Respiratory Depressant Effects of Fentanyl Analogs

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ABSTRACT

Aim: Opioid-related fatalities involving synthetic narcotics have reached unprecedented levels. This study evaluated the respiratory depressant effects of seven fentanyl analogs that have either emerged in the recreational drug marketplace or been identified in toxicological analyses following fatal or non-fatal intoxications and for which their effects on ventilation had not been previously characterized.

Methods: Adult male Swiss Webster mice (n = 8 per group) were administered fentanyl analogs (isobutyrylfentanyl, crotonylfentanyl, *para*-methoxyfentanyl, *para*-methoxybutyrylfentanyl, 3-furanylfentanyl, thiophenefentanyl, benzodioxolefentanyl) and their effects on respiratory rate, tidal volume, and minute volume as compared to mu-opioid-receptor (MOR) agonist standards (fentanyl, morphine, and buprenorphine) were measured using whole-body plethysmography (WBP).

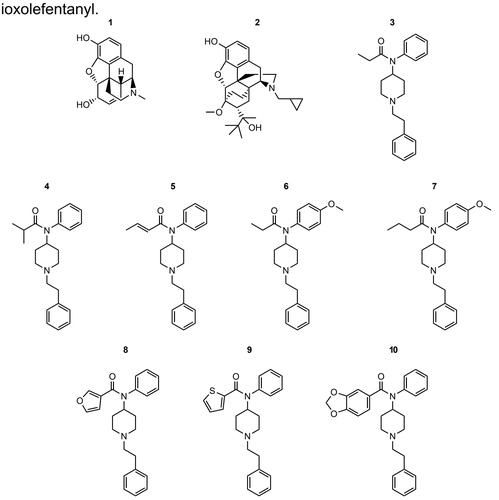
Results: All compounds elicited significant ($p \leq 0.05$) hypoventilation relative to vehicle at least at one dose tested: morphine (1, 3.2, 10, 32 mg/kg), buprenorphine, (0.032, 0.1, 0.32, 1, 3.2 mg/kg), fentanyl (0.01, 0.032, 0.1, 1, 32 mg/kg), isobutyrylfentanyl (0.1, 0.32, 1, 3.2, 10 mg/kg), crotonylfentanyl (0.1, 0.32, 1, 3.2, 10 mg/kg), *para*-methoxyfentanyl (0.1, 0.32, 1, 3.2, 10 mg/kg), *para*-methoxybutyrylfentanyl (0.32, 1, 3.2, 10 mg/kg), 3-furanylfentanyl (0.1, 0.32, 1, 3.2, 10 mg/kg), thiophenefentanyl (1, 3.2, 10, 32, 100 mg/kg), and benzodioxolefentanyl (3.2, 10, 32, 100 mg/kg). The ED_{50} values for hypoventilation showed a rank order of potency as follows: fentanyl (ED_{50} = 0.96 mg/kg) > 3-furanylfentanyl (ED_{50} = 2.60 mg/kg) > crotonylfentanyl (ED_{50} = 2.72 mg/kg) > *para*-methoxyfentanyl (ED_{50} = 3.31 mg/kg) > buprenorphine (ED_{50} = 10.8 mg/kg) > isobutyrylfentanyl (ED_{50} = 13.5 mg/kg) > *para*-methoxybutyrylfentanyl (ED_{50} = 16.1 mg/kg) > thiophenefentanyl (ED_{50} = 18.0 mg/kg) > morphine (ED_{50} = 55.3 mg/kg) > benzodioxolefentanyl (ED_{50} = 10168 mg/kg). A naloxone pretreatment (10 mg/kg) attenuated the hypoventilatory effects of all drugs.

Conclusions: These results establish that the respiratory depressant effects of these fentanyl analogs are at least in part mediated by opioid receptors.

METHODS

Whole-Body Plethysmography: Subjects were adult male Swiss Webster mice and their ventilatory parameters were assessed under 5% CO2 and reverse-light dark conditions. A cumulative dosing design was employed similar to that which was previously described (1). All drugs were administered via the subcutaneous route.

Figure 1. Structures of drugs tested: **(1)** morphine, **(2)** buprenorphine, **(3)** fentanyl, **(4)** isobutyrylfentanyl, **(5)** crotonylfentanyl, **(6)** *para*-methoxyfentanyl, **(7)** *para*-methoxybutyrylfentanyl, **(8)** 3-furanylfentanyl, **(9)** thiophenefentanyl, **(10)** benzodioxolefentanyl.



RESULTS

Morphine, Buprenorphine, and Fentanyl Elicit Respiratory Depression in Mice and Naloxone Attenuates Their Effects

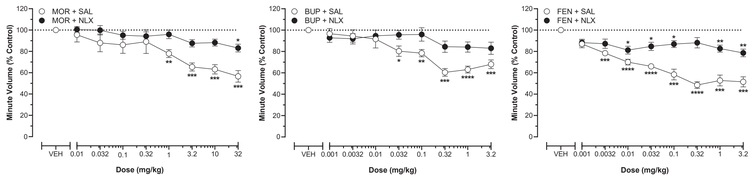


Figure 2. Results from cumulative dose ventilation tests for positive controls (n = 8–16 mice per group). Minute volume as a percentage of control (± SEM) is shown for **(1)** morphine [MOR], **(2)** buprenorphine [BUP], and **(3)** fentanyl [FEN] (left to right) in the presence (filled circles) and absence (unfilled circles) of 10 mg/kg naloxone pretreatment. Significant differences between a drug's dose and within-subject vehicle condition are indicated by asterisks: * $p \leq 0.05$, ** $p \leq 0.01$, *** $p \leq 0.001$, **** $p \leq 0.0001$. SEM: standard error of the mean.

Fentanyl Analogs Elicit Respiratory Depression in Mice and Naloxone Attenuates Their Effects

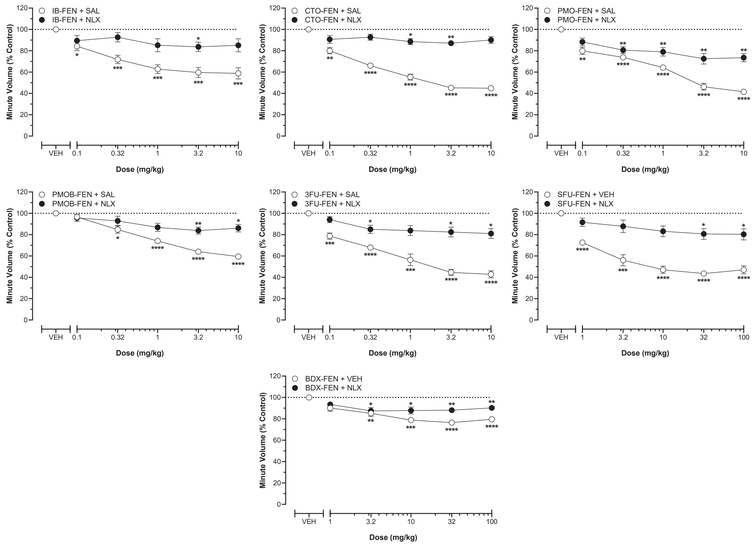


Figure 3. Results from cumulative dose ventilation tests for analogs of fentanyl (n = 8 mice per group). Minute volume as a percentage of control (± SEM) is shown for **(4)** isobutyrylfentanyl [IB-FEN], **(5)** crotonylfentanyl [CTO-FEN], **(6)** *para*-methoxyfentanyl [PMO-FEN], **(7)** *para*-methoxybutyrylfentanyl [PMOB-FEN], **(8)** 3-furanylfentanyl [3FU-FEN], **(9)** thiophenefentanyl [SFU-FEN], **(10)** benzodioxolefentanyl [BDX-FEN] in the presence (filled circles) and absence (unfilled circles) of 10 mg/kg naloxone pretreatment. Significant differences between a drug's dose and within-subject vehicle condition are indicated by asterisks: * $p \leq 0.05$, ** $p \leq 0.01$, *** $p \leq 0.001$, **** $p \leq 0.0001$. SEM: standard error of the mean.

#	Drug	Abbr.	%E_{max} ± SEM	Hypoventilation TD_{50} (mg/kg)	Potency Ratio (Morphine)	Potency Ratio (Fentanyl)	[1]Antinociception ED_{50} (mg/kg)	Protective Index = TD_{50}/ED_{50}
1	Morphine	MOR	56.6 ± 5.42	55.3 (19.3–393)	1.00	0.02	7.82 (5.42–11.0)	7.07
2	Buprenorphine	BUP	60.5 ± 3.65	10.8 (3.61–70.8)	5.10	0.09	0.11 (0.07–0.17)	102
3	Fentanyl	FEN	51.6 ± 4.73	0.96 (0.50–2.24)	57.6	1.00	0.08 (0.04–0.16)	12.0
4	Isobutyrylfentanyl	IB-FEN	58.8 ± 5.30	13.5 (6.28–46.8)	4.10	0.07	0.08 (0.04–0.13)	176
5	Crotonylfentanyl	CTO-FEN	44.9 ± 2.30	2.72 (1.99–3.88)	20.3	0.35	0.23 (0.18–0.29)	12.1
6	*Para*-methoxyfentanyl	PMO-FEN	41.5 ± 2.53	3.31 (2.52–4.52)	16.7	0.29	0.43 (0.23–0.77)	7.74
7	*Para*-methoxybutyrylfentanyl	PMOB-FEN	59.5 ± 2.56	16.1 (10.9–27.1)	3.44	0.06	0.11 (0.05–0.20)	152
8	3-Furanylfentanyl	3FU-FEN	42.9 ± 3.34	2.60 (1.80–3.94)	21.4	0.37	0.51 (0.36–0.74)	5.05
9	Thiophenefentanyl	SFU-FEN	43.5 ± 2.38	18.0 (10.8–32.9)	3.08	0.05	4.66 (3.65–5.95)	3.85
10	Benzodioxolefentanyl	BDX-FEN	76.5 ± 1.98	10168 (1790–276521)	0.0054	0.000094	46.3 (25.8–83.4)	220

Table 1. Results from cumulative dose ventilation tests. Efficacy estimates are expressed as %E_{max} (maximum suppression as percentage of control; the smaller the percentage, the greater the suppression--i.e. 100% = zero suppression, 0% = complete suppression). Potency estimates are expressed as TD_{50} (mg/kg) values for drug alone, drug potency ratio to morphine, and drug potency ratio to fentanyl. Safety estimates are expressed as protective indices (PI = TD_{50}/ED_{50}). Data are mean ± SEM or 95% confidence intervals for n = 8 mice per group. SEM: standard error of the mean. TD_{50}: half-maximal toxic dose. ED_{50}: half-maximal effective dose.

Respiratory Depressant Effects of Fentanyl Analogs Are Opioid Receptor Mediated

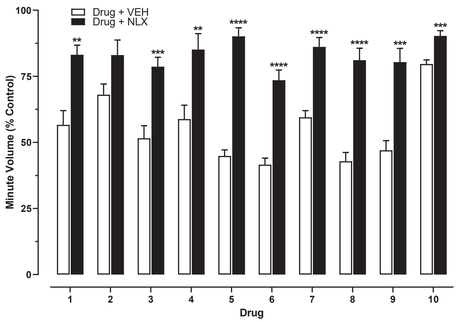


Figure 4. Results from cumulative dose ventilation tests for **(1)** 32 mg/kg morphine, **(2)** 3.2 mg/kg buprenorphine, **(3)** 3.2 mg/kg fentanyl, **(4)** 10 mg/kg isobutyrylfentanyl, **(5)** 10 mg/kg crotonylfentanyl, **(6)** 10 mg/kg *para*-methoxyfentanyl, **(7)** 10 mg/kg *para*-methoxybutyrylfentanyl, **(8)** 10 mg/kg 3-furanylfentanyl, **(9)** 100 mg/kg thiophenefentanyl, **(10)** 100 mg/kg benzodioxolefentanyl accompanied by a pretreatment of either 10 mg/kg naloxone [NLX; filled bars] or saline [VEH; open bars] and the vehicle of the test drug. Each bar represents minute volume (± SEM) as a percentage of control for n = 8 mice per group. Significant differences between agonist + saline (open bars) vs agonist + 10 mg/kg naloxone (filled bars) condition are indicated by asterisks: ** $p \leq 0.01$, *** $p \leq 0.001$, **** $p \leq 0.0001$. SEM: standard error of the mean.

REFERENCES

(1) Varshneya, N. B., Walentiny, D. M., Moisa, L. T., Walker, T. D., Akinfiresoye, L. R., Beardsley, P. M., 2019. Opioid-like antinociceptive and locomotor effects of emerging fentanyl-related substances. Neuropharmacology 151, 171-179.

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