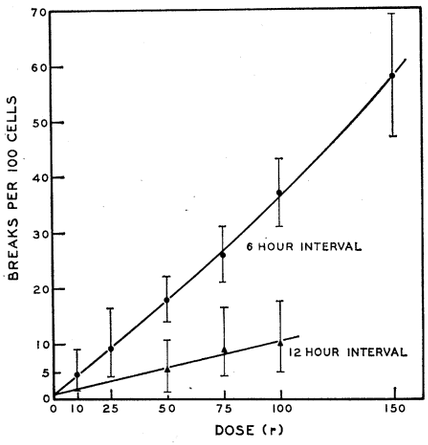

Fig. 2. X-ray-induced chromatid aberration frequencies in the corneal epithelium *in situ*. The measures of significance indicated for each point are the 95-percent probability limits, calculated according to the method of Stevens (*22*).

a small number of cells were observed in metaphase, but no mitotic burst occurred until 24 hours after treatment. However, the highest frequency of aberrations was observed 30 hours after treatment. Data for the cases in which the interval between treatment and fixation was 12 hours indicate that a marked decrease in the frequency of aberrations occurs between hours 6 and 12 after treatment.

Figure 1, *B*, *C*, and *D*, shows some of the typical chromatid-type aberrations observed. Table 1 summarizes the data obtained from all the treatments. A mean breakage rate of $(3.52 \pm 0.19)10^{-3}$ breaks per cell per roentgen was obtained for the corneal epithelium. This figure includes the data for the 6-hour fixation time for the 10-, 25-, 50-, 75-, and 100-r doses and the data for the 30-hour fixation time for the 150-r dose. At the 12-hour fixation time, for the three doses studied, a mean breakage rate of (0.95 ± 0.093) 10^{-3} breaks per cell per roentgen was obtained.

In these irradiated cells of the corneal epithelium of the Chinese hamster there is a direct linear proportionality between the dose and the yield of aberrations over the entire dose range studied, including the 10-r dose (Fig. 2). However, there is some indication that at the high dose (150 r) an exponential relationship begins to enter the dose curve, as expected, because of two-hit aberrations. The linear relationship at the lower dose levels (particularly at the 10-r dose) is of major interest since it signifies that there is no threshold effect down to the 10-r level. This rectilinear relationship at all dose levels with respect to point mutations and one-hit chromosome aberrations was predicted by Muller (*2*) and Sax (*5*), respectively, but the absence of a threshold for x-ray-induced genetic damage has been questioned by many people. Carlson's (*7*) findings in the grasshopper neuroblasts indicated that there was no threshold down to the 7.8-r level, but he was limited to scoring acentric fragments, and his findings did not answer questions pertaining to mammalian tissues. The data reported here, along with Bender's (*13*) findings of linearity between dose and yield of aberrations down to 25 r in the bone marrow of the Chinese hamster, Dubinin's (*18*) findings, and the recent work of Glass and Ritterhoff (*19*), seem to substantiate the view that there is no threshold for chromosome breakage down to the 5-r and 10-r levels.

The mean breakage rate of 0.00352 breaks per cell per roentgen obtained for the corneal epithelium is in very good agreement with the breakage rates obtained by Bender for human epithelioid cells in vitro (*9*, *10*), monkey epithelioid cells in vitro (*10*), human leukocytes in vitro (*1*), and human leukocytes in vivo (*20*) but is lower than the value obtained by Bender for Chinese hamster embryonic fibroblasts in vitro (*10*) and bone marrow cells in vivo (*13*). The differences in the data reported here and other available data on the Chinese hamster may be explained by the apparent instability of the chromosomes in embryonic fibroblast cultures, manifested in a high rate of spontaneous breakage; by differences in the interval of time between treatment and fixation; and by the qualities of the x-rays used in the studies of bone marrow and corneal epithelium. These are matters which more extensive investigation will clarify (*21*).

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General Disruption Resulting from Electrical Stimulus of Ventromedial Hypothalamus

Abstract. Electrical stimulation of the ventromedial nucleus of the hypothalamus caused hungry rats to stop eating. However, control tests showed such stimulation to be potentially noxious and capable of stopping drinking in thirsty rats. Thus the stopping of eating caused by this stimulation may not have been indicative of a primary effect on hunger.

In 1951 Anand and Brobeck (*1*) demonstrated that discrete bilateral electrolytic lesions in the region of the ventromedial hypothalamus resulted in hyperphagia, leading to obesity, whereas such lesions in the lateral hypothalamic region caused long-lasting aphagia and adipsia, leading to death.

The observation that the hunger drive can be elicited by appropriate electrical stimulation of the lateral hypothalamus is a consonant finding (*2*). Recently, the story seems to have been completed by reports that electrical stimulation in the medial hypothalamus of hungry animals inhibits eating (*3–5*). Other observations are reported here to emphasize the need for considerable care in assessing the significance of such an inhibition of eating.

Ten male rats of the Wistar strain were fitted with monopolar electrodes aimed bilaterally at the center of the ventromedial nucleus of the hypothalamus. With reference to the deGroot

(*6*) stereotaxic system, the electrodes were aimed approximately 6 mm anterior to, and 2 mm dorsal to, the interaural line at 0.65 mm on either side of the midline. A continuous 60-cycle sinusoidal current was used unilaterally for stimulation. Animals were selected which indicated by their behavior that a current of less than 50 μa was having some effect at both electrode sites. There were five such animals, in which stimulation usually caused a twitch of the ears or mild exploratory sniffing.

At the end of the experiment it was found that eight of the ten electrodes had entered the medial hypothalamus; four of them had terminated in immediate proximity to the ventromedial nucleus, the other four had terminated at points about 1 mm or less from the nucleus. Although the extent of the current spread with the stimulation technique used is not known, it appears, from the results presented below, that the particular placement within the medial hypothalamus is not critical, given the behavioral criterion for selection used in this experiment.

After recovery from the operation the rats were placed on a feeding schedule (14 g/day of Purina laboratory chow), and given experience in the test box where they received wet mash to eat, together with stimulation of various intensities, so that they would become adapted to the situation prior to formal testing.

Stimulation at low levels of current caused these rats to stop eating and to explore actively. Some animals manifested grooming behavior, both during and after stimulation. After an extended period of stimulation, activity decreased; then, when the current was cut off, there was a marked increase in exploratory behavior, lasting for as long as several minutes, followed by a resumption of eating. In a demonstration of the potent inhibitory effect of the stimulation, a rat that had had nothing to eat for 48 hours was given Noyes pellets (a favorite food of rats), together with stimulation for 30 minutes; it would not eat the pellets. Furthermore, as with Wyrwicka's and Dobrzecka's goats (*3*) and Morgane's rats (*5*), termination of such stimulation in some satiated animals led to eating, apparently on the "rebound."

Although the rats did not seem to be at all upset by stimulation at low levels of current exploratory behavior gave way to extreme restlessness and then to well-directed attempts to escape from the test box as the strength of the current was increased. This suggested the possibility that the cessation of eating was secondary to some other effect. Careful measurements support this belief.

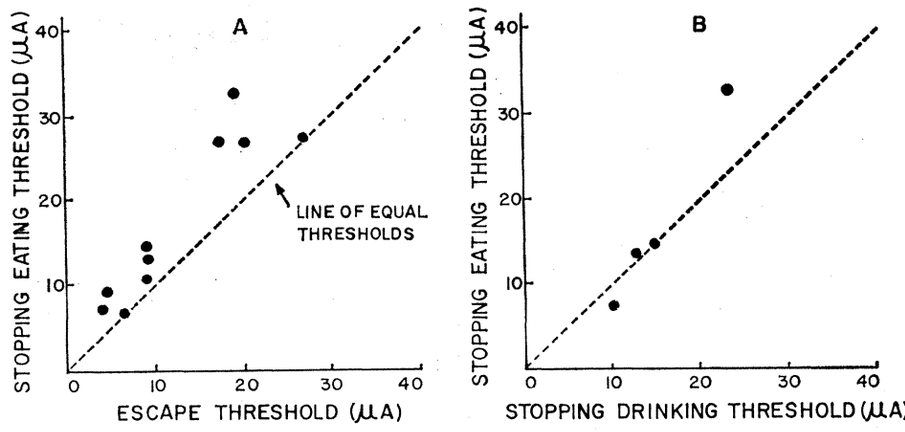

Fig. 1. Relationships between the currents required to inhibit eating and drinking for 10 seconds and the current required to motivate escape behavior.

The magnitude of the current required to inhibit feeding behavior was determined for each electrode by turning on the stimulating current at a level too low to stop the rat from eating wet mash and then advancing the current 1.25 μa after every 10-second period during which eating occurred. The level of current at which the rat first stopped eating for at least 10 seconds was considered the threshold, but in order to provide a more striking demonstration of the inhibitory potency of the stimulus, the current was then further advanced until a 1-minute period of no eating occurred. For the 70 threshold determinations made, the current required in this final minute averaged only 1.7 μa above the threshold current.

Current thresholds for the aversive effects of electrical stimulation were obtained by teaching the rats to press a bar to turn off the seemingly noxious stimulation. Since escape speed (reciprocal latency) was apparently a linear function of current, a straight line was drawn through the points and extrapolated backward to zero escape speed; the current values thereby intercepted were considered the escape thresholds.

Figure 1*A* shows the relation between the hunger-inhibiting and the aversive effects of the stimulation; each point represents the cessation-of-eating and the escape thresholds for a single electrode. Since for every electrode the cessation-of-eating threshold was at least as high as the escape threshold, it is evident that the current required to inhibit eating for 10 seconds was always in a range where it may have had aversive motivational effects. It is therefore possible that ventromedial stimulation prevents feeding behavior by upsetting or distracting animals.

Were this the case, one would expect other activities besides feeding to be "inhibited." In fact, drinking in thirsty rats was readily stopped by the stimulation. The current required to stop drinking for 10 seconds in rats that had had no water for 48 hours was determined for four electrodes in a manner identical to that used in obtaining cessation-of-eating thresholds. Figure 1*B* relates the cessation of drinking threshold to the cessation of eating threshold; current levels which caused the rats to stop eating were generally sufficient to cause them to stop drinking as well.

These results suggest that the stopping of eating caused by stimulation of the ventromedial hypothalamus and adjacent areas may not represent a primary effect on hunger (*7*).

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