rabbit anti-Lee serum (*6*) and not by anti-Asian influenza serum (*7*).

The results are presented in Table 1. In a total of six passages the original allantoic fluid inoculum was diluted cumulatively to $10^{-12.3}$. The EID$_{50}$ titers of the 3rd and 5th tissue culture passage fluids were $10^{-4.3}$ and $10^{-4.1}$ per 0.1 ml, respectively. Previous work with transmissible cell cultures which did not support the growth of influenza virus showed that in all but one case no infectious virus could be detected after 6 days of incubation on the 1st tissue culture passage and that no increase in hemagglutinins occurred (*2*). Although the hemagglutination titers obtained from the fluids of infected MDCK cultures were not uniform, hemagglutinins were present in every passage, and in one case (5th passage) reached a titer of 1:1280. The irregularity in hemagglutination titers may be due to the use for passage of low dilutions of tissue culture fluid pools containing relatively large amounts of noninfectious virus. The cytopathic effect, though somewhat variable in degree, was definitely present in every passage.

A simultaneous attempt to propagate influenza B virus in transmissible cell lines of bovine (*8*), ovine (*8*), porcine (*9*), and caprine (*5*) kidney was unsuccessful, as evidenced by the lack of cytopathic effects, inability to demonstrate hemagglutinins in the tissue culture fluids, and the inability to detect viable virus in 3rd-passage tissue culture fluids by amniotic inoculation of embryonated eggs.

Andrewes (*10*) suggests a possible origin of the Asian influenza virus from an animal reservoir in China. My evidence of the ability of influenza B virus to propagate in cultures of canine kidney cells and the report by Ado and Titova (*11*) of the experimental infection of puppies with an Asian strain of type A influenza virus suggest that the dog may be a good prospect as an animal reservoir of human influenza virus (*12*).

IRVING J. GREEN*
U.S. Naval Medical Research Unit No. 1, University of California, Berkeley

References and Notes

1. T. Francis, Jr., and T. P. Magill, *Science* **82**, 353 (1935); W. J. Mogabgab, I. J. Green, O. C. Dierkhising, *ibid.* **120**, 320 (1954); W. J. Mogabgab, I. J. Green, O. C. Dierkhising, I. A. Phillips, *Proc. Soc. Exptl. Biol. Med.* **89**, 654 (1955); K. K. Takemoto, R. K. Lynt, W. P. Rowe, R. J. Huebner, J. A. Bell, G. W. Mellin, D. J. Davis, *Proc. Soc. Exptl. Biol. Med.* **89**, 308 (1955); W. J. Mogabgab, G. I. Simpson, I. J. Green, *J. Immunol.* **76**, 314 (1956).
2. I. J. Green, M. Lieberman, W. J. Mogabgab, *J. Immunol.* **78**, 233 (1957).
3. V. D. Solov'ev and A. K. Alekseeva, *Probl. Virol.* **4**, 6 (1959).
4. J. Salk, *Am. J. Public Health* **47**, 1 (1957).
5. S. H. Madin and N. B. Darby, Jr., unpublished.
6. Rabbit anti-Lee serum obtained from the Naval Medical Research Unit No. 4, Great Lakes, Ill.
7. Anti-Asian influenza serum obtained from E. H. Lennette, Viral and Rickettsial Disease Laboratory, California State Department of Public Health, Berkeley.
8. S. H. Madin and N. B. Darby, Jr., *Proc. Soc. Exptl. Biol. Med.* **98**, 574 (1958).
9. E. Stice, unpublished.
10. C. H. Andrewes, *Perspectives in Virology* (Wiley, New York, 1959), p. 184.
11. A. D. Ado and S. M. Titova, *Probl. Virol.* **4**, 36 (1959).
12. This work was performed while I was affiliated with the U.S. Medical Research Unit No. 1, University of California. The opinions and assertions are mine and are not to be construed as official or reflecting the views of the Navy Department or the naval service at large.

* Present address: U.S. Naval Medical Research Unit No. 2, Box 14, APO 63, San Francisco, Calif.

16 April 1962

Adjustment of Monkeys to Five Continuous Days of Work

Abstract. The efficiency of well-trained monkeys improved after repeated exposures to a prolonged task. This result suggests that resistance to fatigue may be increased by prior experience with the fatiguing situation.

Experimental studies of fatigue and sleep loss in humans have often revealed surprisingly small decrements in performance, even after several days without sleep (*1*). In our experiments with monkeys we have also observed the same small performance decrements during 5 days of prolonged vigilance. We found, however, that the extent of the decrement was a function of the amount of prior experience with the fatiguing situation.

Four mature male rhesus monkeys, seated in restraining chairs for the duration of the experiment, served as subjects. During 3- to 8-hour sessions these monkeys had been trained to press a lever in order to prevent electric shocks to their feet; any pause in responding longer than 5 seconds produced a 5 to 6 ma shock of approximately 0.5-second duration (*2*). The downward force required to depress the lever was approximately 50 g. All of the monkeys had become highly skilled in the lever-pressing task before they were tested in prolonged sessions; three of them had previously received more than 300 total hours of avoidance practice.

Two of the monkeys were studied in sessions of 48-, 72-, and 120-hour duration before the standard 120-hour test was selected. The other two monkeys were tested after experiments on the first two monkeys had been completed; only 120-hour tests were scheduled for these later animals. Subjects were fed their normal food ration in a 30-minute daily period (9:00 to 9:30 A.M.) during which they did not have to press the lever. Rest periods of varying length (2 to 10 days) alternated with periods on prolonged avoidance. The subjects ate well and appeared to be healthy throughout the course of the experiment.

A safety provision in the circuitry prevented danger to an animal from the frequent shocks that would occur if the subject stopped pressing the lever. The shock circuit was automatically disconnected and the test terminated whenever an animal received 250 shocks within one day.

Over the course of 4 to 6 months each monkey was tested in at least eight sessions of continuous lever-pressing (*3*), interrupted only by the 30-minute daily eating periods and terminated either by (i) the passage of 120 hours, or by (ii) the occurrence of 250 shocks within a single day—whichever occurred first. Each prolonged session was called a test. The total number of daily shocks was used as the measure of efficiency.

Figure 1 compares performance decrements after different amounts of prior experience with the prolonged tests; each curve illustrates the decreasing efficiency of the monkeys as the test continues for 5 days. Data of tests 1 and 2 represent group means for the first two 120-hour tests; data of tests 3 and 4, the next two tests; and so forth.

In the computation of results for the tests shortened by the occurrence of 250 daily shocks before the fifth day (which happened on only three tests during the experiment), monkeys were assigned 250 shocks for every scheduled test day that followed the day on which the 250-shock maximum occurred. This assigned value is probably a very conservative estimate of how many shocks the subject would have received if the test had actually lasted the entire 5 days.

The data show that decrements over the course of the tests are reduced by prior exposure to such tests. By parametric analysis of variance of the group data, differences among the

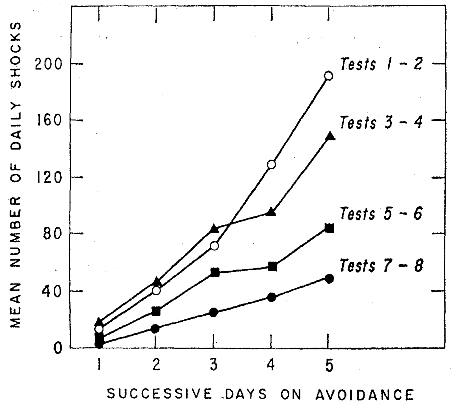

Fig. 1. Mean number of shocks per day on successive prolonged avoidance tests.

successive tests, as well as the "days × tests" interaction, were statistically significant at well beyond the .01 level. During tests 7 and 8 subjects were doing as well on the fourth day of continuous work as they had originally done on the second day (tests 1 and 2). The largest improvements occurred on the fourth and fifth days of the tests (*4*).

Even though on tests 1 to 3 individual monkeys showed marked differences in their ability to maintain performance over the 120 hours, every monkey clearly exhibited an adjustment-to-fatigue similar to that of the averaged results in Fig. 1. During tests 7 and 8 all monkeys were doing well throughout the entire 5 days; the individual differences among the monkeys were much smaller than they had been originally.

Since the monkeys were excellent avoidance performers prior to the first prolonged sessions (and no monkey received more than 20 shocks on the first day of test 1), the effect shown in Fig. 1 was certainly not due to simple acquisition of the lever response. What, then, was the mechanism through which this improvement took place? Do these results illustrate "learning" in the usual sense? Our monkeys may have learned to adjust to the situation by a change in work methods, either through the elimination of unnecessary, tiring movements or by the acquisition of more adaptive responses.

For example, we observed one monkey pressing the lever with different hands, and another monkey closing his eyes between lever presses. We did not, however, notice such techniques in all the subjects. Another possibility is that the monkeys actually learned to "sleep" and lever-press simultaneously (*5*), a phenomenon that has already been examined in humans (*6*). We are currently obtaining continuous electroencephalographic and photographic records of our subjects to determine whether the usual objective indices of sleep are modified during the course of repeated prolonged tests.

It is possible that this adjustment-to-fatigue may be correlated with some physiological or biochemical adaptation, caused by changes in the normal sleeping cycles of the monkeys or by the stressful nature of the task. If the improvement were an indirect effect of such adaptation, it might not be appropriate to apply the term "learning" to the adjustment described here.

Further analysis of adjustment-to-fatigue may prove of value in practical problems which involve prolonged monitoring, such as might occur in industry, communications, and space flight (*7*).

Robert Byck*
Eliot Hearst
Clinical Neuropharmacology Research Center, National Institute of Mental Health, Saint Elizabeths Hospital, Washington 20, D.C.

References and Notes

1. H. L. Williams, A. Lubin, J. J. Goodnow, *Psychol. Monograph* **73**, No. 484 (1959); V. R. Carlson, *Am. J. Psychol.* **74**, 552 (1961).
2. This type of avoidance schedule was first studied by M. Sidman [*Science* **118**, 157 (1953)], and examined in prolonged sessions by J. Boren [*J. Exptl. Analysis Behavior* **3**, 201 (1960)].
3. The first two monkeys tested were given daily intraperitoneal injections (6 to 12 ml) of a 10-percent solution of mixed magnesium and potassium aspartates, as well as placebo injections, during some of the prolonged tests. The specific type of improvement described in this report cannot be attributed to these injections since (i) the other two monkeys (not injected) showed the same effect, and (ii) the improvement in the injected monkeys was still observed months after the injections had been stopped. Data on physiological and behavioral aspects of the aspartates will be reported separately.
4. Later, two of the monkeys were also tested in 8- or 10-day sessions of continuous avoidance. During a total of five such tests neither subject reached the cutoff criterion of 250 daily shocks. One subject received no more than 80 shocks on any day of these extra-long tests.
5. The data from another well-trained monkey that had to make a single lever press to one of two randomly-presented visual stimuli to prevent shock (responses to the second stimulus were punished) suggest that learning to sleep-and-press cannot wholly explain the improvement reported here. This monkey had to keep his eyes open to observe stimuli and still showed an improvement with repeated tests.
6. A. M. Granda and J. T. Hammack, *Science* **133**, 1485 (1961).
7. We acknowledge the expert technical assistance of Gerald Paul and Joe Whitley.
* Present address: Department of Pharmacology, Albert Einstein College of Medicine, Bronx, N.Y.

Electroencephalogram in the Permanently Isolated Forebrain of the Cat

Abstract. The cerebral cortex of cats with chronic or permanent mesencephalic transections of the brain stem showed both high-voltage, slow-wave synchronized electroencephalographic patterns and low-voltage, fast-wave desynchronized rhythms when studied postoperatively. The alternation of sleep and wakefulness electrical activity in the permanently isolated forebrain is contrasted with the observations of Bremer in the acute "cerveau isolé" preparation.

Since the work of Bremer in the "cerveau isolé" preparation (*1*), it has been generally accepted that the cat forebrain with a high transection of the brain stem exhibits a fixed EEG pattern of high-voltage, slow-wave activity similar to that found in one of the stages of sleep in the normal cat. In recent years this stage has been called deep sleep by Dement (*2*) and slow sleep (sommeil lente) by Jouvet *et al.* (*3*). Bremer's observations have been confirmed recently (*3, 4*) in cats with rostropontine and mesencephalic transection. All of these studies have been performed, however, in experimental preparations with survival times of only 3 to 9 days after transection of the brain stem. Only Batsel (*5*) has studied, so far, long-term preparations; his observations, in dogs, are similar to those reported here in the cat.

In addition to the dramatic physiological and behavioral changes seen in high decerebrate cats, one is also impressed by the clear-cut alternation between sleep and wakefulness in these animals.

The explanation of such striking change probably rests in the restablishment of homeostasis and in the lessening of shock-like factors in the long-term preparation. Similarly, the same mechanisms could determine the permanent "slow sleep" pattern of the short-term "cerveau isolé" preparation, and conversely, a long-term isolated forebrain could exhibit both high-frequency, low-voltage waking patterns, and slow, high-voltage sleep rhythms.

Woods and Bard have emphasized the importance of the time factor in neuroendocrine experiments performed in animals with extensive lesions in the nervous system (*6*). Adametz (*7*) has also shown the importance of what he calls "shock factor" (diaschisis) in the behavioral and neurological results of acute experimental procedures.

To explore the problems of sleep-