complexity of understanding determinants of behavior, but also points out the profundity of early experience and early nutrition as major contributors to ultimate adult behavior.

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The Chimpanzee as an Animal Model for Investigating Alcoholism

Abstract. *Young chimpanzees* (Pan troglodytes) *will accept ethanol in quantities sufficient to produce symptoms of withdrawal when ethanol is subsequently discontinued. Mild to severe symptoms of physical dependence, including grand mal seizures, are observed when ethanol is abruptly withdrawn after 6 to 10 weeks of chronic oral intake. In addition, the rate of disappearance of ethanol in blood increased during periods of chronic ingestion, an indication of developing metabolic tolerance. These results suggest that the young chimpanzee may be a suitable model for experimental studies of alcoholism.*

Experimental investigation of the parameters associated with alcoholism has been limited by the lack of suitable animal models. Recently, however, Freund (*1*) reported physical dependence in mice after oral intake of ethanol, and Goldstein and Pal (*2*) obtained withdrawal reactions in mice by forcing them to continuously inhale ethanol while partially blocking ethanol metabolism with daily injections of pyrazole. Physical dependence has also been produced in dogs (*3*) by administration of ethanol through an implanted gastric cannula, and in dogs and rhesus monkeys (*Macaca mulatta*) (*4*) by introduction of aqueous ethanol solutions directly into the stomach, through a gastric catheter. With the exception of Freund (*1*), who reduced body weight to enhance ingestion of a liquid diet containing ethanol, all of these investigators employed forced administration to achieve physical dependence. We have obtained an increasing oral acceptance of ethanol by chimpanzees (*Pan troglodytes*), and have achieved pharmacologically significant concentrations of ethanol in blood and subsequent withdrawal reactions in those subjects. Thus, animal models may be appropriately used in investigations of alcoholism. Specifically, we may use as such a model, one of the species most closely related to man, the chimpanzee.

When chimpanzees were 1 to 7 months old, we offered them doses of ethanol mixed with a liquid diet at scheduled feeding times. Normal weight gains were maintained in these developing animals with a liquid diet based upon a standard nursery formula (SMA, Wyeth) combined with Provimalt (C. B. Fleet Co.), a high protein additive (*5*), and with a multivitamin preparation. Ethanol or isocalorically substituted lactose was then added so that 19 percent of the total daily caloric intake was derived from protein, 16 percent from fat, 20 to 65 percent from carbohydrate, and 0 to 45 percent from ethanol. The animals were offered this mixture in baby bottles four to five times daily, depending upon their age. Since the ethanol dose for each animal is dependent upon his acceptance of the liquid diet, we found it necessary to begin by offering ethanol in concentrations as low as 1 percent (weight to volume). We then gradually increased the concentration to as much as 10 percent, resulting in daily doses of from 2 g/kg of body weight initially to as high as 8 g/kg during the weeks prior to withdrawal. At the higher daily doses (above 5 to 7 g/kg) fluctuating concentrations of ethanol in blood were continuously present, reaching diurnal maximums of 250 to 500 mg per 100 ml of blood. Corresponding diurnal minimums of 50 to 300 mg per 100 ml of blood occurred just before the first morning feeding. Since the animals were not fed between 11 p.m. and 8 a.m., a pattern of withdrawal symptoms began to emerge during the early morning hours if the concentration of ethanol in blood decreased to less than 100 to 150 mg per 100 ml of blood. The observed hyperreflexia, mild tremulousness, and irritability were readily reversed when ethanol was administered in the first morning feeding. Thus, the continual presence of ethanol in the blood appears to be an important factor in the development of physical dependence, an observation consistent with other reports (*1, 6, 7*).

By gradually increasing the dosage, the animals only infrequently refused their ethanol-containing diet. When an occasional refusal did occur, however, the ethanol concentration was temporarily reduced, and a normal level of food consumption was reestablished, so that the possibility that the coercive

effect of food deprivation was responsible for the enhancement of ethanol acceptability was minimized. After periods of abstinence from ethanol (3 to 4 weeks), subjects who previously accepted 10 percent solutions of ethanol generally refused initial concentrations above 3 percent, an indication that the enhancement is partially reversible.

After a 6- to 10-week period, ethanol was abruptly removed from the diet, and the animals were studied for symptoms of physical dependence. We followed this procedure with six chimpanzees and observed withdrawal reactions in each case. General symptoms of hyperreflexia and irritability were most pronounced when the concentration of ethanol in the blood approached zero. Concomitant symptoms of photophobia, rapid respiration, sweaty palms and feet, spastic rigidity, and decreased responsiveness to external auditory stimuli were also noted. Severe symptoms, including tonic and clonic convulsions, occurred in three of the subjects, one of whom died after prolonged and repeated seizures. Of these three animals, two had seizures only after ethanol was no longer detectable in the blood. One of them experienced a single convulsion; the other had five convulsive episodes separated by 2- to 3-hour intervals. The third animal experienced a mild seizure even during the final weeks of ethanol administration as the concentration of ethanol in her blood approached its diurnal minimum (124 mg per 100 ml of blood); further development of the withdrawal syndrome was suppressed by administration of ethanol. She became convulsive during withdrawal while a moderate amount of ethanol still remained in the blood (130 mg per 100 ml of blood). The convulsive episodes became prolonged and temporally less discrete after the disappearance of blood ethanol. This animal experienced severe hyperthermia (107.6°F, rectal), extremely rapid respiration (greater than 200 breaths per minute), and tachycardia (greater than 230 beats per minute); she died 32 hours after the final dose of ethanol. A postmortem examination revealed marked congestion and edema of the brain as well as scattered areas of bronchopneumonia.

The withdrawal syndrome demonstrated by the chimpanzee parallels that described for humans by Victor (*8*). In general, seizures in the chimpanzees were of the grand mal type and lasted approximately 4 to 7 minutes. The symptoms exhibited by the animal that died were similar to those that characterize the delirium tremens described by Victor, although we cannot establish the occurrence of hallucinatory behavior in the nonhuman subject. Preliminary data also indicate that, during withdrawal, the chimpanzee exhibits hyperthermia and alkalosis, both of which have been reported to be correlates of the human syndrome of alcohol withdrawal (*8, 9*).

Two primary criteria for establishing addiction of an organism to ethanol are (i) the development of dependence and (ii) the development of tolerance to the drug. In addition to demonstrating the development of physical dependence, we have obtained evidence of metabolic tolerance in chimpanzees after periods of ethanol ingestion. The concentration of ethanol in the blood was determined using a modification of the gas chromatographic procedure described by Roach and Creaven (*10*). Rates of disappearance of ethanol were determined by the method of least squares with values ($n = 4$ to 10) greater than 20 mg per 100 ml of blood (*11*) from the linear descending limb of the blood ethanol curve after peak amounts in excess of 100 to 125 mg per 100 ml of blood. Linear correlation coefficients of these disappearance rates were statistically reliable ($r = .92$ to .99, $P < .01$).

Figure 1 illustrates the relations between the rate of ethanol disappearance, the dose level, and the concentration of ethanol in the blood, as functions of time. Since the chimpanzees gradually accepted increasingly higher concentrations of ethanol, the observed increase in ethanol disappearance rate may be both time- and dose-related. Combining the data from all subjects in Fig. 1, we calculated correlation coefficients for the following combinations of variables: daily dose and time ($r = +.67$, $P < .01$), disappearance rate and time ($r = +.62$, $P < .01$), and disappearance rate and daily dose ($r = +.89$, $P < .001$). Since all correlations were statistically reliable, we analyzed the partial correlations to determine the relation between two of these variables while eliminating the influence of the third (*12*). If the influence of time is statistically eliminated, the partial correlation between daily dose and disappearance rate remains significant ($r = +.81$, $t = 5.44$, $P < .001$). If daily dose is eliminated, however, the partial correlation between disappearance rate and time is no longer statistically significant ($r = +.07$, $t = .27$). This suggests that, within the time period of the experiment, the observed metabolic

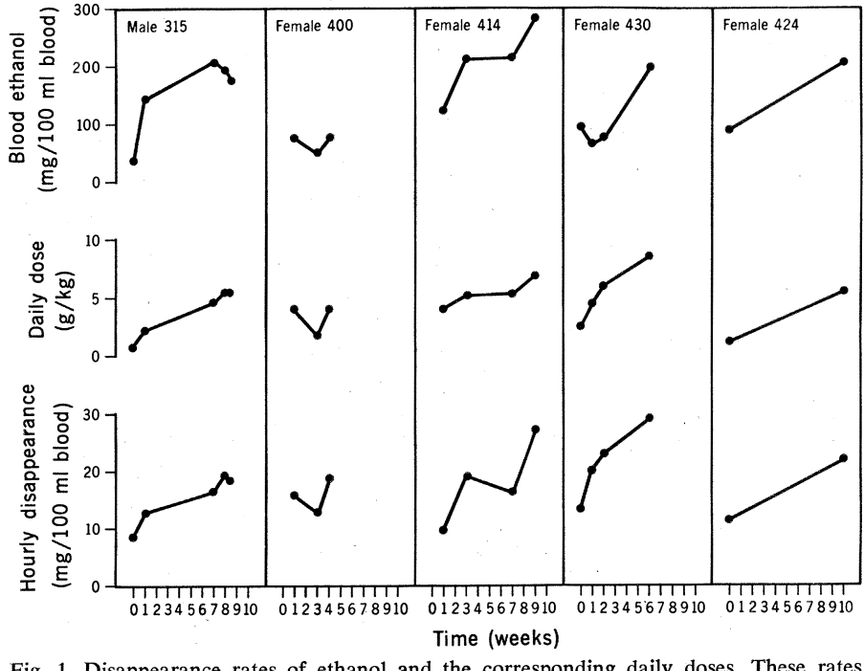

Fig. 1. Disappearance rates of ethanol and the corresponding daily doses. These rates were measured at various times during the first 6 to 10 weeks of ethanol ingestion. Data were not obtained from one subject. Maximum concentration of ethanol in blood from each disappearance curve are included for comparative purposes. Grand mal seizures were observed in females 414, 430, and 424 upon abrupt withdrawal of ethanol.

tolerance is dose-related rather than time-related. The increase in disappearance rate during periods of chronic intake is partially reversed during subsequent periods of abstinence. Of the five surviving animals, four had decreased rates of disappearance of ethanol after periods in which no ethanol was administered. Metabolic tolerance has also been described in man during, and immediately following, chronic ethanol ingestion (*7, 13*); however, no significant differences in disappearance rates were found between alcoholics and nonalcoholics when both groups had abstained from ethanol for at least 3 weeks (*14*). Our observation of partial reversibility of metabolic tolerance in chimpanzees is consistent with those findings.

We have also obtained liver biopsy specimens from each of the chimpanzees both before and after the periods of ethanol administration (*15*). Histologic and ultrastructural examination indicated marked fatty infiltration after chronic ethanol ingestion. Normal hepatocytes were again predominant, however, in biopsies taken after periods of abstinence, an indication that the fatty changes are reversible. Electron microscopy of hepatic tissue revealed increased amounts of smooth endoplasmic reticulum, large fat droplets, and equivocal enlargement of the mitochondria. These changes in chimpanzee liver tissue are consistent with those observed in man after ethanol ingestion (*16*).

The development of alcoholism in humans is probably associated with biological and biochemical processses, as well as with social and behavioral factors. Systematic and controlled experimentation with animal models may elucidate the mechanisms underlying the development of this disease process. The observed similarities between the response of the chimpanzee and that of man to ethanol intake suggest that the chimpanzee may be useful in investigating the etiology of alcoholism.

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Mating Behavior and Life Habits of the Sweet-Bay Silk Moth (Callosamia carolina)

Abstract. *The mating activity of the sweet-bay silk moth* (Callosamia carolina) *was confined to the midday period between 10 a.m. and 3 p.m. The closely related tulip-tree silk moth* (Callosamia angulifera), *with which* Callosamia carolina *is often confused, is entirely nocturnal. Reproductive isolation between these sibling species is most probably geared to these differences in circadian activity. Larvae of* Callosamia carolina *thrived only on the leaves of* Magnolia virginiana, *upon which the species was double-brooded annually.*

The silk moths of the family Saturniidae have fascinated people by their beautiful color patterns, large size, general nocturnal activity, and habit of spinning cocoons composed of silk. The reproductive biology of three silk moths in the genus *Callosamia* has up to now been unclear. The best known of these three is the promethea moth (*C. promethea* Drury), which is broadly distributed over the eastern half of the United States. The other two are the tulip-tree silk moth (*C. angulifera* Walker) and the sweet-bay silk moth (*C. carolina* Jones) whose ranges primarily encompass the South Atlantic and Gulf Coast states. The adult coloration of all three species is remarkably similar, and males and females of each species exhibit a strong sexual dimorphism. Males differ in color from the female and are darker.

Jones (*1*) published the first descriptive note on the sweet-bay, or Carolina, silk moth in 1908, where he described the species as a "variety" of the tulip-tree silk moth (*Callosamia angulifera*). Because of similarities in wing coloration and scale patterns, the sweet-bay silk moth was confused with or lumped with the better known tulip-tree silk moth (*C. angulifera*). Kimball (*2*), in 1965, pointed out some of the morphological features that separate the two species. However, no one has yet reported any features of the life history of *C. carolina* that would set it apart as a species distinct from *C. angulifera.* I conducted a study of *C. carolina* in Florida over an 18-month period in 1970–71 to clarify its relation to the other two species.

A field survey in several parts of central Florida beginning in April 1970 revealed the distribution of *C. carolina.* This search indicated that the species only occurs in or near swamp habitats where the sweet-bay or white-bay tree (*Magnolia virginiana*) is prevalent. Experiments have indicated that this species of tree is the only food plant readily accepted by larvae of *C. carolina* in Florida. Other members of the family Magnoliaceae, including tulip-tree (*Liriodendron tulipifera*) and southern magnolia (*Magnolia grandiflora*), failed to sustain larval growth beyond the first or second instars. A few *C. carolina* from South Carolina have been reared, in captivity, on tulip-tree. However, it has recently been found (*4*) that newly hatched larvae from South Carolina, when placed on tulip-tree, died in the first instar. Thus, it seems doubtful that *C. carolina* ever utilizes the tulip-tree in nature.

Sweet-bay trees tend to occur, in Florida, in high-density stands that are