and mamillary body (Table 1). Pituitary stalk had a larger concentration of hormone. Two additional hypothalamic nuclei and five areas of brain outside the hypothalamus were assayed and did not contain detectable vasopressin. Immunoreactive oxytocin was found only in the hypothalamic areas that contained vasopressin and was present in lesser amounts. Posterior pituitary glands from four subjects were available for assay and contained 5120 ± 1780 ng of vasopressin (mean ± standard error) and 2970 ± 932 ng of oxytocin.

The results reported here are in accord with the concept that both supraoptic and paraventricular nuclei are major sources of both vasopressin and oxytocin in humans. In addition, the results agree with previous findings in the rat (*3*) that both hormones are present in additional hypothalamic areas but not generally throughout the brain. The hormone-containing areas may be part of the neurosecretion system supplying the posterior pituitary or may serve some other brain function (*7*). Hormone in hypothalamic areas could be contained in axons of passage through nuclear areas, be synthesized in multiple hypothalamic nuclei, or be taken up in these areas after being synthesized elsewhere. There are two reasons for believing that I measured immunoreactive vasopressin and oxytocin in human brain tissue rather than a general artifact of human postmortem brain tissue: the hormones were found in some tissue samples but not others, and immunoreactivity in serial dilutions of tissue samples was identical with that obtained with synthetic hormone.

There was no direct correlation between the hormone concentration and the time interval between death and freezing of the brain tissue. Hormone concentration in areas of two brains obtained 3 and 5.5 hours after death were not consistently different from those in two brains obtained at 12 hours after death. Although the results in Table 1 are qualitatively meaningful in delineating hormone-containing areas, the quantitative relationships are suspect because of possible postmortem degeneration. For instance, median eminence had a lesser concentration of vasopressin than either supraoptic or paraventricular nuclei in contrast to previous findings in the rat where median eminence had the highest concentration of any hypothalamic area (*3*).

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Morphine Tolerance: Is There Evidence for a Conditioning Model?

Siegel reports that a Pavlovian interpretation can account for tolerance to the analgesia produced by small doses of morphine (*1*). He shows that animals repeatedly exposed to morphine paired with one environment and test situation show less of an analgesic response to morphine when tested in the presence of those same cues than when tested in the presence of different cues. Thus, he concludes that the presence of stimuli reliably associated with systemic morphine administration is crucial to the development of tolerance to the analgesic effects of morphine. We believe that Siegel's experiments are inconclusive.

Siegel did not distinguish adequately between Pavlovian contingencies and the possibly independent process of behavioral tolerance. The well-documented phenomenon of behavioral tolerance, extensively studied by the "hot plate" test (*2–4*), refers to the fact that powerful interactions can occur between the administration of drugs and the test situations used to evaluate drug effects. Thus, prior experience in the test apparatus is a significant determinant of the amount of tolerance produced by certain drug regimens. For example, animals repeatedly injected with morphine and tested on the hot plate show a greater reduction in analgesia than animals injected with equivalent doses of morphine but tested only once on the hot plate at the end of the injection regimen.

Siegel argues that his work has extended previous findings "by demonstrating that the display of tolerance is specific to the environment in which the drug has been administered, and that 'morphine tolerant' rats, when assessed for the effects of the narcotic in an environment other than that in which they became tolerant, evidence a relatively nontolerant response" (*1*). Yet, in that experiment he did not distinguish exposure to the environmental cues associated with morphine administration from exposure to the test procedures used to evaluate morphine analgesia. That is, when rats were tested in a novel environment to determine whether they became relatively nontolerant, they were also tested with the analgesiometric device with which they had no prior experience, thereby also preventing any manifestation of behavioral tolerance to the test situation. Similarly, when rats were tested in the same environment in which they had previously received the drug in order to determine if they were relatively more tolerant, they were also tested with the analgesiometric device with which they had prior experience. This procedure maximized the chances of observing behavioral tolerance. Therefore, Siegel's experimental design did not distinguish between differences in analgesia attributable either to changes in general environmental cues or to the presence or absence of prior experience with the test apparatus (that is, behavioral tolerance). The experiment shows only that behavioral tolerance to the analgesic effect of morphine can develop after repeated testing with the paw pressure analgesiometer as well as with the hot plate.

In order to provide support for the role of Pavlovian contingencies in the development of behavioral tolerance, one should show that identifiable conditioned stimuli contribute to the reduced analgesia resulting from repeated pairings of morphine administration with the same test situation. One should, for example, show that, in the same animals repeatedly tested on the hot plate, the presentation of one environmental cue associated with repeated morphine administrations [that is, a conditioned stimulus or (CS) (+)] results in a greater decrement in morphine analgesia than the presentation of a different environmental cue associated with repeated saline administra-

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tions [that is, a CS (−)]. An additional consideration in the design of such experiments relates to the inclusion of control groups exposed to nonfunctional analgesiometric devices. While such procedures may control for practice effects (*1*), they do not control for the contribution of behavioral tolerance to a reduction in morphine analgesia, since behavioral tolerance can result even from repeated exposure to a nonfunctional hot plate (*2, 3*).

To argue that cues associated with the test situation were the relevant conditioned stimuli in Siegel's experiment would be to equate the demonstration of Pavlovian conditioning with the demonstration of behavioral tolerance. Yet, there are data which suggest that behavioral tolerance may not be adequately explained by Pavlovian contingencies. Massed trials are more effective than spaced trials in producing behavioral tolerance (*4*), whereas a Pavlovian interpretation would predict the opposite. Moreover, the demonstration of behavioral tolerance, even after exposure to a nonfunctional test apparatus, does not eliminate the possibility that the performance of certain responses emitted in the test situation contributes to behavioral tolerance.

In an earlier report, Siegel showed that rats exhibiting tolerance to morphine after repeated testing on the hot plate also exhibit a hyperalgesic response when given saline instead of a narcotic (*5*). This hyperalgesic response, suggesting the presence of an increased sensitivity to pain, might conceivably represent a compensatory conditioned response that could account for the development of analgesic tolerance. Yet, Siegel has provided no evidence that the presence of this hyperalgesic response is necessary or sufficient (or both) even for the production of behavioral tolerance to the hot plate. Moreover, he has not demonstrated that a hyperalgesic response develops after pairings of morphine administration with testing with the analgesiometer or with the nonfunctional hot plate, manipulations he reported (*1*). Thus, Siegel has presented a Pavlovian model of narcotic tolerance without offering good evidence either for the role of conditioned stimuli (environmental cues) or conditioned responses (hyperalgesia) in the development of tolerance.

The construction of a valid Pavlovian model of narcotic tolerance requires attention to additional issues. First, Siegel did not show that his effects were not restricted to situations characterized by drug-test interactions. All his data are the product of designs that involve repeated testing (*1, 5, 6*). Yet, even in the absence of repeated testing, tolerance to analgesic effects of morphine can be produced by dose regimens quite similar to those used by Siegel (*3*). Thus, questions remain as to the contribution of classical conditioning contingencies to the tolerance observed when procedures that provide no opportunity for behavioral tolerance are used. Second, studies of tolerance should give explicit recognition to the possible development of "pharmacological" or "physiological" tolerance; for example, tolerance which could be attributed neither to Pavlovian training nor to drug-test interactions. Siegel's own data seem to suggest the presence of physiological tolerance (*1*). He reports that an initial dose of morphine in test-naive rats produces approximately a 975-gram analgesiometer withdrawal threshold. Other groups of rats also having no previous exposure to the analgesiometer (exposed only to repeated morphine injections and hot plate testing) show only 245- and 316-g withdrawal thresholds when injected with morphine. Thus it seems that the group repeatedly injected with morphine are tolerant, as compared to animals receiving their first dose of morphine, even though the repeatedly injected groups had never experienced pairings of morphine administration with the analgesiometer test and environment.

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As was first suggested by Dews (*1*), drug-induced behavioral effects may become attenuated over the course of repeated drug administrations because the organism learns a behavioral strategy that compensates for drug-induced impairments. Dews' example illustrates the operation of such behavioral tolerance: The alcoholic learns to adopt a broad-based gait; the experienced drinker does not remain erect because the alcohol is, necessarily, pharmacologically less effective in altering balance. Rather, he has acquired behavior which compensates for an effect of the drug because he has practiced this behavior while drugged. Most discussions of behavioral tolerance have interpreted the concept in this manner (*2, 3*), and have acknowledged that behavioral tolerance is relevant "only if the animal has had certain behavioral experiences under the drug" (*3*, p. 85). Tolerence attributable neither to such acquired behavioral proficiency in coping with drug-induced impairments, nor to traditional pharmacological mechanisms, was described in experiments by Mitchell and his colleagues (*4*), some of which are cited by Hayes and Mayer (*5*) to support their criticisms of the conditioning model of tolerance. Mitchell *et al.* called this new type of tolerance "behavioral tolerance." This appropriation of existing terminology to label a new phenomenon appears to be the source of confusion.

Mitchell's experiments demonstrated that rats (and humans) display greater analgesic tolerance to the last of a series of morphine injections if this final injection (test injection) is conducted in the context of the same environmental cues as the prior injections (pretest injections). That is, same-tested subjects were more tolerant than different-tested subjects. The similarity of environmental cues between pretest and test injections for same-tested subjects was maximized by confronting them with the analgesia-assessment apparatus after all injections, but the apparatus was nonfunctional (that is, the plate was not heated) until the test session. Thus, prior to tolerance assessment, both groups had the same amount of experience in making the analgesia-indicant response (namely, none).

Clearly, in Mitchell's experiments, the greater tolerance of same-tested subjects cannot be due to their greater practice in performing the test response while drugged. Hayes and Mayer are correct in indicating that these earlier experiments demonstrated that tolerance may result from "prior experience with the test apparatus." However, since the apparatus was, for the relevant groups in the cited experiments, nonfunctional, Hayes and Mayer are promulgating a terminological confusion by stating that Mitchell's data (and related findings of my own) may be due to "the well-documented phenome-

non of behavioral tolerance" (*5*). As discussed elsewhere (*6*), such an expanded concept of behavioral tolerance is confusing. To avoid such confusion, it would be appropriate to distinguish the "Mitchell effect" from "behavioral tolerance."

To explain the Mitchell effect, I suggested (*7*) that environmental cues present at the time of pharmacological stimulation (CS) become associated with the systemic effect of the drug (UCS). When the drug is administered in the context of environmental cues that have, in the past, been paired with the drug, drug compensatory conditional responses (CR's) attenuate the effect of the drug and are partially responsible for tolerance. The model is based on many experiments concerning the conditioning of drug effects (*8, 9*).

Hayes and Mayer suggest that the model may be inadequate because "Massed trials are more effective than spaced trials in producing behavioral tolerance . . . whereas a Pavlovian interpretation would predict the opposite." This criticism is unwarranted. Massed and spaced trials in Pavlovian conditioning refer to intertrial intervals of seconds and minutes, respectively (*10*). In the drug tolerance work cited by Hayes and Mayer, the interval between injections was varied over a range of weeks [for example, tolerance was more rapid when successive drug administrations occurred 1 or 2 weeks apart than when they occurred 3 weeks apart (*11*)]. There is no empirical or theoretical justification for the assertion that intertrial intervals of 1 to 2 weeks should lead to poorer Pavlovian conditioning than an intertrial interval of 3 weeks. Moreover, it is not established that tolerance is facilitated by such "massed" trials; indeed, some investigators have reported the opposite effect (*12*).

Hayes and Mayer also state that all my data "are the product of designs that involve repeated testing." They are incorrect. This is a further manifestation of their failure to distinguish between testing the effect of a drug and mere exposure to apparatus that will subsequently be used to test the effect of the drug (behavioral tolerance as opposed to the Mitchell effect).

I do not understand the force of Hayes and Mayer's comments about the adequacy of my published demonstrations of a hyperalgesic CR. It is true that additional research is needed. However, morphine-compensatory CR's have been reported in many experiments (*9*).

Hayes and Mayer suggest that their criticisms of the conditioning model are relevant to my other experiments in the area (*13*). However, they do not explain how any alternative model can explain these demonstrations that a variety of nonpharmacological manipulations, known to be effective in generally affecting the strength of CR's (extinction, partial reinforcement, and CS habituation) similarly affect the display of morphine analgesic tolerance; nor do they recognize that the results of these experiments show that mere exposure to the test apparatus may either facilitate or hinder the development of tolerance, in a manner readily predictable by the conditioning model, but not by alternative formulations. Also, work from other laboratories indicates that many procedures which are effective in retarding (*14*) or facilitating (*15*) morphine tolerance similarly affect conditioning.

One design of an experiment that Hayes and Mayer find appropriate for assessing the conditioning model of tolerance has, in fact, been completed (*16*). Furthermore, behavioral tolerance interpretations of this experiment are especially implausible; the role of environmental cues in morphine tolerance, and the existence of a compensatory CR, are demonstrated with a "nonbehavioral" effect of the drug (temperature alteration). Additional recent work has demonstrated that the Mitchell effect does not depend on having the analgesiometric test apparatus as part of the pretest administration environment. An arbitrary audio and visual cue can serve equally well as a CS for the elicitation of a tolerant response (*17*); thus any criticism of the conditioning model which emphasizes the effects of pretest experience with the test apparatus cannot be relevant in interpreting the results of this experiment.

Where appropriate, I have acknowledged the role of pharmacological mechanisms of a nonassociative nature, as well as of associative factors in studies of tolerance (*9, 16*), just as the contribution of learning to tolerance has been acknowledged by others who approached the issue from a pharmacological perspective (*18*).

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Sympatric Speciation: Evidence?

The claims made by Tauber and Tauber (*1*) seem to exceed the information which can be derived from the facts given. From reading the title and abstract, I was led to believe that they had evidence indicating that sympatric speciation must have occurred to account for the existence of *Chrysopa carnea* and *Chrysopa downesi*. However, their data merely indicate that it is possible to interpret this speciation event as having occurred without the necessity of geographic isolation. The evidence does not refute the equally plausible hypothesis that geographic isolation could account for the same speciation event.

The allelic differences at the three loci described explain why these two popu-

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