Limits to equipotent molar ratios

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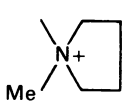
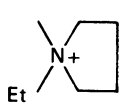
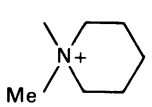
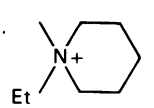
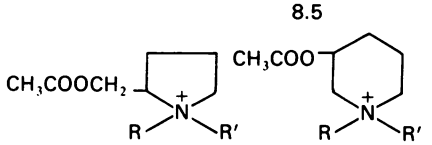
Some compounds are remarkably weak but still appear to be full agonists. Examples are shown in Table 1, where the numbers indicate the mean equipotent molar ratios relative to acetylcholine on the guinea-pig isolated ileum in Tyrode's solution at 37°C. With the very weak compounds, however, there is a large variation in the results and it is possible that they may be acting indirectly, perhaps by releasing acetylcholine (Henderson, Ariëns, Ellenbroek & Simonis, 1968). It is therefore necessary to consider likely limits to equipotent molar ratios and whether high ratios by themselves indicate that compounds are not acting directly on receptors.

If efficacy is unaltered, the equipotent molar ratio depends only on the difference in affinity. If efficacy is reduced, the limit to the equipotent molar ratio depends on the proportion of receptors occupied by the stronger agonist (Stephenson, 1956). For acetylcholine and the guinea-pig ileum this appears to be around 2% (Van Rossum, 1966), varying from preparation to preparation, so the equipotent molar ratio could be at least 50 times the difference in affinity without any detectable change in slope. Although in some molecules the insertion of a methylene group can reduce affinity 40-fold (Abramson, Barlow, Franks & Pearson, 1974) or even 100-fold (Barlow & Casy, 1975), the replacement of trimethylammonium by triethylammonium in antagonists did not reduce affinity more than 2-fold and replacement by ethylpiperidinium did not reduce it more than 4.5-fold (Abramson *et al.* 1974). The values for the acetoxy compounds with these onium groups in Table 1 seem therefore to be too large for an action exclusively at receptors.

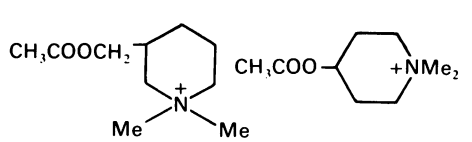
In tissues where more receptors are occupied the limit will be lower. On the frog rectus, which has a very low receptor reserve (Michelson & Shelkovnikov, 1976), the triethylammonium and ethylpiperidinium compounds are partial agonists (Barlow, Bremner & Soh, 1977).

Table 1 Equipotent molar ratios relative to acetylcholine on the guinea-pig isolated ileum at 37°C

$CH_3COOCH_2CH_2-$	$-\overset{+}{N}Me_3$	$-\overset{+}{N}Me_2Et$	$-\overset{+}{N}MeEt_2$	$-\overset{+}{N}Et_3$
	1	2.8 BS 2.5 HI	380 BS 700 HI	4025 BB 1700 HI
				
	2.6 C 3.6 S 3.9 BB 8.5	171 BB 217	3520 BB 3410	5620 BB 15200



R=R′=Me	2925 ±18 (5)	R=R′=Me	156 ±2 (6)
R=Me, R′=Et	11030 ±200 (3)	R=Me, R′=Et	8860 ±100 (2)



	1700 ±12 (3)		77 ±3 (4)

Results are taken from Holton & Ing (1949; HI), Barlow, Scott & Stephenson (1963; BS*), Cho, Jenden & Lamb (1972; C), Schwarzenfeld & Whittaker (1977; S), Barlow, Bremner & Soh (1977; BB*): all other results are from Abramson (1964*) and include mean ± s.e. mean and number of estimates in the lower part of the table. The asterisk indicates studies made in the presence of hexamethonium.

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A study in the guinea-pig of the pharmacokinetics and pharmacodynamics of cytosine arabinoside

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Cytosine arabinoside (Ara-C) is a nucleoside analogue with useful antileukaemic activity in man (Armentrout & Burns, 1974). However, it has a plasma half-life of 6–20 min (Momparler, 1974), being excreted in the urine and deaminated to an inactive metabolite – uracil arabinoside. Ara-C itself is inactive, but after phosphorylation to the active metabolite cytosine arabinoside triphosphate (Ara-CTP) inhibits DNA polymerase (Furth & Cohen, 1968). I have studied the relationsip *in vivo* in the guinea-pig between plasma Ara-C concentration and bone marrow intracellular Ara-CTP, and *in vitro* the relationship between Ara-C concentration, intracellular Ara-CTP and inhibition of DNA synthesis.

Twenty male Dunklin Hartley guinea-pigs, weight 350–450 g, were injected i.v. through indwelling jugular venous cannulae with [^{3}H]-Ara-C 2 mg/kg. This dose produces bone marrow depression when given daily i.v. for seven days. Serial blood samples were taken to measure plasma Ara-C levels. At various times up to 5 h after injection the animals were killed and bone marrow Ara-CTP measured. [^{3}H]-Ara-C was measured after separation on Ag50W-1X4 200–400 mesh ion-exchange columns. [^{3}H]-Ara-CTP was measured following separation on PEI ion-exchange plates or by column chromatography on Ag 1 × 8 200–400 mesh columns.

For *in vitro* studies, guinea-pig bone marrow was suspended in Eagle's medium and incubated with 100 nM [^{3}H]-thymidine (TdR). The effect of Ara-C (10 nM to 100 μM) on the inhibition of TdR incorporation into DNA was studied. In a parallel series of incubations using [^{3}H]-Ara-C and unlabelled TdR, the [^{3}H]-Ara-CTP production was measured.

In vivo, the plasma Ara-C levels declined in a biphasic manner after the initial distribution phase. The first phase had a half-life of 27 ± 6 min and the second phase 60 ± 8 minutes. An hour after the injection of [^{3}H]-Ara-C the plasma Ara-C level had fallen to 439 ± 8.5 nM whearas the bone marrow Ara-CTP concentration had risen to 1.8 ± 0.34 μM, and then declined exponentially with a half-life of 143 ± 33 minutes. By 5 h the plasma Ara-C concentration was 62 ± 6 nM. *In vitro* this level of Ara-C produced 50 ± 3% inhibition of DNA synthesis. However, the bone marrow Ara-CTP level *in vivo* was 810 ± 90 nM and