approximately one-tenth as active as the terminal epoxides. The methylenedioxybenzyloxy analogs of IIa, IIb, IIc, and III, prepared from piperonyl alcohol by analogous methods, were uniformly one-tenth as active on both test insect species.

The exceptionally high order of activity of these compounds prompted me to examine the effect of their vapors on *Tenebrio*. Thus, 20 freshly molted *Tenebrio* pupae confined in a pint jar, exposed only to the vapors of 0.1 to 0.5 mg of IIa or IIc coated on the bottom quarter of each jar, molted to pupal adult intermediates and second pupae. Thus, the potential utility of the present compounds to control sensitive pest species is compounded by the additional possibility of using them as fumigants.

As candidates for the control of insect pests, these new hybrid compounds would appear to be superior to methyl *trans,trans*-10,11-epoxyfarnesenate and the cecropia hormones by virtue of their greater biological activity and ease of synthesis. Although some of these compounds display biological activity in the picogram range, they are not directly toxic to insects up to a million times the concentration required to prevent metamorphosis. Thus, they do not kill insects in the manner of ordinary insecticides but are effective by deranging development, through interference with metabolic processes that are able to proceed only in the relative absence of JH. Since these compounds are not directly toxic to insects in the conventional sense, imaginative and timely applications to pest species must be developed to fully utilize their practical potential.

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References and Notes

1. W. S. Bowers, *Science* **161**, 895 (1968).
2. ———, M. J. Thompson, E. C. Uebel, *Life Sci.* **4**, 2323 (1965).
3. H. Roller, K. H. Dahm, C. C. Sweeley, B. M. Trost, *Angew. Chem.* **79**, 190 (1967).
4. A. S. Meyer, H. A. Schneiderman, E. Hanzmann, J. H. Ko, *Proc. Nat. Acad. Sci. U.S.* **60**, 853 (1968).
5. M. Julia, S. Julia, R. Guegan, *Bull. Soc. Chim. Fr.* (1960), p. 1072.
6. W. S. Bowers, H. M. Fales, M. J. Thompson, E. C. Uebel, *Science* **154**, 1020 (1966).
7. K. Slama, M. Suchy, F. Sorm, *Biol. Bull.* **134**, 154 (1968).
8. I thank Dr. H. Roller for the pure natural isomer of the major cecropia juvenile hormone (compound IVb).

24 December 1968

Oyster Ciliary Inhibition by Cystic Fibrosis Factor

Abstract. *Ciliary inhibition in oysters serves as an assay in identifying a serum factor in cystic fibrosis patients and heterozygotes. Serums from 47 patients with cystic fibrosis and 19 heterozygotes caused ciliary cessation within 35 minutes, whereas serums from only 2 of 64 individuals without cystic fibrosis inhibited ciliary activity within this time.*

Serums from patients with cystic fibrosis (CF) inhibit ciliary synchrony of rabbit tracheal explants incubated for 4 to 6 days in culture (*1*). Concentrated preparations of serums from known CF heterozygotes also cause cilia rhythm to become asynchronous. The serum factor responsible was described by Spock *et al.* (*1*) to be heat-labile and nondialyzable. Additional evidence of an abnormal factor in cystic fibrosis was found by Mangos *et al.* (*2*), who described an inhibitor of sodium transport present in sweat and saliva from CF patients; this may or may not be the same as the serum abnormality.

We have tested the effect of serum from CF patients on the ciliary activity of oyster gills, a readily available source which eliminates lengthy incubation. Gill tissue from fresh oysters (*Crassostrea virginica*) was removed, and vertical sections (3 by 3 mm) were suspended in filtered seawater in a hanging drop preparation (*3*). After cilia were observed to be active in seawater (under phase-contrast microscopy), the seawater was replaced by serum, and the preparation was tightly sealed with wax and examined at various times. The time in which the cilia stopped beating was recorded for each serum. Sealed cilia preparations in seawater remained active for approximately 1 hour. The typical reaction produced by serum from CF patients on oyster tissue was an immediate expulsion of debris from tubules between the gill mounds followed by cessation of the mound cilia. In Fig. 1, the cilia in normal serum are compared to gill cilia exposed for 15 minutes to serum from CF patients. Typical accumulation of debris covers the cilia and mounds in the serum preparation from CF patients.

The time required for ciliary cessation was approximately the same on duplicate runs of the same individual's serum; however, the age and condition of the serum affected the obtaining of repeatable results. After serum had been frozen and thawed a number of times, the capacity to stop ciliary action was lost. Hemolyzed serum failed to give repeatable results. Because of the variation in ciliary activity of oyster gills, it was necessary to use controls with each series of unknown serums, including seawater, serum from normal individuals, and serum from CF patients. Unknowns were examined every 5 minutes and tested in duplicate on gill cilia from different oysters.

In a study in which the identity of all samples was unknown to the examiners, the effects of serums from 47 CF patients, 19 CF heterozygotes, 25 allergic children, and 39 healthy individuals were observed (*4*). Serums from 62 individuals without CF did not inhibit ciliary action, which persisted for 40 to 50 minutes (Fig. 2). The serums from children with allergic rhinitis and bronchial asthma failed to inhibit the oyster cilia; movement persisted from 40 to 50 minutes. Serum from normal individuals did not overlap CF homozy-

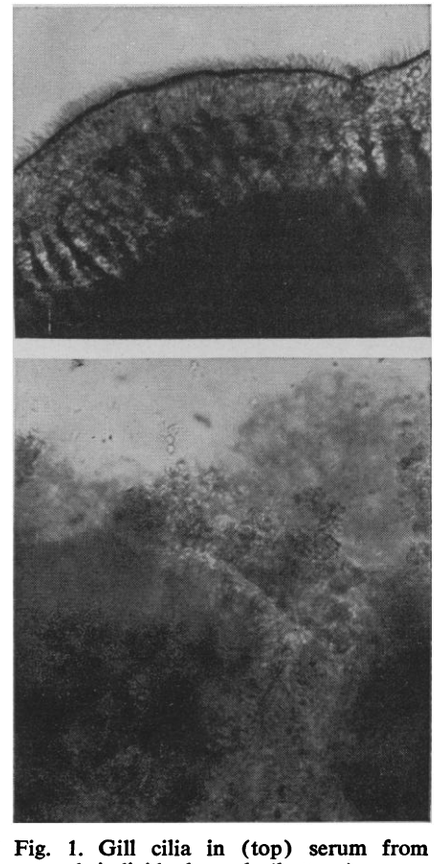

Fig. 1. Gill cilia in (top) serum from normal individuals and (bottom) serum from CF patients. Typical accumulation of debris covers the cilia and mounds in the serum preparation from CF patients (phase contrast, × 160).

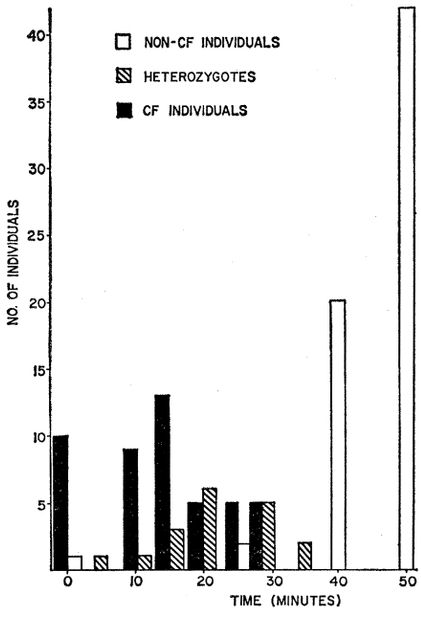

Fig. 2. Number of individuals (CF patients, heterozygotes, and normal individuals) whose serums stopped oyster cilia at intervals from 0 to 50 minutes.

gotes and heterozygotes except for the serums from two healthy individuals which caused the cilia to stop in less than 30 minutes. It is not known whether these two individuals are heterozygotes for cystic fibrosis; however, serums from 19 heterozygotes did cause the ciliary action to stop in less than 5 to 35 minutes. Although the population sampled was small, the results from the serums of these two individuals without CF are compatible with the estimated heterozygote frequency of approximately 2 to 10 percent. None of the serums from CF patients or their parents permitted ciliary action as long as serums from the normals. Serums from 47 CF patients stopped ciliary action in periods ranging from the time of exposure to within 30 minutes after contact. Neither age, sex, nor clinical severity of CF influenced the time of ciliary cessation.

When cystic fibrosis serum was diluted with seawater at ratios of 1:1, 1:2, and 1:3, the first, but not the second concentration, caused the cilia to stop before 35 minutes. The inhibitory activity was present in serum from CF patients after dialysis against seawater in Visking tubing 8/32. Saliva from CF patients, but not from normal individuals, also inhibited ciliary action. Twenty minutes of exposure to saliva from CF patients caused ciliary cessation.

Additional results indicate that the factor in CF patients and heterozygotes responsible for cessation in oyster cilia shares properties in common with the CF factor found by Spock *et al.* (*1*) and by Mangos *et al.* (*2*). It is nondialyzable, heat-labile, and has a molecular weight between 75,000 and 180,000 as indicated by gel filtration. Mangos and McSherry found significant inhibition of transductal reabsorption of sodium in the rat parotid after it was exposed to saliva and sweat from CF patients, in addition to solutions of the following compounds (all bases)—polylysine, polyornithine, and protamine sulfate (*2*). The effects of these compounds were also observed on oyster cilia preparations. After exposure to dilute concentrations of these solutions (*5*), oyster ciliary action stopped immediately after administration of polylysine and polyornithine. Protamine sulfate did not stop the cilia before 40 minutes; but immediately after exposure, there was a moderate expulsion of debris from the gill mounds, an indication of tissue injury produced by this compound. Although the mechanism of these basic compounds and the CF factor which inhibits ciliary activity is not yet understood, it is interesting that the same synthetic compounds which mimic the inhibitory effect of saliva and sweat from CF patients on sodium reabsorption in the parotid also inhibit oyster ciliary action.

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References and Notes

1. A. Spock, H. M. C. Heick, H. Cress, W. S. Logan, *Pediat. Res.* **1**, 173 (1967).
2. J. A. Mangos and N. R. McSherry, *Science* **158**, 135 (1967); *Pediat. Res.* **2**, 378 (1968).
3. L. H. Lockhart and B. H. Bowman, in preparation.
4. Serums from CF patients attending clinics at the Texas Rehabilitation Hospital, Houston, and the University of Minnesota Medical School, Minneapolis, were provided by Drs. G. Harrison, R. Doggett, and W. J. Warwick, respectively. Controls included serums from 13 healthy children in the Head Start Programs of LaMarque and Texas City, Texas; 26 healthy adults; and 25 children suffering from allergic rhinitis and bronchial asthma were provided by Dr. A. Goldman.
5. Polylysine hydrobromide, molecular weight 40,000 to 100,000 and polyornithine hydrobromide, molecular weight 60,000 to 120,000 (Pierce Chemical Company) were prepared to the concentration of 1 mg/ml with filtered seawater. Protamine sulfate (Upjohn) was prepared as 1 mg/ml in normal saline.
6. Supported by PHS grant HD 03321; by special project 409, Children's Bureau of the Department of Health, Education, and Welfare; and by grant allocation from The National Foundation. We thank L. Nicholson and S. Anderson for technical assistance.

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Averaged Evoked Responses in Vigilance and Discrimination: A Reassessment

Abstract. *With the use of monopolar recordings for averaged evoked responses, detected signals in a vigilance task are associated with a late positive component which is absent for undetected signals as well as nonsignals. Bipolar recordings obscure the late positive component associated with detected signals. The data suggest that the late positive component represents cerebral processes associated with evaluation of unpredictable changes in stimulation.*

Although human averaged evoked responses (AER) vary in a lawful manner with changes in stimulus parameters (*1*), and in some instances with perceptual variables (*2*), as yet no portion of an AER can be specifically attributed to processes underlying the perception or evaluation of a stimulus. Discrimination tasks, in which errors of stimulus perception occur, provide an opportunity to observe differences in AER to stimuli which are physically identical but are perceived differently. Several experiments of this sort have been reported, but only one by Haider *et al.* (*3*) assessed the AER to both correct and incorrect responses to the *discriminanda.* In this study the observer was required to detect small changes in stimulus intensity (signals) randomly interspersed among more frequent standard stimuli (nonsignals). The forms of the AER for detected and undetected signals did not differ, although the amplitude was greater for the detected signals. Since the AER amplitude to nonsignals in successive blocks of stimuli over the entire experiment tended to decrease and correlated with the proportion of signals detected within each block, the changes in the signal AER appeared to reflect alterations in subjects' alertness rather than processes specifically related to detection of the signals.

In contrast to these results of Haider *et al.* (*3*), we found (*4*) that, when subjects were reading, unpredictable onset of stimulation as well as stimulus changes randomly embedded in a series of standard stimuli elicited AER containing a prominent late positive component (LPC), usually highest between 300 and 350 msec after presentation of the stimulus. Although the standard stimuli were ignored by the subjects, the unpredictable stimulus changes