Effect of paroxetine or quetiapine combined with oxycodone vs oxycodone alone on ventilation during hypercapnia: a randomized clinical trial

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Effect of Paroxetine or Quetiapine Combined With Oxycodone vs Oxycodone Alone on Ventilation During Hypercapnia
A Randomized Clinical Trial

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IMPORTANCE Opioids can cause severe respiratory depression by suppressing feedback mechanisms that increase ventilation in response to hypercapnia. Following the addition of boxed warnings to benzodiazepine and opioid products about increased respiratory depression risk with simultaneous use, the US Food and Drug Administration evaluated whether other drugs that might be used in place of benzodiazepines may cause similar effects.

OBJECTIVE To study whether combining paroxetine or quetiapine with oxycodone, compared with oxycodone alone, decreases the ventilatory response to hypercapnia.

DESIGN, SETTING, AND PARTICIPANTS Randomized, double-blind, crossover clinical trial at a clinical pharmacology unit (West Bend, Wisconsin) with 25 healthy participants from January 2021 through May 25, 2021.

INTERVENTIONS Oxycodone 10 mg on days 1 and 5 and the following in a randomized order for 5 days: paroxetine 40 mg daily, quetiapine twice daily (increasing daily doses from 100 mg to 400 mg), or placebo.

MAIN OUTCOMES AND MEASURES Ventilation at end-tidal carbon dioxide of 55 mm Hg (hypercapnic ventilation) using rebreathing methodology assessed for paroxetine or quetiapine with oxycodone, compared with placebo and oxycodone, on days 1 and 5 (primary) and for paroxetine or quetiapine alone compared with placebo on day 4 (secondary).

RESULTS Among 25 participants (median age, 35 years [IQR, 30-40 years]; 11 female [44%]), 19 (76%) completed the trial. The mean hypercapnic ventilation was significantly decreased with paroxetine plus oxycodone vs placebo plus oxycodone on day 1 (29.2 vs 34.1 L/min; mean difference [MD], −4.9 L/min [1-sided 97.5% CI, −∞ to −0.6]; $P = .01$) and day 5 (25.1 vs 35.3 L/min; MD, −10.2 L/min [1-sided 97.5% CI, −∞ to −6.3]; $P < .001$) but was not significantly decreased with quetiapine plus oxycodone vs placebo plus oxycodone on day 1 (33.0 vs 34.1 L/min; MD, −1.2 L/min [1-sided 97.5% CI, −∞ to 2.8]; $P = .28$) or on day 5 (34.7 vs 35.3 L/min; MD, −0.6 L/min [1-sided 97.5% CI, −∞ to 3.2]; $P = .37$). As a secondary outcome, mean hypercapnic ventilation was significantly decreased on day 4 with paroxetine alone vs placebo (32.4 vs 41.7 L/min; MD, −9.3 L/min [1-sided 97.5% CI, −∞ to −3.9]; $P < .001$), but not with quetiapine alone vs placebo (42.8 vs 41.7 L/min; MD, 1.1 L/min [1-sided 97.5% CI, −∞ to 6.4]; $P = .67$). No drug-related serious adverse events were reported.

CONCLUSIONS AND RELEVANCE In this preliminary study involving healthy participants, paroxetine combined with oxycodone, compared with oxycodone alone, significantly decreased the ventilatory response to hypercapnia on days 1 and 5, whereas quetiapine combined with oxycodone did not cause such an effect. Additional investigation is needed to characterize the effects after longer-term treatment and to determine the clinical relevance of these findings.

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Supplemental content

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Ventilation in humans is tightly controlled by feedback mechanisms involving carbon dioxide.[1,2] When chemical receptors in the brain and the carotid body sense increased carbon dioxide, ventilation increases to remove carbon dioxide from the body.[1,2] Opioids decrease this ventilatory response to hypercapnia (**Figure 1**),[2-5] which can lead to severe respiratory depression and death.[6] Some other drugs, such as benzodiazepines, have minimal effects on ventilation on their own at standard doses, but can exacerbate opioid-induced respiratory depression.[7]

In 2016, the US Food and Drug Administration (FDA) required that drug labeling for benzodiazepines and opioids include boxed warnings about increased potential for respiratory depression with their simultaneous use.[7] Following this labeling change, the FDA took proactive steps to review whether other drugs that might be used in place of benzodiazepines (as prescribed or off-label) may exacerbate opioid-induced respiratory depression and conducted in vivo rat studies with 14 drugs from diverse pharmacological classes.[8] The selective serotonin reuptake inhibitor (SSRI) paroxetine and the atypical antipsychotic quetiapine exacerbated oxycodone-induced respiratory depression.[8] To further investigate these findings, this clinical trial involving healthy participants assessed whether paroxetine-oxycodone or quetiapine-oxycodone combinations decreased the ventilatory response to hypercapnia compared with oxycodone alone.

Key Points

Question Do specific psychotropic drug–opioid combinations further decrease the ventilatory response to hypercapnia beyond the respiratory depression caused by opioids alone?

Findings In this randomized crossover clinical trial that included 25 healthy participants, paroxetine plus oxycodone significantly decreased mean hypercapnic ventilation compared with placebo plus oxycodone on day 1 (29.2 vs 34.1 L/min, respectively) and day 5 (25.1 vs 35.3 L/min, respectively), whereas quetiapine plus oxycodone did not significantly decrease mean hypercapnic ventilation compared with placebo plus oxycodone on day 1 (33.0 vs 34.1 L/min, respectively) or day 5 (34.7 vs 35.3 L/min, respectively).

Meaning Further research is needed to determine the clinical relevance of the finding of decreased ventilation during hypercapnia when paroxetine is combined with oxycodone under experimental conditions.

Methods

Study Setting and Dates

A randomized, double-blind, 3-way crossover trial involving healthy participants at a clinical pharmacology unit (Spaulding Clinical Research, West Bend, Wisconsin) from January to May 2021 evaluated the effects of paroxetine or quetiapine combined with oxycodone, compared with oxycodone alone, on the ventilatory response to hypercapnia (Figure 1). The Advarra Institutional Review Board approved this study (https://www.advarra.com). All participants provided written informed consent. The protocol and statistical analysis plan are available in Supplement 1.

Participants and Randomization

Participants were recruited by standard approaches for healthy volunteer clinical pharmacology studies (ie, online advertising and emails or texts to individuals in the site's database). Self-identified race and ethnicity were collected in an open-ended format by clinical staff as recommended by the FDA's guidance document *Collection of Race and Ethnicity Data in Clinical Trials*.[9] Key inclusion criteria were ages 18 to 50 years, nonsmoking, and negative test results for alcohol or illicit drugs. Participants were excluded if they had a history of sleep disorders, panic disorder, panic attacks, generalized anxiety disorder, hypoventilation syndrome, or sleep apnea; used opioid or psychotropic drug within 60 days of the study start; had a Mallampati score (predicts difficult tracheal intubation) greater than 2; or could not tolerate the ventilatory assessment procedure during screening.

Participants were randomized to 1 of 6 treatment sequences (Figure 1) using a random number generator in R statistical software. Randomization was conducted in block sizes of 6 for the first 18 participants, and the remaining 2 participants were randomly assigned in 2 of the 6 treatment sequences. Replacement participants were assigned to the treatment sequence of the participant they replaced.

Study Procedures and Interventions

Participants checked in to the clinic the day before the study started, received study drugs on days 1 through 5 (oxycodone on days 1 and 5, and paroxetine or quetiapine [or matched placebos] on days 1 through 5), and checked out on day 6. (See Figure 1 for study drug dosing details.) This was repeated twice with 7 days of washout between periods. Study drugs were administered to align the time of maximum concentration for all drugs at the 5-hour time point (paroxetine at 0 hours, oxycodone at 3 hours, quetiapine at 3 and 14 hours).Each period included 16 ventilatory assessments (0 [predose], 4, 5, 6, 8, and 24 hours on days 1 and 5 and 0, 4, 5, and 6 hours on day 4) and 26 blood samples (0 [predose], 3, 4, 5, 6, 8, 9, 12, and 24 hours on days 1 and 5 and 0, 3, 4, 5, 6, 8, 9, and 12 hours on day 4). Plasma concentrations of paroxetine, quetiapine, oxycodone, and selected metabolites were measured by validated liquid chromatography and tandem mass spectrometry (eMethods 1 in Supplement 2).

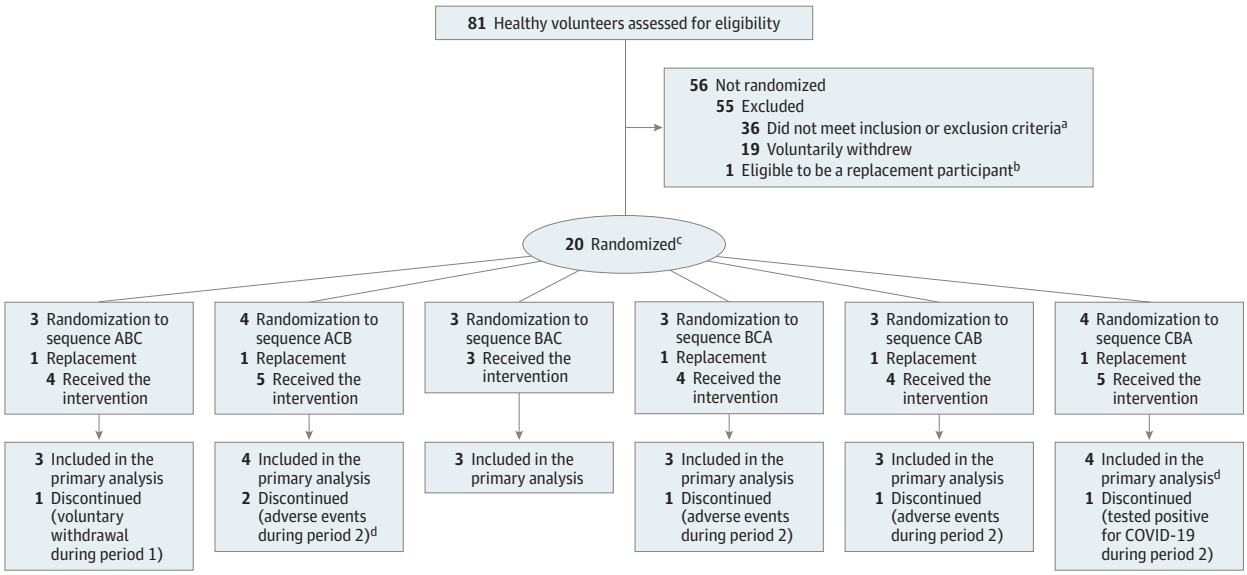
Participant safety was monitored with clinical laboratory tests, vital signs, electrocardiograms, and physical examinations. Continuous pulse oximetry and telemetry were performed on days when oxycodone was administered, and naloxone was available for participants with severe respiratory depression. Criteria for discontinuation of the study drugs included apnea defined as discontinuation of rhythmic breathing for more than 90 seconds, end-tidal carbon dioxide higher than 67.5 mm Hg, or oxygen saturation less than 85% lasting more than 2 minutes.

Ventilatory Assessments

During each assessment, participants sat in an upright position with a fitted mask attached to a pneumotachometer and

Figure 1. Flow of Participants in Study, Interventions and Overall Study Design

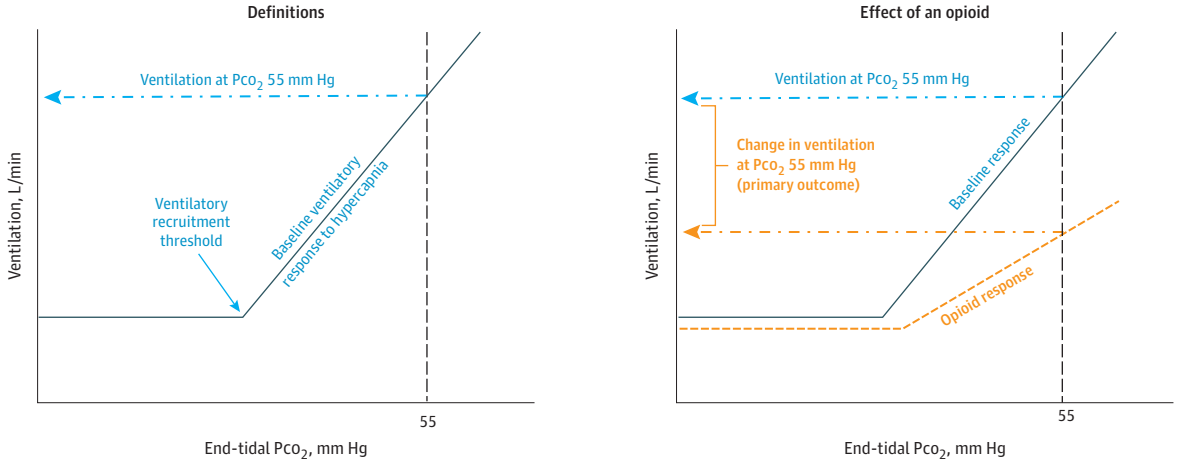
A Flow of participants in the study



B Study drug interventions

Treatment group	Study drugs[e]				
	Day 1	Day 2	Day 3	Day 4	Day 5
A	Placebo + oxycodone 10 mg	Placebo	Placebo	Placebo	Placebo + oxycodone 1 mg
B	Paroxetine 40 mg + oxycodone 20 mg	Paroxetine 40 mg	Paroxetine 40 mg	Paroxetine 40 mg	Paroxetine 40 mg + oxycodone 20 mg
C	Quetiapine 50 mg 2/d + oxycodone 10 mg	Quetiapine 100 mg 2/d	Quetiapine 150 mg 2/d	Quetiapine 200 mg 2/d	Quetiapine 200 mg + oxycodone 10 mg

C Illustration of the ventilatory response to hypercapnia at baseline[f]



[a] Ten participants had a Mallampati score greater than 2, (predicts difficult tracheal intubation); 5, deemed unlikely to comply with protocol; 5, tested positive for alcohol or illicit drugs; 7, abnormal medical history, laboratory results, or physical examination findings.

[b] Participant was not needed as a replacement.

[c] Five participants replaced the 6 who did not complete all treatment periods. The study design planned for 5 replacements.

[d] One participant was included in the primary analysis for only day 1, after which the participant discontinued.

[e] See the Methods section for timing of study drug administration. Participants received 4 mg of ondansetron 30 minutes before each dose of oxycodone on days 1 and 5 only to prevent nausea and vomiting.

[f] Ventilation increases at an approximately linear rate after carbon dioxide (P_{CO_2}) is higher than the ventilatory recruitment threshold (VRT). The opioid causes small decreases in ventilation below the VRT, shifts the VRT to the right, and decreases the rate of rise in ventilation as P_{CO_2} increases further.[2-4,6]

went through preparatory steps of relaxed breathing (5 minutes of room air then 3 minutes of 100% oxygen), hyperventilation to decrease end-tidal carbon dioxide (1-2 minutes 100% oxygen), followed by rebreathing.[10,11] Upon switching the circuit to the rebreathing bag (7% carbon dioxide, 93% oxygen), participants were instructed to take 3 deep breaths and then breathe normally. This causes approximate equilibration of carbon dioxide in mixed venous blood, arterial blood, brain, and lung with the rebreathing bag.[1,11] Subsequently, carbon dioxide increases at an approximately linear rate as exhaled carbon dioxide is rebreathed through the closed circuit, which increases ventilation above a certain carbon dioxide threshold (Figure 1).[1,12] The procedure continued until end-tidal carbon dioxide was approximately 55 mm Hg (see eMethods 2 in Supplement 2 for additional details). Rebreathing data were reviewed by 2 independent assessors blinded to study treatment and time of assessments to evaluate completeness of data for study outcomes (statistical analysis plan in Supplement 1 and eTable 1 in Supplement 2). Deidentified participant data are available in Supplement 3 and Supplement 4. The eDictionary in Supplement 2 contains a list of variable names and definitions for the data sets.

Outcomes and Sample Size Calculation

The primary end point was the minute ventilation when end-tidal carbon dioxide was 55 mm Hg (Figure 1), which has been used in prior drug-induced respiratory depression studies.[4,5] The primary outcome comparisons were performed between paroxetine or quetiapine combined with oxycodone vs placebo combined with oxycodone, assessed separately on days 1 and 5. Day 1 was included because quetiapine can cause more sedation after 1 dose than after 5 days of dosing,[13] and it was not known if a similar pattern would be observed with ventilation. Comparisons between paroxetine or quetiapine alone vs placebo on day 4 were secondary outcomes.

Additional secondary outcomes included the maximum plasma concentration and area under the curve (AUC) for plasma concentration vs time of oxycodone when combined with paroxetine or quetiapine compared with oxycodone with placebo. Multiple exploratory outcomes (eTable 2 in Supplement 2) were assessed as specified in the protocol and statistical analysis plan, including pharmacokinetic parameters for paroxetine and quetiapine, additional respiratory measurements including during relaxed room-air breathing, sedation assessments, and pharmacokinetic-pharmacodynamic (concentration-response) modeling. Although reporting summary statistics for exploratory outcomes was prespecified, comparisons between study treatments for the exploratory outcomes were a post hoc assessment. In addition, study drug maximum plasma concentration and AUC were compared based on cytochrome-P450 2D6 (CYP2D6) metabolizer phenotype status as a post hoc assessment.

Sample size requirements were calculated based on 2 primary outcomes (day 1 and day 5) and adjusted for multiplicity ($\alpha = .025$). The assessments with paroxetine or quetiapine were considered as separate experiments. A sample size of 20 participants was determined to have 90% power at a 1-sided significance level to detect a 4-L/min decrease in the primary end point (ventilation at 55 mm Hg end-tidal carbon dioxide) assuming a standard deviation of 5 L/min, based on prior opioid ventilatory studies.[4,5] A 4-L/min decrease was the estimated approximate effect size from 10 mg of oxycodone and would indicate that paroxetine or quetiapine was further decreasing hypercapnic ventilation by a similar amount.[4,5] The protocol allowed for enrollment of up to 5 replacement participants to account for discontinuations.

Statistical Analysis

All participants who completed paired rebreathing assessments with placebo plus oxycodone and at least 1 of the other 2 study treatments (paroxetine plus oxycodone or quetiapine plus oxycodone) for day 1 or day 5 were included in the primary analysis without imputation of missing data. Study treatments were compared using a linear mixed-effects model with baseline ventilation at an end-tidal carbon dioxide of 55 mm Hg as a continuous variable; treatment, sequence, and period as categorical variables; and participant as a random effect. A similar analysis was performed on day 4 as a secondary outcome. For pharmacokinetic analyses, all concentrations less than the lower limit of quantitation were considered 0. Maximum oxycodone concentration and AUC were log-transformed and the values between study treatments were compared using a linear mixed-effects model on days 1 and 5 with treatment as a categorical variable and participant as a random effect. Pharmacokinetic-pharmacodynamic modeling included drug concentration as a continuous variable and random effects by participant on the intercept and concentration variable (eMethods 3 in Supplement 2). Demographics are reported with standard descriptive statistics.

A 1-sided *P* value was used to assess the primary outcomes because the study aim was to evaluate whether the study drugs decreased ventilation, and a value <.025 was considered significant based on Bonferroni correction for 2 primary outcomes. A 1-sided *P* value <.025 was also considered significant for the secondary ventilation outcome, and these outcomes are reported with 1-sided upper 97.5% CIs. For secondary and exploratory outcomes assessing pharmacokinetics, a difference in exposure was concluded if the 2-sided 90% CI of the geometric mean ratio [GMR] excluded 1, which is standard in pharmacokinetic studies.[14] Post hoc comparisons are reported with 2-sided 95% CIs and a difference was reported if the CIs excluded 0. Secondary and exploratory CIs are not adjusted for multiplicity, and all analyses except for primary outcomes should be interpreted as exploratory because of the potential for type I error due to multiple comparisons. Statistical analyses were performed in R (version 4.1.2; The R Project for Statistical Computing).

Results

Study Participants

Twenty-five participants (20 originally randomized and 5 replacement participants; Figure 1) were enrolled (median age, 35 years [IQR, 30 to 40 years]; 11 female [44%]). **Table 1** contains additional participant characteristics, including resting

respiratory measurements and CYP3A4 and CYP2D6 metabolizer phenotypes. Nineteen participants completed the trial and 1 additional participant completed through day 1 of period 2 and had placebo plus oxycodone data available (Figure 1). Primary outcomes data were available for 20 participants on day 1 and 19 participants on day 5.

Primary Outcomes

The mean ventilation at 55 mm Hg end-tidal carbon dioxide with the paroxetine plus oxycodone combination on day 1 was 29.2 L/min (95% CI, 25.7 to 32.7); with quetiapine plus oxycodone, 33.0 L/min (95% CI, 30.0 to 36.0); with placebo plus oxycodone, 34.1 L/min (95% CI, 31.1 to 37.2). The day 5 values were 25.1 L/min (95% CI, 21.2 to 29.0) with paroxetine plus oxycodone; 34.7 L/min (95% CI, 30.9 to 38.5) with quetiapine plus oxycodone, and 35.3 L/min (95% CI, 31.4 to 39.2) with placebo plus oxycodone (**Table 2**).

Compared with placebo plus oxycodone, paroxetine plus oxycodone significantly decreased ventilation on day 1 (mean difference [MD], −4.9 L/min [1-sided 97.5% CI, −∞ to −0.6]; $P = .01$) and on day 5 (MD, −10.2 L/min [1-sided 97.5% CI, −∞ to −6.3]; $P < .001$), while quetiapine plus oxycodone did not significantly decrease ventilation on day 1 (MD, −1.2 L/min [1-sided 97.5% CI, −∞ to 2.8]; $P = .28$) or day 5 (MD, −0.6 L/min [1-sided 97.5% CI, −∞ to 3.2]; $P = .37$).

Secondary Outcomes

Figure 2 and eFigure 1 in Supplement 2 show pharmacodynamic and pharmacokinetic data across days 1, 4, and 5 (for individual pharmacodynamic data, see eFigures 2 and 3 in Supplement 2). On day 4, the oxycodone administered on day 1 had washed out, allowing for a comparison between effects of paroxetine and quetiapine alone and placebo. Mean ventilation at 55 mm Hg end-tidal carbon dioxide on day 4 was 32.4 L/min (95% CI, 28.2 to 36.5) with paroxetine alone, 42.8 L/min (95% CI, 38.7 to 46.8) with quetiapine alone, and 41.7 L/min (95% CI, 37.7 to 45.6) with placebo (Table 2). Compared with placebo, paroxetine alone significantly decreased ventilation (MD, −9.3 L/min [1-sided 97.5% CI, −∞ to −3.9]; $P < .001$), whereas quetiapine alone did not significantly decrease ventilation (MD, 1.1 L/min [1-sided 97.5% CI, −∞ to 6.4]; $P = .67$).

Paroxetine did not significantly increase oxycodone maximum plasma concentration (GMR, 1.06 [90% CI, 0.96 to 1.17]) or AUC (GMR, 1.03 (90% CI, 0.91 to 1.17) on day 1 but did significantly increase oxycodone maximum plasma concentration (GMR, 1.30 [90% CI, 1.19 to 1.43]) and AUC (GMR, 1.10 [90% CI, 1.02 to 1.19]) on day 5 (Table 2). Quetiapine did not significantly increase oxycodone AUC on day 1 (GMR, 1.06 [90% CI, 0.98 to 1.15]) but did significantly increase oxycodone maximum plasma concentrations on days 1 (GMR, 1.25 [90% CI, 1.14 to 1.37]) and 5 (GMR, 1.39 [90% CI, 1.22 to 1.57]) and AUC on day 5 (GMR, 1.27 [90% CI, 1.19 to 1.36]).

Exploratory Outcomes

Figure 3 displays the oxycodone-alone concentration-response model and the day 5 primary end point observed data for the drug combinations. Multidrug concentration-response analysis (eTable 3 in Supplement 2) showed that increasing concentrations of paroxetine and oxycodone were each associated with decreased hypercapnic ventilation (paroxetine slope, −0.13 L/min per ng/mL [95% CI, −0.17 to −0.09]; oxycodone slope, −0.24 L/min per ng/mL [95% CI, −0.35 to −0.12]), whereas an increasing concentration of quetiapine or its metabolite norquetiapine was not associated with decreased hypercapnic ventilation (quetiapine slope, 0.015 L/min per ng/mL [95% CI, 0.007 to 0.022]; norquetiapine slope, −0.015 L/min per ng/mL [95% CI, −0.038 to 0.001]; oxycodone slope, −0.25 L/min per ng/mL [95% CI, −0.34 to −0.16]).

eFigure 4 in Supplement 2 displays respiratory outcomes when participants were breathing room air at rest (ventilation, respiratory rate, end-tidal carbon dioxide, and oxygen saturation), participant-reported sedation, and slope of the hypercapnic ventilatory response curve on day 5. Comparisons between treatment groups for these outcomes are described in the Post Hoc Assessments section below. Additional exploratory pharmacokinetic (eFigure 5 and eTables 4 and 5) and pharmacodynamic outcomes (eFigure 6, eTables 6 and 7) are displayed in Supplement 2.

Table 1. Study Participant Demographics and Baseline Characteristics

Characteristic	No. (%) (N = 25)
Age, median (IQR), y	35 (30-40)
Sex	
Male	14 (56)
Female	11 (44)
Race, No. (%)[a]	
Asian	2 (8)
Black or African American	12 (48)
White	11 (44)
Hispanic or Latino ethnicity	5 (20)
Body weight, median (IQR), kg	68 (61-81)
BMI, median (IQR)	24.8 (22.0-26.1)
Resting respiratory measurements, median (IQR)	n = 24
Minute ventilation, L/min	7.8 (7.3-9.0)
Respiratory rate, breaths/min	15 (12-17)
Tidal volume, L	0.61 (0.54-0.68)
End-tidal carbon dioxide, mm Hg	37.1 (35.6-39.0)
Oxygen saturation, %	97.1 (95.9-98.0)
CYP3A4 metabolizer phenotype	
Extensive metabolizers	25 (100)
CYP2D6 metabolizer phenotype	
Extensive metabolizers	19 (76)
Intermediate metabolizers	6 (24)

Abbreviations: BMI, body mass index, calculated as weight in kilograms divided by height in meters squared; CYP, cytochrome P450.

[a] Self-identified race and ethnicity were reported by participants in an open-ended format.

Post Hoc Assessments

Compared with placebo plus oxycodone at the 5-hour time point on day 5, paroxetine plus oxycodone increased resting end-tidal carbon dioxide (41.4 vs 37.4 mm Hg; MD, 4.0 mm Hg [95% CI, 2.4 to 5.6 mm Hg]), decreased resting oxygen saturation (95.5% vs 96.6%; MD, −1.1% [95% CI, −2.1% to −0.1%],

Table 2. Primary and Secondary Outcomes

	No. of participants	Mean (2-sided 95% CI)		Mean difference (1-sided 97.5% CI)	*P* value[a]
Primary outcomes					
Ventilation at 55 mm Hg end-tidal Pco_2, L/min		Paroxetine + oxycodone	Placebo + oxycodone		
Day 1	20	29.2 (25.7 to 32.7)	34.1 (31.1 to 37.2)	−4.9 (−∞ to −0.6)	.01
Day 5	19	25.1 (21.2 to 29.0)	35.3 (31.4 to 39.2)	−10.2 (−∞ to −6.3)	<.001
		Quetiapine + oxycodone	Placebo + oxycodone		
Day 1	20	33.0 (30.0 to 36.0)	34.1 (31.1 to 37.2)	−1.2 (−∞ to 2.8)	.28
Day 5	19	34.7 (30.9 to 38.5)	35.3 (31.4 to 39.2)	−0.6 (−∞ to 3.2)	.37
Secondary outcomes					
Ventilation at 55 mm Hg end-tidal Pco_2, L/min		Paroxetine	Placebo		
Day 4	19	32.4 (28.2 to 36.5)	41.7 (37.7 to 45.6)	−9.3 (−∞ to −3.9)	<.001
		Quetiapine	Placebo		
Day 4	19	42.8 (38.7 to 46.8)	41.7 (37.7 to 45.6)	1.1 (−∞ to 6.4)	.67
		GM (CV %)		GMR (2-sided 90% CI)	*P* value[b]
Oxycodone maximum plasma concentration, ng/mL		Paroxetine + oxycodone	Oxycodone + placebo		
Day 1	19	19.1 (25)	18.0 (30)	1.06 (0.96 to 1.17)	.33
Day 5	19	23.4 (26)	18.0 (26)	1.30 (1.19 to 1.43)	<.001
		Quetiapine + oxycodone	Oxycodone		
Day 1	20	22.9 (28)	18.3 (30)	1.25 (1.14 to 1.37)	<.001
Day 5	19	24.9 (26)	18.0 (26)	1.39 (1.22 to 1.57)	<.001
Oxycodone AUC, ng/mL × h		Paroxetine + oxycodone	Oxycodone		
Day 1	20	107 (29)	104 (20)	1.03 (0.91 to 1.17)	.64
Day 5	19	112 (30)	102 (24)	1.10 (1.02 to 1.19)	.05
		Quetiapine + oxycodone	Oxycodone		
Day 1	20	113 (25)	107 (25)	1.06 (0.98 to 1.15)	.24
Day 5	19	129 (23)	102 (24)	1.27 (1.19 to 1.36)	<.001

Abbreviations: AUC, area under the curve; CV, coefficient of variation; GM, geometric mean; GMR, geometric mean ratio; Pco_2, end-tidal partial pressure of carbon dioxide.

[a] A 1-sided *P* value <.025 was considered significant for the primary and secondary ventilation outcomes.

[b] A difference in exposure was concluded if the 2-sided 90% CI of the geometric mean ratio excluded 1 (2-sided *P* < .1).

and decreased the slope of the hypercapnic ventilatory response curve (1.00 vs 1.44 L/min per mm Hg; MD, −0.44 L/min per mm Hg [95% CI, −0.85 to −0.03]); quetiapine plus oxycodone increased resting end-tidal carbon dioxide (40.4 vs 37.4 mm Hg; MD, 3.0 mm Hg [95% CI, 1.4 to 4.6]), decreased resting oxygen saturation (95.2% vs 96.6%; MD, −1.4% [95% CI, −2.4% to −0.4%]), and increased participant-reported sedation (40 vs 25 mm; MD, 15 mm [95% CI, 3 to 28]). Additional post hoc comparisons for exploratory outcomes are shown in eTables 4 through 7 in Supplement 2. eTable 8 in Supplement 2 shows paroxetine, quetiapine, and oxycodone maximum plasma concentration and AUC by CYP2D6 metabolizer phenotype. The 95% CIs of the GMR crossed 1 for all comparisons of CYP2D6 extensive vs intermediate metabolizer status.

Adverse Events

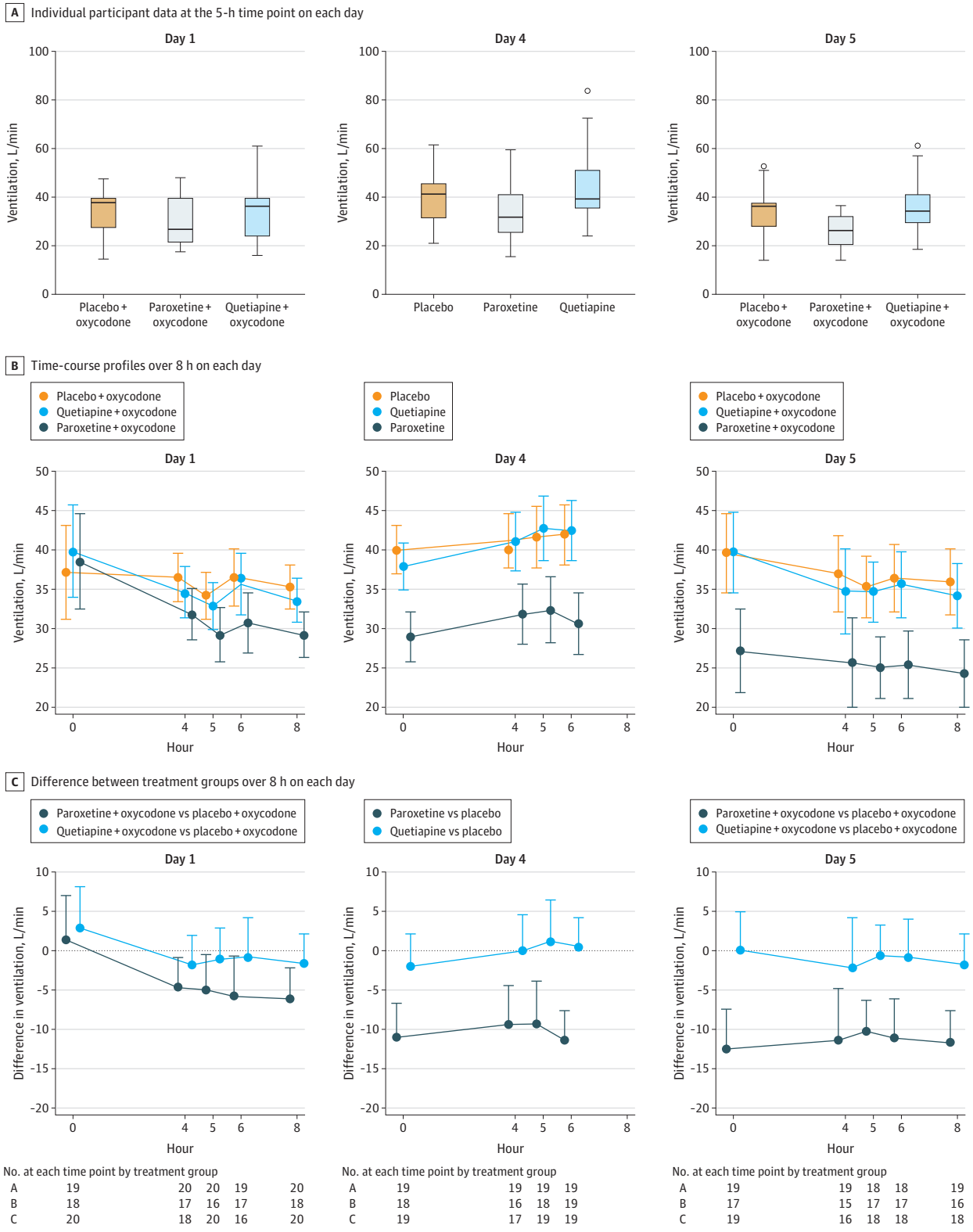
No serious adverse events occurred. Twenty-two participants (88%) experienced 1 or more adverse events. The most commons adverse events were nausea (64%), dizziness (52%), headache (48%), somnolence (32%), and fatigue (32%). eTable 9 in Supplement 2 contains the incidence and number of adverse events by treatment group.

Discussion

In this randomized, double-blind, crossover clinical trial involving healthy participants, paroxetine (40 mg daily for 5 days) combined with oxycodone (10 mg on days 1 and 5) compared with oxycodone alone decreased ventilation when end-tidal carbon dioxide was 55 mm Hg. In contrast, quetiapine (increasing daily doses from 100 mg to 400 mg) combined with oxycodone did not decrease ventilation when end-tidal carbon dioxide was 55 mm Hg.

The finding that paroxetine combined with oxycodone, compared with oxycodone alone, decreased the ventilatory response to hypercapnia is concerning because this is the primary feedback mechanism for the body to rescue itself from opioid-induced respiratory depression.[2,6] The secondary outcomes supported that paroxetine decreased the ventilatory response to hypercapnia through a direct pharmacodynamic effect rather than by a pharmacokinetic interaction because paroxetine had a similar effect on its own compared with placebo on day 4. Furthermore, exploratory concentration-response modeling supported that the increase in oxycodone

Figure 2. Minute Ventilation at End-Tidal Carbon Dioxide of 55 mm Hg

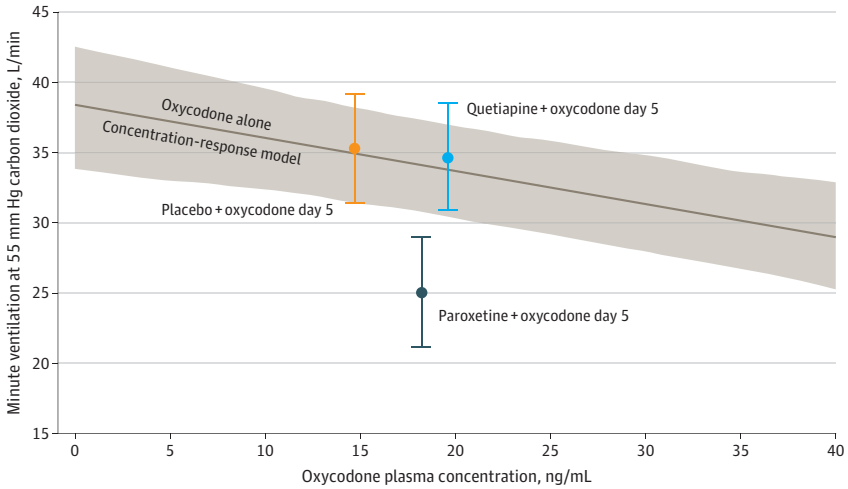


A, Bars indicate medians; box borders, IQRs; and circles, outside the range. Whiskers extending from box borders to the last observation within 1.5 × the IQR.

B, For dosing administration, see the Figure 1. Data points indicate model-estimated means and whiskers 2-sided 95% CIs.

C, The primary outcome comparisons at 5 hours are on days 1 and 5; secondary outcomes, day 4, the secondary outcome comparison. Data points indicate the model-estimated mean difference; whiskers, the upper 1-sided 97.5% CIs.

Figure 3. Oxycodone Concentration-Response Model and Observed Data for Drug Combinations



The oxycodone concentration–response model is based on a linear mixed-effect model with all data from oxycodone alone. The downward sloping black line indicates the prediction; the shaded region, 95% CI (mean slope, −0.29 L/min per ng/mL [95% CI, −0.47 to −0.11); mean intercept, 39.8 L/min [95% CI, 34.0 to 45.7]; see eMethods 3 and eTable 3 in Supplement 2). Data points represent the observed data from the 5-hour time point on day 5 (primary end point) for mean ventilation at 55 mm Hg carbon dioxide (values in Table 2) and geometric mean oxycodone plasma concentration with placebo plus oxycodone was 14.7 ng/mL (coefficient of variation [CV], 31%); oxycodone concentration with paroxetine, 18.2 ng/mL (CV, 21%); and oxycodone concentration with quetiapine, 19.6 ng/mL (CV, 21%).

concentration with paroxetine did not explain the observed effect of paroxetine on the primary outcome (Figure 3 and eTable 3 in Supplement 2). This study included exploratory outcomes of resting respiratory measures while participants breathed room air for 5 minutes prior to the rebreathing procedure. When performing post hoc comparisons at the primary end point time on day 5, neither drug combination significantly decreased resting minute ventilation; however, both drug combinations significantly increased resting end-tidal carbon dioxide (by ≈ 3-4 mm Hg) and decreased resting oxygen saturation (by ≈ 1.1%-1.4%).

In the nonclinical study that motivated this clinical trial,[8] quetiapine caused a substantially larger increase in oxycodone maximum plasma concentration than what was observed in this clinical trial, likely explaining the different respiratory effects observed with the quetiapine-oxycodone combination in the nonclinical study vs this clinical trial. This was likely due to interspecies differences in pharmacokinetics and that substantially higher doses of each drug were administered. The nonclinical study findings with paroxetine were similar to those observed in this trial. Review of older literature identified additional nonclinical studies supporting a relationship between certain systemically administered drugs that affect serotonin and ventilatory depression.[15-20] Inhibition of serotonin synthesis increased baseline ventilation and the ventilatory response to carbon dioxide, which was reversed by administering a serotonin precursor.[18-20] Furthermore, morphine-induced respiratory depression was enhanced by drugs that increase serotonin, including monoamine oxidase inhibitors and the SSRI fluoxetine.[18,19] Other studies identified a relationship between paroxetine or fluoxetine alone and decreased ventilation.[21-24] Additional studies have shown that specific types of serotonin neurons increase their firing rate in response to hypercapnia and that activation of specific serotonin receptor subtypes stimulates ventilation.[25] However, paroxetine does not bind to serotonin receptors at clinically relevant concentrations but rather is highly selective for inhibiting the serotonin transporter, leading to its SSRI properties.[26]

Regarding clinical data, a retrospective analysis of patients referred to a sleep clinic found that SSRIs, compared with a norepinephrine-dopamine reuptake inhibitor, were associated with impaired breathing and worse nocturnal oxygen saturation.[27] Several previous studies involving patients with panic disorder used inhalation of carbon dioxide as a trigger for anxiety and panic symptoms. In addition to finding that multiple SSRIs[28-31] and certain tricyclic antidepressants[28,29] decreased hypercapnia-induced anxiety, a subset of studies using the carbon dioxide rebreathing method found that chronic treatment with SSRIs or certain tricyclic antidepressants decreased the ventilatory response to hypercapnia in this population.[32,33] In overdose, paroxetine and other SSRIs are not known to cause severe respiratory depression or death on their own,[34] suggesting that ventilatory depressant effects may plateau after exceeding a certain exposure, which is consistent with the findings from the nonclinical study with paroxetine alone.[8]

Sound data regarding concomitant medications can be difficult to obtain on patients who overdose while taking opioids because information often relies on death certificates, which vary by death investigation practice (eg, performing comprehensive postmortem drug testing) and reporting practice (eg, focusing on a single lethal drug or listing multiple drugs).[35] Retrospective analyses of administrative health care data that grouped all antidepressants together identified prior antidepressant prescription as a predictor of opioid overdose or

serious opioid-induced respiratory depression, and antidepressant use was included in a developed risk index.[36,37] However, these studies[36,37] did not evaluate the causal link between antidepressants and overdose and were limited by potential treatment and outcome misclassification. An additional recent retrospective analysis with similar limitations and the potential for unmeasured confounding variables found that use of SSRIs that inhibit oxycodone metabolism (paroxetine or fluoxetine; inhibit CYP2D6) at the time of oxycodone initiation was associated with a small but significantly higher risk of opioid overdose compared with the use of other SSRIs.[38] The results from this clinical trial confirmed that paroxetine caused a relatively small increase in oxycodone concentration; however, quetiapine, which inhibits CYP3A4, also increased oxycodone plasma concentration without affecting the primary outcome.

This clinical trial is a part of the FDA's proactive work to address the opioid crisis and help reduce opioid overdoses and deaths and more specifically to determine whether drugs that might be used in place of benzodiazepines may also exacerbate opioid-induced respiratory depression.[8] The findings may have important clinical implications for patients taking paroxetine, or potentially other SSRIs, who concomitantly use opioids, but further research is needed to determine this. SSRIs take approximately 3 weeks to reach maximal therapeutic effect, which correlates with the time required for presynaptic inhibitory serotonergic receptors to densensitize.[39,40] Some prior nonclinical studies suggest different effects of SSRIs on respiration over a similar time frame.[21] Further clarifying the potential time-dependent risks of SSRIs when combined with opioids will be important because treating co-occurring mental health conditions is a critical part of addressing the opioid crisis.

Limitations

This study has several limitations. First, it is not known if the findings with paroxetine will extend to other SSRIs; however, as reviewed in this article, the effects may be due to paroxetine's primary mechanism of action common among SSRIs. Second, the study was conducted in a controlled setting with procedures to increase end-tidal carbon dioxide. Although this differs from what patients would experience, the method allows testing drug combinations at doses that do not lead to severe respiratory depression when breathing room air while still assessing ventilatory effects as carbon dioxide increases, which reflects the physiology of severe respiratory depression seen with opioid overdoses.[2,6] Third, the study involved healthy participants with 5 days of dosing; thus, it is not known if the paroxetine effect on ventilation would persist with longer-term treatment. However, clinical studies discussed earlier that involved patients referred to a sleep clinic and with panic disorder suggest that SSRIs affect ventilation after longer-term treatment.[27,33]

Conclusions

In this preliminary study that involved healthy participants, paroxetine combined with oxycodone, compared with oxycodone alone, significantly decreased the ventilatory response to hypercapnia on days 1 and 5, whereas quetiapine combined with oxycodone did not cause such an effect. Additional investigation is needed to characterize the effects after longer-term treatment and to determine the clinical relevance of these findings.

ARTICLE INFORMATION

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REFERENCES

1. Duffin J. Measuring the respiratory chemoreflexes in humans. *Respir Physiol Neurobiol*. 2011;177(2):71-79. doi:10.1016/j.resp.2011.04.009

2. Pattinson KT. Opioids and the control of respiration. *Br J Anaesth*. 2008;100(6):747-758. doi:10.1093/bja/aen094

3. Ladd LA, Kam PC, Williams DB, et al. Ventilatory responses of healthy subjects to intravenous combinations of morphine and oxycodone under imposed hypercapnic and hypoxaemic conditions.

Br J Clin Pharmacol. 2005;59(5):524-535. doi:10.1111/j.1365-2125.2005.02368.x

4. van der Schrier R, Jonkman K, van Velzen M, et al. An experimental study comparing the respiratory effects of tapentadol and oxycodone in healthy volunteers. *Br J Anaesth*. 2017;119(6):1169-1177. doi:10.1093/bja/aex295

5. van der Schrier R, Roozekrans M, Olofsen E, et al. Influence of ethanol on oxycodone-induced respiratory depression. *Anesthesiology*. 2017;126(3):534-542. doi:10.1097/ALN.0000000000001505

6. Gross JB. When you breathe IN you inspire, when you DON'T breathe, you…expire. *Anesthesiology*. 2003;99(4):767-770. doi:10.1097/00000542-200310000-00002

7. Woodcock J to Wen L and Alexander-Scott L. August 31, 2016. Re: Docket No. FDA-2016-P-0689. US Food and Drug Administration, Silver Spring, MD. Accessed February 23, 2022. https://www.regulations.gov/document/fda-2016-P-0689-0003

8. Xu L, Krishna A, Stewart S, et al. Effects of sedative psychotropic drugs combined with oxycodone on respiratory depression in the rat. *Clin Transl Sci*. 2021;14(6):2208-2219. doi:10.1111/cts.13080

9. *Collection of Race and Ethnicity Data in Clinical Trials Guidance for Industry and Food and Drug Administration Staff*. Published October 26, 2016. Accessed February 23, 2022. https://www.fda.gov/media/75453/download

10. Read DJ. A clinical method for assessing the ventilatory response to carbon dioxide. *Australas Ann Med*. 1967;16(1):20-32. doi:10.1111/imj.1967.16.1.20

11. Rebuck AS. Measurement of ventilatory response to CO_2 by rebreathing. *Chest*. 1976;70(1)(suppl):118-121. doi:10.1378/chest.70.1_Supplement.118

12. Duffin J. Role of acid-base balance in the chemoreflex control of breathing. *J Appl Physiol (1985)*. 2005;99(6):2255-2265. doi:10.1152/japplphysiol.00640.2005

13. Datto C, Berggren L, Patel JB, Eriksson H. Self-reported sedation profile of immediate-release quetiapine fumarate compared with extended-release quetiapine fumarate during dose initiation. *Clin Ther*. 2009;31(3):492-502. doi:10.1016/j.clinthera.2009.03.002

14. Bioavailability studies submitted in NDAs or INDs—general considerations. US Food and Drug Administration. Published 2019. Accessed April 9, 2019. https://www.fda.gov/regulatory-information/search-fda-guidance-documents/bioavailability-studies-submitted-ndas-or-inds-general-considerations

15. Bianchi AL, Denavit-Saubié M, Champagnat J. Central control of breathing in mammals. *Physiol Rev*. 1995;75(1):1-45. doi:10.1152/physrev.1995.75.1.1

16. Mueller RA, Lundberg DB, Breese GR, et al. The neuropharmacology of respiratory control. *Pharmacol Rev*. 1982;34(3):255-285.

17. Armijo JA, Flórez J. The influence of increased brain 5-hydroxytryptamine upon the respiratory activity of cats. *Neuropharmacology*. 1974;13(10-11):977-986. doi:10.1016/0028-3908(74)90089-6

18. Flórez J, Delgado G, Armijo JA. Adrenergic and serotonergic mechanisms in morphine-induced respiratory depression. *Psychopharmacologia*. 1972;24(2):258-274. doi:10.1007/BF00403646

19. Meldrum MJ, Isom GE. Role of monoaminergic systems in morphine-induced respiratory depression. *Neuropharmacology*. 1981;20(2):169-175. doi:10.1016/0028-3908(81)90200-8

20. Mueller RA, Lundberg D, Breese GR. Evidence that respiratory depression by serotonin agonists may be exerted in the central nervous system. *Pharmacol Biochem Behav*. 1980;13(2):247-255. doi:10.1016/0091-3057(80)90081-7

21. Annerbrink K, Olsson M, Hedner J, Eriksson E. Acute and chronic treatment with serotonin reuptake inhibitors exert opposite effects on respiration in rats. *J Psychopharmacol*. 2010;24(12):1793-1801. doi:10.1177/0269881109106908

22. Henderson DR, Konkle DM, Mitchell GS. Effects of serotonin re-uptake inhibition on ventilatory control in goats. *Respir Physiol*. 1999;115(1):1-10. doi:10.1016/S0034-5687(98)00103-0

23. Morrison JL, Chien C, Riggs KW, et al. Effect of maternal fluoxetine administration on uterine blood flow, fetal blood gas status, and growth. *Pediatr Res*. 2002;51(4):433-442. doi:10.1203/00006450-200204000-00007

24. Olsson M, Annerbrink K, Bengtsson F, Hedner J, Eriksson E. Paroxetine influences respiration in rats. *Eur Neuropsychopharmacol*. 2004;14(1):29-37. doi:10.1016/S0924-977X(03)00044-0

25. Corcoran AE, Hodges MR, Wu Y, et al. Medullary serotonin neurons and central CO_2 chemoreception. *Respir Physiol Neurobiol*. 2009;168(1-2):49-58. doi:10.1016/j.resp.2009.04.014

26. Bourin M, Chue P, Guillon Y. Paroxetine: a review. *CNS Drug Rev*. 2001;7(1):25-47. doi:10.1111/j.1527-3458.2001.tb00189.x

27. Robillard R, Saad M, Ray LB, et al. Selective serotonin reuptake inhibitor use is associated with worse sleep-related breathing disturbances in individuals with depressive disorders and sleep complaints. *J Clin Sleep Med*. 2021;17(3):505-513. doi:10.5664/jcsm.8942

28. Bertani A, Perna G, Arancio C, Caldirola D, Bellodi L. Pharmacologic effect of imipramine, paroxetine, and sertraline on 35% carbon dioxide hypersensitivity in panic patients. *J Clin Psychopharmacol*. 1997;17(2):97-101. doi:10.1097/00004714-199704000-00006

29. Perna G, Bertani A, Gabriele A, Politi E, Bellodi L. Modification of 35% carbon dioxide hypersensitivity across one week of treatment with clomipramine and fluvoxamine. *J Clin Psychopharmacol*. 1997;17(3):173-178. doi:10.1097/00004714-199706000-00006

30. Bertani A, Caldirola D, Bussi R, Bellodi L, Perna G. The 35% CO_2 hyperreactivity and clinical symptomatology in patients with panic disorder after 1 week of treatment with citalopram. *J Clin Psychopharmacol*. 2001;21(3):262-267. doi:10.1097/00004714-200106000-00003

31. Perna G, Bertani A, Caldirola D, Di Pasquale D, Migliarese G, Bellodi L. Modulation of hyperreactivity to 35% CO_2 after one week of treatment with paroxetine and reboxetine. *J Clin Psychopharmacol*. 2004;24(3):277-282. doi:10.1097/01.jcp.0000125682.97466.3c

32. Gorman JM, Browne ST, Papp LA, et al. Effect of antipanic treatment on response to carbon dioxide. *Biol Psychiatry*. 1997;42(11):982-991. doi:10.1016/S0006-3223(97)00160-1

33. Bocola V, Trecco MD, Fabbrini G, et al. Antipanic effect of fluoxetine measured by CO_2 challenge test. *Biol Psychiatry*. 1998;43(8):612-615. doi:10.1016/S0006-3223(97)00221-7

34. Gummin DD, Mowry JB, Beuhler MC, et al. 2020 annual report of the American Association of Poison Control Centers' National Poison Data System (NPDS). *Clin Toxicol (Phila)*. 2021;59(12):1282-1501. doi:10.1080/15563650.2021.1989785

35. Hedegaard H, Bastian BA, Trinidad JP, Spencer M, Warner M. Drugs most frequently involved in drug overdose deaths: United States, 2011-2016. *Natl Vital Stat Rep*. 2018;67(9):1-14.

36. Zedler B, Xie L, Wang L, et al. Development of a risk index for serious prescription opioid-induced respiratory depression or overdose in Veterans' Health Administration patients. *Pain Med*. 2015;16(8):1566-1579. doi:10.1111/pme.12777

37. Zedler BK, Saunders WB, Joyce AR, Vick CC, Murrelle EL. Validation of a screening risk index for serious prescription opioid-induced respiratory depression or overdose in a US commercial health plan claims database. *Pain Med*. 2018;19(1):68-78. doi:10.1093/pm/pnx009

38. Yunusa I, Gagne JJ, Yoshida K, Bykov K. Risk of opioid overdose associated with concomitant use of oxycodone and selective serotonin reuptake inhibitors. *JAMA Netw Open*. 2022;5(2):e220194. doi:10.1001/jamanetworkopen.2022.0194

39. Nutt DJ, Forshall S, Bell C, et al. Mechanisms of action of selective serotonin reuptake inhibitors in the treatment of psychiatric disorders. *Eur Neuropsychopharmacol*. 1999;9(suppl 3):S81-S86. doi:10.1016/S0924-977X(99)00030-9

40. Czachura JF, Rasmussen K. Effects of acute and chronic administration of fluoxetine on the activity of serotonergic neurons in the dorsal raphe nucleus of the rat. *Naunyn Schmiedebergs Arch Pharmacol*. 2000;362(3):266-275. doi:10.1007/s002100000290