THE EFFECT OF AIR QUALITY ON SLEEP AND COGNITIVE PERFORMANCE IN SCHOOL CHILDREN AGED 10–12 YEARS: A DOUBLE-BLINDED, PLACEBO-CONTROLLED, CROSSOVER TRIAL

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Abstract

Objectives: To investigate the effect of CO_2 during sleep on next-morning cognitive performance in young schoolchildren, the authors performed a double-blind fully balanced crossover placebo-controlled study. **Material and Methods:** The authors included 36 children aged 10–12 years in the climate chamber. The children slept at 21°C in 6 groups each at 3 different conditions separated by 7 days in a random order. Conditions were as follows: high ventilation with CO_2 at 700 ppm, high ventilation with added pure CO_2 at 2000–3000 ppm, and reduced ventilation with CO_2 at 2–3000 ppm and bioeffluents. Children were subjected to a digital cognitive test battery (CANTAB) in the evening prior to sleep and on the next morning after breakfast. Sleep quality was monitored with wrist actigraphs. **Results:** There were no significant exposure effects on cognitive performance. Sleep efficiency was significantly lower at high ventilation with CO_2 at 700 ppm which is considered to be a chance effect. No other effects were seen, and no relation between air quality during sleep and next-morning cognitive performance was observed in the children emitting an estimated 10 l_{CO_2}/h per child. **Conclusions:** No effect of CO_2 during sleep was found on next day cognition. The children were awakened in the morning,

and spent from 45–70 min in well-ventilated rooms before they were tested. Hence, it cannot be precluded that the children have benefitted from the good indoor air quality conditions before and during the testing period. The slightly better sleep efficiency during high CO_2 concentrations might be a chance finding. Hence, replication is needed in actual bedrooms controlling for other external factors before any generalizations can be made. Int J Occup Med Environ Health. 2023;36(2)

INTRODUCTION

Poor ventilation may lead to reduced cognitive performance among adults at work or children at school [1]. While it has been proposed that the observed effects on cognitive performance may be caused by high concentrations of carbon dioxide (CO_2) from human metabolism and other bioeffluents (organic contaminants emanating from the human body) [2–5], the underlying mechanisms, including possible interactions with other components in indoor air, remain unresolved. Hence, more research in this area is needed.

If poor ventilation can affect cognition during the daytime, it may be hypothesized that poor indoor air quality during sleep may impact cognitive performance the following day. This hypothesis could be highly relevant to test in school children, as they are more susceptible to air pollutants due to children's increased breathing rates compared to adults [6]. In a study of 500 bedrooms of Danish children, only 32% had CO_2 levels <1000 ppm [7], and concentration of CO_2 in bedrooms can easily exceed 2500 ppm due to closed doors and windows [8]. By comparison, the concentration of CO_2 in outdoor air is approx. 400 ppm and recommendations for workplaces in Denmark are to keep CO_2 concentrations <1000 ppm.

To the best of the authors' knowledge, there is so far only 1 published study of the potential effects of bedroom air quality on daytime cognitive performance in university students [8]. This study recruited university students sleeping in their dormitory rooms. The study found no statistically significant effect of bedroom air quality during sleep on next-morning logical thinking. However, when combining the data from the main experiment with data from a pilot experiment, they found statistically significant effects indicating that better ventilation was associated with improved sleep efficiency (the proportion of time in bed spent asleep) and improved cognitive performance the following day. In the main and pilot experiments, air quality, however, was controlled either mechanically using an inaudible fan or by opening a window. As suggested by the improved sleep efficiency found in the experiment, the impact of air quality on cognitive performance, could perhaps, at least partly be explained by improved sleep quality. It is well-established that sleep deprivation has a negative effect on various aspects of cognitive performance, e.g., impaired executive functions, working memory, and reaction time [9], with even minor restrictions in sleep leading to poorer performance [10].

On this background, the present study investigated whether CO_2 levels alone or in combination with other bioeffluents, both considered major sources of pollution during the night, influence sleep quality and next-morning cognitive performance in schoolchildren.

The authors hypothesized that:

– H1. Children exposed to a well-ventilated environment during sleep would exhibit better next-morning cognitive performance compared to their performance following sleep in air with high levels of CO_2 alone or CO_2 together with other bioeffluents.

– H2. Children would perform better on cognitive tests following a night in high levels of CO_2 alone compared with the performance after a night in high levels of CO_2 and bioeffluents.

– H3. Children would evidence better sleep in a well-ventilated environment compared with high levels of CO_2 alone or CO_2 plus bioeffluents.

MATERIAL AND METHODS

The present study was designed as a double-blind, placebo-controlled, and fully balanced crossover trial.

Participants

Children aged 10–12 years were recruited from 2 local schools in Aarhus, Denmark. Forty-four children and their parents agreed to participate in a pre-examination (Table 1).

Thirty-six children were enrolled in the study and allocated to 6 groups of 6 children. The parents of the children signed an informed consent, and the children received DKK 500 (approx. USD 80) per night as compensation. One child was excluded because of difficulty completing the cognitive test. The study was approved by the Regional Committee on Health Research Ethics for Central Denmark Region (Registration No. 1-10-72-203-17).

Procedure

All groups participated for 3 nights. Most visits were separated by 7 days, except for 2 groups participating 2 nights separated by 6 days, and 1 group participating 2 nights separated by 14 days; the between-group differences were due to school holidays.

At each exposure condition (condition 1, 2 and 3, see below), the children were assigned the same seat during cognitive testing and the same bunk bed for sleeping.

Experimental conditions

The experiments took place in a 72.9 m³ (2574.4 ft³) climate chamber (5.4 × 5.4 × 2.5 m [17.7 × 17.7 × 8.2 ft]) made of stainless steel. The chamber was empty except for 4 metal bunk beds and CO_2 measurement sensors. The beds were equipped with standard mattresses, linen

and duvets (Figure 1). The children were allowed to bring a teddy bear or a pillow for comfort, but all other personal belongings were kept outside of the chamber. During all conditions, temperature was held constant around 21°C (Table 2) by ventilation and regulation of wall temperatures. It was not possible to keep humidity at a constant level during condition 3 due to low ventilation. The ozone concentration in the climate chamber was <1 ppb.

The 3 experimental sleeping conditions were as follows (Figure 2):

– In condition 1, the children slept in a chamber ventilated with a clean air change rate of 2.8/h (per child ventilation rate of 9.5 l/s).

– In condition 2, the children slept in a chamber ventilated with a clean air change rate of 2.8/h as in condition 1, so the clean air supply rate per child was approx. 9.5 l/s. In this condition, pure CO_2 was dosed. The change in CO_2 (Figure 2) was at the same pace as in condition 3.

– In condition 3, the children slept in a chamber ventilated with clean air at an air change rate of 0.2/h (per child ventilation of 0.7 l/s). This condition corresponded to poorly ventilated bedrooms and the CO_2 concentration increased to 3000 ppm (Figure 2).

Under each condition, the room air was mixed by a fan with an insignificant noise level establishing a vertical upwards circulation to align the amounts of CO_2 and bioeffluents at the top and bottom bunks (Figure 1). No draft or high air velocities were detected at the beds. Carbon dioxide was monitored using 4 calibrated monitors. The main monitor, Vaisala Indigo 201 GMP252 (Vantaa, Finland), monitored CO_2 via tubes placed in the 4 corners of the chamber at a height of 0.5 m above the floor. Vaisala GMP 220 monitored CO_2 in the supply air. Six sampling lines (plastic tubes) were attached to the headboards of the upper and lower bunks to monitor CO_2 at each bed area using Innova LumaSense 1412i with an Innova 1303 multiplexer. The sampling lines were

Table 1. The demographics and normal sleep habits of the children enrolled to the study on the effect of CO_2 during sleep on next-morning cognitive performance, Denmark

Participant No.	Participants (N = 36)						
	gender	group	school	age [years]	sleep habits at home		
					sleep time [h]	bedtime	wake up time
1	female	1	1	12	10.25	8:30 p.m.	6:45 a.m.
2	female	1	1	10	10.5	8:30 p.m.	7:00 a.m.
3	female	1	1	11	10.5	8:30 p.m.	7:00 a.m.
4	female	2	1	12	9	9:00 p.m.	6:45 a.m.
5	male	3	1	12	9.5	9:00 p.m.	6:30 a.m.
6	female	2	1	12	8.5	9:00 p.m.	7:00 a.m.
7	female	4	2	11	9.75	8:45 p.m.	6:45 a.m.
8	male	6	2	12	9.5	9:10 p.m.	6:40 a.m.
9	female	4	2	12	8	9:30 p.m.	6:50 a.m.
10	female	4	2	12	–	–	–
11	male	3	1	12	10	8:30 p.m.	6:30 a.m.
12	male	5	2	12	9	9:00 p.m.	6:30 a.m.
13	male	6	2	12	9	9:30 p.m.	6:30 a.m.
14	male	6	2	12	9.5	9:00 p.m.	6:30 a.m.
15	female	4	2	12	10.5	8:15 p.m.	6:45 a.m.
16	female	4	2	12	9.5	9:30 p.m.	7:00 a.m.
17	male	6	2	11	9	9:20 p.m.	6:30 a.m.
18	male	6	2	12	8.5	10:00 p.m.	6:36 a.m.
19	female	2	1	11	9.75	9:00 p.m.	7:00 a.m.
20	male	3	1	11	9	8:00 p.m.	6:30 a.m.
21	male	1	1	13	10	9:00 p.m.	7:00 a.m.
22	male	6	2	12	9.5	9:00 p.m.	7:00 a.m.
23	female	4	2	12	9	8:30 p.m.	6:00 a.m.
24	female	5	2	12	9.5	9:30 p.m.	7:00 a.m.
25	female	2	1	12	9.5	8:00 p.m.	7:00 a.m.
26	male	5	2	12	9	8:30 p.m.	6:30 a.m.
27	male	1	1	12	10	9:00 p.m.	7:00 a.m.
28	female	5	2	12	8.67	9:00 p.m.	6:40 a.m.
29	male	3	1	12	10.5	8:00 p.m.	6:30 a.m.
30	female	2	1	11	8.5	10:00 p.m.	6:30 a.m.
31	female	2	1	11	8.25	8:30 p.m.	6:30 a.m.
32	female	5	2	11	9.75	8:30 p.m.	6:45 a.m.

Table 1. The demographics and normal sleep habits of the children enrolled to the study on the effect of CO_2 during sleep on next-morning cognitive performance, Denmark – cont.

| Participant No. | Participants (N = 36) | | | | | | |
| | gender | group | school | age [years] | sleep habits at home | | |
					sleep time [h]	bedtime	wake up time
33	male	1	1	12	9.5	9:00 p.m.	7:00 a.m.
34	male	3	1	13	9.5	9:15 p.m.	6:45 a.m.
35	male	3	1	11	9.25	8:45 p.m.	6:15 a.m.
36	male	5	2	12	9.5	9:00 p.m.	6:30 a.m.

Group 1–6 – children enrolled in the study were allocated to 6 groups of 6 children.
School 1, 2 – children were recruited from 2 local schools in Aarhus, Denmark.

placed behind the heads of the children to avoid monitoring misleadingly high CO_2 concentrations due to exhaled air.

Volatile organic compounds and carbonyl measurements

Organic gases were collected from 2:00 am to 6:00 am using 2 Tenax TA adsorbent tubes (Gerstel, Mülheim an der Ruhr, Germany) volatile organic compounds (VOCs) (flow rate 21 ml/min on average), while carbonyl compounds were sampled as 2,4-dinitrophenyl hydrazones with two LpDNPH S10 cartridges (Sigma-Aldrich, Darmstadt, Germany) (flow rate 2 l/min). The VOC adsorbent tubes were analyzed by thermal desorption (Gerstel, Mülheim an der Ruhr, Germany) and gas chromatography – mass spectrometry (GC-MS) with an Agilent 7890B GC (Agilent, Santa Clara, CA, USA) and 5977A MSD with a Restek Rtx-200MS column (Restek, Centre County, PA, USA) (30 m × 0.25 mm × 0.25 μm) and helium carrier gas. Thermal desorption was 20–300°C, GC temperature program was 10°C min^{-1} from 35°C to 300°C.

The following VOCs were quantified: 1-butanol, 2-butanone, α-pinene, β-pinene, d-limonene, hexanal, nonanal, decanal, tetradecane, pentadecane, hexadecane, dodecane, toluene, m- and o-xylene, and 1,2,4-trimethylbenzene. An indoor air standard certified reference material

Figure 1. The exposure chamber equipped for overnighting used in the study on the effect of CO_2 during sleep on next-morning cognitive performance in schoolchildren (N = 36), Denmark

(Sigma-Aldrich, Germany) and hexanal were used for calibration (3–60 ng/μl, 1.0 μl split ratio 9:1).

Carbonyl LpDNPH cartridges (Thermo Fisher Scientific, Waltham, MA, USA) were extracted in 4.00 ml acetonitrile and analyzed using high performance liquid chromatography (Dionex Ultimate 3000 HPLC, Thermo Fisher

Table 2. Chamber environment from 9 p.m. to 6 a.m. mean temperature (Tp), relative humidity (RH), carbon dioxide (CO_2) and total volatile organic compounds (VOCs) in the chamber during 6 sessions for each condition in the study on the effect of CO_2 during sleep on next-morning cognitive performance in schoolchildren (N = 36), Denmark

Ventilation	Tp [°C]		RH [%]		CO_2 [ppm]		Total VOCs [μg/m³]	
	M	SD	M	SD	M	SD	M	SD
Good								
low CO_2	21.3	0.2	46.0	0.6	681	7	21.7	3.6
high CO_2	21.4	0.2	46.4	0.9	2842[b]	108	20.1	2.1
Poor – CO_2+ bioeffluents	21.9[a]	0.2	54.8[a]	6.4	2873[a]	124	72.0[a]	14.9

[a] p < 0.05 Tukey's multiple comparisons group vs. good ventilation – low CO_2.
[b] p < 0.05 Tukey's multiple comparisons group vs. good ventilation – high CO_2.
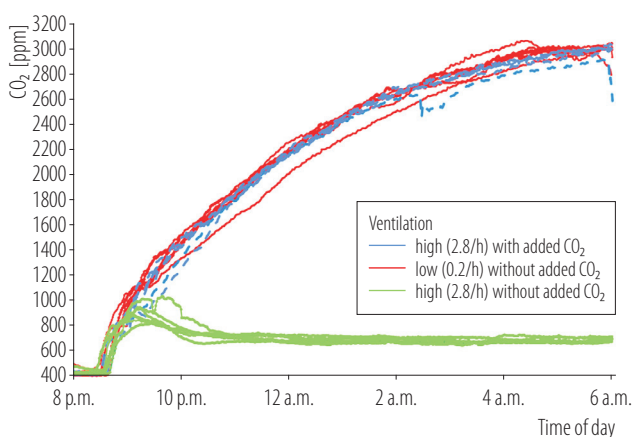

Figure 2. The environmental conditions under the 3 different scenarios (the children entered the chamber 8:30 p.m.; the children were told to go to sleep 9:30 p.m.; the children were awakened 6:00 a.m.) in the study on the effect of CO_2 during sleep on next-morning cognitive performance in schoolchildren (N = 36), Denmark

Scientific, USA) with UV-detection at 360 nm, an Acclaim 120 C18 column (3 μm particles, 4.6 mm × 150 mm, Thermo Fisher Scientific) and eluent of 45% acetonitrile in MilliQ water. Hydrazones of formaldehyde, acetone, acetaldehyde, acrolein, propionaldehyde and crotonaldehyde (Sigma-Aldrich , Germany) were quantified ($R^2 > 0.99$, 0.1–5 μg/ml). The detection limit was ≤0.02 mg/ml. Acrolein and crotonaldehyde were not detected above the detection limit.

Two technicians watched the children through the night. To ensure double blinding, the technicians were not involved in the assessment of cognitive performance, sleep, or in the statistical analyses.

Outcome assessments

Cognition

The Connect Research edition from *Cambridge Neuropsychological Test Automated Battery* (CANTAB) was used for cognitive testing. The children went through the cognitive tests prior to the inclusion in order to train and to assure that they were able to perform the test. During the exposure visits each child was tested in the evening and again next morning. Six children were tested simultaneously in a room with partition walls to prevent distractions. They completed CANTAB on iPads validated for the purpose, using headphones to receive test instructions. The children were instructed to stay in their seats until everybody had finished in order to minimize distraction. Three investigators were present to supervise the children.

Cambridge Neuropsychological Test Automated Battery (CANTAB) is designed to minimize practice effects (better performance due to increasing familiarity with a test) using parallel modes and/or stimuli randomization. To minimize bias from remaining practice effects, the authors created a training baseline test, which was excluded from the analysis. Hence, the test performed at the first night in

the chambers was used as baseline, and the authors also included the order of visits in the analysis. This procedure was implemented, as several studies have shown that the largest practice effects tend to occur between the first and second test session [11].

The CANTAB test battery consisted of 5 selected tests arranged in the following order:
1) *Motor Screening Task* (MOT),
2) *Reaction Time* (RTI),
3) *Spatial Working Memory* (SWM),
4) *One-Touch Stockings of Cambridge* (OTS),
5) *Rapid Visual Information Processing* (RVP).

The selected tests represent cognitive functions known to be influenced by sleep and were especially chosen to match the age of the children.

In MOT, children have to tap on a cross appearing in different locations on the screen. The RTI is a test of reaction time and sustained attention. The SWM is a test of working memory and strategy. The OTS is a test of planning ability and working memory. The RVP is a test of sustained attention and continuous performance.

Finally, a composite score was created by calculating Z-scores for the 10 outcomes and using the sum of the Z-scores to get a measure of total performance in CANTAB.

Sleep and sleep quality

Sleep was monitored with wrist actigraphy (WA) using Fitbit Alta HR sleep trackers (Fitbit, San Francisco, CA, USA). The actigraph was placed on the wrist of the non-dominant hand after dinner, and the children wore them during the entire night. The devise estimates sleep onset, sleep stages (light sleep, deep sleep, and REM-sleep) and time awake based on movement and heart rate patterns. Using data from the WAs and recorded bedtime information (the time children were instructed to go to sleep and were awoken), we calculated sleep onset latency (SOL) (the time it takes to fall asleep) and sleep efficiency (SE), as:

$$(TST/TiB) \times 100 \qquad (1)$$

where:
TST – total sleep time,
TiB – time in bed.

Time in bed included SOL hence the calculated sleep efficiency was affected not only by short awakenings during night but also the time necessary to fall asleep.

In the morning, children rated their sleep quality by completing a brief questionnaire, all questions using a child-friendly scale consisting of 3 options: a green satisfied face (2), a yellow neutral face (1), or a red dissatisfied face (0).

In order to examine whether the children slept differently in the climate chamber compared to sleeping at home, children were subsequently asked to wear WAs at home once the experiments were completed. Twenty-eight children (88%) agreed to this with 1 participant being excluded due to vacation. Participants wore the WAs for 5 consecutive nights (Sunday–Thursday) and completed a sleep diary consisting of bedtime, time they went to sleep, wake up time, optional comments.

Outcomes

The primary outcome measure, determined before the start of the study was reaction time as captured by the "median for simple reaction time with 1 target button" (RTISMDRT) estimated by CANTAB. Secondary outcome measures were the CANTAB composite score of 10 items as Z-scores, the median for 5-choice reaction time with 5 target buttons (RTIFMDRT), and SE. Only the primary hypothesis was tested. All other tests were performed for hypothesis-generating purposes only. Hence, the authors did not perform any formal correction for multiple testing.

Statistical analysis

The cognitive outcomes were analyzed as the difference between morning and evening measurements and

the sleep outcomes were analyzed directly. For each outcome, the authors used a linear mixed model with exposure (3 levels), group (6 levels) and visit (3 levels) as fixed effects and a random effect at the individual level. Restricted maximum likelihood estimation and the Kenward-Roger degrees of freedom method were used. An overall F-test for no differences between exposure groups was performed and predicted outcome means were estimated for each exposure group.

Model fit was assessed by inspecting quantile plots for the residuals. No substantial departures from normality were observed. All analyses were performed using Stata/IC 15.0.

Ethics approval statement

The study was approved by the Regional Committee on Health Research Ethics for Central Denmark Region (Registration No. 1-10-72-203-17). The study was reported to the Danish Data Protection Agency (journal No. 2016-051-000001/641). The study was conducted in accordance with The Declaration of Helsinki and written consent was obtained from all participants prior to participation.

RESULTS

Exposure conditions

Similar levels of CO_2 were obtained during the 2 high CO_2 exposure conditions (Figure 2). Within the same exposure conditions, the authors obtained similar build up curves of CO_2 along the time of exposure across the different nights. Mean temperature and humidity were kept constant between the different exposures except for slightly higher temperatures (0.5–0.6°C) and humidity (8.4–8.8%) during condition 3 (Table 2). Temperature and humidity were also stable during each night with variation coefficients ranging between 0.01–0.02 for temperature. The experiments were performed during the period of March–June when the mean (range) outdoor temperatures were 10.3°C (2.6–16.7) during "clean air days," 9.0°C (0.7–17.3) during "clean air + CO_2 days," and 11.3°C (−0.6–18.3) "CO_2 and bioeffluent days," thus quite similar. The authors found significantly higher mean temperature (by 0.6°C), RH (by 8.4%) and VOCs (by 50 μg/m³) when ventilation was reduced compared with the other 2 conditions (Table 2). Slightly higher temperature was caused by the limitations in the capacity of the system conditioning the room temperature when air supply to the chamber was set to a minimum.

VOCs and carbonyls

Small carbonyl compounds (formaldehyde, acetaldehyde, and acetone) were the most prominent VOCs with average total carbonyl concentrations of 14.5±2.1 μg/m³ during high ventilation (with and without added CO_2) and 52.7±17.5 μg/m³ during low ventilation, with acetone constituting more than 70% of the total.

The average concentration of VOCs collected by Tenax tubes was 6.3±1.5 μg/m³ during high ventilation and increased to 19.3±3.3 μg/m³ during low ventilation experiments. Major bioeffluents observed were nonanal and decanal, which are emitted from skin by, e.g., ozonolysis of unsaturated fatty acids in skin oil [12]. The odor threshold of these compounds is about 3–6 μg/m³ [13]. The average total VOC concentrations of both carbonyl compounds collected by DNPH-tubes and VOC collected by Tenax tubes were 20.1±2.1 μg/m³, 21.7±3.6 μg/m³ and 72.0±14.9 μg/m³ for the 2 well-ventilated conditions and the low ventilated conditions respectively (Table 2).

Participants

One participant dropped out after the first night in the chamber. Due to late cancellation, the authors were unable to enroll a replacement from the waiting list. For 3 children, sufficient WA sleep data could not be obtained for 1 night, and for 1 participant there were no data for any of the 3 nights. Both happened due to a technical error of the WA and these data were excluded.
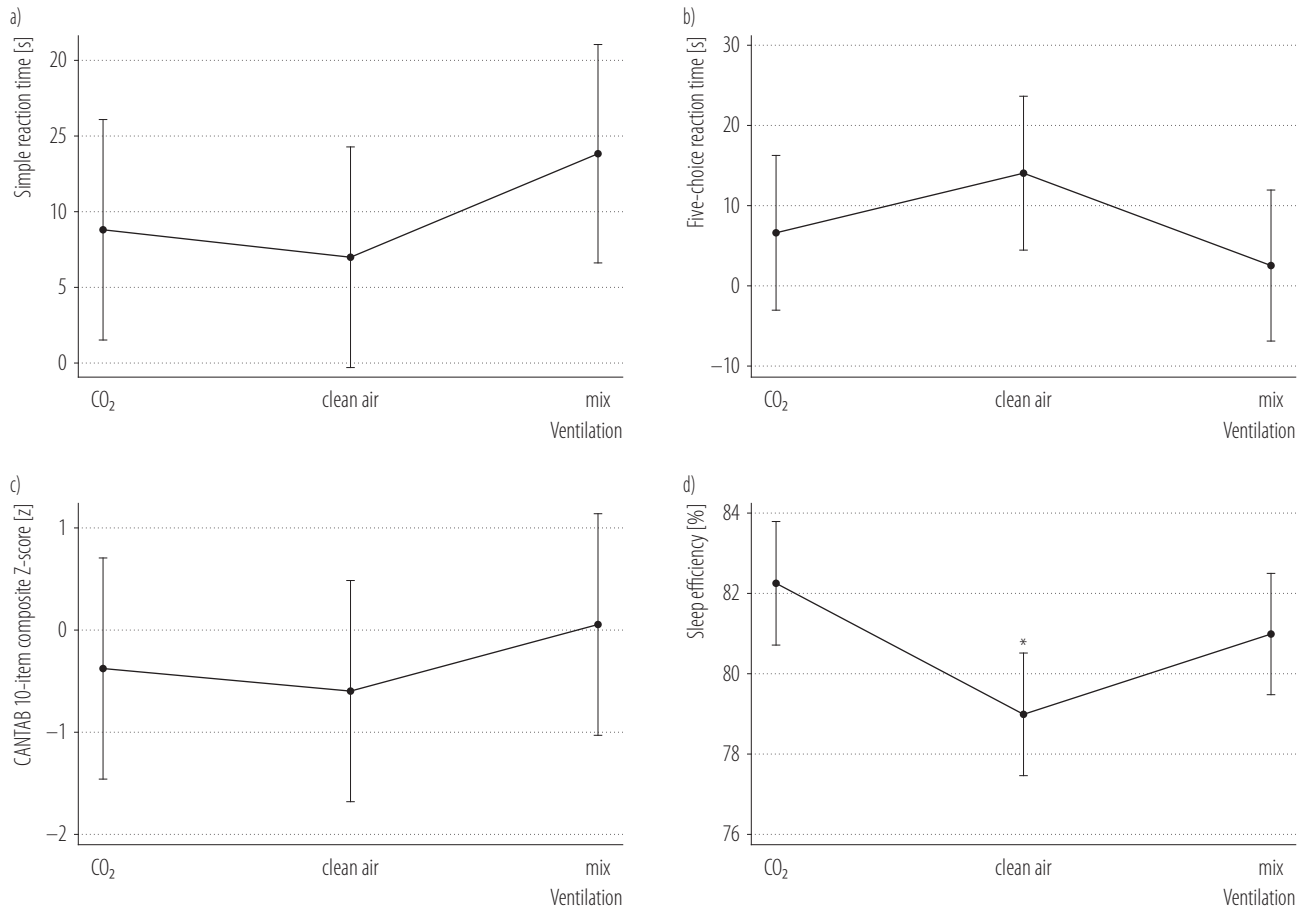
CO$_2$ – CO$_2$ good ventilation added CO$_2$; clean air – good ventilation; mix – low ventilation with high CO$_2$ and bioeffluents.
Graph shows marginal means (95% CI). A lower value indicates a poorer performance.
* p < 0.05.

Figure 3. Cognitive outcomes and sleep efficiency on the different exposure days on next-morning cognitive performance in schoolchildren (N = 36):
a) simple reaction time, b) five-choice reaction time, c) CANTAB 10-item composite Z-score, d) sleep efficiency

Cognitive performance

Due to technical difficulties, 1 participant failed to complete OTS and RVP on the second morning and was thus excluded from the analysis of these outcomes.

There were no significant effects of exposure on the primary outcome measure RTISMDRT, nor on any of the secondary outcome measures: RTIFMDRT or CANTAB 10-item composite Z-score (Figure 3). There were no significant effects of exposure on any of the remaining CANTAB outcomes. No group effects were observed. A period effect (whether a participant gradually became better or worse during the test

sessions) was found for RVPA (p = 0.001), but all exposure effects were adjusted for period effects in the analysis.

Although the difference did not reach statistical significance, the children performed better overall on the morning after sleeping in the chamber with high ventilation where the concentration of CO$_2$ was low (800 ppm). This morning 6 out of 10 CANTAB outcomes:

– median for simple reaction time with one target button (RTISMDRT);

– working memory between errors for 4, 6 and 8 boxes (SWMBE468);

– working memory between errors for 12 boxes (SWMBE12);
– working memory strategy score for trials with 6 and 8 boxes (SWMSX);
– sustained attention 0/1 answers (RVPA);
– sustained attention median reaction time for correct answers (RVPMDL) were better compared to the mornings after the 2 exposures with high concentrations of CO_2 (with and without other bioeffluents).

Sleep and sleep quality

The secondary outcome measure, sleep efficiency, was unexpectedly significantly lower when sleeping in the chamber with high ventilation and low CO_2 <800 ppm compared to exposures with CO_2 added to the chamber with high ventilation (3000 ppm) (3.25, p < 0.001) and with CO_2 together with other bioeffluents but at low ventilation (3000 ppm) (1.98, p < 0.001). There was no significant difference between sleep efficiency during the 2 nights with high CO_2.

Sleep onset latency was significantly longer when sleeping in a chamber with low CO_2 compared to exposures with high CO_2 levels either added 19.2 min longer (p < 0.001) or due to low ventilation rate 10.5 min longer (p < 0.029), data not shown.

No effects of exposure conditions were found on light, deep, or REM sleep.

Because of the significant exposure effect on sleep efficiency, the authors analyzed whether sleep efficiency affected cognition. However, no statistically significant effects were observed on any of the cognitive test outcomes.

Self-reported sleep quality was not associated with any cognitive outcomes (data not shown).

The results of the 2 exposures with high CO_2 levels were combined and compared to low level CO_2 exposure in a post hoc analysis for the cognitive outcomes and sleep. No significant effects were found either for any of the cognitive outcomes or sleep.

Sleep results from the chambers and at home are shown in Figure 4. The children had poorer sleep efficiency the first night in the chamber compared to second night, third night and at home (p < 0.001). Further, sleep efficiency was significantly lower on the second night compared to at home. Sleep latency was significantly longer on the first night (p < 0.001) (data not shown). The children had significantly less light sleep on the first, second and third night compared to at home (p < 0.001). The amount of light sleep did not differ significantly between the first, second or third night. The amount of REM sleep on the first night was significantly lower compared to third night and when sleeping at home (p < 0.005). There was no significant difference between REM sleep on the first and the second night. The children had significantly less deep sleep on the first night compared to the second night (p = 0.01) and when sleeping at home (p = 0.02).

The authors estimated emission rates of CO_2 from sleeping children in the chamber using measured CO_2 and outdoor air supply rate; the authors used the measurements from the latter part of the night once CO_2 concentration reached steady-state. The emission rates were around 10 l/h per child. The rates were similar to those measured by Fan et al. [14] for young adults and they matched the prediction based on the basal metabolic rate [15]. The authors could not draw credible conclusions as to whether there were differences in emission rates between different conditions because of the accuracy of measurements. Such differences were earlier observed for people awake and attributed to changes in respiration [16]. These results require confirmation in future experiments.

DISCUSSION

In the present investigation of the effect of CO_2 with or without other bioeffluents during the night on sleep quality and next-morning cognitive performance of schoolchildren, the authors were able to achieve the desired exposures with a fully balanced design (6 groups × 3 exposures). However, contrary to the hypotheses, the authors
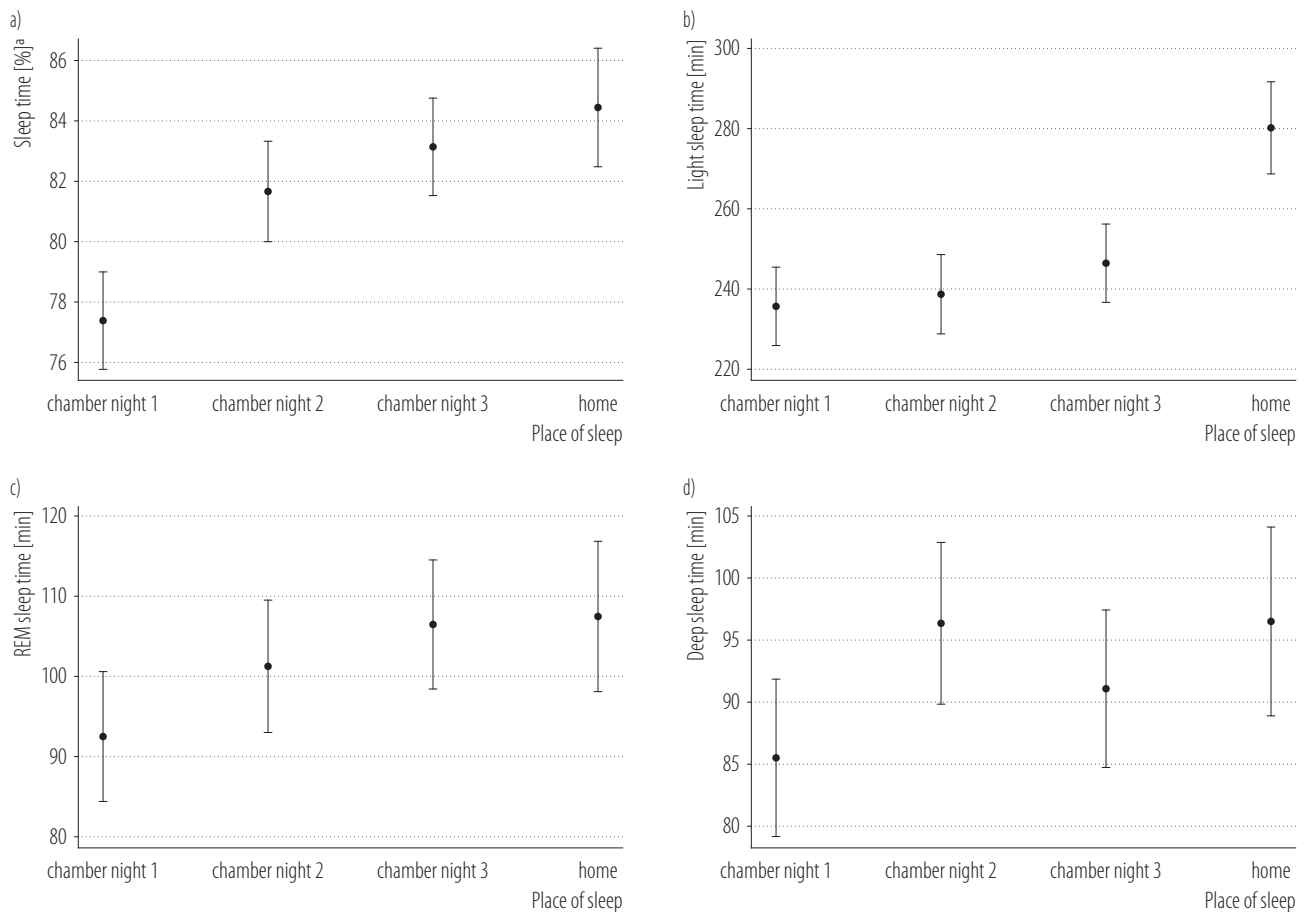
a Percentage of sleep time out of total time in bed.
Graph shows marginal means (95% CI).

Figure 4. Sleep quality according to day in the chamber night 1–3 and at home: a) sleep efficiency, b) light sleep, c) REM sleep, and d) deep sleep in the study on the effect of CO_2 during sleep on next-morning cognitive performance in schoolchildren (N = 36)

observed no significant difference in the next-morning cognitive functioning after sleeping in a chamber with high ventilation and low CO_2 (800 ppm) in comparison with the 2 high CO_2 situations. Cognitive performance was better on 6 out of 10 CANTAB outcomes on that morning but not significantly so. One plausible explanation for this result is that poor indoor air quality during sleep does not disturb sleep to a degree that will affect next-morning cognitive performance. However, other studies have shown an effect of poor air quality on several cognitive functions both when performing schoolwork or office work [1–3,10,17,18]. In these experiments, the tasks were performed directly

under these poor air quality conditions. Hence, they consequently examined a direct effect on performance. In the present study, the authors examined the indirect effect of poor air quality. The children slept under the different air quality conditions, they were awakened in the morning, and spent from 45–70 min in well-ventilated rooms before they were tested. Hence, it cannot be precluded that the children have benefitted from the good indoor air quality conditions before and during the testing period. Except for sleep efficiency and sleep onset latency (they were unexpectedly poorer under the high ventilation rate condition), there were no effects of poor air quality on sleep quality

either measured with actigraphy or subjectively assessed by the children participating in the experiments.

Contrary to the authors' secondary hypothesis regarding the association between better sleep efficiency and clean air, the children in the present study revealed lower sleep efficiency during exposure to clean air. This finding is in contrast with recommendations about sleeping in well-ventilated rooms and may be spurious. While the authors have no clear explanation for this counter-intuitive finding, 1 possible explanation could be that higher CO_2 levels are more similar to the children's home environment as indicated by Bekö et al. [7], and so children felt more confident under these conditions and more calm which promoted their sleep. The authors' finding is in direct contrast to the recent study by Xu et al. [19] showing a decrease in sleep quality within the same CO_2 exposure range. The study by Xu et al. was performed in 12 college students sleeping alone for a duration of 9 nights under 3 different conditions of 800–3000 ppm CO_2 in a randomized sequence at a temperature of 25–27°C. This design is well-suited to minimize the first night effect, as each session is repeated 3 times, thus increasing the signal-to-noise ratio. However, some of the observed differences between the studies might relate to the effect of age difference and co-sleeping in the authors' study. Co-sleeping with others provide a more safe sleep environment with a much stronger positive impact on sleep efficiency than changes in air quality [20]. However, most of the children were used to sleeping alone, and the highest sleep efficiency was found during exposure to high ventilation and added CO_2 and not during exposure to low ventilation with higher amounts of bioeffluents. There was also no significant difference in sleep efficiency when sleeping at high CO_2 either in conditions 2 and 3.

It is important to notice that the calculation of sleep efficiency is affected by sleep latency. The longest sleep latency was observed during exposure with low level of CO_2. This is interesting as the concentration of CO_2 increased throughout the night and was not much different at the onset

(Figure 2). The difference in CO_2 concentration between conditions with high and low CO_2 only reached approx. 800 ppm during the period 9:00–9:45 p.m. (mean sleep onset) (Figure 2). Sixty-nine percent of sleep onsets were observed before 10:00 p.m., and the mean sleep latency was 44.1 min (SD = 27.1). The low sleep efficiency observed during exposure to high ventilation and a CO_2 concentration approx. 700 ppm compared to about 1500 ppm during the high CO_2 concentrations would therefore relate to a CO_2 difference of only 800 ppm. There was a slightly higher temperature during the low ventilation situation However, this is not substantiated in the data as the authors did not find a difference between sleep efficiency during the 2 conditions with high CO_2 concentrations where the temperatures likewise were 0.6°C different. The improved SOL during the night with high CO_2 and bioeffluents was unexpected, and the authors do not have a clear explanation for this finding. As children were randomized to the 3 exposure conditions, improved SOL is not expected to be caused by an adaptation to the environment in the course of experiments. The authors observed a slightly but statistically significant higher mean temperature and relative humidity when the target CO_2 – level was reached in the reduced ventilation condition.

In general, children slept better at home compared with the nights in the chamber. However, this is likely to be less important for the interpretation of the results, as the authors primarily wanted to compare sleep changes across different conditions in the chamber. Nevertheless, a considerable "first-night" effect was found as indicated by significantly lower sleep efficiency and longer sleep latency during the first night in the chamber as well shorter REM and NREM (deep) sleep. The first-night effects term is usually used in studies using comprehensive polysomnography and may be caused by the discomfort of wearing electrodes and cables, being under surveillance and/or sleeping in a new environment [21]. Most likely, the wrist actigraph was not the cause of the observed first-night effect since it hardly had any effect on the ability to lie down comfortably or falling asleep, and studies test-

ing for first night effects at home of other types of actigraphs have not reported such effects [22]. Hence, the first night effect observed was most likely due to other factors including changes in the sleep environment, differences in sleep schedules, the presence of other children in the chamber, and the awareness of being under constant surveillance.

A possible solution to minimize the first-night effect would have been to arrange 4 nights in the chamber with the first night being without any exposure. However, it was decided to keep the number of nights in the chamber to a minimum to reduce the burden on the children and their families and minimize dropouts. The influence of the first-night effect was also kept to a minimum by the fully balanced study design and by controlling for period effects in the statistical analyses.

Limitations

Testing

The authors found a strong first night effect in all the groups. Even with a fully randomized and balanced design as the present the first night effect potentially could obscure the ability to find the real effects of the conditions on cognition and sleep efficiency.

Children slept only 1 night under reduced air quality conditions before cognitive testing. It is therefore unclear whether sleeping repeatedly in bedrooms with poor ventilation will result in poor sleep and consequently impaired cognitive functioning. As sleep quality parameters generally were better at home, future studies should take this into account and either perform interventions in the usual sleeping environment or use more sessions to precondition the children to the new environment.

Wrist actigraphy

One limitation of using WA to measure sleep is the missing disclosure of algorithms used to decide sleep and awake time. Fitbit Alta HR has been shown to overestimate total sleep time, sleep efficiency, and deep sleep duration com-

pared with polysomnography in children with suspected central disorders of hypersomnolence [23]. Studies of WA devices with technology similar to Alta HR have also shown a tendency to overestimate total sleep time compared to other actigraphs or polysomnographic recordings [24]. Nevertheless, this tendency has been shown to be constant across different measures [24], which makes it possible to use WA to compare within-subject changes of sleep across different nights. In addition, WA are easy to use and do not seem to interfere with sleep [22].

CONCLUSIONS

Sleeping in the climate chamber together with 5 other children at CO_2 concentrations of 2–3000 ppm had no significant effect on next-morning cognitive performance in children aged 10–12 years emitting an estimated $10\,l\,CO_2/h$ per child when compared with sleeping in a chamber with low CO_2 of 700 ppm. Whether the CO_2 was added to clean air or was part of the air when ventilation was reduced in the chamber did not influence the results. Sleep efficiency measured with wrist actigraphy was highest when children slept in the chamber with reduced ventilation and increased levels of CO_2 and other bioeffluents. Adaptation to the experimental sleep setting, as demonstrated by distinctly lower sleep quality during the first night could be part of the reason for the observed result. The present findings require replication and validation in actual bedrooms before any general conclusions are drawn regarding ventilation requirements in bedrooms.

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