

CONDITIONED NALORPHINE-INDUCED
ABSTINENCE CHANGES: PERSISTENCE
IN POST MORPHINE-DEPENDENT MONKEYS¹

STEVEN R. GOLDBERG² and CHARLES R. SCHUSTER³

THE UNIVERSITY OF MICHIGAN MEDICAL SCHOOL

Every tenth lever-press of three morphine-dependent rhesus monkeys was reinforced with food. A red light, initially a neutral stimulus, was presented every third or fourth session for 5 min before and 5 min after an intravenous injection of nalorphine, a morphine antagonist that produces an immediate abstinence syndrome in morphine-dependent monkeys. After several pairings, conditioned suppression of lever pressing, heart-rate decrease, vomiting, and excessive salivation were observed during the red-light period before nalorphine injection. No conditioned electrocardiogram, respiration or temperature changes occurred. After 10 red light-nalorphine pairings, morphine administration was completely discontinued and monkeys were then tested monthly for persistence of the conditioned responses. The red light paired with saline injection continued to suppress lever pressing and to produce heart-rate decreases after 60 to 120 days of complete abstinence from morphine. Subsequently, daily presentations of the red light-saline injection complex rapidly extinguished these conditioned responses. Nevertheless, they could be rapidly reinstated by additional nalorphine injections.

After a period of chronic morphine administration, failure to continue periodic administration of drug to an organism results in severe physiological and behavioral disturbances several hours after the last dose of drug. This complex of signs and symptoms, termed the morphine abstinence syndrome, indicates that the organism has become physically dependent on the drug (Tatum, SeEVERS, and COLLINS, 1929; SeEVERS and DENEAU, 1963). It has been frequently suggested that conditioning factors arising during periods of abstinence distress may contribute to the perpetuation of drug-taking behavior. For example, IRWIN and SeEVERS (1956), WIKLER (1965), and GOLDBERG and SCHUSTER (1967) found that certain aspects of the morphine abstinence syndrome can be classically conditioned. Further, GOLDBERG, WOODS, and SCHUSTER (1969) showed that stim-

uli paired with the morphine abstinence syndrome can produce large conditioned increases in morphine self-administration responding. To allow rapid manipulation of the morphine-dependent state in the majority of the preceding studies, the abstinence syndrome was precipitated by the administration of a morphine antagonist, nalorphine, rather than by discontinuing morphine administration. The intravenous injection of this narcotic antagonist in a morphine-dependent organism precipitates within seconds an acute abstinence syndrome that lasts for several hours (WOODS, 1956).

Previous experiments (GOLDBERG and SCHUSTER, 1967, 1969) have shown that intravenous injections of low nalorphine doses (0.1 to 1.6 mg/kg) fail to produce either observable physiological effects or changes in food-reinforced fixed-ratio responding in non-dependent rhesus monkeys. In monkeys physically dependent on morphine, however, intravenous injection of a low nalorphine dose (approximately 0.2 mg/kg) produces vomiting (emesis), excessive salivation, heart-rate change, and complete suppression of food-reinforced fixed-ratio responding (GOLDBERG and SCHUSTER, 1967). In the latter experiment, an initially neutral stimulus (tone) acquired the ability to produce conditioned emesis, salivation, heart rate deceleration, and sup-

¹This paper is based on a dissertation submitted to the Graduate School of The University of Michigan by the senior author in partial fulfillment of the requirements for the Ph.D. degree. A preliminary report of this work was presented at the Spring Meeting of the Federation of American Societies for Experimental Biology, 1969. *Federation Proceedings*, 1969, 28, 512. The work was supported by NIMH grant 5R-10MH-12084.

²Reprints may be obtained from Steven R. Goldberg, Department of Pharmacology, Harvard Medical School, 25 Shattuck St., Boston, Massachusetts 02115.

³Now at Department of Psychiatry, University of Chicago Medical School, Chicago, Illinois 60637.

pression of food-reinforced fixed-ratio responding after several pairings with intravenous injections of this low nalorphine dose. The present experiment utilized a similar conditioning procedure in additional physically dependent monkeys while monitoring respiration rate, respiration amplitude, temperature, and electrocardiogram (EKG), in addition to heart rate. To determine whether conditioned changes acquired in this manner might be dependent on the drug state of these animals, the persistence of conditioned changes was studied following long periods of complete abstinence from morphine. Finally, the effect of nalorphine on post morphine-dependent subjects and its ability to reinstate conditioning in the absence of physical dependence were explored.

METHOD

Subjects

Four male and two female adult rhesus monkeys, weighing between 3.7 and 5.8 kg, with no previous experimental history served; three of them (M2113, M2115, and M2116) had been physically dependent on morphine for approximately three months before the start of the experiment. During this time, they were maintained on 8 mg/kg a day of morphine sulfate, given as a subcutaneous injection of 2 mg/kg every 6 hr. A lower maintenance dose of morphine than that used in previous work (12 mg/kg a day; Goldberg and Schuster, 1967) was chosen for these subjects to diminish health problems during subsequent termination of the morphine regimen. The remaining three subjects (M2126, M2195, and M2196) had been maintained for two months on a dose regimen of 12 mg/kg a day of morphine sulfate, given as a subcutaneous injection of 3 mg/kg every 6 hr. Before the start of the experiment, morphine administration was discontinued for monkeys M2126, M2195, and M2196 and they were then allowed a 60-day period of abstinence from morphine.

All monkeys were surgically prepared with chronic jugular catheters (Schuster and Brady, 1964). In addition, monkeys M2113, M2115 and M2116 had a chronic 18-gauge stainless steel needle with a sealed tip stereotaxically implanted in the area of the preoptic nucleus. Thermistor probes placed within the implanted needle were then used to measure temperature.

All subjects were reduced to 85% of their free-feeding weights immediately before the experiment and trained to press a lever under a fixed-ratio 10 schedule of food reinforcement (every tenth response was reinforced with a pellet of food; FR 10).

Apparatus

Experimental sessions were conducted once a day during which monkeys were kept in Plexiglas restraining chairs (similar to Foringer Primate Cockpits, cat. #1206 M1) enclosed in sound-attenuating isolation booths. The remainder of the time they were maintained in community cages containing running water and wood-shavings litter, but no food. The Plexiglas chairs were equipped with a mouth-operated food lever (Thompson, Schuster, Dockens, and Lee, 1964). Dietrich and Gambrell 0.7-g monkey food pellets (Foringer Cat. #1281) were presented by a Foringer pellet dispenser. A stimulus light panel was mounted on the ceiling of the isolation booth. During the experimental session, subjects could be observed through a wide-angle viewing lens. A white noise generator, sufficient to mask equipment noise for the experimenter, operated continuously during all experimental sessions. Injections were administered from outside the isolation booth by syringes connected by a polyethylene catheter to the chronic jugular catheter in the monkey. Nalorphine injection solutions were prepared daily by adding nalorphine HCl to 1.0 cc of physiological saline. Physiological saline solution (0.9%) was used for saline injections (1.0 cc) and to flush out the catheter after each injection. Wound clips attached to the area of the right shoulder and left waist of the six monkeys served as electrocardiogram leads and connecting leads led from the isolation booth to a Grass Polygraph. A Yellow Springs Instrument Co. Thermistor Probe, placed within the implanted needle of monkeys M2113, M2115, and M2116, served as a temperature transducer and its leads were also run from the isolation booth to the Grass Polygraph. Finally, to monitor the respiration of monkeys M2113, M2115, and M2116, the skin on either side of the chest was punctured with Grass needle electrodes (to measure changes in resistance), the electrodes taped in place, and the leads run to the Grass Polygraph. Apparatus in the isolation booth was connected by cables to auto-

matic programming as well as recording equipment.

Procedure

Monkeys M2113, M2115, and M2116 were tested once a day, 1 to 2 hr after a morphine injection. Initially, they were trained to press the lever under an FR 10 schedule of food reinforcement. After surgical implantation of jugular catheters, the monkeys were placed on a 2-hr, three-component schedule of reinforcement, in which the first two components were chained. The schedule continued unchanged throughout the experiment. Each 2-hr session was started by a 30-min fixed-interval (FI 30-min) component which was followed by a 60-min FR-10 food component. The session ended with a 30-min S^A component, which allowed time for abstinence irritability to subside before handling the monkeys. On this schedule, the monkeys' first response on the lever after 30 min produced a stimulus in the presence of which every tenth response was reinforced with a pellet of food. At the end of 60 min of the FR-10 period, they were advanced into the 30-min S^A period during which responses had no consequences, after which they were removed from the situation. During the FI period, the discriminative stimuli were two 6-w, 110-v blue lights; during the FR-10 food period, they were two 6-w, 110-v white lights. A houselight was illuminated throughout the session. Water was continuously available during the session. Lever-press responses, heart rate, EKG, temperature, respiratory rate, and respiratory amplitude were monitored during all test sessions.

After stabilization of lever-press response rate on this schedule, a visual stimulus (25-w, 110-v, red light) was presented every third or fourth session. The red light was presented 10 to 30 min after the start of the FR-10 food component for 5 min before and after an intravenous injection of 1.0 cc of saline through the jugular catheter. During several adaptation sessions, neither the red light nor the injection procedure disrupted the monkeys' FR response rate, heart rate, respiratory rate, respiratory amplitude, or temperature, thus establishing the red light and saline injection (L+S) procedure as neutral stimuli. Conditioning training was then begun, during which the red light was presented every third or fourth session, 5 min before and after an intravenous

injection of 0.2 mg/kg (1.0 cc) of nalorphine. This dose of nalorphine was established as an optimal dose for conditioning in a previous study (Goldberg and Schuster, 1967). Two or three control sessions with no light or injection presentations were interspersed between conditioning sessions with red light-nalorphine injection (L+N) pairings. After 10 conditioning sessions, monkeys were returned to their home cage and the morphine administration regimen was abruptly and completely discontinued (withdrawal). During the first weeks of morphine abstinence, experimental sessions were not conducted and the monkeys were allowed continuous access to food and water. At the end of the third week of abstinence from morphine, they were once again deprived of food and daily control sessions were started after they were reduced to 85% of their free-feeding weights. On the thirtieth day of abstinence, a session was conducted in which an L+S pairing was presented during the FR-10 food component. Following this initial test for the persistence of the conditioned response, L+S pairings were presented during the FR-10 food component once every 30 days. On all other days control sessions were conducted. This was continued until the conditioned suppression of lever-press responding began to disappear, or until the one hundred twentieth day (4 months) after the chronic morphine regimen was terminated. At this time, daily sessions with L+S pairings were conducted until the conditioned response to both the red light and saline injection was extinguished. Sessions with L+N pairings were then conducted, every third or fourth day, to determine whether the conditioned response could be reinstated by nalorphine injections following a prolonged period of morphine abstinence.

The influence of past experience with nalorphine-induced abstinence on the responses of monkeys M2115 and M2116 to nalorphine during reconditioning was explored in a control experiment. The subjects were three monkeys (M2126, M2195, and M2196) that had been completely abstinent from morphine for 60 days, following a two-month maintenance period on 12 mg/kg a day of morphine sulfate. These monkeys had never experienced nalorphine-induced abstinence while dependent on morphine. They worked on a schedule identical to that described in this experiment. Four sessions were conducted in which a saline in-

jection was given through the catheter during the FR-10 food component. After these saline control sessions, injections of 0.2 mg/kg of nalorphine were tested during the FR-10 food component. If this dose of nalorphine produced changes in food-reinforced fixed-ratio behavior or heart rate, it was repeated daily to determine whether the effect would disappear with repeated presentations. Respiration and temperature were not monitored.

The results presented in this report focus on the FR-10 food component because no systematic changes in FI response rate were observed during the experiments.

RESULTS

Conditioning, persistence, extinction and reconditioning. Figure 1 shows cumulative response records of selected sessions for M2113, M2115, and M2116. More prolonged pausing after reinforcement was found in control sessions before conditioning training than is usual on this type of schedule, but performance was stable over sessions. Session 5 was the

last of the adaptation trials with L+S presentations. No disruption of the monkeys' FR responding was produced by either the red light or the injection of saline. In Session 6, the first conditioning trial with a L+N presentation, the monkeys responded normally during the 5-min red-light period before the nalorphine injection. After the injection, responding was abruptly suppressed and continued so for the rest of the session. Emesis and excessive salivation, indicated by arrows in Fig. 1, were observed in the three monkeys within 1 min after the nalorphine injection. In Session 15, the tenth conditioning trial, the conditioned suppression of lever-press responding observed in Sessions 8 to 15 was demonstrated during the 5-min red-light period before the nalorphine injection. Conditioned emesis and excessive salivation were also observed with M2113 after the onset of the red light, before the injection of nalorphine. Monkeys M2115 and M2116 showed emesis and excessive salivation only after the injection of nalorphine.

In the first of the two or three control sessions that followed the fifth, sixth, and eighth

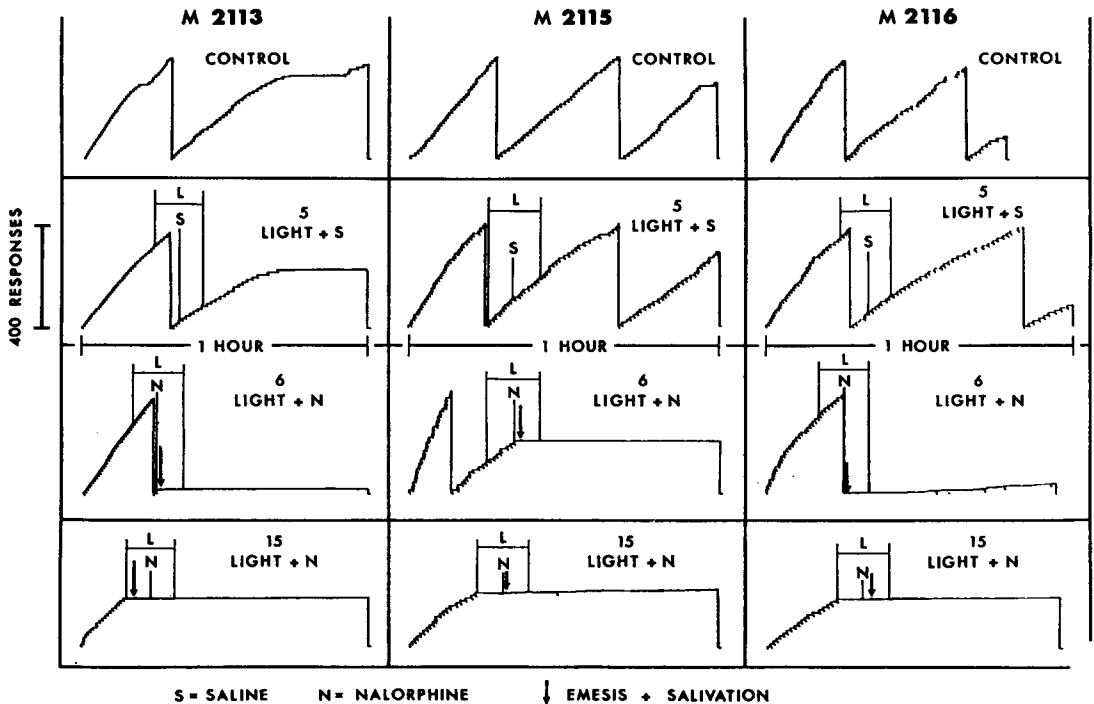


Fig. 1. Cumulative response curves from M2113, M2115, and M2116. Each segment shown is a complete FR-10 food-component record extracted from a 2-hr session. A control session before light-injection pairings is shown first; 5 was a session establishing light and saline injection (L + S) as neutral stimuli; 6 was the first conditioning session with a light-nalorphine injection presentation (L+N); 15 was the tenth conditioning session. Arrows indicate observation of emesis and salivation.

conditioning sessions, M2113 and M2116 exhibited what appeared to be conditioned responses, though neither the red light nor an injection was presented. This response can be seen in Fig. 2. During Session 1, the last control session after the fifth conditioning session, no disruption of responding occurred. In Session 2, the first control session following the sixth conditioning session with a L+N presentation, both monkeys showed suppression of lever-press responding. Emesis, salivation, and a large fall in heart rate were also observed with M2113 at this time. By the following control session (Session 3) these responses were no longer seen. Monkey M2115 failed to show either disruption of lever-press responding or physiological changes during any of the control sessions.

Following Session 15, the chronic morphine regimen of the three monkeys was discontinued. The health of M2113 declined at this time and it failed to survive the first two weeks of abstinence. Monkeys M2115 and M2116 were tested with L+S presentations at

30-day intervals after the morphine regimen was terminated. The results of these presentations are shown in Fig. 3. Session 16 is the first session with a L+S presentation, 30 days after chronic morphine administration ceased. The monkeys responded normally for food until the onset of the red light. Lever-press responding was then almost completely suppressed and remained so until several minutes after the red-light period ended. Session 17 is the second session with a L+S presentation conducted 60 days after chronic morphine treatment ceased. Response rate of M2116 was completely suppressed during the entire red-light period. Response rate of M2115 was not suppressed during the red-light period until 2 min before the saline injection. It was then almost completely suppressed until the injection of saline, and remained suppressed throughout the remainder of the red-light period. Following Session 17, daily extinction sessions with L+S presentations were begun with M2115. Test L+S presentations were continued at 30-day intervals with M2116, until

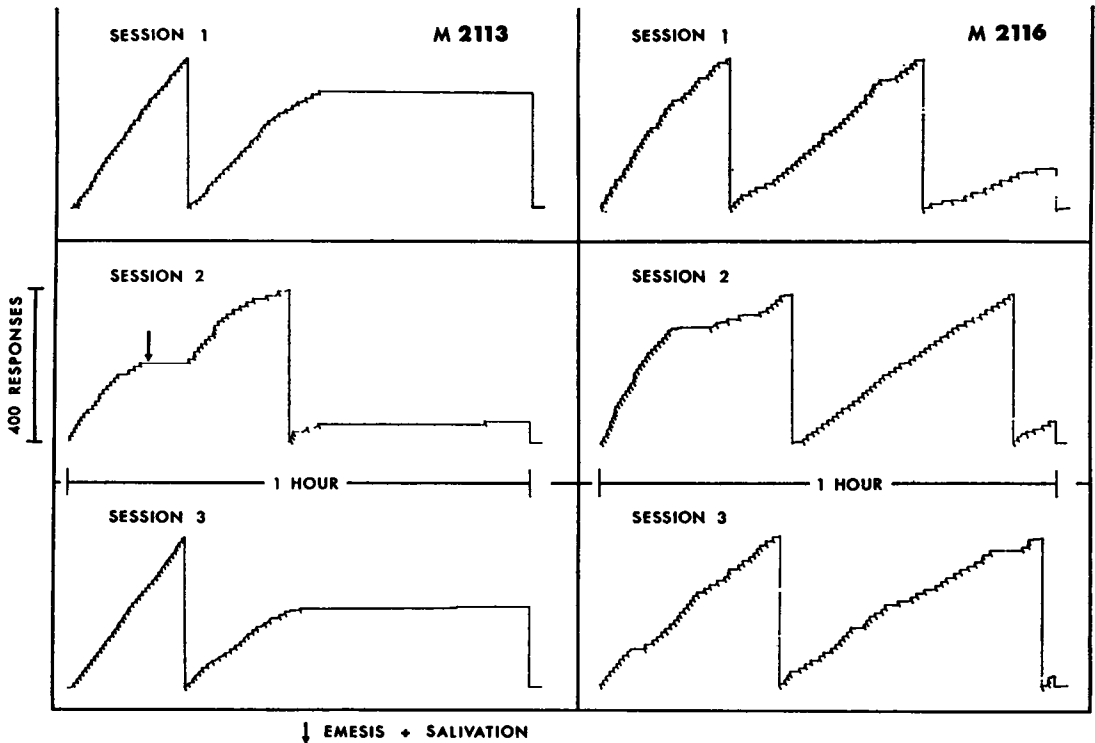


Fig. 2. Cumulative response curves from M2113 and M2116. Each segment shown is a complete FR-10 food-component record extracted from a 2-hr session. Session 1 is the last control session after the fifth conditioning session; 2 was the first control session following the sixth conditioning session; 3 was the second control session following the sixth conditioning session. Arrows indicate observation of emesis and salivation.

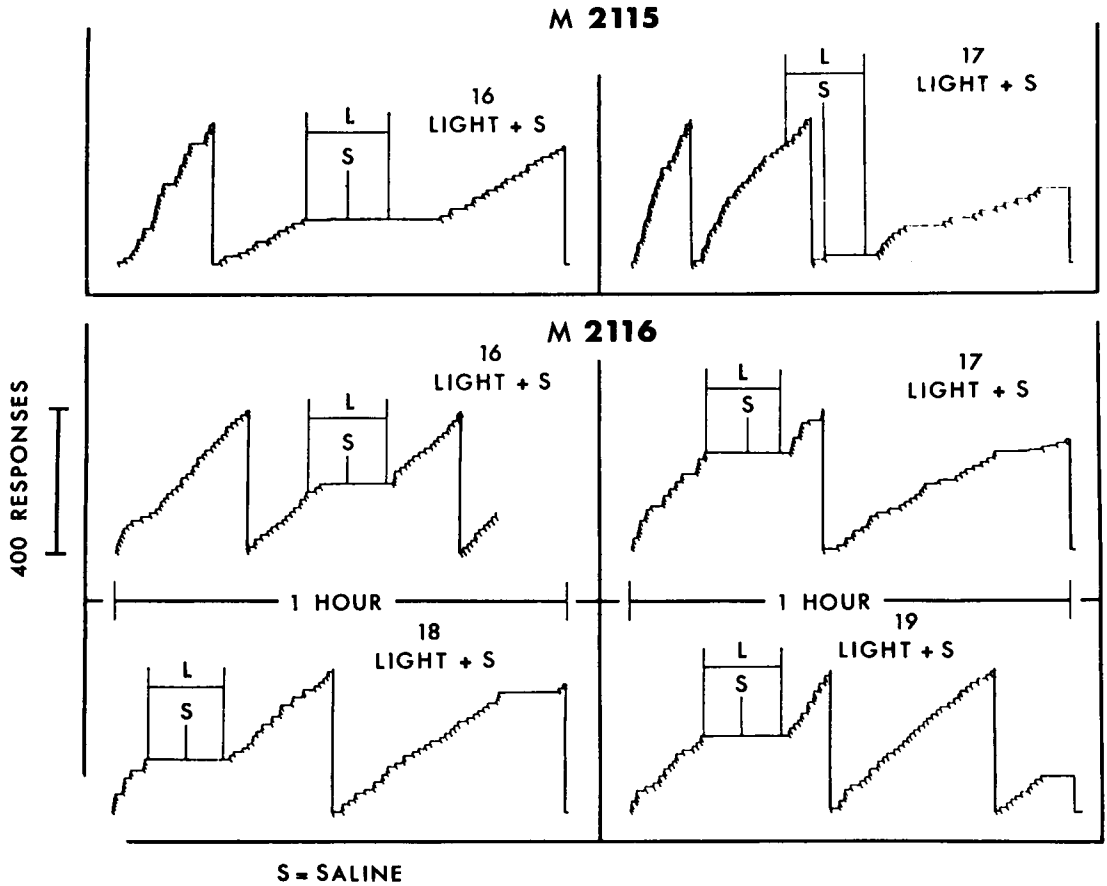


Fig. 3. Cumulative response curves from M2115 and M2116. Each segment shown is a complete FR-10 food-component record extracted from a 2-hr session. Sessions 16 to 19 were monthly sessions, following a 30-day period of abstinence from morphine, which tested the persistence of the light and saline injection (L+S) as conditioned stimuli.

120 days after chronic morphine treatment ceased. Session 18 is the third test session with a L+S presentation conducted after 90 days of abstinence from morphine, and Session 19 is the fourth test session with a L+S presentation conducted after 120 days of abstinence from morphine. In both sessions, lever-press responding was suppressed during the entire red-light period. Following Session 19, daily extinction sessions with L+S presentations were begun with M2116.

After extinction of the conditioned response to the red light and saline injection, sessions with L+N presentations were conducted every third or fourth day. Figure 4 shows selected cumulative response records from M2115 and M2116. Session 28 (M2115) and Session 25 (M2116) were the final extinction sessions with L+S presentations. The L+S presentation pro-

duced no change in the monkeys' FR response rate compared to control sessions. On the control session, after the final extinction session with a L+S presentation, responding was stable and response rate was at normal levels, indicating that the conditioning and extinction procedure had not interfered with the monkeys' baseline FR performance. Session 29 (M2115) and Session 26 (M2116) were the first reconditioning sessions with a L+N presentation after the extinction sessions. The monkeys responded normally during the red-light period before the injection. After the nalorphine injection, the responding of both monkeys was almost completely suppressed for the remainder of the session and emesis and excessive salivation were observed. Session 31 (M2115) and Session 28 (M2116) were the third reconditioning sessions. These sessions show the re-

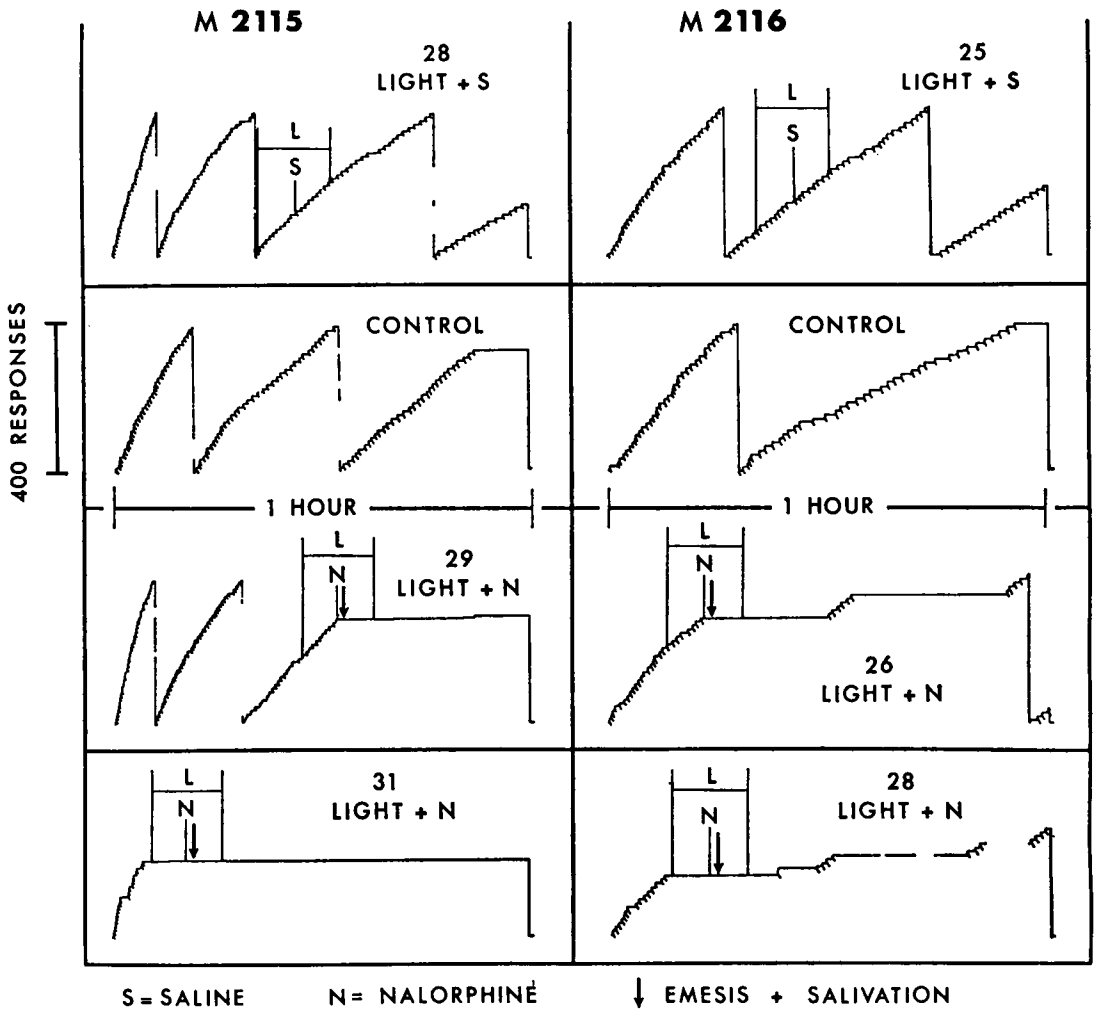


Fig. 4. Cumulative response curves from M2115 and M2116. Each segment shown is a complete FR-10 food-component record extracted from a 2-hr session. Session 28 for M2115 and 25 for M2116 was the final extinction session with a light-saline injection presentation (L+S); a control session prior to reconditioning sessions is shown next; Session 29 for M2115 and 26 for M2116 was the first reconditioning session with a light-nalorphine presentation (L+N); Session 31 for M2115 and 28 for M2116 was the third reconditioning session. Arrows indicate observation of emesis and salivation.

instatement of conditioned suppression of lever-press responding during the red-light period before injection. Emesis and excessive salivation were observed in both monkeys only after the nalorphine injection.

Figures 5, 6, and 7 show the development, persistence, extinction, and reconditioning of the behavioral and physiological responses in M2113, M2115, and M2116. The percentage change in heart rate and FR response rate from the 5-min period before the red-light onset to the 5-min period during the red-light presentation, before the injection of nalor-

phine or saline, is shown in Fig. 5 for M2113, in Fig. 6 for M2115, and in Fig. 7 for M2116.

The first five sessions established the red light and injection of saline as neutral stimuli. After Session 6, the first conditioning session, FR response rate during the red light decreased and was almost completely suppressed by Session 8 for M2113, by Session 10 for M2115, and by Session 12 for M2116.

In Sessions 9 to 15 for M2113, and 11 to 15 for M2115, heart rate declined during the 5-min red-light period before the nalorphine injection. Tachograph recordings of beat-to-

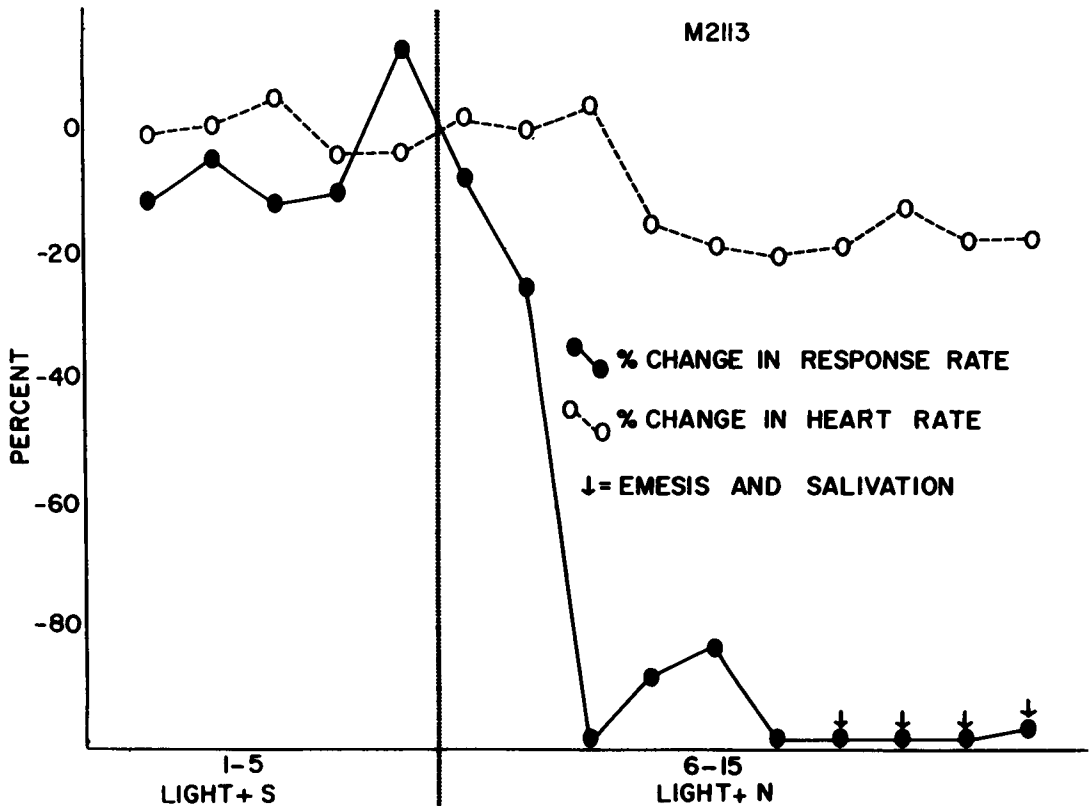


Fig. 5. M2113. Percentage change in heart rate and food-reinforced FR response rate from the 5-min period preceding the light onset to the 5-min period during the light illumination and before injection. Numerals 1 to 5 designate sessions establishing the light and saline (S) injection as neutral stimuli; 6 to 15 were the conditioning sessions during which nalorphine (N) was administered. Arrows mark sessions when emesis and salivation were observed in the light period before injection of saline or nalorphine. Each conditioning session was followed by two to three control sessions not indicated on the graph.

beat heart rate during this 5-min period failed to show any increases in rate. The decreases in heart rate in M2113 and M2115 were slow in onset with maximal decreases occurring in the last 2 min of the red-light period before the injection of nalorphine. Conditioned heart rate changes developed more slowly than the conditioned suppression of food-lever responding. No heart rate changes were observed in M2116 during the red-light period before the injections of nalorphine or saline. After the injection of nalorphine, heart rate of all three monkeys rose from a normal FR-10 food period range of 180 to 210 beats per minute to 240 to 260 beats per minute.

Emesis and excessive salivation were observed in M2113 during the red-light period preceding the injection of nalorphine in Sessions 12 to 15 (Fig. 5). Conditioning of emesis and salivation developed more slowly than

that of either food-response suppression or heart rate decreases. No emesis or excessive salivation was observed in M2115 and M2116 during the red-light periods preceding any of the injections of nalorphine or saline. In the 5-min period after the injection of nalorphine, all three monkeys showed emesis, excessive salivation, an increase in respiratory rate from a range of 15 to 25 per min to 40 to 60 per min, a decrease in respiratory amplitude of approximately 50%, and an average fall in temperature of 1° C from a normal FR-10 food-period level of 38° C to 39° C. No changes in respiration or temperature were observed in any of the monkeys, except after nalorphine injections. At no time in the experiment was any change seen in the shape of the EKG.

Sessions 16 and 17 for M2115 (Fig. 6) and Sessions 16 through 19 for M2116 (Fig. 7) were test sessions with L+S presentations conducted

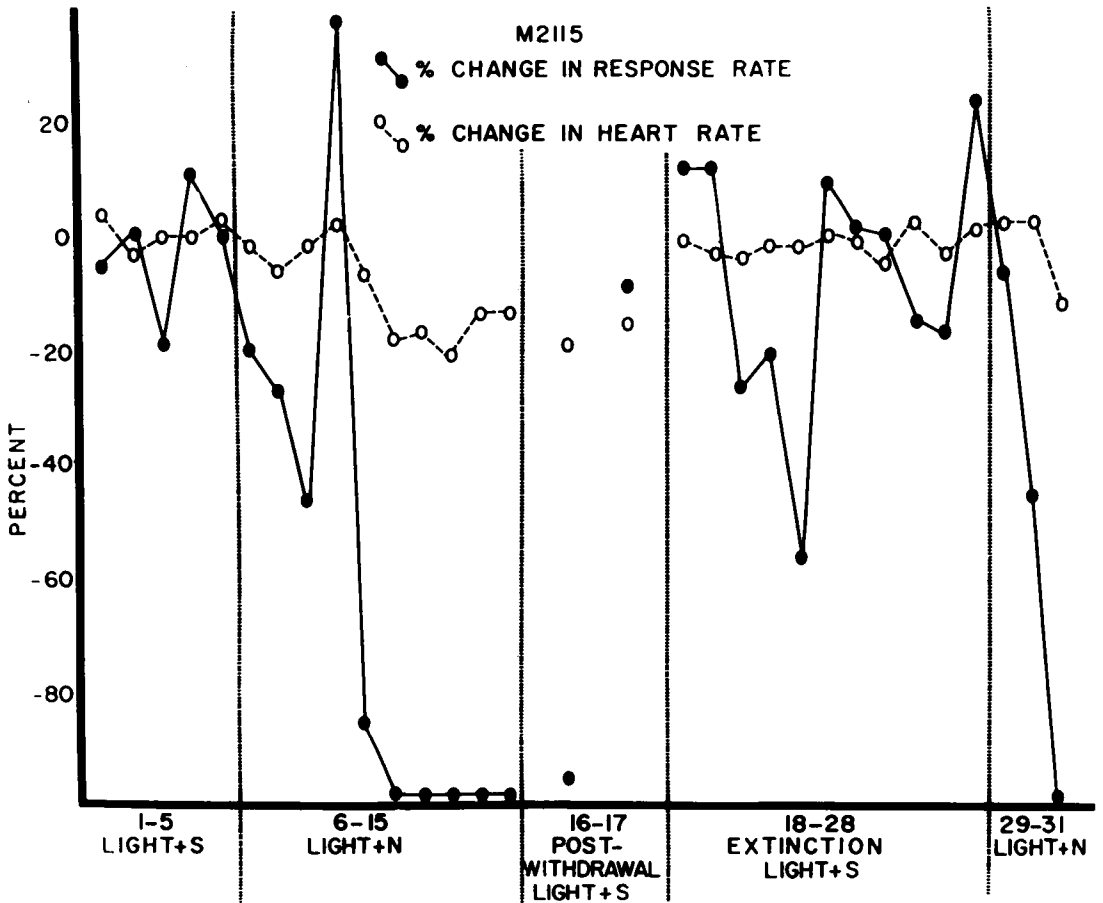


Fig. 6. M2115. Percentage change in heart rate and food-reinforced FR response rate from the 5-min period preceding the light onset to the 5-min period during the light illumination and before injection. Numerals 1 to 5 designate sessions establishing the light and saline (S) injection as neutral stimuli; 6 to 15 were the conditioning sessions during which nalorphine (N) was administered. After Session 15, the tenth conditioning session, chronic morphine treatment was discontinued (withdrawal); 16 and 17 were post-withdrawal sessions with light-saline injection presentations, conducted on the thirtieth and sixtieth day of morphine abstinence; 18 to 28 were daily extinction sessions with light-saline injection presentations; 29 to 31 were reconditioning sessions with light-nalorphine injection presentations. Each conditioning and reconditioning session was followed by two to three control sessions not indicated on the graph.

at 30-day intervals after morphine treatment (post-withdrawal) ceased. In Session 16, after 30 days of abstinence from morphine, M2115 showed suppression of FR response rate and a fall in heart rate during the red-light period before and after the saline injection. In Session 17, after 60 days of abstinence from morphine, heart rate decreases were still seen with M2115, but FR response rate in the 5-min red-light period before saline injection was at normal control levels. The FR response rate of M2116 remained almost completely suppressed throughout the red-light period in Sessions 16 through 19, test sessions with L+S presenta-

tions, conducted after 30, 60, 90, and 120 days of abstinence from morphine.

In extinction sessions 18 to 28 for M2115 (Fig. 6), and 20 to 25 for M2116 (Fig. 7), the FR response rate and heart rate during the red light rose to a level approximating that observed in the initial sessions with L+S presentations (1 to 5). Reconditioning sessions with L+N presentations were then conducted and results closely paralleled those in the initial conditioning sessions. Additional reconditioning sessions with L+N pairings were not conducted because of problems with the jugular catheters of both animals.

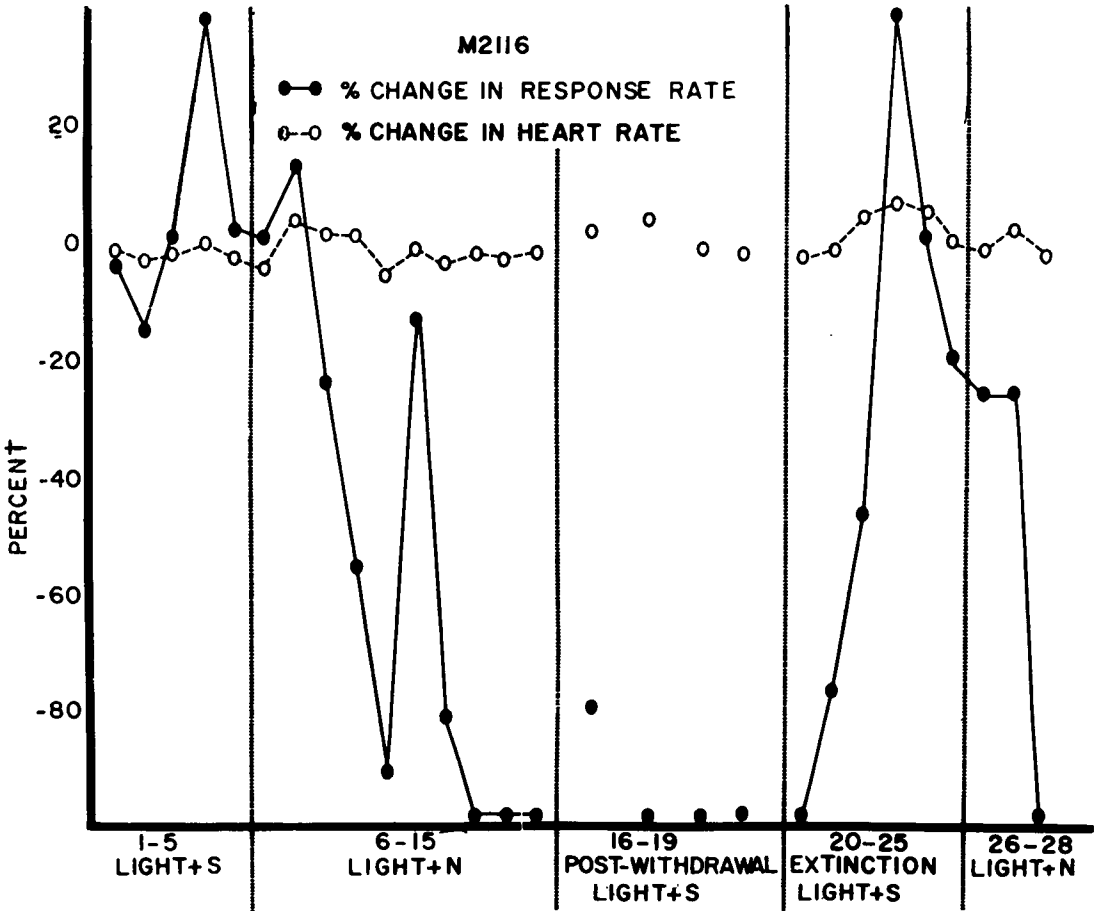


Fig. 7. M2116. Percentage change in heart rate and food-reinforced FR response rate from the 5-min period preceding the light onset to the 5-min period during the light illumination and before injection. Numerals 1 to 5 designate sessions establishing the light and saline (S) injection as neutral stimuli; 6 to 15 were the conditioning sessions during which nalorphine (N) was administered. After Session 15, the tenth conditioning session, chronic morphine treatment was discontinued (withdrawal); 16, 17, 18, and 19 were post-withdrawal sessions with light-saline injection presentations, conducted on the thirtieth, sixtieth, ninetieth, and one hundred twentieth day of morphine abstinence; 20 to 25 were daily extinction sessions with light-saline injection presentations; 26 to 28 were reconditioning sessions with light-nalorphine injection presentations. Each conditioning and reconditioning session was followed by two to three control sessions not indicated on the graph.

Nalorphine sensitivity of post-dependent monkeys. Figure 8 shows cumulative response records for the three monkeys (M2126, M2195, and M2196) that were tested with nalorphine 60 days after chronic morphine treatment ceased. These monkeys had never received nalorphine while dependent on morphine. The control session to the left in Fig. 8 was the last day of the four sessions in which saline was given during the FR-10 food period. Saline injections did not disrupt the FR lever-press responding of the three monkeys. The session to the right in Fig. 8 was the first ses-

sion with an injection of 0.2 mg/kg nalorphine. The nalorphine injections immediately suppressed FR response rates of all three monkeys. This suppression lasted only 4 to 8 min for M2195 and M2196. Monkey M2126 continued to show suppression of responding the remainder of the session. No emesis, excessive salivation, or heart rate changes were seen after the injections of nalorphine. After several repetitions of this dose of nalorphine, the suppression of responding in all three of the monkeys continued to be seen.

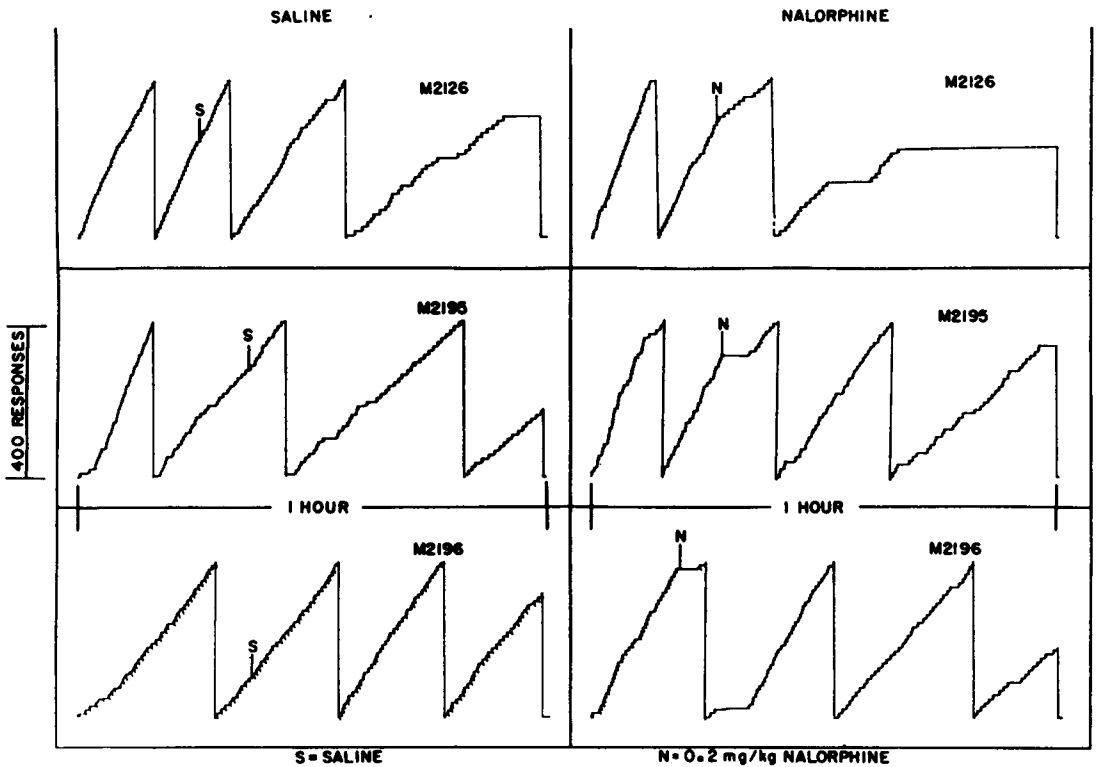


Fig. 8. Cumulative response records from post morphine-dependent monkeys M2126, M2195, and M2196. Each segment shown is a complete FR-10 food-component record extracted from a 2-hr session. Sessions to the left are control sessions with saline (S) injections before test sessions with nalorphine (N) injections. The sessions to the right are the first test sessions with injections of nalorphine.

DISCUSSION

Intravenous injection of 0.2 mg/kg nalorphine to morphine-dependent rhesus monkeys produced suppression of food-reinforced fixed-ratio responding, heart rate increase, respiratory rate increase, respiratory amplitude decrease, emesis, salivation, and a fall in brain temperature. After several pairings with nalorphine injections, initially neutral stimuli (red light and injection procedure) acquired the ability to elicit conditioned suppression of fixed-ratio responding for food, emesis, salivation, and heart rate decrease, confirming previous findings (Goldberg and Schuster, 1967). Although respiration and temperature changes were prominent features of the response to nalorphine by the morphine-dependent monkeys, no conditioned respiration or temperature changes were observed. Therefore, it was not the nalorphine-induced abstinence syndrome that was conditioned, but rather, certain aspects of this syndrome.

The finding in the present experiment, that the heart rate response to nalorphine was always an increase while the conditioned response was a decrease, is consistent with previous findings of Goldberg and Schuster (1967) and with those of investigators using a similar paradigm with electric shock in monkeys (Brady, 1967) and in rats (deToledo and Black, 1966; Parrish, 1967). Stebbins and Smith (1964) and Nathan and Smith (1968) reported conditioned heart rate increases, rather than decreases, to a stimulus preceding electric shock in monkeys. These inconsistencies in direction of heart rate change might be explained by the recent finding of Brady, Kelly, and Plumlee (1969) that monkeys pass through two phases during acquisition of conditioned heart rate change: an initial phase of heart rate deceleration with no accompanying blood pressure change, and a final phase of both heart rate and blood pressure increase. Brady *et al.* (1969) also noted that conditioned heart rate changes were acquired less rapidly than conditioned

suppression of food-reinforced lever-press responding. Similar differences in speed of acquisition were found in previous experiments with rats (deToledo and Black, 1966; Parrish, 1967) and in the present experiment with monkeys M2113 and M2115 (Fig. 5 and 6). In addition, conditioned emesis and salivation, seen with M2113, were acquired less rapidly than either suppression of food-reinforced responding or heart rate decrease (Fig. 5). Thus, it appears that conditioning of the different responses proceeds independently, rather than, as previously suggested (Goldberg and Schuster, 1967), conditioned suppression of fixed-ratio responding for food being dependent on conditioning of the physiological signs of the abstinence syndrome.

In the present experiment, stimuli that acquired conditioned properties in the morphine-dependent monkeys retained their ability to elicit conditioned suppression of fixed-ratio responding and heart rate decrease after 60 to 120 days of complete morphine abstinence. Wikler and Pescor (1967) have described similar persistence of a conditioned abstinence sign ("wet-dog shakes") in post-dependent rats. These findings demonstrate that, after acquisition, abstinence-associated conditioning can persist after long periods of morphine deprivation. The conditioned-abstinence responses of post-dependent monkeys M2115 and M2116 in the present experiment rapidly extinguished, however, after several daily presentations of the conditioned stimuli (red light and injection procedure, in the absence of nalorphine (Fig. 6 and 7). This rapid course of extinction is in marked contrast to the earlier results of Goldberg and Schuster (1967), obtained when extinction training was conducted immediately after conditioning training while monkeys were still physically dependent on morphine. Under these conditions, 40 to 45 daily extinction sessions were required to extinguish fully the conditioned responses. In previous behavioral studies, the passage of time between conditioning and extinction training has been shown to have little effect on the course of extinction (Pavlov, 1927; Skinner, 1950). When conditioned responses are acquired under a particular drug state, however, changing the drug state may result in reduced responding, an effect often referred to as "state dependent learning" (Overton, 1968). For example, Belleville (1964) found

that a lever-pressing response in rats, acquired under morphine, extinguished more rapidly under saline than under the same drug state (morphine). Similar effects have been observed with thioridazine in rats using a conditioned suppression procedure similar to that in the present experiment (Heistad and Torres, 1959). Such findings suggest that the change in the drug state of post-dependent monkeys M2115 and M2116 was primarily responsible for the more rapid observed course of extinction than that seen in monkeys maintained dependent on morphine.

The failure of previous studies (Goldberg and Schuster, 1967, 1969) to demonstrate either disruption of fixed-ratio responding for food or physiological responses to intravenous injections of 0.1 to 1.6 mg/kg nalorphine in non-dependent rhesus monkeys, indicates that the actions of 0.2 mg/kg nalorphine injections in the morphine-dependent subjects of the present experiment were based upon their ability to antagonize certain of morphine's effects and produce an immediate abstinence syndrome. After 70 to 125 days of abstinence from morphine, however, injections of 0.2 mg/kg nalorphine continued to produce marked physiological and behavioral responses in M2115 and M2116 and rapidly reinstated conditioned suppression of fixed-ratio responding and conditioned heart-rate deceleration (Fig. 4, 6, and 7). Irwin and Seevers (1956) described similar reinstatement of conditioned responses in post-dependent rhesus monkeys and suggested that this response to nalorphine was a conditioned effect resulting from past experience with nalorphine-induced abstinence. Recent findings that post morphine-dependent rhesus monkeys with no history of nalorphine-induced abstinence show an increased sensitivity to nalorphine when compared to non-dependent monkeys (Goldberg and Schuster, 1969) suggest the alternative explanation that increased nalorphine sensitivity may result from persistence of physiological changes after periods of morphine dependence. This analysis is supported by the present finding that post-dependent monkeys M2126, M2195, and M2196, which had never received nalorphine while dependent on morphine, showed immediate suppression of fixed-ratio responding that lasted several minutes after intravenous injections of 0.2 mg/kg nalorphine (Fig. 8). Nalorphine's effects on these monkeys, how-

ever, was not as marked as on the post-dependent monkeys with prior conditioning training (M2115 and M2116). No emesis or salivation was seen and the fixed-ratio response suppression was more transient. Consequently, the nalorphine sensitivity of M2115 and M2116, after several months of abstinence, is probably related to both the prior conditioning history of these monkeys and the persistence of physiological changes long after the last period of morphine dependence.

Stimuli that acquire conditioned properties as a function of pairing with aversive stimuli elicit markedly different behavior under different environmental conditions (Hunt, 1959). Although stimuli paired with nalorphine injections elicited suppression of fixed-ratio responding for food in the present experiment, this was probably due to a more general effect of disrupting ongoing behavior. For example, if the experimental contingencies are rearranged so that the monkey is engaged in intravenous morphine self-administration behavior, then presentation of conditioned stimuli associated with the nalorphine-induced abstinence syndrome facilitates behavior rather than suppressing it (Goldberg *et al.*, 1969). This would suggest that abstinence-associated conditioning may play an important role in the maintenance of opioid-taking behavior. The implications of conditioning history for the treatment of human opioid abuse have been recently reviewed by Wikler (1965) and Goldberg (1970). The persistence of abstinence-associated conditioning in the post morphine-dependent monkeys of the present experiment suggests a possible mechanism for the relapse of post-dependent subjects to opioid-taking behavior after periods of abstinence.

REFERENCES

- Belleville, R. E. Control of behavior by drug-produced internal stimuli. *Psychopharmacologia*, 1964, 5, 95-105.
- Brady, J. V. Emotion and the sensitivity of psychoendocrine systems. In D. C. Glass (Ed.), *Neurophysiology and emotion*. New York: The Rockefeller University Press, 1967. Pp. 70-96.
- Brady, J. V., Kelly, D., and Plumlee, L. Autonomic and behavioral responses of the rhesus monkey to emotional conditioning. *Annals of the New York Academy of Sciences*, 1969, 159, 959-976.
- deToledo, L. and Black, A. H. Heart rate changes during conditioned suppression in rats. *Science*, 1966, 152, 1404-1406.
- Goldberg, S. R. Relapse to opioid dependence: the role of conditioning. *Advances in Mental Science*, 1970, 2, (in press).
- Goldberg, S. R. and Schuster, C. R. Conditioned suppression by a stimulus associated with nalorphine in morphine-dependent monkeys. *Journal of the Experimental Analysis of Behavior*, 1967, 10, 235-242.
- Goldberg, S. R. and Schuster, C. R. Nalorphine: increased sensitivity of monkeys formerly dependent on morphine. *Science*, 1969, 166, 1548-1549.
- Goldberg, S. R., Woods, J. H., and Schuster, C. R. Morphine: conditioned increases in self-administration in rhesus monkeys. *Science*, 1969, 166, 1306-1307.
- Heistad, G. T. and Torres, A. A. A mechanism for the effect of a tranquilizing drug on learned emotional responses. *University of Minnesota Medical Bulletin*, 1959, 30, 518-527.
- Hunt, H. F. Effects of drugs on emotional responses and abnormal behavior in animals. In J. O. Cole and R. W. Gerard (Eds.), *Psychopharmacology*. Washington, D.C.: N.A.S.-N.R.C., 1959, No. 583. Pp. 268-283.
- Irwin, S. and Seevers, M. H. Altered response to drugs in the post addict (*Macaca mulatta*). *Journal of Pharmacology and Experimental Therapeutics*, 1956, 116, 31-32.
- Nathan, M. A. and Smith, O. A. Jr. Differential conditional emotional and cardiovascular responses—a training technique for monkeys. *Journal of the Experimental Analysis of Behavior*, 1968, 11, 77-82.
- Overton, D. A. Dissociated learning in drug states (state dependent learning). In D. H. Efron, J. O. Cole, J. Levine, and J. R. Wittenborn (Eds.), *Psychopharmacology*. P.H.S. Publication No. 1836, Washington, D.C.: U.S. Government Printing Office, 1968. Pp. 918-930.
- Parrish, J. Classical discrimination conditioning of heart rate and bar-press suppression in the rat. *Psychonomic Science*, 1967, 9, 267-268.
- Pavlov, I. P. *Conditioned reflexes* (trans. and ed. by G. V. Anrep). London: Oxford University Press, 1927.
- Schuster, C. R. and Brady, J. V. The discriminative control of a food-reinforced operant by interoceptive stimulation. *Pavlov Journal of Higher Nervous Activity*, 1964, 14, 448-458.
- Seevers, M. H. and Deneau, G. A. Physiological aspects of tolerance and physical dependence. In W. S. Root and F. G. Hoffman (Eds.), *Physiological pharmacology*. New York: Academic Press, 1963, 1, Pp. 565-640.
- Skinner, B. F. Are theories of learning necessary? *Psychological Review*, 1950, 57, 193-216.
- Stebbins, W. C. and Smith, O. A. Cardiovascular concomitants of the conditioned emotional response in the monkey. *Science*, 1964, 144, 881-883.
- Tatum, S. L., Seevers, M. H., and Collins, K. H. Morphine addiction and its physiological interpretation based on experimental evidences. *Journal of Pharmacology and Experimental Therapeutics*, 1929, 36, 447-475.
- Thompson, T., Schuster, C. R., Dockens, W., and Lee, R. Mouth-operated food and water manipulanda

- for use with monkeys. *Journal of the Experimental Analysis of Behavior*, 1964, 7, 171-172.
- Wikler, A. Conditioning factors in opiate addiction and relapse. In D. M. Wilner and G. G. Kassebaum (Eds.), *Narcotics*. New York: McGraw-Hill, 1965. Pp. 85-100.
- Wikler, A. and Pescor, F. T. Classical conditioning of a morphine abstinence phenomenon, reinforcement of opioid-drinking behavior and "relapse" in morphine-addicted rats. *Psychopharmacologia*, 1967, 10, 255-284.
- Woods, L. A. The pharmacology of nalorphine (*N*-allylnormorphine). *Pharmacological Reviews*, 1956, 8, 175-198.

Received 22 December 1969.