INVERTEBRATE PHARMACOLOGICAL ASSAY OF NOVEL, POTENT GLUTAMATE ANTAGONISTS: ACYLATED SPERMINES

Ian S. Blagbrough*, Mary Bruce, Barrie W. Bycroft*, Alan J. Mather*, and Peter N.R. Usherwood, Department of Pharmaceutical Sciences* and Department of Zoology, University of Nottingham, Nottingham NG7 2RD UK

The discovery of new drugs to combat disease associated with the L-glutamate gated ion channels of mammalian central nervous system (and the development of compounds which may have potential as novel pesticides by antagonism of related channels in invertebrates) require a pharmacological screen that is sensitive to micromolar concentrations of drug/toxin. Our investigation of peptidergic amine action on an insect nerve-muscle system (Usherwood and Blagbrough 1989a and 1989b) has demonstrated reversible, non-competitive antagonism of quisqualate-sensitive glutamate receptors (quis-GluR) at micromolar concentrations. Quis-GluR are present in both vertebrates and invertebrates.

Substituted polyamines were tested on the metathoracic retractor unguis nerve muscle preparations of the locust Schistocerca gregaria (Usherwood and Machili 1968). Quis-GluR antagonism was inferred from the depression of the twitch contraction evoked by stimulation (at 0.22 Hz or at 0.7 Hz) of the excitatory (glutamatergic) motoneurones which innervate the retractor unguis muscle. In some experiments the postjunctional quis-GluR of this muscle were excited by bath application of L-glutamate; in others the receptor channel was opened by the endogenous agonist (L-glutamate). Dose response curves were constructed from the twitch contraction data in the presence and absence of antagonist. IC-50 values for the inhibition by peptidergic amines and acylated spermines were standardised using the synthetic toxin philanthotoxin-343 (Eldefrawi et al 1988; Blagbrough et al 1989).

Table 1. Quis-GluR antagonism by acylated spermines.

Acylated Polyamine	IC-50 (μM)
N-(2-Hydroxyphenylacetyl)-spermine	62
N-(4-Hydroxyphenylacetyl)-spermine	8.7
N-(4-Hydroxycinnamoyl)-spermine	60
N-(4-Hydroxyphenylpropanoyl)-spermine	6.0
Philanthotoxin-343	23

The above derivatives and analogues of arthropod venom toxins (Table 1) have been shown to be potent non-competitive antagonists of L-glutamate gated non-specific cation channels on insect skeletal muscle. The potential for such antagonists in the discovery and development of new pharmaceutical agents and novel pesticides is under active investigation.

We thank the British Technology Group for generous financial support, and Dr. T. Smith for his interest in our work.

Blagbrough, I.S. et al (1989) J. Pharm. Pharmacol. 41: Suppl. 95P
Eldefrawi, A.T. et al (1988) Proc. Natl. Acad. Sci. USA 85: 4910-4913
Usherwood, P.N.R., Blagbrough, I.S. (1989a) in "Progress and Prospects in Insect Control BCPC Mono. No. 43" (Ed. N.R. McFarlane), BCPC, 45-58
Usherwood, P.N.R., Blagbrough, I.S. (1989b) in "Insecticide Action From Molecule to Organism" (Eds. T. Narahashi & J.E. Chambers), Plenum, 13-31
Usherwood, P.N.R., Machili, P. (1968) J. exp. Biol. 49: 341-361