Biosensor-based monitoring of ethanol toxicokinetics in the brain of freely-moving animals

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Abstract

Alcohol has been a continuous presence in the evolution of mankind and indeed, nowadays, represents the most widespread psychotropic agent in Western societies. As many substances of abuse, ethanol activates the mesolimbic pathway, so alcohol addiction has become one of the most important health issues. Given these implications, it has become of primary importance the real-time monitoring of ethanol toxicokinetics and its effects in definite brain regions. Recently, implantable amperometric biosensors have offered a valid alternative to microdialysis for monitoring brain metabolites. In this study, we present the characterization of an implantable amperometric biosensor for the real-time measurement of brain ethanol by improving a prior implantable design and integrating it in a biotelemetric device, to consent freely movements to the animals. The biosensor was then implanted in in the Shell of the Nucleus Accumbens by stereotaxic procedures in rats already conditioned to carry ethanol self-administration. After the surgery, we waited for the animal's recovery with water and food ad libitum. The night before the experiments, the animals were left without water for 12 hours. The following day the biosensor was polarized and amperometric signal from ethanol was monitored and recorded up to its stabilization. At this stage, the animals had at their disposal, for 30 minutes, a solution containing 10% of ethanol. In this period, we were able to detect a significant increase of the current derived from ethanol when the animal swallowed the ethanol solution. Ethanol is one of the most widespread psychotropic agents in western society. While its psychoactive effects are mainly associated with GABAergic and glutamatergic systems, the positive reinforcing properties of ethanol are related to activation of mesolimbic dopaminergic pathways resulting in a release of dopamine in the nucleus accumbens. Given these neurobiological implications, the detection of ethanol in brain extracellular fluid (ECF) is of great importance. In this study, we describe the development and characterization of an implantable biosensor for the amperometric detection of brain ethanol in real time. Ten different designs were characterized in vitro in terms of Michaelis–Menten kinetics (VMAX and KM), sensitivity (linear region slope, limit of detection (LOD), and limit of quantification (LOQ)), and electroactive interference blocking. The same parameters were monitored in selected designs up to 28 days after fabrication in order to quantify their stability. Finally, the best performing biosensor design was selected for implantation in the nucleus accumbens and coupled with a previously developed telemetric device for the real-time monitoring of ethanol in freely moving, untethered rats. Ethanol was then administered systemically to animals, either alone or in combination with ranitidine (an alcohol dehydrogenase inhibitor) while the biosensor signal was continuously recorded. The implanted biosensor, integrated in the low-cost telemetry system, was demonstrated to be a reliable device for the short-time monitoring of exogenous ethanol in brain ECF and represents a new generation of analytical tools for studying ethanol toxicokinetics and the effect of drugs on brain ethanol levels. Ethanol is one of the most widespread psychotropic agents in western society. 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Among the techniques that aim to measure ethanol concentration in the brain, microdialysis has been the most widely used, but because of its invasiveness, associated with low temporal resolution, and the necessity of using connecting tubes to carry out the experiments, it is not particularly suitable for clinical trials. Recently, electrochemical biosensors, also minimally invasive, have been developed, which offer the possibility of monitoring the real-time variations of ethanol concentrations in the brain of animal models due to the very small dimensions of the transducer electrode. Recently, non-invasive methods have been used for

the direct monitoring of alcohol in the brain, which use spectroscopic techniques such as magnetic resonance spectroscopy and magnetic resonance imaging or positron emission tomography, which are principally used to monitor ethanol metabolites. The aim of this review is to discuss all the techniques used to monitor brain ethanol and highlight their strengths and weaknesses.

Biography:

Gaia Rocchitta completed her PhD in Neuroscience at School of Medicine of Sassari University (Italy) in 2004 and her postdoctoral studies at School of Chemistry & Chemical Biology, University College, Dublin (Ireland) in 2006. She currently is a tenured researcher and lecturer in pharmacology and nutraceutical at School of Medicine of Sassari University (Italy). She has published more than 50 papers in peer-reviewed journals.