Respiratory depression by analgesics at 41 bar

K. FURSET, L. AANDERUD, and I. TYSEREBOTN

Department of Physiology (K.F.I.T.) University of Bergen and Department of Anesthesiology (L.A.), Haukeland Hospital, N-5000 Bergen, Norway

Furset K, Aanderud L, Tyssenbotn I. Respiratory depression by analgesics at 41 bar. Undersea Biomed Res 1989; 16 (3):219–226.—The effects of morphine and fentanyl on respiration and tissue CO₂, measured transcutaneously were studied at surface and at 41 bar ambient pressure in conscious, trained rats. Morphine and fentanyl were given in equianalgesic doses i.v., 7 and 0.025 mg/kg, respectively. Fentanyl caused a rapid but brief respiratory depression which was the same at 1 and 41 bar, and essentially the same results were found in the morphine groups, although there was a longer latency and duration of action. No statistical differences in the degree of respiratory depression were found at 41 bar compared to 1 bar for either analgesic.

carbon dioxide  
high pressure  
analgesics  
rats  
respiratory depression  
transcutaneous measurement

Increased ambient pressure may interfere with the action of drugs, and reversal of anesthesia is well documented in such conditions (1–4). Less is known, however, about the effects of the commonly used analgesic drug morphine and the synthetic opiate fentanyl during pressure exposure, due to lack of suitable methods for testing pain in animals under high pressure. Use of the hot-plate method for measuring the analgesic effect in experimental animals is difficult to interpret correctly because the high ambient temperature required in a He atmosphere interferes with the test stimulus (5, 6). The effect of morphine at 48 bar was recently studied using the formalin test, in which the behavioral response to a s.c. injection of formalin in the hindpaw was evaluated (7). No reduction in the analgesic effect was found. To our knowledge, no data exist on the respiratory depressant effect of morphine or fentanyl at increased ambient pressure.

The aim of the present investigation was to study the respiratory depression of morphine and fentanyl by noting the reduction of respiratory rate and secondary increase in transcutaneous measured Pco₂ (Prco₂) at 41 bar. The doses used of the 2 drugs were equivalent as to the analgesic effects in rats according to Janssen et al. (8).
METHODS

Thirty-two male Wistar rats, weighing 280–350 g, were individually adapted to the experimental conditions for 30 min daily for 18 days (9). The adaptation included several compressions to 2 bar for 10 min.

Tissue CO₂ was measured transcutaneously (PtcO₂) by a Kontron Ag-AgCl CO₂ sensor (Kontron Instruments Ltd, Zurich, Switzerland) attached to depilated skin on the abdomen of the conscious rats (Fig. 1). The experimental set-up and the reliability of the method in hyperbaric conditions have been discussed in a previous report (10).

The respiratory frequency (RF) was measured by projecting a light beam at a specific angle against the rat thorax. The light reflected from the body was monitored by a photo cell connected to an amplifier. The frequency of the changes in intensity of reflected light was recorded (Fig. 1).

The pressure chamber

A 30-liter steel pressure chamber with an internal diameter of 28 cm was used. The front port contained a window allowing continuous monitoring of the rats by a video camera. All electrical penetrations and gas outlets and inlets were placed in the rear chamber port. An electric fan ventilated the chamber atmosphere through soda lime to avoid CO₂ elevation in the atmosphere and to secure proper gas mixing. The temperature was maintained at 22°C during control measurements at 1 bar, and was increased to 33 ± 0.5°C at 41 bar to maintain thermoneutrality of the animals (11). The temperature was stabilized by an external water jacket.

Surgery

The day before the experiment, the right external jugular vein was cannulated by a PE-50 catheter (Intramedic, Clay Adams, Parsippany, NJ) with Hypnorm anesthesia 1 ml/kg i.p. (fluanisone 10 mg/ml and fentanyl 0.315 mg/ml, Mekos, Hålsingborg, Sweden).

The abdominal skin was carefully depilated with cream (calciumthioglycolate, Vyzo, Coflett-Marwell Hauge, Oslo, Norway) left on the skin for 5 min then cleaned.
with acetone. The CO₂ sensor was fixed to this area before the experiments, as described previously (10).

**Experimental procedure**

The rats were placed in the restraining cage after appropriate application of the CO₂ electrode. The catheter in the jugular vein was connected to a steel cannula penetrating the rear chamber port of the pressure chamber. A plastic syringe on the outside was driven by a high-pressure pump calibrated to infuse morphine or fentanyl against the 41 bar chamber pressure at the same rate as used in the 1 bar situation.

Four series of experiments were performed. The rats in the 2 control series (1 bar) were placed in the closed pressure chamber for 2 h (equal to the compression time in the pressure series). During this period, air was flushed through the chamber to simulate compression of the control animals. All animals were immobilized in a snug-fitting restraining cage identical to the cage used for adaptation (Fig. 1), and the infusion of drugs was started after all animals had been an identical time in restraint. The animals were continuously observed by a video camera.

**Control experiments at 1 bar**

In series 1 the animals \( (n = 8) \) were given morphine (7 mg/kg), and in series 2 the animals \( (n = 8) \) were given fentanyl (0.025 mg/kg), both infused into the jugular vein at a rate of 0.4 ml/min. The PrCO₂ was measured continuously for 35 min with the PCO₂ sensor fixed to the abdominal skin. During the same period the RF was continuously measured. After the experiments were completed, the animals were killed by a lethal dose of pentobarbital.

**Experiments at 41 bar**

Before compression, 0.1 bar O₂ was added. In series 3 \( (n = 8) \) and 4 \( (n = 8) \) the animals were compressed at a rate of 0.3 bar/min by adding pure He to the pressure chamber until 41 bar was reached. After an adaptation period of 10 min at 41 bar, the rats were given the same amount of morphine (series 3) and fentanyl (series 4) with the same rate of infusion as in series 1 and 2. The PrCO₂ and RF were monitored continuously for 35 min after drug infusion as described for the 1-bar experiments. After the measurements were completed the animals were killed by rapid decompression. Statistics were performed by Wilcoxon's signed rank and Student's t test; \( P \) values less than 0.05 were considered significant.

**RESULTS**

**Control experiments at 1 bar**

All animals seemed calm and relaxed during the 2 h they were restrained in the experimental situations, indicating that the habituation program was sufficient. After infusion of either morphine or fentanyl, the animals seemed drowsy, whether at 1 bar or at 41 bar.
Series 1. Effect of morphine at 1 bar

At 1 bar the $P_{\text{rCO}_2}$ and RF remained stable for the first 2 h at $6.20 \pm 0.25$ (mean ± SEM), and $94 \pm 5$ min, respectively (Fig. 2 A). Morphine, 7 mg/kg, was then infused i.v. at a rate of 0.4 ml/min for 6.5 min. The rats seemed drowsy before the infusion was completed. The $P_{\text{rCO}_2}$ began increasing $3.2 \pm 0.34$ min after the start of the infusion and reached a maximal value ($1.35 \pm 0.13$ kPa increase) after $24.7 \pm 2.2$ min. The $P_{\text{rCO}_2}$ decreased gradually during the rest of the experiment (Fig. 3 A). The RF fell ($P < 0.05$) during the first 5 min of the morphine infusion (Fig. 2 A) and remained below control value for the rest of the experimental period.

---

Fig. 2. A, RF/min (mean ± SEM) before and after infusion of morphine at 1 and 41 bar. B, RF/min (mean ± SEM) before and after infusion of fentanyl at 1 and 41 bar. Asterisk denotes significant differences between RF at 1 and 41 bar. * = $P < 0.05$, ** = $P < 0.01$. Plus signs denote significant differences in RF before and after (5 min) start of morphine or fentanyl infusion at 1 or 41 bar. (+ = $P < 0.05$, ++ = $P < 0.01$).
Series 2. Effects of fentanyl at 1 bar

The $P_{TCO_2}$ and RF remained stable ($6.1 \pm 0.27$ kPa and $95 \pm 2$/min) during the 2 h in the chamber before infusion (Figs. 2 B and 3 B). Fentanyl 0.025 mg/kg was then given i.v. The rats seemed drowsy before the infusion was completed. The $P_{TCO_2}$ began increasing $2.4 \pm 0.35$ min after the start of the infusion, and reached its maximal increase of $1.60 \pm 0.17$ kPa $15.4 \pm 0.6$ min after start of infusion. After reaching its maximum the $P_{TCO_2}$ fell gradually during the experiments (Fig. 3 B). The RF fell ($P$
< 0.01) within 5 min after the fentanyl infusion (Fig. 2 B), but was not significantly changed 10 min later and during the rest of the experiment, compared to control RF.

**Measurements during compression**

The animals also seemed relaxed during and after compression to 41 bar. During the compression phase an increase of PtcCO₂ of 0.48 ± 0.16 and 0.54 ± 0.11 kPa was observed in experimental groups 3 and 4, respectively. The RF decreased during compression by 15–20/min (P < 0.05) in the 2 groups of animals (Fig. 2 A and B), possibly due to the increased breathing gas density at 41 bar.

**Series 3. Morphine experiments at 41 bar**

After a compression to 41 bar (2 h) and a stabilization period of 10 min, the PtcCO₂ was 6.35 ± 0.10 kPa and RF was 76 ± 6.4. Morphine was then infused i.v. at 41 bar at the same rate and in the same amount as at 1 bar. The PtcCO₂ began to increase 4.3 ± 0.3 min after the start of the infusion (Fig. 3 A) and had reached its maximal elevation of 1.26 ± 0.07 kPa after 24.5 ± 2.0 min. RF decreased after 5 min, but not significantly (Fig. 2 A), and remained at this level during the hyperbaric exposure.

**Series 4. Fentanyl experiments, 41 bar**

Fentanyl was infused i.v. at 41 bar after compression (2 h) and a stabilization period of 10 min. PtcCO₂ and RF were 6.24 ± 0.24 kPa and 80 ± 3.3/min before the infusion. The PtcCO₂ started to increase 2.1 ± 0.3 min after the start of the infusion (Fig. 3 B) and had reached the maximal kPa increase of 2.04 ± 0.19 after 15.3 ± 1.3 min. RF fell significantly (P < 0.05) during the first 5 min and remained low for 5–10 min (Fig. 2 B), then returned to the same frequency as before the infusion.

**DISCUSSION**

**Comparison of the action of fentanyl and morphine**

The present results agree with earlier studies on fentanyl and morphine (8, 12–15). It is generally accepted that the action of fentanyl is more rapid than morphine and is of shorter duration. The PtcCO₂ values increased faster in the fentanyl groups than in the morphine groups, independent of the ambient pressure. The respiratory depression assessed by RF reached its maximum at 5 min for both drugs in all groups, but subsided more rapidly in the fentanyl series.

**Effect of fentanyl and morphine during hyperbaric exposure**

Only limited information is available on the analgesic actions of morphine and fentanyl during pressure exposure. Greenbaum and Evans (5) found that the analgesic effect of morphine at 600 feet of seawater was the same as at surface when tested in mice by the hotplate method. No reduction in the analgesic effect was found in a
recent report (7) using the formalin test in rats at 48 bar. The pharmacokinetics of morphine in rats is not altered by high pressure (16).

Doses of morphine and fentanyl used in the present study are equivalent to the ED$_{50}$ doses used in the tail withdrawal test described by Janssen et al. (8).

We have recently shown that measurements of transcutaneous PCO$_2$ can be performed using a CO$_2$ electrode applied on the skin of rats during hyperbaric exposure at least to 41 bar (10). This method is accurate in determining PCO$_2$ alterations in the tissue as well as atmospheric PCO$_2$ in hyperbaric environments up to 41 bar. A high correlation between Pr$_{TCO_2}$ and PCO$_2$ in arterial blood (0.93) was also found at 1 bar in awake rats (10) and humans (17, 18). The efficiency of the electrode to detect abrupt changes in ventilation was documented in our previous study (10).

Side effects of strong analgesics are well known, respiratory depression being the most serious. This respiratory depression first causes an increase of the arterial CO$_2$, followed by increased tissue CO$_2$ concentration and increased arterial bicarbonate. The results of the present study indicate that the effect on the respiratory center is of a similar degree for the given doses of the two drugs, but the onset of the respiratory depression is different. The effect of fentanyl was detected much sooner than the effect of morphine, which agrees with previous reports describing the effect on animals (8, 12, 13) and man (14, 15). The fall in the RF was most pronounced after 5 min in both groups at pressure as well as at 1 bar. RF was lower at 41 than at 1 bar before drug infusion, probably due to increased breathing gas density, because using high-density breathing, RF has been shown to decrease during pressure exposure as well as at 1 bar (19). The increase in Pr$_{TCO_2}$ reached its maximum value after 15 min in the fentanyl groups, whereas the maximal changes in Pr$_{TCO_2}$ after morphine infusion occurred later (after 25 min) and was slightly less pronounced. The fall in respiratory frequency is also comparable for the two drugs, which indicates that the depressant effect of morphine and fentanyl on the CNS is similar at normal and high ambient pressure. This conclusion is strengthened by the finding that the time for maximal respiratory depression measured by Pr$_{TCO_2}$ was the same in the 1- and 41-bar groups.

In conclusion, the present data indicate that the drugs fentanyl and morphine exert the same degree of respiratory depression at 1 and 41 bar in rats. Further studies in humans may be warranted.

---

We thank Anette Leganger, Gerd S Salvesen, Otto Furset, Diacor AS, Oslo and Janssen Pharma AS, Oslo, for technical assistance.

This study was supported by grants from The Norwegian Research Council for Science and the Humanities (The Hyperbaric Medical Research Program; grant 13.91.99-116).—Manuscript received July 1988; accepted February 1989.

REFERENCES


