trophic component of at least some forms of obesity may thus be due to an abnormality in the mechanisms which constitute adipocyte resistance to enlargement. Whether there is some neural or humoral mediation between adipocyte size and feeding behavior is not known, but some such association now seems likely.

IRVING M. FAUST
PATRICIA R. JOHNSON
JULES HIRSCH
Rockefeller University, New York 10021

References and Notes

1. A. Gasnier and A. Mayer, *Ann. Physiol. Physicochim. Biol.* **15**, 145 (1939).
2. G. C. Kennedy, *Proc. R. Soc. London Ser. B* **140**, 578 (1953).
3. R. A. Liebelt, *Ann. N.Y. Acad. Sci.* **110**, 723 (1963); _____, S. Ichinoe, N. Nicholson, *ibid.* **131**, 559 (1965).
4. R. Schemmel, O. Mickelsen, S. A. Pierce, J. T. Johnson, R. G. Schirmer, *Proc. Soc. Exp. Biol. Med.* **136**, 1269 (1971); C. Chlouverakis and J. Hojnicki, *Metabolism* **23**, 133 (1974); I. M. Faust, thesis, Cornell University (1973).
5. J. G. Kral, *Am. J. Physiol.* **231**, 1090 (1976); I. M. Faust, P. Johnson, J. Hirsch, *ibid.*, p. 538.
6. J. Hirsch and P. W. Han, *J. Lipid Res.* **10**, 77 (1969).
7. M. R. C. Greenwood and J. Hirsch, *ibid.* **15**, 474 (1974).
8. R. Schemmel, O. Mickelsen, J. L. Gill, *J. Nutr.* **100**, 1041 (1970).
9. The high-fat diet was composed (percentages by weight) of Crisco (55), casein (25), dextrose (13), salts (4), Nutritional Biochemicals Vitamin Mix (2.82), L-cystine (0.12), and L-cysteine-HCl (0.06).
10. J. Hirsch and E. Gallian, *J. Lipid Res.* **9**, 110 (1968).
11. We have found that when inguinal depots are removed from 3-week-old rats, complete regeneration will occur over a period of about 7 months, if the rats are fed a high-fat diet [I. M. Faust, P. R. Johnson, J. Hirsch, *Science* **197**, 391 (1977)]. At the time the rats in the present experiment were killed (3 months after inguinal depot removal) inguinal depot regeneration was already clearly noticeable.

12. Supported in part by NSF grant PCM 76-09324 and a grant from the Howard M. Pack Foundation. The Osborne-Mendel rats were generously donated by C. Reader of the National Institute of Cancer Research. We thank J. Monahan and R. Kava for their excellent technical assistance.

21 December 1976; revised 14 March 1977

A Critical Period for Acoustic Trauma in the Hamster and Its Relation to Cochlear Development

Abstract. *Young hamsters pass through a developmental stage during which they are unusually susceptible to acoustic trauma. This sensitive period occurs after apparent structural and functional maturation of the ear and appears to be dependent on unidentified developmental changes within the cochlea.*

Exposure to loud sound can produce permanent damage in human and animal cochleas (*1, 2*). Recent research on the phenomenon of priming for audiogenic seizures (*3, 4*) and on acoustic trauma in young guinea pigs (*5*) suggests that young mice from certain inbred strains and young guinea pigs are particularly susceptible to acoustic trauma. The data in these studies do not provide any evidence on the locus of the developmental changes presumed to underlie the observed changes in susceptibility. The object of this study was to examine susceptibility of the hamster ear to acoustic trauma as a function of age. The data show that (i) young hamsters pass through a critical period of susceptibility to noise trauma and (ii) the developmental events underlying this phenomenon appear to occur in the cochlea.

Hamsters (*Mesocricetus auratus*) were obtained from a commercial dealer (*6*) at specific ages or were bred in the laboratory. Animals were exposed to an octave-band noise (5 to 10 khz, 125 db re 20 $\mu N/m^2$) for 2.5 minutes at one of the following ages: 11, 15, 19, 23, 27, 31, 40, 48, 55, 62, or 75 days after birth. Five days after noise exposure, cochlear microphonic (CM) responses were measured in exposed animals and in control animals of the same ages. The procedures used for noise exposure and CM recording have been described (*4*) and will be mentioned briefly here.

Hamsters were anesthetized with urethane (1.5 mg per gram of body weight, injected intraperitoneally) and placed in a head holder after tracheal cannulation. The ear canal was excised at the level of the tympanic ring, and the auditory bulla was exposed. The tip of a sound speculum containing a calibrated probe tube was sealed over the tympanic ring, and a silver ball electrode was placed onto the round window through a fenestra in the bulla. The intensity of sound (in decibel sound pressure level) necessary to produce a criterion CM response of 1.0 μv was determined at frequencies between 0.5 and 20.0 khz. All surgery and CM recording were conducted using appropriate double-blind procedures.

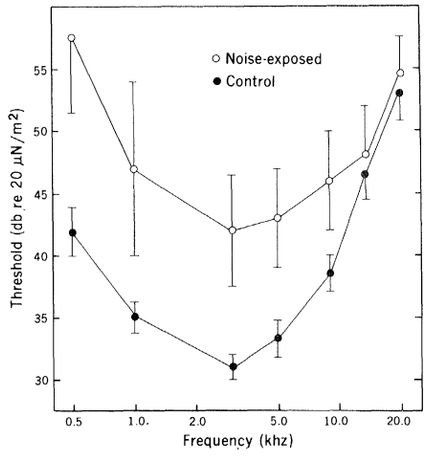

Fig. 1. Mean (± S.E.) 1.0 μv threshold versus frequency for six 45-day-old control animals and nine 45-day-old animals exposed to noise at 40 days of age.

Mean thresholds at each frequency were determined for control and noise-exposed animals in each age group. Examples in one age group are presented in Fig. 1. These data typify the pattern of threshold loss seen in all age groups in which significant threshold losses were observed (*7*). The difference between means for control and noise-exposed animals at a given frequency provided a measure of threshold loss at that frequency in a particular age group. These measures of threshold loss (in decibels) were averaged over all test frequencies (0.5,1,3,5,9,13.5, and 20 khz) in each age group. Threshold loss, measured 5 days after noise exposure, was dependent on developmental age (Fig. 2A). Although the amount of threshold loss within a given age group depended on frequency, the general relationship between threshold loss and age at noise exposure was similar at all frequencies tested (*7*).

It seems unlikely that a critical period of susceptibility to acoustic trauma has any adaptive advantage for the young hamster. Rather, we interpret the existence of the phenomenon as indicating that some developmental change is occurring between 27 and 55 days of age, a correlate of which is enhanced susceptibility to noise-induced CM deficit, a deficit that presumably reflects permanent damage to the organ of Corti (*8*).

It might be expected that a given noise exposure would not be maximally effective in producing cochlear damage before about 20 days after birth. Structural development of the hamster ear has been studied by Stephens (*9*), who reported that the middle and inner ear appear mature by light microscopy at about 30 days after birth. At 15 days, the malleoincudal joint has not yet ossified, and mesenchyme continues to be abundant in the middle ear. Thus, there is no reason to expect the middle-ear transmission system fully to function before 20 days of age. However, given the apparent structural maturation of the ear by 20 days, there is no obvious reason to predict that the effectiveness of noise exposure in inducing threshold loss should decrease

between 55 and 75 days of age. Several possible explanations can be considered.

The simplest explanation might be that the resonant frequency of the hamster's ear canal changes during development. To determine the nature of this change, ear canal length was measured in six 45-day-old animals and in six 80-day-old animals. The mean lengths were 6.4 mm [standard error (S.E.) = 0.4 mm] and 8.7 mm (S.E. = 0.5 mm), respectively, which would give rise to resonant frequencies of 13,656 and 10,075 hertz. Thus, the decline in noise effectiveness cannot be explained by a change in ear canal resonance relative to the frequency content of the damaging noise.

A developmental change in the efficiency of the acoustic reflex (*10*) in attenuating transmission through the middle ear after 55 days of age could reduce susceptibility to noise trauma in older animals. This possibility was evaluated by studying an additional ten animals as controls for possible middle-ear muscle effects during noise exposure (*10*). All animals in this group were deeply anesthetized with Nembutal (50 mg/kg, intraperitoneally) at 75 days of age. Five were exposed to the noise band as in the previous experiment, and five unexposed animals served as controls for possible anesthetic effects on threshold sensitivity.

The mean threshold loss (averaged across all test frequencies) was calculated for the two anesthetized groups; the difference between these two values was 2.0 db. This difference may be compared with the mean threshold loss of − 0.4 db in the normal 75-day-old groups (Fig. 2A). An analysis of variance (*11*) over all thresholds in the four 75-day-old groups indicated that neither the noise effect nor the noise-anesthesia interaction was significant [F (1, 12) = 2.4, $P > .05$, and F (1, 12) = 0.04, $P > .05$]. Thus, middle-ear muscle activity does not appear to determine the critical period.

Any developmental change in either the middle-ear cavity or ossicular chain that resulted in a decrease in the efficiency of sound transmission through the middle ear after 55 days of age could lead to the observed decrease in threshold loss. However, any such explanation would predict that CM threshold sensitivity in normal animals would decrease after day 55. The development of CM thresholds at 3, 9, and 20 khz in control animals is shown in Fig. 2B. The CM thresholds are relatively stable after about 31 days of age, and the sensitive period for acoustic trauma is not paralleled by a period of unusually low thresholds (*12*). We conclude, therefore, that the locus of the developmental process underlying the critical period for acoustic trauma is the cochlea. Furthermore, the critical period occurs after the apparent maturation of the cochlea, as indicated by light microscopy (*9*) or by electrophysiological measures of function (this study). It would therefore seem that further developmental changes occur in the hamster cochlea for at least 40 days after apparent structural maturation at 20 days of age; the nature of this developmental change remains to be determined.

Short exposures to particularly intense sound produce immediately apparent cochlear damage which is presumably the result of mechanical disruption of the organ of Corti (*13*). Exposure to less intense sound can also permanently damage the organ of Corti, but the structural changes are not complete until about 1 month after exposure (*14*). The physiological basis of these long-term changes is not fully understood. Proposed mechanisms include changes in the permeability of the reticular lamina (*15*), metabolic exhaustion of hair cells (*15*), and disturbances in cochlear blood supply (*16*). Developmental changes within the cochlea could conceivably affect any one of these hypothesized mechanisms, but further experiments are required to describe the specific patterns of cochlear damage produced by noise exposure during the critical period.

The question of the differential sensitivity of young animals to acoustic trauma is of considerable practical importance in considering problems of hearing conservation in young children. Data from our study, together with CM data on mice (*4*) and histological data on guinea pigs (*5*), suggest that young animals pass through a period of heightened susceptibility to acoustic trauma shortly after apparent structural maturation of

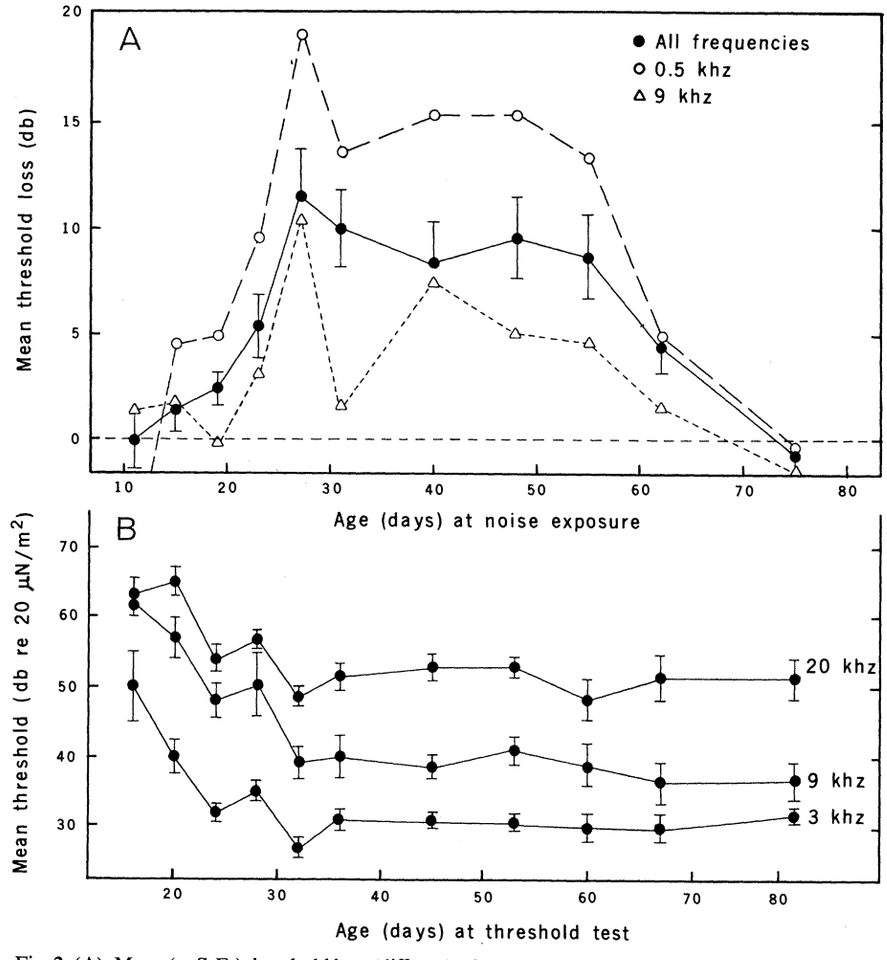

Fig. 2. (A). Mean (± S.E.) threshold loss (difference between mean threshold in control and noise-exposed groups) at 0.5 khz and 9 khz, and averaged over all test frequencies, as a function of age at noise exposure. Numbers of animals in successive control groups are 6, 6, 5, 5, 11, 7, 6, 8, 6, 4, and 4; numbers in successive noise-exposed groups are 7, 6, 7, 8, 8, 7, 9, 6, 5, and 6. (B) Mean (± S.E.) 1.0-μv thresholds in control groups. The time axis for (A) is age at noise exposure, and that for (B) is displaced by 5 days relative to the upper axis so that the curves are directly comparable.

the cochlea. If such a period occurs during human auditory development, then its onset would probably occur prenatally (*17*). Sound levels in incubators used for premature babies have been described in detail (*18*), but the consequences of these noise exposures on subsequent auditory development have not been described. The question of possible damaging consequences of high-level amplification from hearing aids in young children is also of concern to audiologists (*18*). It will be important to verify the existence of a critical period for acoustic trauma in other species and to determine whether the human auditory system passes through such a developmental stage.

GREGORY R. BOCK
JAMES C. SAUNDERS
Department of Otorhinolaryngology and Human Communication, University of Pennsylvania, Philadelphia 19104

References and Notes

1. J. D. Miller, *J. Acoust. Soc. Am.* **56**, 729 (1974); H. Spoendlin, in (*2*), pp. 69–86.
2. D. Henderson, R. P. Hamernik, D. S. Dosanjh, J. H. Mills, Eds., *Effects of Noise on Hearing* (Raven, New York, 1976).
3. G. R. Gates and C. S. Chen, *Exp. Neurol.* **41**, 457 (1973); C. S. Chen, G. R. Gates, G. R. Bock, *ibid.* **39**, 277 (1973).
4. J. C. Saunders and K. A. Hirsch, *J. Comp. Physiol. Psychol.* **90**, 212 (1976).
5. S. A. Falk, R. V. Cook, J. D. Haseman, G. M. Sanders, *Laryngoscope* **84**, 444 (1974).
6. Outbred hamsters of the LVG:LAK strain were obtained from the Lakeview Hamster Colony, Newfield, N.J.
7. The largest threshold losses occur at frequencies below the frequency range of the traumatic noise. When threshold losses are measured behaviorally, maximum threshold loss usually occurs at a frequency slightly above that of the traumatic noise. However, when threshold losses are measured by recording cochlear microphonics from the round window, maximum threshold loss is usually observed in lower frequencies [J. D. Durrant, in (*2*), pp. 179–196].
8. We are currently examining the anatomical correlates of acoustic trauma in young hamsters (L. R. Rowe, J. C. Saunders, G. R. Bock, in preparation). We have not used noise-test intervals of longer than 5 days, but several considerations lead us to believe that the threshold shifts observed in this study are permanent and not temporary. The noise exposure was very brief (2.5 minutes), and we know of no suggestion in the literature on temporary threshold shift in humans or animals that such brief exposures might lead to long-lasting temporary threshold shifts. Furthermore, even when such shifts are induced by exposure durations of several days, most of the temporary component of the threshold shift recovers within 5 days [W. D. Ward, A. Glorig, D. L. Sklar, *J. Speech Hearing Res.* **15**, 603 (1972); W. Melnick and M. Maves, *Ann. Otol. Rhinol. Laryngol.* **83**,820 (1974)].
9. C. B. Stephens, *Acta Oto-Laryngol.* Suppl. 296 (1972).
10. J. Wersäll, *ibid.*, Suppl. 139 (1958).
11. This analysis uses a 2 × 2 × 7 factorial design, the factors being noise, anesthetic presence, and test frequency. Subjects were repeated on the frequency factor [B. J. Winer, *Statistical Principles in Experimental Design* (McGraw-Hill, New York, 1962), pp. 337–345].
12. Developing thresholds and increasing acoustic trauma correspond during the first 30 days (Fig. 2). As threshold sensitivity increases toward its mature value, mean threshold loss rises toward its maximum value. This reciprocal relationship can be considered to reflect the fact that the effectiveness of a sound in producing acoustic trauma is closely related to its sensation level (its magnitude in relation to hearing threshold) rather than to its absolute intensity level. Developmental changes in middle-ear transfer characteristics could play a significant role during this phase of threshold development.
13. H. Spöendlin, *Acta Oto-Laryngol.* **71**, 166 (1971).
14. C. W. Stockwell, H. W. Ades, H. Engström, *Ann. Otol. Rhinol. Laryngol.* **78**, 1144 (1969).
15. B. A. Bohne, in (*2*), pp. 41–67.
16. J. E. Hawkins, *Ann. Otol. Rhinol. Laryngol.* **80**, 903 (1971).
17. J. C. Sanders and G. R. Bock, in *Studies on the Development of Behavior and the Nervous System*, vol. 4, *Early Influences*, G. Gottlieb, Ed. (Academic Press, New York, in press).
18. J. H. Mills, *J. Acoust. Soc. Amer.* **58**, 767 (1975); W. F. Rintlemann and F. H. Bess, in *Childhood Deafness* (Grune & Stratton, New York, in press).
19. Supported by an otolaryngology training grant (National Institute of Neurological and Communicative Diseases and Stroke 1T01NS-15769) to J. B. Snow, Jr., and by a grant from the Deafness Research Foundation. We thank R. Stanek and E. Seifter for their assistance. This research was conducted in the Auditory Research Laboratory, Philadelphia General Hospital.

7 July 1976; revised 23 November 1976

Suprachiasmatic Nuclear Lesions Do Not Abolish Food-Shifted Circadian Adrenal and Temperature Rhythmicity

Abstract. *Daytime restriction of food and water availability in nocturnal animals phase shifts the circadian periodicity of plasma corticosteroid concentrations and body temperature. These shifted rhythms persist in animals with lesions of the suprachiasmatic nuclei who are arrhythmic under normal conditions. These findings suggest the existence of an additional "clock" that may be involved in the generation of the rhythm.*

It has been suggested (*1*) that the suprachiasmatic region of the rat brain is a central pacemaker (or biological clock) responsible for the generation of several biological rhythms. Destruction of this region is associated with a loss of rhythmicity of drinking behavior, locomotor activity, sleep and wakefulness, and adrenal cortical activity, as well as with a loss of estrus cyclicity. We reported previously (*2*) that there is a 12-hour phase shift in the circadian periodicity of plasma corticosteroid concentrations and body temperature in rats maintained under normal lighting conditions, but in which access to food and water is restricted to a 2-hour period (0930 to 1130). The present studies were designed to determine whether such phase shifting

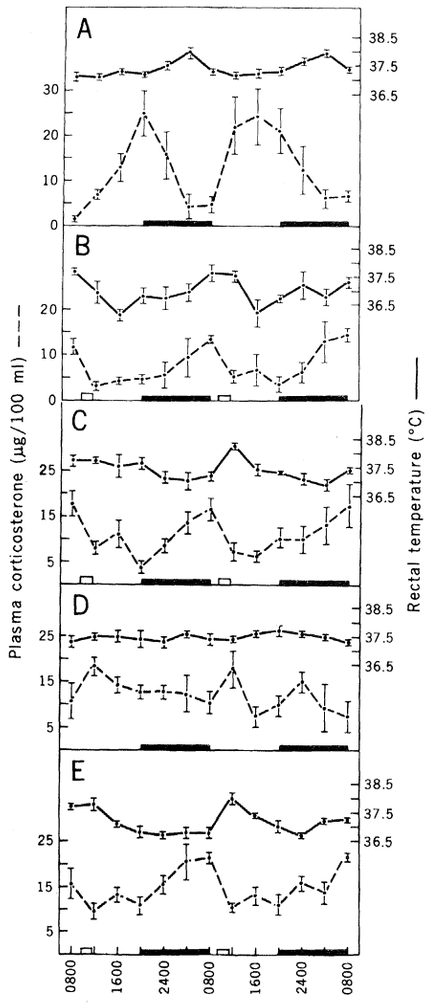

Fig. 1. Circadian periodicity (over a 48-hour period) of body temperature and plasma corticosteroid concentrations in adult female Sprague-Dawley rats. (A) Rats (N = 4) given unrestricted access to food and water and sham lesions; periodicity studied 2 weeks after the lesions were made. (B) Rats (N = 4) on restricted feeding schedule studied 2 weeks after sham lesions were made. (C) Rats (N = 7) on restricted feeding schedule studied 2 weeks after SCN lesions were made. (D) Rats (N = 5) on unrestricted feeding schedule studied 2 weeks after SCN lesions were made. (E) The same rats as in (D) studied 2 weeks later when they had been changed to the restricted feeding schedule. Vertical bars indicate ± standard error. Solid horizontal black bars indicate darkness. Open horizontal bars indicate time of daily access to food and water in animals on restricted feeding schedule. (B) and (C) show that SCN lesions do not change the shifts in the circadian patterns of body temperature and plasma corticosteroid concentrations induced by the restricted feeding schedule. The arrhythmic pattern in animals on the unrestricted schedule and with SCN lesions (D) is shifted by restricted feeding to a pattern (E) almost identical to that in the animals shown in (B) and (C). Patterns of body temperature and plasma corticosterone concentrations obtained from individual animals in (A), (B), (C), and (E) were similar to those depicted for the group.