The enhancing effect of phosphate on the constriction response is not specific for LPS. It could be demonstrated also with phenyl-*p*-quinone at doses of 20 and 40 μg per mouse.

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Interaction between doxapram and pentobarbitone in the mouse

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The classification of doxapram as a non-specific analeptic (e.g. Goodman & Gilman, 1975) has been challenged by Cohn (1974) and Kraynack, Cohn & others (1976), who found that doxapram potentiated amylobarbitone sleeping time in the rat. This result was in contrast to those for other 'analeptics' such as nikethamide, which had no effect on amylobarbitone sleeping time and picrotoxin which reduced amylobarbitone sleeping time.

The present study was designed to determine whether there was an interaction between the barbiturates and doxapram in mice and if so to examine some possible mechanisms for this interaction.

Female mice (25–30 g) of the Manchester Strain were used and all drugs were administered intraperitoneally.

After injection of pentobarbitone sodium, 30 mg kg^{-1} the time to onset of ataxia and loss of righting reflex (induction time) was noted. Sleeping time was measured as the duration of loss of righting reflex. Finally the time to full recovery, i.e. no discernible ataxia or sedation, was noted. Since this last parameter was extremely subjective a second observer with no knowledge of which drugs had been injected assessed the time to full recovery.

In separate experiments, respiratory rates were measured using the method of Bousfield & Rees (1969). Blood barbiturate concentrations were measured using the method of Brodie, Burns & others (1953). Each determination was made on the pooled blood from 4 mice killed 30 min after the pentobarbitone injection. LD50 values for doxapram were calculated using the method of Litchfield & Wilcoxon (1949). 20 mice were used for each dose of drug and observations were continued for 24 h.

Mice receiving both pentobarbitone sodium and doxapram hydrochloride were given both drugs simultaneously. In control experiments when SKF 525A was used, this drug was injected 45 min before the barbiturate.

Doxapram caused a dose-dependent increase in the hypnotic action of pentobarbitone (Table 1). The lowest dose used, 10 mg kg^{-1} doxapram caused a significant prolongation of the time necessary for full recovery, whilst the highest dose used, 100 mg kg^{-1}, decreased induction time, increased sleeping time and prolonged the time to full recovery. Mice given these doses of doxapram alone did not become noticably ataxic, nor did they loose their righting reflex.

Potentiation of the actions of pentobarbitone by doxapram was also seen in the respiratory rate measurements. Table 2 shows the change in respiratory rate in mice treated with 100 mg kg^{-1} doxapram and 30 mg kg^{-1} pentobarbitone 30 min after injection. Doxapram also

Table 1. *Effects of doxapram and SKF 525A on the hypnotic activity of an intraperitoneal injection of 30 mg kg^{-1} pentobarbitone in the mouse.*

	Time (min): mean (range) n = 18		
	Induction†	Sleep	Full recovery
Doxapram 100 mg kg^{-1}	6·9 (3·3–∞)**	38·8 (0–140)**	All > 240**
Concurrent controls	8·8 (6·6–∞)	10·0 (0–20·4)	59 (47–70)
Doxapram 50 mg kg^{-1}	7·1 (5·0–∞)	26·9 (0–90)*	(180–>240)**
Concurrent controls	7·9 (4·7–13·2)	16·8 (2·9–42)	62·5 (45–85)
Doxapram 25 mg kg^{-1}	9·7 (7·0–14·0)	16·3 (9·1–32·0)	150·5 (90–205)**
Concurrent controls	10·4 (6·0–15·1)	15·0 (7·0–23·9)	76·9 (60–93)
Doxapram 10 mg kg^{-1}	10·6 (7·1–14·9)	7·9 (4·0–17·3)	63·9 (53–75)**
Concurrent controls	9·8 (8·0–13·0)	7·1 (4·1–15·0)	52·9 (41–75)
SKF 525A 10 mg kg^{-1}	7·2 (4·9–10·5)**	86·8 (55–144)**	All > 240**
Concurrent controls	10·6 (6·0–∞)	13·5 (0–43·6)	79·0 (63–91)

* $0·1 < P > 0·5$; ** $P < 0·05$ compared with concurrent control values using the Mann Whitney 'U' test.
† Means exclude mice not losing their righting reflex.

* Correspondence.

Table 2. *Effect of 100 mg kg^{-1} doxapram and 10 mg kg^{-1} SKF 525A on the respiratory rate and blood barbiturate concentration in mice treated with 30 mg kg^{-1} pentobarbitone for 30 min.*

	Change in respiratory rate breaths min^{-1}†	Pentobarbitone concn μg ml^{-1}
Saline + pentobarbitone	−26·1 ± 9·9	18·7 ± 1·4
Doxapram + pentobarbitone	−50·7 ± 6·9*	23·5 ± 0·8*
SKF 525A + pentobarbitone	−78·3 ± 10·4*	29·7 ± 3·9*

Results are expressed as means ± s.e. of not less than 8 determinations.
* $P < 0.05$ significance from saline + pentobarbitone results.
† Calculated from preinjection control values.

increased the duration of the respiratory depression caused by pentobarbitone and this roughly paralleled the increase in sleeping time. Respiratory rate depression with pentobarbitone was also increased by 50 mg kg^{-1} doxapram ($P < 0.05$), although none of the doses of doxapram used had any significant effects on respiratory rate when administered alone. It has been shown that although doxapram is a respiratory stimulant its major effect is on tidal volume and it has little effect on respiratory rate in most animal species (Luscombe & Nicholls, 1971; Kenneth & Wang, 1974).

Table 2 also shows the blood pentobarbitone concentration 30 min after injection in mice treated with 100 mg kg^{-1} doxapram. These mice had significantly higher blood barbiturate concentrations than mice treated with pentobarbitone alone.

Both Tables 1 and 2 also show the effect of 10 mg kg^{-1} SKF 525A on the responses to pentobarbitone measured in this study. The results obtained were qualitatively similar in all respects to those obtained with the higher dose of doxapram. Although the sleeping time was increased more by SKF 525A than by doxapram the barbiturate blood concentrations were correspondingly greater.

The experiments in which the effects of pentobarbitone on the lethal dose of doxapram were examined showed that the LD50 value for doxapram alone (267·6 mg kg^{-1}, 95% confidence limits 193–298) was not significantly altered in mice treated with 30 mg kg^{-1} pentobarbitone (270·9 mg kg^{-1}, 95% confidence limits 239–347). A higher dose of pentobarbitone, 60 mg kg^{-1}, did reduce the LD50 value for doxapram to 160 mg kg^{-1} (95% confidence limits 145–185). In conjunction with this dose of pentobarbitone, doxapram's potentiating action could increase the brain concentration of pentobarbitone to lethal concentrations as the LD50 value for pentobarbitone alone in mice is 130 mg kg^{-1} (Barnes & Eltherington, 1973). Thus whilst doxapram potentiates the actions of pentobarbitone, there is no evidence that pentobarbitone increases the toxicity of doxapram. This contrasts with the interaction between doxapram and narcotic analgesics (Gregoretti & Pleuvry, 1977; Pleuvry, 1978) where morphine and other opiates enhance the toxicity of doxapram in rodents.

In summary, doxapram appears to potentiate the actions of pentobarbitone in mice through a mechanism involving increased blood concentrations of the barbiturate. There were no obvious differences between this action of doxapram and a similar action by SKF 525A which is known to inhibit the mixed function oxidase system necessary for the metabolism of barbiturates (Axelrod, Reichenthal & Brodie, 1954).

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