiles, accumulation of fatty acids and acid phosphate [2] may influence the nutritional suitability of grain for the granary weevil. In fact, female and male *S. granarius* are retained for significantly longer periods of time on stored kernels than on newly harvested seeds of wheat [3]. Therefore we investigated the significance of the major components of newly harvested as well as stored wheat kernels for the stimulation of feeding in both sexes of *S. granarius*.

Wheat grains aged one week (=fresh wheat) or one year after harvest (=stored wheat) were extracted for 24 h in either pentane or water at 20±2 °C. After evaporation of the solvent, the extracts were dried to constant weight and tested in graded amounts on mated granary weevils being ≈ 1 month after pupal-adult ecdysis. Before the experiments, the insects were kept without food for ≈ 3 days and sexed according to the shape of their rostrum [4]. The test arena consisted of a glass ring (diam. 10 cm, ht. 3.5 cm) with a layer of thin porous tissue paper on the basis being suspended 3.5 cm above a hot stage maintained at 29±0.1 °C. Ten male or female weevils were conditioned to the above environment for 2–4 h, whereupon 10 µl of an extract or solvent were deposited in the center of the arena and the behavior responses of the insects observed for 20 min at 24–25 °C and 200 lux (four repetitions per sex, extract and dosage).

In the vicinity of either aqueous or pentane extracts of wheat (fresh and stored) the male and female granary weevils displayed a typical sequence of behavior [5] which – according to the dosage – consists of raising and waving of the antennae, elevation of the head and thorax, arrestance and intermittent and/or continuous probing by directing the rostrum to the impregnated region. In both sexes of *S. granarius* the time of arrestance was positively correlated with the extent of probing.

The water extracts of fresh and stored wheat were definitely more active than the corresponding pentane extracts in inducing arrestance of male and female *S. granarius* (Fig. 1). The prolonged arrestance due to the water extracts is caused partly by the fact that water alone acts as a feeding aggregant for both sexes. Figure 1a shows that the average time of arrestance of male weevils in response to pentane and water extracts of both kinds of wheat clearly depends on the dosage offered. Interestingly, the time of male arrestance caused by pentane extract of stored wheat was 3.7–11.8 times longer (according to the dosage) than the one induced by pentane extract of fresh wheat. On the other hand, such a gap in activity between the aqueous extracts of fresh and stored wheat was not evident.

Female granary weevils were found to respond to both wheat extracts by longer periods of arrestance than the male weevils (Fig. 1b). The ratio of arrestance of females/males ranged from 0.9 to 2.2 for stored wheat and 1.3–5.9 for fresh wheat in the case of pentane extracts, while it was 1.2–1.6 for stored wheat and 0.8–2.1 for fresh wheat in the case of water extracts. When dosages ranging from 1–10^3 µg were offered, pentane (but not water) extracts of stored wheat caused 2.5–4.3 times longer periods of female arrestance than pentane extracts of fresh wheat. The extensive periods of arrestance along with repeated probings of female *S. granarius* on both wheat extracts indicate that the latter may also contain oviposition attractants.

It is thus clear that pentane-soluble phagostimulants accumulate in wheat seeds during storage, whereas water-soluble phagostimulants are present in the grain kernels at harvest as well as after one year's storage. The chemical composition of cereal seeds is known to change during storage, mainly because of microbial activity which in turn leads i.a. to an increase in free fatty acids, particularly linoleic, oleic, and palmitic acid [2]. The latter were shown to exert phagostimulation in several insect species infesting stored foodstuffs [1]. Among the water-soluble components of wheat some cations, mono- and disaccharides are also known to be phagostimulants [1]. Air passed over newly harvested wheat was found to be twice as attractive to granary weevils as that passed over stored wheat [6]; this is probably due to

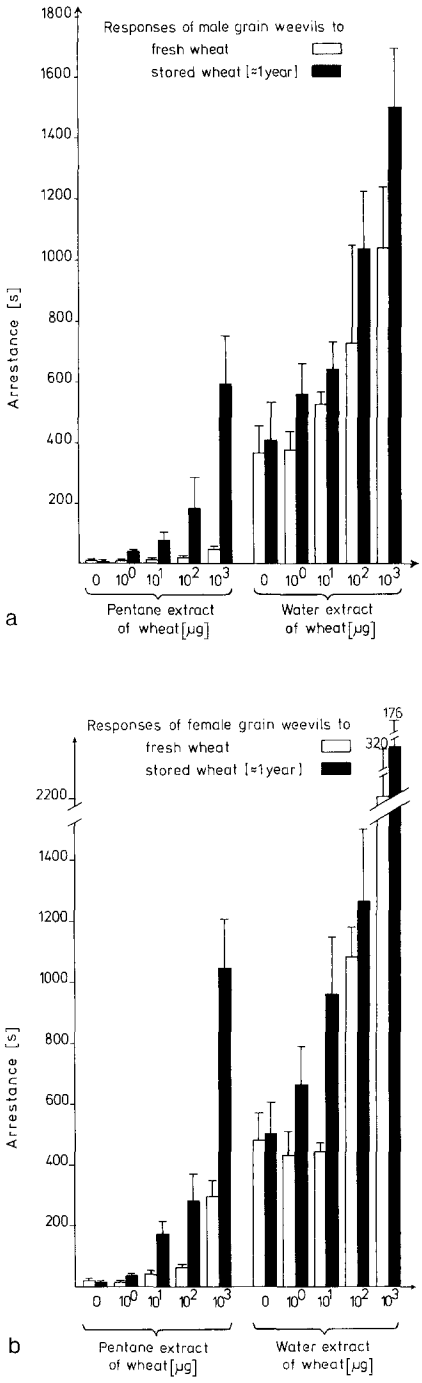

Fig. 1. Retention time of (a) male (b) female *Sitophilus granarius* (≈ 1 month old) on graded amounts of different wheat extracts for a 20-min period. Each column represents the average time of arrestance (including probing) of 10 granary weevils in 4 repetitions

substantial evaporation of water, lower alcohols, aldehydes, and ketones from newly harvested wheat. The accumulation of phagostimulatory components in stored wheat is likely to have played an important role in the process of domestication and spread of infestations by granary weevils. The study is being continued in order to elucidate the function of the phagostimulants involved.

Received October 22, 1980

1. Levinson, H.Z., Levinson, A.R.: Ent. Exp. Appl. *24*, 505 (1978)
2. Rohrlich, M., Thomas, B., in: Handbuch der Lebensmittelchemie, Bd. 5, S. 179. Berlin-Heidelberg-New York: Springer 1967; Zeleny, L., Coleman, D.A.: Cereal Chem. *15*, 580 (1938)
3. Kanaujia, K.R., Levinson, H.Z.: unpublished results
4. Halstead, D.G.H.: Bull. Ent. Res. *54*, 119 (1963)
5. Levinson, H.Z.: Z. angew. Entomol. *84*, 1 (1977)
6. Donat, H.J.: ibid. *65*, 1 (1970)

Different Ecdysteroid Titers in Spring- and Summer Generations of the Swallowtail, *Iphiclides podalirius*

K. Scheller and T.A. Wohlfahrt
Zoologisches Institut der Universität, D-8700 Würzburg

J. Koolman
Physiologisch-Chemisches Institut der Universität, D-3550 Marburg

The postembryonic development of insects is under the control of morphogenetic hormones: the ecdysteroids and juvenile hormones [1]. These hormones are controlled by the nervous system. Exogenous conditions, such as day length, temperature, or humidity, are capable of modifying via the nervous system the duration of development as well as certain behavioral patterns or morphological properties. This might be achieved by changes in hormonal conditions, e.g., by a change in the hormone titer.

The postembryonic development of the swallowtail *Iphiclides podalirius* is determined by day length [2]. Short-light days induce diapause and the appearance of a spring generation in the following year. Long-light days induce a second generation, the summer generation. Males of *Iphiclides* of spring- and summer generation have been found to differ by a number of morphological traits [3]. The differences between the two types are related to alterations of the physiological strategies of the caterpillars as responses to their internal milieu. Therefore, it was of interest to determine whether the titer of ecdysteroids is different in both types of butterflies.

Both, hemolymph which was separated from hemocytes by centrifugation and the remainder insect were homogenized separately in methanol and analyzed by radioimmunoassay with an ecdysone-specific antiserum (DUL-1) after a procedure described elsewhere [4]. The age of the insects after ecdysis from 4th to 5th instar was 7 days (insect A and B) and 7.75 days (C and D). Control insects started pupation at day 11.5 (spring generation), and day 10 (summer generation).

The values for the ecdysteroid concentrations in the hemolymph as well as in the homogenate (Table 1) are in the same range as those published for other lepidopteran insects [5]. It is remarkable that the concentration in the hemolymph is clearly lower than in the remainder animal. Obviously, there must be at least one tissue with a high ecdysteroid content. It is well known that the hemolymph titer of ecdysteroids may differ from the average content in the whole insect, reflecting an uneven distribution of the hormone [6]. This is understood to be due to carrier proteins and/or intracellular receptor molecules.

Table 1. Comparison of the ecdysteroid concentrations in 5th instar larvae of *Iphiclides*. The concentrations were expressed as equivalents of ecdysone. Each value represents the average of 3 determinations. The standard deviation was less than 10%

Insect (single determinations)		Hemolymph [n*M*]	Remainder caterpillar (homogenate) [pmol/g]
Spring generation	A	12.8	21.5
	B	12.5	23.8
Summer generation	C	2.0	33.0
	D	4.5	21.6

The hormone titers do not significantly differ in the homogenates of the spring- and summer generations. However, the ecdysteroid concentration in the hemolymph of the spring generation is about 3 times higher than that of the summer generation. We assume that this difference is significant and typical for the different generations. The question whether ecdysteroids are reponsible for the different phenotypes of *Iphiclides* must be left to further experiments which are underway.

Received October 1, 1980

1. Riddiford, L.M.: Ann. Rev. Physiol. *42*, 511 (1980)
2. Wohlfahrt, Th.A.: Verh. Dtsch. Zool. Ges. *1954*, 133 (1955)
3. Wohlfahrt, Th.A.: Spixiana *2*, 113 (1979)
4. Reum, L., Koolman, J.: Insect Biochem. *9*, 135 (1979)
5. Böhm, G.-A.: Zool. Jb. Physiol. *83*, 361 (1979)
6. Koolman, J., Scheller, K., Bodenstein, D.: Experientia *35*, 134 (1979)

Giant Human Erythrocytes by Electric-Field-Induced Cell-to-Cell Fusion

P. Scheurich and U. Zimmermann

Arbeitsgruppe Membranforschung am Institut für Medizin, Kernforschungsanlage Jülich, D-5170 Jülich

Cell-to-cell fusion plays an important role in several biological phenomena including fertilization, endo- and exocytosis, and the biogenesis of muscle fibers [1]. Furthermore, the in vitro induction of cell fusion has attracted increasing interest in the last decade for both membrane research and somatic hybridization [2].

In general, cells can be fused either by addition of fusogenic substances such as po-