100,000 *g* for 30 minutes. The fraction lying at the 0.9/1.45 M interface contained glial cells while the fraction at the 1.45/2.0 M interface contained neurones.

Microscopy revealed some possible contamination of the neuronal fraction by glia or debris but no neurones could be seen in the glial fraction. The respiratory rates of the two fractions were similar (500 nmol O_2.mg protein $^{-1}h^{-1}$) but the carbonic anhydrase (4.4 ± 0.2 μmol CO_2.mg protein $^{-1}h^{-1}$) and Na^+,K^+-ATPase (7.7 ± 2.3 μmol.Pi.mg protein $^{-1}h^{-1}$) activities of the glial fraction were two fold and seven fold higher respectively than those of the neuronal fraction. A manometric method (modification of Meldrum & Roughton 1933) was used for carbonic anhydrase and the method described previously (Gilbert & Wyllie, 1976) for ATPase. Sodium-activated magnesium-dependent ATPase activity (Gilbert & Wyllie, 1975) was evident in both fractions but only the activity of the neuronal fraction was sensitive to inhibition by the anticonvulsant sodium valproate (1mM).

The Na^+,K^+-ATPase activity of the glial fraction was markedly dependent on the medium potassium concentration in the range 0–20 mM whereas the activity of the neuronal fraction was not, and, in addition, the glial fraction exhibited potassium-activated ATP-hydrolysing activity which was not significant in the neuronal fraction.

These results suggest there are differences in enzyme activities in glial cells and neurones which may be linked to ion pumps; that the sensitivities of the cells to drugs differ, and in particular, that activities are detectable in the glial fraction which are compatible with the hypothesis that these cells are specialised for removing potassium ions from the interstitial fluid.

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Actions of γ-acetylenic GABA on single central neurones in rat

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γ-Acetylenic GABA (GAG; RMI 71645) is a potent enzyme-activated irreversible inhibitor of GABA-transaminase. It increases brain GABA concentrations (Jung, Lippert, Metcalf, Schechter, Böhlen & Sjoerdsma, 1977) and has a significant anticonvulsant effect (Schechter, Tranier, Jung & Sjoerdsma, 1977). However, at high dose levels (~250 mg/kg i.p. or 50 μg and above when administered by injection into the cerebral ventricles), GAG produces marked excitation, myoclonic jerks and occasionally convulsions (Palfreyman, Huot, Lippert & Schechter, 1978). We have investigated the effects of GAG on the activity of single neurones to see if an explanation for this stimulant effect can be found.

Extracellular recordings were made from GABA-sensitive cells in the medullary reticular formation and the nucleus accumbens of urethane-anaesthetised rats. Drugs were applied iontophoretically from multibarreled micropipettes. The majority of cells were firing spontaneously but some were driven by the continuous application of (±)-homocysteic acid.

GAG reduced the firing rate of approximately 25% of the cells studied. This effect was rapid in onset and its time-course was similar to that of the ejection current.

In a slightly larger proportion of neurones GAG clearly and reversibly antagonised the actions of GABA, an effect which sometimes considerably outlasted the period of GAG application. This antagonism could be overcome if the GABA ejection current was increased, indicating that the antagonism was competitive.

GAG's depression of neuronal firing rate would be expected to contribute to the anticonvulsant effects previously attributed solely to the established action of GABA-transaminase inhibition. The behavioural excitation seen at high dose levels, on the other hand, might reflect GABA antagonism at the single-cell level as described here.

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The action of N-methyl-D-aspartic and kainic acids on motoneurones with emphasis on conductance changes

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Interactions of putative blockers with neuronal responses to excitatory amino acid analogues seem to indicate that there are two distinct agonist receptors (see review by Watkins, 1978): (a) a 'glutamate' receptor (with which the conformationally restricted analogue kainic acid preferentially interacts), (b) an 'aspartate' receptor (with which N-methyl-D-aspartate (NMDA) preferentially interacts). We have investigated the membrane conductance (G_M) changes underlying the response to these analogues.

L-glutamate (1 M), L-aspartate (1 M), kainate (20 mM), NMDA (0.2 M) and D-homocysteate (0.2 M) (all *ca* pH 8, ejected as anions) were applied iontophoretically from the outer barrels of a coaxial electrode to cat lumbar motoneurones (the screened central barrel recording intracellularly).

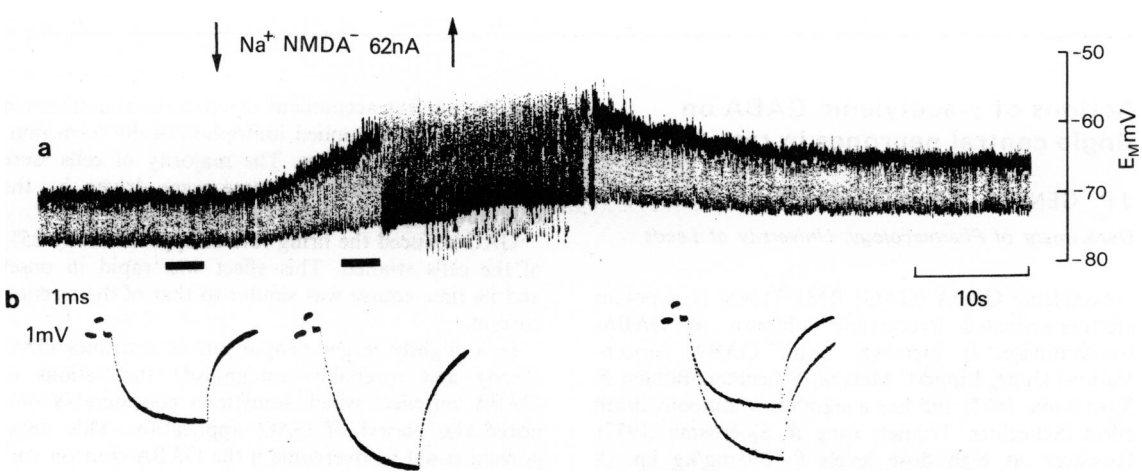

Figure 1 Depolarizing response of a lumbar motoneurone in a decerebrated cat to a current balanced iontophoretic application of N-methyl-D-aspartate (NMDA). The membrane potential record (E_M) is modulated by conductance measuring pulses (−3 nA, 12 ms constant current pulses injected through the screened recording electrode) and the AHPs of NMDA evoked firing (wide, dark band). High frequency repetitive firing resulted when NMDA had depolarized the cell by 9 mV and continued for 12 s after the ejecting current was turned off. The membrane potential subsequently recovered. Conductance measuring pulses below the potential record were averaged during the periods shown by the black bars (30 samples) and are shown superimposed on the right. Just before the NMDA induced firing, G_M had decreased to 62% of the control value.