

### Introduction

Carbon dioxide ( $CO_2$ ) is commonly used to kill rats. When provided the opportunity to escape (e.g. in aversion testing) all rats choose to avoid  $CO_2$ , but the concentration avoided is variable. When rats are unable to escape (i.e. during acute exposure) rats also vary in their behavioral response. These results suggest that rats, like humans, may vary in their sensitivity to  $CO_2$ . The aim of this study was to assess if these differences are consistent within rats, and if the responses during forced exposure relate to those during aversion testing.

# Methods





Active responses during acute exposure and in aversion-avoidance tests were highly variable between rats but consistent within rats. Active responses during forced exposure to CO<sub>2</sub> were related to the strength of aversion in the aversion-avoidance test. We conclude that rats vary in sensitivity to CO<sub>2</sub>. Further research is needed to determine if this variability in rats, as in human subjects, is related to differences in the affective responses evoked by  $CO_2(e.g. anxiety, dyspnea)$ .

## Variability in rat behavior during exposure to CO<sub>2</sub>

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During acute exposure tests rats were exposed twice to  $CO_2$  gradual fill (18% volume min<sup>-1</sup>). Active responses consisted of rearing, locomotion, and escape attempts frequencies. The passive response was time immobile (i.e. freezing time). All responses were standardized as z-scores, and these values were added together to provide sum score in the case of the active responses.

> were tested twice in the Rats aversion-avoidance test, assessing motivation to avoid CO2 gradual fill (18% volume min-1) in a preferred dark chamber versus escaping to a bright chamber (1650 lux). The  $CO_2$ concentration at which rats left the dark chamber was recorded.







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highly Active responses were consistent (n=11) rat within between the first and second exposure to  $CO_2$  (r = 0.84, p < 0.01). Passive responses were not consistent between exposures (r = -0.38, p = 0.26).

Rats were highly variable in the  $CO_2$ concentration at which they escaped from the dark to the light chamber, but individual rats were consistent in this response across two tests (p <0.01, r =0.81; n = 11).

The mean (across the two tests)  $CO_2$  concentration at which rats escaped from the dark to the light chamber was negatively correlated with the mean (again averaged across the two tests) active response during acute exposure (r *= -0.55, p = 0.08; n = 11).*