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The Effects of Prior Fentanyl Administration and of Pain on Fentanyl Analgesia: Tolerance to and Enhancement of Narcotic Analgesia[1]

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ABSTRACT

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The studies reported here were aimed at verifying some of the hypotheses which can be predicated on the assumption that tolerance does not develop to the physiological actions of narcotic analgesic drugs. Experiment 1 identified the time course of the analgesic effects of 0.0025, 0.005, 0.01 and 0.02 mg/kg of fentanyl in the tail-flick procedure. The analgesic effect of subcutaneously injected fentanyl reached its peak intensity within 15 to 30 min and gradually declined 45 to 60 min after injection. It also appeared that the procedure of repeatedly determining postdrug latencies in the same animal acts to inflate the measure of the analgesic effect of the drug. The inflationary effect was found to be inversely proportional to the dose within the 0.0025 to 0.02 mg/kg dose-range and caused a marked distortion of the dose-response curve. The main experiment analyzed the effects of mechanical pain and of antecedent fentanyl administration on the analgesic response to 0.01, 0.02 and 0.04 mg/kg of fentanyl. Antecedent fentanyl administration acted *to reduce* the magnitude of analgesia up to 3.1-fold indicating that tolerance had developed. The reduction appeared to reflect mainly a decreased peak intensity of analgesia; it was also proportional to the dose being tested, thus causing a flattening of the linear dose-response curve in a log-log plot. Antecedent exposure to mechanical pain acted *to magnify* analgesia 3.7-fold. The magnification appeared to reflect mainly a prolonged duration of analgesia; it did not co-vary with the test dose and caused an upward parallel shift of the dose-response curve. Antecedent exposure both to fentanyl and to mechanical pain neutralized the reducing and the magnifying effects of these conditions when applied alone, so that in this case the overall analgesic effect of 0.01 to 0.04 mg/kg of fentanyl was similar to that in a normal control group. The present findings are consistent with the hypothesis that antecedent exposure to pain and to narcotics may act mutually antagonistically in determining the magnitude of narcotic analgesia.

A number of theories have evolved to explain the phenomenon of tolerance to narcotic analgesic drugs (Clouet and Iwatsubo, 1975; Cochin, 1972; Goldstein, 1974; Takemori, 1975). These theories have taken into account various properties of the phenomenon and their diversity is likely to have originated from the fact that so many different processes contribute to the development of tolerance. Among these processes are drug metabolism and distribution, drug-receptor interactions, cellular adaptations, neurohormonal and immunological processes (Cochin, 1972) and conditioning (Siegel, 1977). One basic assumption which is inherent in the current theories of tolerance is that the phenomenon would consist primarily of a decrease in the physiological actions of narcotics after antecedent exposure to these drugs. The ability to induce tolerance is thus said (*e.g.*, Mushlin *et al.*, 1976) to be a pharmacological property of the narcotic analgesics.

Paradoxical as it may seem, however, it appears (Colpaert, 1978) equally possible to predict the development of tolerance to narcotic analgesia from a hypothetical model which explicitly assumes that there is no decrease in the physiological actions of narcotics with repeated exposure. In the terms of this model, tolerance to narcotic analgesia does not result from an inherent pharmacological property of the narcotic analgesics, but reflects the operating characteristics of the neural systems involved in pain processing. The merit of this model, if any, resides in the new working hypotheses on analgesia which it may generate. Two of these hypotheses are: 1) nociceptive stimulation alone should augment the analgesic response to narcotic drugs and 2) whereas antecedent treatment with narcotics reduces the analgesic response (tolerance), this tolerance should be attenuated by coupling the treatment with nociceptive stimulation. In support of the second hypothesis, it has been shown (Colpaert *et al.*, 1978) that tolerance to the analgesic effect of 0.04 mg/kg

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of fentanyl (Janssen *et al.*, 1963) fails to develop if the antecedent fentanyl injections are associated with brief episodes of mechanical pain. Also, chronic treatment with the long-acting narcotic bezitramide (Janssen *et al.*, 1971) produces neither tolerance to 1.25 mg/kg of bezitramide nor cross-tolerance to 10 mg/kg of morphine, if the chronic treatment is associated with a proposed experimental condition of chronic pain (Colpaert, 1979). An important limitation of both studies, however, is that tests for tolerance were conducted on only single doses of the narcotic analgesics.

This study presents an analysis of the effects of nociceptive stimulation on the magnitude of fentanyl analgesia in the rat and on the development of tolerance to this analgesia. The antecedent exposure to a narcotic drug consisted of eight fentanyl injections. The antecedent exposure to nociceptive stimulation consisted of brief episodes of mechanically produced pain. The tail-flick procedure was used to determine the analgesic response of the animals to the fentanyl doses 0.01, 0.02 and 0.04 mg/kg.

Materials and Methods

Animals. The animals were experimentally naive male Wistar strain rats weighing 240 to 320 g. The laboratory was air conditioned (21 ± 1°C; relative humidity 65 ± 5%) and continuously illuminated. The rats were kept in individual rodent living cages equipped with a grid floor and had free access to dry standard laboratory food and tap water. The animals were used only once.

Analgesia assay. The tail-flick procedure used here has been described in detail elsewhere (Janssen *et al.*, 1963). The rat was placed in a standard rat holder with the tail hanging freely outside the holder. A reading consisted of dipping the distal 5 cm of the tail into a warm (55 ± 1°C) water bath and determining the reaction time for its withdrawal to the nearest 0.1 sec. To avoid tissue damage, the cut-off time was limited to 30.0 sec. All readings were taken by a single observer who was blind to the treatment conditions.

Time-effect study. Experiment 1 determined the time-effect relationship for the analgesic effects of 0.0025 to 0.02 mg/kg of fentanyl. The results of this experiment were to serve as a basis for selecting adequate doses and postdrug test intervals for the second experiment.

As nociceptive stimulation may interfere with narcotic analgesia (see "Introduction"), and because any analgesia assay necessarily involves at least some exposure to a nociceptive stimulus, time-effect studies were carried out in two separate groups.

In the Repeated Readings groups (total n = 36), readings were performed 15 min before as well as 5, 10, 15, 20, 25, 30, 45 and 60 min after the injection of one of four fentanyl doses (*i.e.*, 0.0025, 0.005, 0.01 and 0.02 mg/kg). Nine rats were used per dose.

In the Single Readings group (total n = 288), readings were made 15 min before as well as at one of the eight postdrug intervals. Each of the four dosegroups (n = 72) was further divided into eight subgroups of nine rats; these eight subgroups were used to determine reaction times at the various postdrug intervals. Thus, rats in the Single Readings group were subjected to only one postdrug reading as compared to the eight readings that were carried out in the Repeated Readings group.

Nociceptive stimulation and narcotic analgesia. For the purpose of experiment 2, 176 rats were randomly assigned to one of four groups (n = 44 per group).

One group (saline control; S) was injected twice daily (8:00 A.M. and 3:00 P.M.) with saline for 4 consecutive days. The second group (fentanyl; F) was injected at similar times with 0.04 mg/kg of fentanyl. The third group (fentanyl-clips; FC) was similarly injected with 0.04 mg/kg of fentanyl but, in addition, received exposure to nociceptive stimulation. That is, 5 min after each of the eight fentanyl injections, an alligator clip (pressure, 0.5 kg; part no. 930. 120-000, Hirschmann, West-Germany), which the animals were unable to remove, was applied to each of the hindpaws. The two clips remained in place until 75 min after injection. The fourth group (saline-clips; SC) received saline injections and was similarly exposed to the nociceptive stimulation produced by the clips.

On the fifth day (8:00 A.M.) of the experiment, each of the four groups was divided into four subgroups of 11 rats. These subgroups were randomly allocated to one of the four treatment conditions, *i.e.*, saline and 0.01, 0.02 and 0.04 mg/kg of fentanyl. By using the analgesia assay described above, readings were then carried out in all rats, once before (t = 15 min), as well as 15, 30, 45 and 60 min after the administration of the appropriate treatment condition. Thereafter (3:00 P.M.; 24 hr after the last application of the clips), the diameter of all rats' hindpaws was measured by means of an apparatus described elsewhere (Awouters *et al.*, 1976); the left and right diameters were averaged to yield a single value per animal.

Drug administration. Different concentrations of fentanyl citrate were freshly prepared as aqueous solutions. Injections of saline or fentanyl were carried out subcutaneously at a constant volume of 1 ml/100 g b.wt.

Statistical analysis. To avoid the risk of making any incorrect assumptions about the distribution of the data, and to eliminate the confounding effect of possible differences in variability, the data analyses employed nonparametric statistics. The Wilcoxon matched-pairs signed-ranks test and the Mann-Whitney U test (Siegel, 1956) were used for comparisons of related and independent samples respectively. The probability level for rejection of the null hypothesis was .05.

Results

Time-effect study. The results of the time-effect study are summarized in figure 1. In both the Repeated and the Single Readings group, 0.0025 to 0.02 mg/kg of fentanyl caused the postdrug latencies to increase in a dose-related manner. In all cases except one (Single Readings groups; 0.0025 mg/kg of fentanyl; 5-min interval) the median postdrug reaction time exceeded the median predrug value. Peak increases in postdrug latencies occurred 15 to 30 min after drug treatment, indicating that subcutaneous fentanyl reaches peak effect no sooner than 15 min after injection. Between 30 and 45 min after injection, median latencies invariably decreased; however, latencies at 45 and 60 min always exceeded the predrug level, suggesting that residual drug effects may persist for up to 60 min after fentanyl administration.

Although predrug latencies were almost indentical, it is apparent that the postdrug latencies in the Repeated Readings group generally exceeded those in the Single Readings group. The difference was statistically significant (two-tailed; $P \leq .05$) at several of the postdrug intervals with the doses 0.0025 and 0.005 mg/kg (fig. 1). To further analyze this difference, analgesic effects as demonstrated by the median values depicted in figure 1 were computed according to the method introduced by Winter and Flataker (1950). This was done by expressing area A as a percentage of area B (Sewell and Spencer, 1976). Area A represents the area (shaded areas in fig. 1), in minute-seconds, comprised between the time-effect polygon on the one hand and the predrug base line on the other. Area B is comprised between the predrug base line and the zero level. The insert in figure 1 shows that the percentage of analgesic effect in the Repeated Readings group exceeds that in the Single Readings group at all four doses. The difference between the two groups amounts to a factor 2.9, 2.0, 1.3 and 1.3 at the doses 0.0025 to 0.02 mg/kg, respectively. It thus appears that the procedure of determining postdrug latencies repeatedly in the same animals tends to inflate the measurement of analgesic drug effects in the tail-flick procedure.

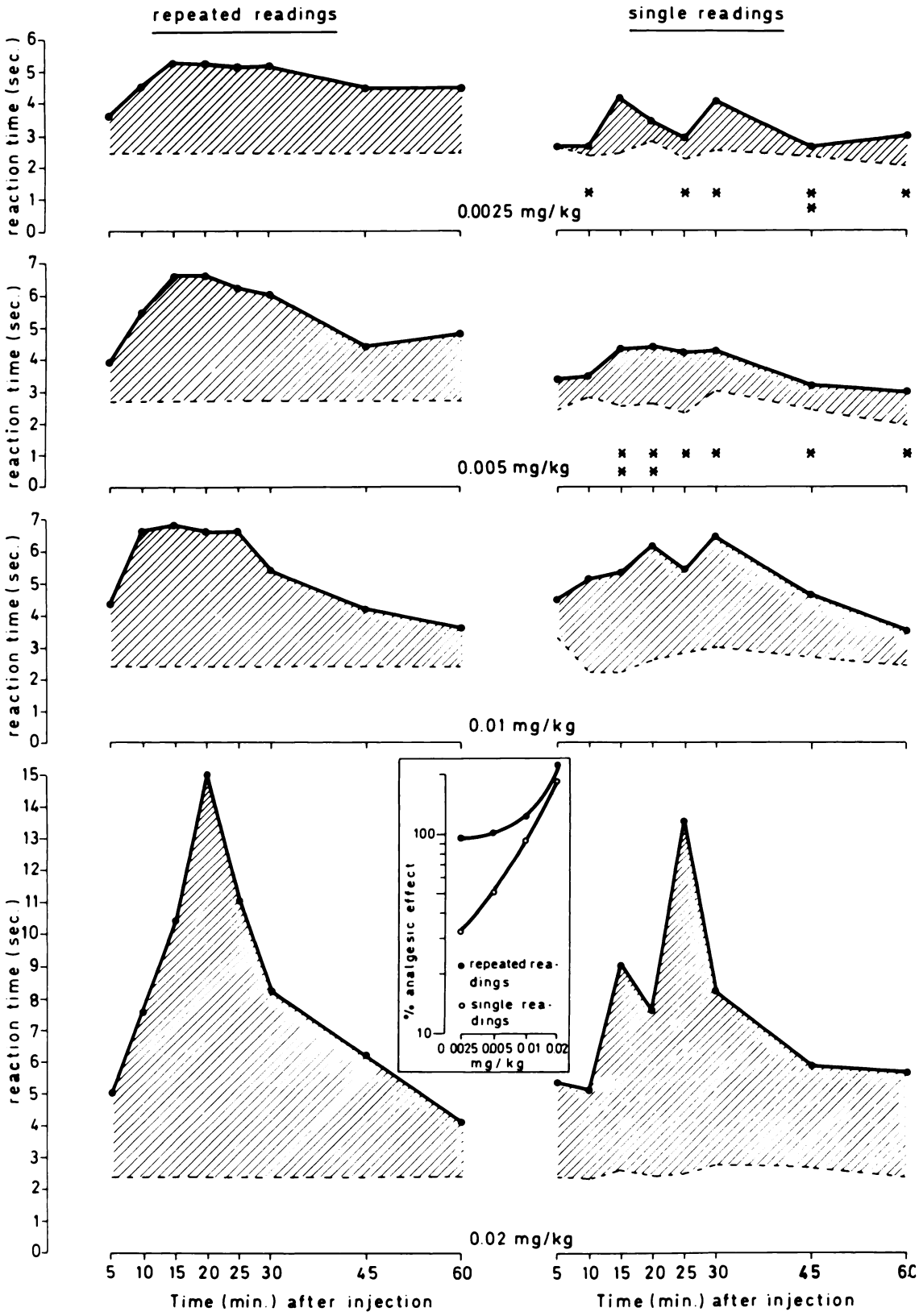

Fig. 1. Effects of 0.0025 to 0.02 mg/kg of fentanyl on tail-flick latency in the rat. Each data point represents the median reaction time (in seconds) of 9 animals. In the Repeated Readings condition, tail-flick readings were carried out once before and eight times after drug injection. In the Single Readings condition, readings were carried out once before and once after drug injection; different subgroups of rats were used at different postdrug intervals. The asterisks refer to the difference in postdrug latencies between the Repeated and the Single Reading groups and indicate two-tailed probability to be $P \leq .05$ (*) or $P \leq .01$ (**) (Mann-Whitney *U* test). The broken line represents the median predrug latency. For the single Readings group, the broken line connects the median predrug latencies in the different subgroups which were used for determining postdrug latencies at different intervals. The insert presents a log-log plot of percentage of analgesic effect as a function of fentanyl dose.

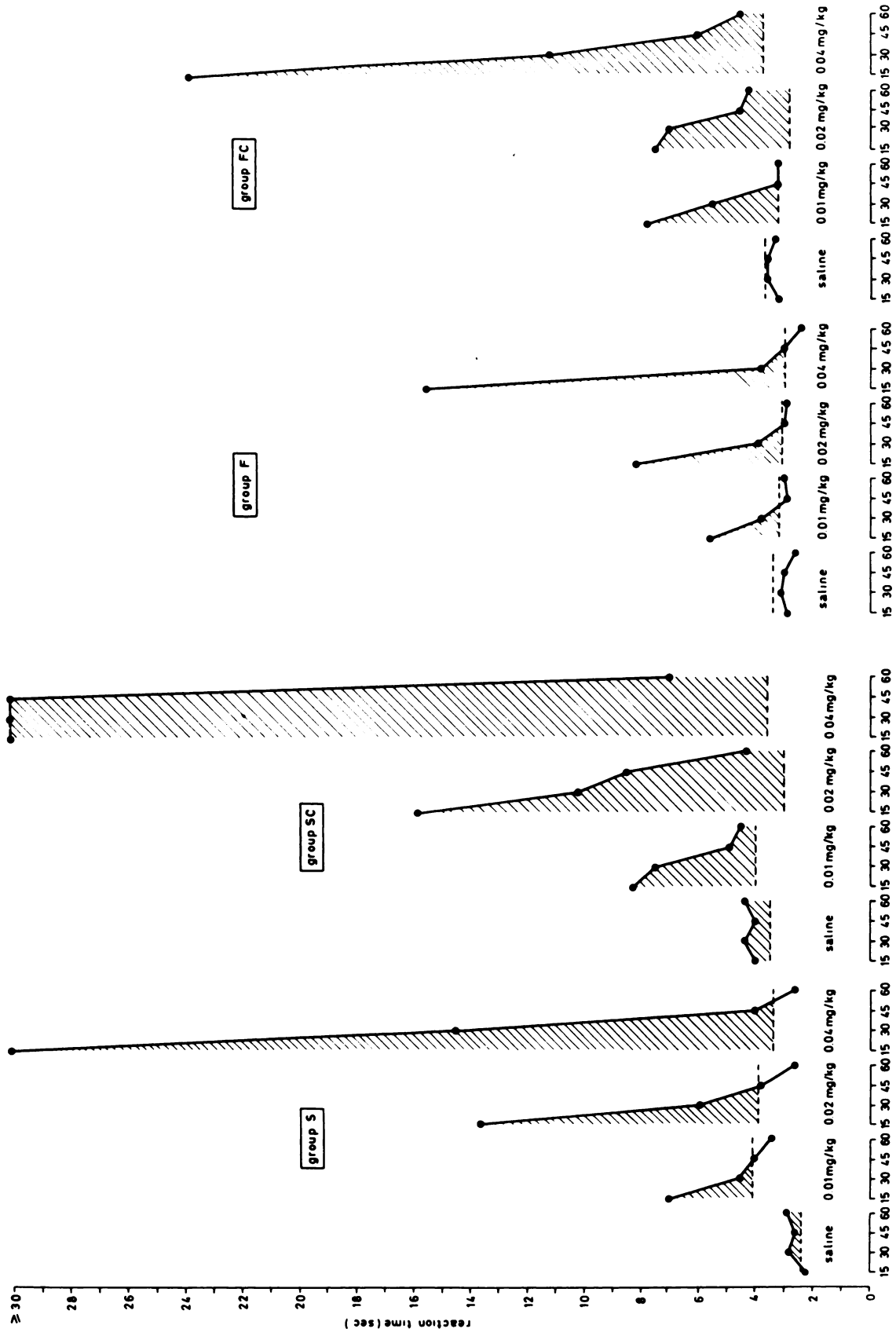

Fig. 2. Effects of saline and of 0.01, 0.02, and 0.04 mg/kg of fentanyl on tail-flick latency in the rat. Each data point represents the median reaction time (in seconds) of 11 animals. The S and F group had received eight injections of saline or 0.04 mg/kg of fentanyl before the test. The SC and FC groups consist of rats which, in addition to these eight injections, had been subjected to nociceptive stimulation produced by crocodile clips. The broken lines represent median predrug latencies for each of the four subgroups of the four main groups.

With regard to experiment 2, the results of the time-effect study suggest the following. 1) To reduce the inflationary effect of repeated readings, the number of postdrug readings in the same animals should be as small as reasonably possible. 2) As the inflationary effect is mostly marked at these lower doses, 0.0025 and 0.005 mg/kg of fentanyl should not be used in the second experiment. Since, in addition, the 30-second cut-off sets a limitation to the analysis of very high doses, the doses finally selected were 0.01, 0.02 and 0.04 mg/kg. 3) The 15-min postdrug interval appears adequate to detect possible changes in the peak effect of fentanyl. Readings at 30, 45 and 60 min appear adequate in delineating the subsequent decay of the effect, and hence might allow the detection of possible changes in the duration of fentanyl analgesia.

Nociceptive stimulation and narcotic analgesia. The main experiment determined the effect of mechanically produced nociceptive stimulation and repeated injection with 0.04 mg/kg of fentanyl on the analgesic effects of 0.01 to 0.04 mg/kg of fentanyl. The experiment involved four groups, *i.e.*, S, F, FC and SC. Gross observation of the overt behavior of the animals during the first 4 days of the experiment indicated the SC group to respond to the nociceptive stimulation with signs reminiscent of intense pain, *i.e.*, squealing, urinating, defaecating and vigorous biting on the clips and the grids. Although similar reactions were observed in the FC group, these were greatly reduced in terms of both intensity and frequency.

The median predrug latency (all 44 rats combined) in the F groups (3.2 sec; 95% confidence limits, 3.0–3.6) was slightly lower than that in the S group (3.4 sec; 2.7–3.9), whereas that in the SC group was slightly higher (3.5 sec; 3.0–3.9). The median latency in the FC group was statistically significant (Mann-Whitney *U* test; two-tailed, $P > .05$).

The results of the postdrug readings are summarized in figure 2. Fentanyl produced a dose-dependent increase in the latency to tail withdrawal in each of the four main groups. In each of the 12 drug-treated subgroups, the peak increase in latency occurred at the 15-min postdrug interval. This peak increase was followed by a gradual shortening of latency to a level (at 60 min) which was generally lower than the predrug base line in groups S and F, but higher in the groups that had been exposed to nociceptive stimulation. No such increases occurred after saline in that none of the four postinjection readings significantly exceeded the preinjection base line in either the S, F, SC or FC group (two-tailed, $P > .05$; Wilcoxon *U* test).

A first analysis of this data is directed toward estimating the overall analgesic effect so that both the intensity and the duration of the effect are taken into account. Thus, the area defined by the postdrug polygon (shaded areas in fig. 2) was expressed as a percentage of the area defined by the predrug base line. The percentage of analgesic effect demonstrated by the median latencies (fig. 2) is plotted in figure 3. A percentage of analgesic effect was also computed for each of the 176 rats individually and the statistical analyses are based on these individual values. It was found that, as compared with the S group, the analgesic effect in the F group was smaller at the fentanyl doses 0.04 and 0.02 mg/kg but essentially similar at the 0.01 mg/kg dose. The SC group exceeded the S group at all doses, although this difference was significant at the doses 0.02 and 0.04 mg/kg only. At none of the three fentanyl doses did the analgesic effect in the FC group differ significantly from that in the S group. Comparisons between experimental groups indicated that both the SC and the FC group exceeded the F group at all three doses. As compared with the SC group, the

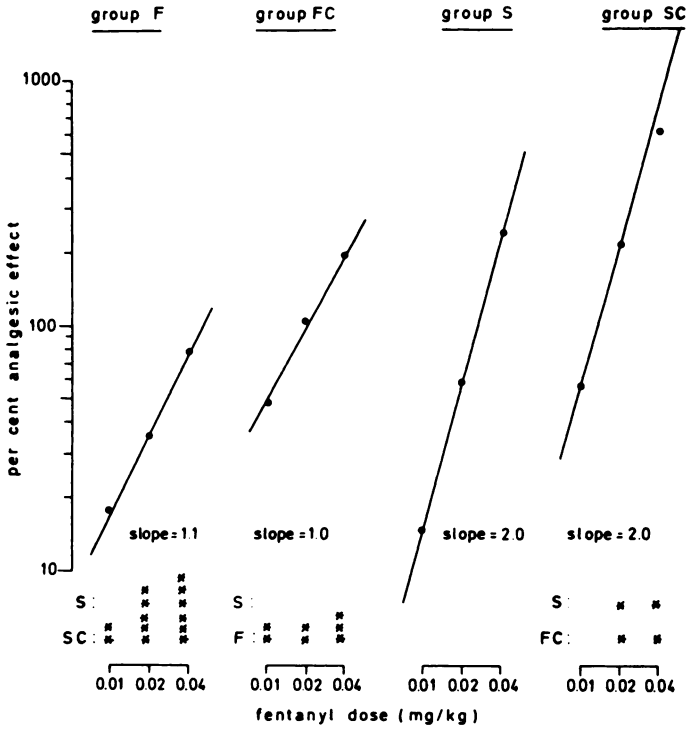

Fig. 3. Log-log plot of percentage of analgesic effect as a function of fentanyl dose. Results are based on the data presented in figure 2. The slope is the parameter b in the regression equation as it is represented by the straight line through the data points. The asterisks indicate two-tailed probability to be $P \leq .05$ (*), $P \leq .01$ (**) or $P \leq .001$ (***) (Mann-Whitney U test).

FC group showed less effect at the doses 0.04 and 0.02 mg/kg but not at the lower dose. In terms of the area described by the postinjection readings, the median response to saline amounted to 0% in the F and FC groups and to 10.9 and 20% in the S and SC groups, respectively. None of the comparisons among these responses to saline yielded statistical significance ($P > .05$).

The lines shown in figure 3 are the regression lines as they were computed on the basis of all three data points in the groups F, FC and S; in the SC group, only the data points at 0.01 and 0.02 mg/kg were used because the one at 0.04 mg/kg obviously presents a gross underestimation of the analgesic effect (fig. 2). The slope of these lines (the parameter b in the regression equation $y = a + bx$) was found to be 2.0 in both the S and the SC group. The slope in the F group appeared to have flattened to a value of 1.1, and a similarly shallow slope (1.0) was also obtained in the FC group. Statistical analysis (Draper and Smith, 1966) revealed no difference (two-tailed, $P > .05$) between the F and the FC groups nor between the S and the SC groups. However, the slope in the FC groups was significantly less steep ($P < .05$) than that in the S and the SC groups, and both the S ($P < .01$) and the SC group ($P < .05$) had steeper slopes than the F group.

The second analysis was carried out to determine to what extent the above described changes in overall analgesic effect were due to changes in the peak intensity and/or the duration of the analgesic effect of fentanyl. To this end, comparisons of latencies were made for the interval at which peak effect occurred (*i.e.*, 15 min), as well as for one of the intervals (*i.e.*, 45 min) at which latencies would seem to be particularly sensitive to the duration of the analgesic effect.

Fifteen minutes after injection, reaction times in the F group were significantly (two-tailed, $P < .05$; Mann-Whitney *U* test)

lower than in the S group at the doses 0.04 and 0.02 mg/kg but not at the lower dose. Similar differences occurred between the FC and the S groups, but all peak latencies in the SC group were essentially similar to those in the S group. Furthermore, the latencies in the F group were lower than those in the FC group at the 0.04 mg/kg dose and also lower than those in the SC group at 0.04 and 0.02 mg/kg. The SC group exceeded the FC group at 0.04 and 0.02 mg/kg but not at 0.01 mg/kg.

Forty-five minutes after injection, latencies in the F group were similar to those in the S group except at the 0.01 mg/kg dose. At this point, however, the SC, but also the FC group, now had significantly higher latencies after 0.04 mg/kg than the S group. The FC group also exceeded the F groups at 0.04 and 0.02 mg/kg but had shorter latencies than the SC group at 0.04 and 0.01 mg/kg. The SC group by far exceeded the F group at all doses.

At the end of the experiment, the hindpaws of those animals in which the clips had been applied were severely inflamed. The inflammation was apparent from an increase in the paw diameter. Although the diameter was 8.5 mm (median value) in both the S (95% confidence limits, 8.4–8.6) and the F group (8.5–8.7; $P > .05$) it had significantly ($P < .001$) increased in the SC (10.5 mm; 10.4–10.7) as well as in the FC group (10.9 mm; 10.6–11.1)

Discussion

The first experiment reported here examined the time course of the analgesia produced by 0.0025 to 0.02 mg/kg of fentanyl. By using a modification (Janssen *et al.*, 1963) of the tail-flick procedure, it was found (fig. 1) that subcutaneously injected fentanyl reaches peak effect within 15 to 30 min after administration. This peak is followed by a gradual decline of the latency, although residual drug effects may still be apparent 45 min after injection. The experiment also revealed that the procedure of determining tail-flick latencies at different postdrug intervals in the same animal acts to significantly inflate the measure of the analgesic action of the drug. The inflationary effect caused the magnitude of fentanyl analgesia to be overestimated by a factor 2.9 at the 0.0025 mg/kg dose; the factor amounted to 2.0 at 0.005 mg/kg and to 1.3 at both 0.01 and 0.02 mg/kg. Due to its inverse relation to the dose, the inflationary effect also caused a marked distortion of the dose-response curve (fig. 1, insert). Although the reason for this effect cannot be determined from the present experiments, the data do seem to rule out tissue damage and subsequent local desensitization as viable causes of the effect. This is because the higher doses of the analgesic yielded longer latencies which in turn should have resulted in more tissue damage; hence, the effect would be expected to relate in a manner which is directly, rather than inversely, proportional to the dose of the analgesic.

The main experiment analyzed the effects of nociceptive stimulation and of antecedent fentanyl administration on the analgesic response to 0.01, 0.02 and 0.04 mg/kg of fentanyl in the rat. In the saline (S-) control group, 0.01 to 0.04 mg/kg of fentanyl produced a dose-dependent increase in tail-flick latency. The overall (intensity × duration; Winter and Flataker, 1950) analgesic effect ranged from 14.8% to 0.01 mg/kg to 243.6% at 0.04 mg/kg. The dose-response data points could be fitted almost perfectly by a linear function in a log-log plot, and the slope of the calculated regression line was 2.0 (fig. 3).

The overall analgesic effect of 0.02 and 0.04 mg/kg of fentanyl in the fentanyl (F-) group was significantly lower than that in the S group, thus indicating that the antecedent fentanyl injections had been adequate in establishing tolerance to narcotic analgesia. The extent to which tolerance occurred was proportional to the dose being tested, so that the dose-response curve, although still linear, had become less steep (slope = 1.1; fig. 3). This result is consistent with earlier evidence (Cox *et al.*, 1975; Theiss *et al.*, 1975) that the dose-response curve for narcotic analgesia may have a lower slope in tolerant as opposed to nontolerant animals. Further analysis of the data suggests that the tolerance which developed in the F group may mainly reflect a reduced maximum intensity and possibly also a shortened duration (fig. 2) of the fentanyl effect.

The data in the saline-clips (SC-) group indicate that the repeated exposure to nociceptive stimulation had acted to magnify the analgesic response to fentanyl 3.7-fold. The increase in effect was equally large at all doses, so that the dose-response curve in the SC group represents an upward shift parallel (slope = 2.0) to the curve in the S group. This magnification of analgesic effect may mainly reflect a prolonged duration of analgesia; the peak intensity of analgesia was also higher but this difference failed to reach statistical significance.

The overall analgesic effect in the FC- group was similar to that in the normal control group but significantly exceeded that in the F group. The latter result confirms and extends earlier data (Colpaert, 1979; Colpaert *et al.*, 1978) indicating that nociceptive stimulation antagonizes, and may even prevent, the development of tolerance to narcotic analgesia. However, the analgesic effect in the FC group was significantly smaller than that in the SC group, thus suggesting that antecedent administration of narcotics may prevent the magnification of narcotic analgesia which otherwise occurs on exposure to nociceptive stimulation. The slope of the dose-response curve in the FC group (slope = 1.0) was shallower than that in the S or SC groups but was parallel to that in the F group (fig. 3). Interestingly, the FC dose-response curve can be understood as reflecting a combination of the processes through which the F and SC curves came to differ from that of the normal control group. That is, similar to the F curve, the FC curve represents a flattening of the normal curve; such a flattening reflects a decreased relative effectiveness of higher as opposed to lower doses. Also, similar to the SC curve, the FC curve may have been subject to an upward parallel shift as a result of which the FC group shows a 2.7 times larger analgesic response than the F group.

Further analysis suggests that the FC group showed a lower peak intensity of analgesia than the S group. However, this decreased peak intensity was compensated for by a prolonged duration of effect, so that the overall analgesic effect of any dose in the FC group did not differ significantly from that in the S group (fig. 3). In this respect, too, the results in the FC group combine the processes through which the F and the SC groups came to deviate from normal (*i.e.*, decreased peak and prolonged duration, respectively).

The present data on narcotic analgesia (experiment 2) are thus consistent with a proposed model (Colpaert, 1978) describing the operating characteristics which a neuronal system may demonstrate in processing nociceptive stimuli. Also consistent with this model is the finding (experiment 1) that prior exposure to the nociceptive stimulus used for assaying narcotic analgesia acts to inflate the measure of this analgesia. That the inflationary effect of repeated postdrug readings relates inversely to the dose being tested may then indicate that higher doses suppress the effects of presenting a nociceptive stimulus and, hence, its impact on the response to a subsequent one.

The model referred to above does not specify which physiological mechanisms may be instrumental in the processes which it describes. Current research on endogenous opioid peptides (Hughes and Kosterlitz, 1977; Terenius, 1978) may point to these substances as possible candidates for a role in establishing different nociceptive thresholds as a result of prior exposure to pain and to narcotic drugs. However, the differences among groups in predrug response were not significant in the present study and naloxone has been found not to exert any detectable effect in a previous study (Colpaert, 1979) in which such differences did reach statistical significance. In addition, the analytical method of Winter and Flataker (1950) used here serves to control for the possibly confounding effect of differences in nociceptive threshold on the measure of drug-produced analgesia.

Since several of the present differences in analgesic response were mainly due to differences in the duration of the response, changes in drug metabolism and distribution may alternatively account for at least part of these differences. Studies on the pharmacokinetics of fentanyl (Hess *et al.*, 1971, 1972; Maruyama and Hosoya, 1969; Novack *et al.*, 1978; Van Wijngaarden and Soudijn, 1968) suggest that the duration of the analgesic action of fentanyl is limited by its disappearance from the brain as a result of redistribution caused by rapid biotransformation and excretion. It is not known, however, how prior exposure to pain and fentanyl may affect the metabolism and excretion of the drug and further studies are required to determine the extent to which this hypothesis may account for the data presented here.

In conclusion, the present data show that antecedent exposure to narcotics acts *to reduce* narcotic analgesia (the phenomenon of tolerance); the reduction is proportional to the dose being tested and may reflect mainly a decreased peak intensity of analgesia. Antecedent exposure to nociceptive stimulation acts *to magnify* narcotic analgesia. In contrast with the above mentioned reduction, however, this magnification does not covary with dose and may reflect mainly a prolonged duration of analgesia, more so than an increased peak intensity. Antecedent exposure both to narcotics and to nociceptive stimulation yields an analgesic response which appears to combine these processes.

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References

AWOUTERS, F., DEPROOST, L. M. C., LENAERTS, F. M. AND NIEMEGEERS, C. J. E.: A new apparatus for measuring diameter changes of the rat paw and tibiotarsal joint. Arzneim. Forsch. **26:** 37–40, 1976.

CLOUET, D. H. AND IWATSUBO, K.: Mechanisms of tolerance to and dependence on narcotic analgesic drugs. Annu. Rev. Pharmacol. **15:** 49–71, 1975.

COCHIN, J.: Some aspects of tolerance to the narcotic analgesics. *In* Drug Addiction I. Experimental Pharmacology, ed. by J. H. Singh, L. Miller and H. Lal, pp. 365–375, Futura Publishing Co., Inc., Mount Kisco, NY, 1972.

COLPAERT, F. C.: Narcotic cue, narcotic analgesia, and the tolerance problem: The regulation of sensitivity to drug cues and to pain by an internal cue processing model. *In* Stimulus Properties of Drugs: Ten Years of Progress, ed. by F. C. Colpaert and J. A. Rosecrans, pp. 301–321, Elsevier/North-Holland Biomedical Press, Amsterdam, 1978.

COLPAERT, F. C.: Can chronic pain be suppressed despite purported tolerance to narcotic analgesia? Life Sci. **24:** 1201–1210, 1979.

COLPAERT, F. C., NIEMEGEERS, C. J. E. AND JANSSEN, P. A. J.: Nociceptive stimulation prevents development of tolerance to narcotic analgesia. Eur. J. Pharmacol. **49:** 335–336, 1978.

COX, B. M., GINSBURG, M. AND WILLIS, J.: The offset of morphine tolerance in rats and mice. Br. J. Pharmacol. **53:** 383–391, 1975.

DRAPER, N. R. AND SMITH, H.: Applied Regression Analysis, John Wiley and Sons, Inc., New York, 1966.

GOLDSTEIN, A.: Opiate receptors. Life Sci. **14:** 615–623, 1974.

HESS, R., HERZ, A. AND FRIEDEL, K.: Pharmacokinetics of fentanyl in rabbits in view of the importance for limiting the effect. J. Pharmacol. Exp. Ther. **179:** 474–484, 1971.

HESS, R., STIEBLER, G. AND HERZ, A.: Pharmacokinetics of fentanyl in man and the rabbit. Eur. J. Clin. Pharmacol. **4:** 137–141, 1972.

HUGHES, J. AND KOSTERLITZ, H. W.: Opioid peptides. Br. Med. Bull. **33:** 157–161, 1977.

JANSSEN, P. A. J., NIEMEGEERS, C. J. E. AND DONY, J. G. H.: The inhibitory effect of fentanyl (R 4263) and other morphine-like analgesics on the warm water induced tail withdrawal reflex in rats. Arzneim. Forsch. **13:** 502–507, 1963.

JANSSEN, P. A. J., NIEMEGEERS, C. J. E., SCHELLEKENS, K. H. L., MARSBOOM, R. H. M., HERIN, V. V., AMERY, W. K. P., ADMIRAAL, P. V., BOSKER, J. T., CRUL, J. F., PEARCE, C. AND ZEGVELD, C.: Bezitramide (R 4845), a new potent and orally long-acting analgesic compound. Arzneim. Forsch. **21:** 862–867, 1971.

MARUYAMA, Y. AND HOSOYA, E.: Studies on the fate of fentanyl. Keio J. Med. **18:** 59–70, 1969.

MUSHLIN, B. E., GRELL, R. AND COCHIN, J.: Studies on tolerance. I. The role of the interval between doses on the development of tolerance to morphine. J. Pharmacol. Exp. Ther. **196:** 280–287, 1976.

NOVACK, G. D., BULLOCK, J. L. AND EISELE, J. H.: Fentanyl: Cumulative effects and development of short-term tolerance. Neuropharmacology **17:** 77–82, 1978.

SEWELL, R. D. E. AND SPENCER, P. S. J.: Antinociceptive activity of narcotic agonist and partial agonist analgesics and other agents in the tail-immersion test in mice and rats. Neuropharmacology **15:** 683–688, 1976.

SIEGEL, S.: Nonparametric Statistics for the Behavioral Sciences, McGraw-Hill Book Company, New York, 1956.

SIEGEL, S.: Morphine tolerance acquisition as an associative process. J. Exp. Psychol. Anim. Behav. Processes **3:** 1–13, 1977.

TAKEMORI, A. E.: Neurochemical bases for narcotic tolerance and dependence. Biochem. Pharmacol. **24:** 2121–2126, 1975.

TERENIUS, L.: Endogenous peptides and analgesia. Annu. Rev. Pharmacol. Toxicol. **18:** 189–204, 1978.

THEISS, P., PAPESCHI, R. AND HERZ, A.: Effects of morphine on the turnover of brain catecholamines and serotonin in rats—chronic morphine administration. Eur. J. Pharmacol. **34:** 263–271, 1975.

VAN WIJNGAARDEN, I. AND SOUDIJN, W.: The metabolism and excretion of the analgesic fentanyl (R 4263) by Wistar rats. Life Sci. **7:** 1239–1244, 1968.

WINTER, C. A. AND FLATAKER, L.: Studies on Heptazone (6-morpholino-4,4-diphenyl-3-hepatone hydrochloride) in comparison with other analgesic drugs. J. Pharmacol. Exp. Ther. **98:** 305–317, 1950.

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