Conclusions

• The present study has successfully developed a method to simultaneously measure extracellular glutamate, transmitter levels and locomotor behaviour in a novel model for schizophrenia.

• Acute ketamine administration induced hyperlocomotion in mice in a dose-dependent manner. A 30 mg/kg dose produced an immediate tonic response (visual observation) and an increase in grooming activity.

• The presence of drug in the extracellular space in a concentration dependent manner confirms that only a limited concentration of ketamine is required in the extracellular space to induce a maximal locomotor effect and any further blockade of NMDA receptors may act to further depress locomotor activity and produce other effects. Therefore selecting the dose of ketamine to produce a maximal locomotor effect without causing any unwanted side-effects remains crucial in designing this animal model.

• A dose between 3 and 10 mg/kg is a range optimal for inducing hyperactivity in mice without inducing pronounced side-effects.

• Despite discrepancies in the literature, NMDA antagonism increased glutamate levels in rat cortex and this has been correlated with activated NMDA receptors. Antagonism of NMDA receptors increased glutamate levels in rat cortex and this has been correlated with activated NMDA receptors.

• The locomotor data of ketamine obtained from LABORAS and EthoVision systems are qualitatively similar.

• The quantitative difference can be attributed to technical issues in measuring locomotor activity and recording the locomotor profile.

Figure 3. Effect of ketamine on grooming activity. Treatment occurred at 0 min.

Figure 4. Effect of ketamine on LMA: a comparison of LABORAS and EthoVision systems.

Introduction

Current treatments for schizophrenia do not possess complete therapeutic efficacy, nor are they devoid of unwanted side-effects. The need to create better and safer treatments is met by a need to develop animal models mimicking the core pathophysiological deficits of the disease. Psychotomimetic-induced models in mice are widely accepted and useful, however, for ethical reasons in mice a single model at 3 presentation some components of the striatal activation are not possible. One of the major limitations of the use of psychotomimetics is the requirement for a single presentation of a drug to induce a measurable effect in the brain. Here we report that positive symptoms of schizophrenia stem from the activity of glutamatergic NMDA receptors (Curti et al 2006). NMDA receptor antagonists such as ketamine have also been shown to induce a behavioral symptomatology in rodents that is characterized by not only locomotor hyperactivity but also altered feeding, sleep and stereotypic behavior.

Whilst the behavioral and some of the neurochemical effects induced by NMDA antagonist receptor agonists can be observed in the present study by altering this model by developing a method to freely moving mice in which both parameters can be measured simultaneously in the same animal. The behavioral and neurochemical effects are compared to the pharmacokinetic profile of ketamine. This present study is the first to our knowledge, which attempts to develop valid reliable animal models of schizophrenia. The similarities and differences between the behavioral and neurochemical changes induced by NMDA antagonists and the drug pharmacokinetics is measured simultaneously in the same animal.

Methods

Animals and Apparatus: Male C57 mice (Harlan, UK), weighing 20-35g at the start of the experiment were used. Procedures were conducted in accordance with the UK home office (scientific Procedures Act, 1986) and all animals were subjected to a minimum of 1 week handling prior to experimentation. Food and water were provided ad libitum. The experimental cages were made of clear plexiglass and the apparatus was designed to allow for the simultaneous measurement of both locomotor activity and microdialysis. Locomotor activity was recorded using LABORAS (Laboratory Animal Behavior Observation, Registration And Analysis System, version 2.1, Maha. B. Inc. Hollandla, Netherlands), an automated behavior registration system. LABORAS recorded the behavioral profile of each mouse via a sensory platform on which its cage sit. The platform translated the mechanical oscillations caused by the mouse’s movement to sensor signals which were amplified, shaped, and converted to digital signals. The frequency and amplitude of the sensor signals were converted to a locomotor activity index.

References

Knable MB & Weinberger DR (1997) Dopamine, the prefrontal cortex and schizophrenia. Biological Psychiatry, 42: 697-706.


Basal and N-Methyl-D-Aspartate LY379268

• The locomotor data of ketamine obtained from LABORAS and EthoVision systems are qualitatively similar.

• The quantitative difference can be attributed to technical issues in measuring locomotor activity and recording the locomotor profile.

Experimental procedure: On the morning of the experiment, mice were attached to the microdialysis system. Microdialysis probes were inserted and flushed with artificial cerebrospinal fluid (aCSF: 147mM NaCl; 3mM KCl; 1.2mM CaCl2; and 1.2mM MgCl2) at a flow rate of 1.25 μl per minute, 30 min interval. A 2-hour stabilisation period was given prior to collection of samples. Following the collection of 3 baseline samples, 6 sample fractions were collected after reversible pharmacological intervention. Samples were collected between 10 and 120 min post-treatment.

After completion of experiments probe placement was validated. The microdialysis experiments were performed simultaneously with recording of locomotor activity.

Schematic representation and typical probe placement of the mouse mPFC

Experimental protocol for combined LMA and microdialysis

Figure 1. Effects of ketamine on extracellular glutamate levels in mPFC, profile superimposed with LMA

Figure 2. Ketamine levels mPFC dialysates

• Data (n=6) are expressed as mean ± SEM and corrected for delay (10 min) in sample recovery time to compensate the length of tubing between probe and collection vial. The in-vitro recovery of ketamine through the dialysis membrane is 7.37%. Ketamine administered at 0 min.

• A dose dependent increase in extracellular drug concentration was noted.

• The decrease in locomotor response despite the increase in grooming activity was observed.

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