Biochemistry and Biophysics Reports 6 (2016) 197–202

Contents lists available at ScienceDirect

Biochemistry and Biophysics Reports

journal homepage: www.elsevier.com/locate/bbrep

Potentiating effect of glabridin from *Glycyrrhiza glabra* on $GABA_A$ receptors

Katrin M. Hoffmann, Leopoldo Beltrán, Paul M. Ziemba, Hanns Hatt, Günter Gisselmann *

Ruhr-University Bochum, Department of Cell Physiology, Bochum, Germany

ARTICLE INFO

Article history:
Received 22 December 2015
Received in revised form
7 April 2016
Accepted 14 April 2016
Available online 16 April 2016

Keywords:
$GABA_A$ Receptor
Glabridin
Insomnia
Anxiety
Xenopus Oocytes
Glycyrrhiza glabra

ABSTRACT

Extracts from *Glycyrrhiza* are traditionally used for the treatment of insomnia and anxiety. Glabridin is one of the main flavonoid compounds from *Glycyrrhiza glabra* and displays a broad range of biological properties. In the present work, we investigated the effect of glabridin on $GABA_A$ receptors. For this purpose, we employed the two-electrode voltage-clamp technique on *Xenopus laevis* oocytes expressing recombinant $GABA_A$ receptors. Through this approach, we observed that glabridin presents a strong potentiating effect on $GABA_A$ $\alpha1\beta(1-3)\gamma2$ receptors. The potentiation was slightly dependent on the β subunit and was most pronounced at the $\alpha1\beta2\gamma2$ subunit combination, which forms the most abundant $GABA_A$ receptor in the CNS. Glabridin potentiated with an EC_{50} of 6.3 ± 1.7 μM and decreased the EC_{50} of the receptor for GABA by approximately 12-fold. The potentiating effect of glabridin is flumazenil-insensitive and does not require the benzodiazepine binding site. Glabridin acts on the β subunit of $GABA_A$ receptors by a mechanism involving the M286 residue, which is a key amino acid at the binding site for general anesthetics, such as propofol and etomidate. Our results demonstrate that $GABA_A$ receptors are strongly potentiated by one of the main flavonoid compounds from *Glycyrrhiza glabra* and suggest that glabridin could contribute to the reported hypnotic effect of *Glycyrrhiza* extracts.

1. Introduction

$GABA_A$ receptors are chloride-selective, heteropentameric ionotropic receptors that mediate fast inhibitory synaptic transmission in the central nervous system. They constitute a target for the majority of clinically relevant anesthetics, e.g., propofol and etomidate [1]. They are also targets for many neuroleptic, anxiolytic and anticonvulsant drugs [2,3]. $GABA_A$ receptors are composed of different combinations of the following subunits: α1–6, β1–3, γ1–3, δ, ε, π and ϑ [2]. The most prominent native receptors in the CNS are the post-synaptically localized heteromultimers of α, β, and γ subunits. The GABA-binding pocket is formed by the α/β-subunit interface, whereas the benzodiazepine-binding pocket is located at the α/γ interface.

More than ten distinct modulatory binding sites in $GABA_A$ receptors are currently known, and these constitute the target of many sedative, anticonvulsive, anxiolytic, antiepileptic, and hypnotic compounds of different chemical classes [4]. Benzodiazepine-site agonists act mainly on γ2-containing $GABA_A$ receptors. In contrast, several other modulators, such as the general anesthetics propofol or etomidate, target the β subunit [5]. Flavonoid modulation of $GABA_A$ receptors has been the focus of intense research for many years. The mechanism of potentiation is complex, only partially understood, and includes flumazenil-sensitive modulation at the benzodiazepine binding site, flumazenil-insensitive modulation at other sites and second-order modulation of benzodiazepine potentiation [6].

Glabridin is a polyphenolic flavonoid compound from liquorice (*Glycyrrhiza glabra*, *Fabaceae*) and is one of the main components of the flavonoid fraction. It has a wide range of biological properties, ranging from neuroprotective to skin-whitening (reviewed by [7]), and is used in dietary supplements, foods and cosmetic products. Liquorice extracts show hypnotic-sedative actions in animal models and are traditionally used for the treatment of insomnia and anxiety [7–9]. Prior research showed that low μM concentrations of glabridin strongly potentiate GABA-induced currents in rat dorsal raphe neurons [10]. However, until recently, no data regarding the effect of glabridin on recombinant $GABA_A$ receptors has been published, and the mechanism of potentiation has not yet been studied. In the present work, we addressed these questions and studied the effect of glabridin on different subtypes of heterologously expressed $GABA_A$ receptors.

* Correspondence to: Ruhr-University Bochum, Department of Cellysiology, Universitätsstr. 150, 44780 Bochum, Germany.
E-mail address: guenter.gisselmann@rub.de (G. Gisselmann).

http://dx.doi.org/10.1016/j.bbrep.2016.04.007

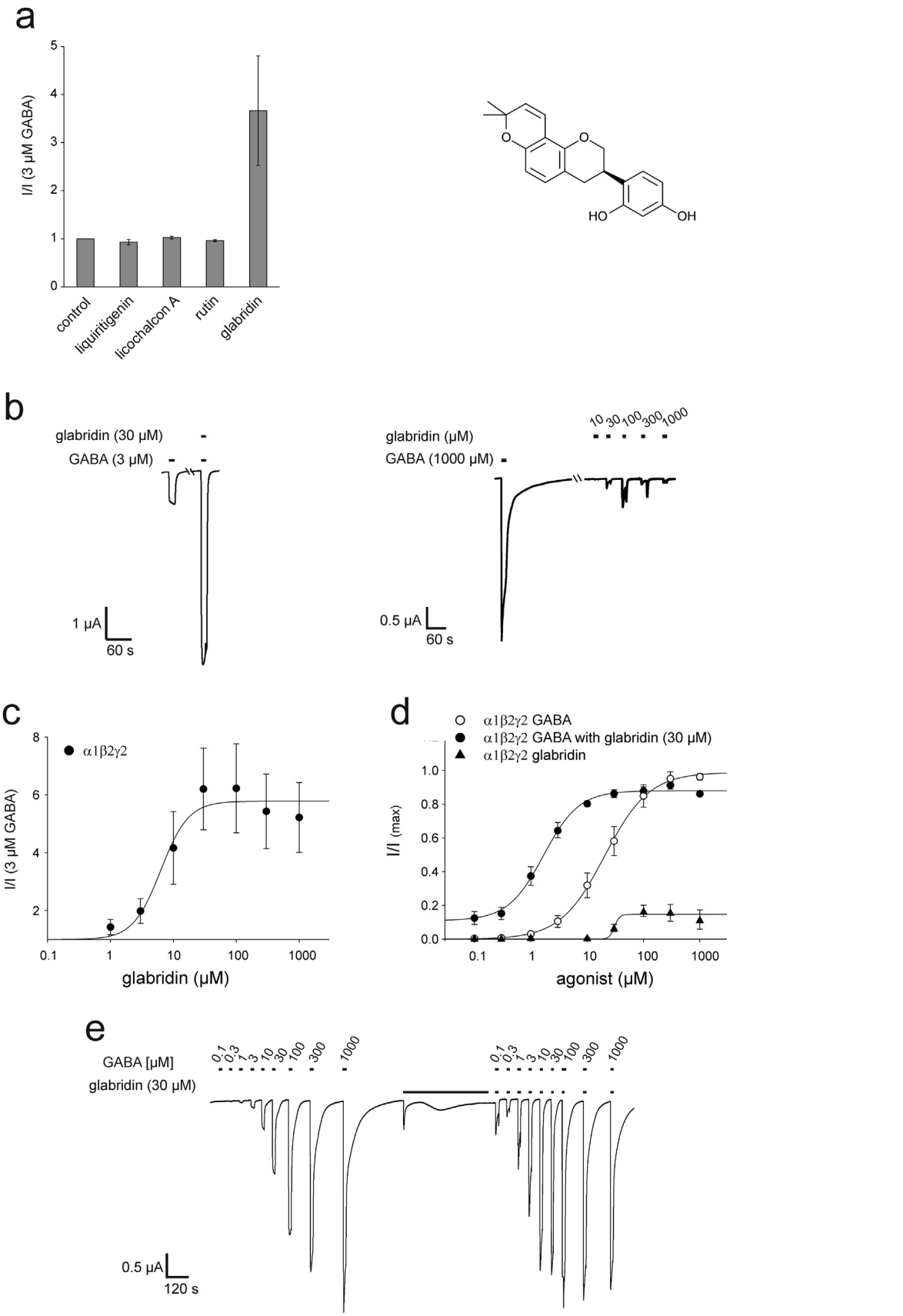

Fig. 1. Modulating effect of glabridin on recombinant $GABA_A$ receptors. (a) Several components (10 µM) were screened for the potentiation of GABA-induced currents (left, n=3–4). The chemical structure of glabridin (right). (b) Representative voltage-clamp recording of a *Xenopus* oocyte expressing the α1β2γ2 $GABA_A$ subtype exposed to 3 µM GABA in the absence and presence of glabridin 30 µM (left). Higher concentrations of glabridin leads to an activation of the $GABA_A$ receptor. (c) Dose-response relationship for the effect of glabridin on GABA induced currents (n of 4–6 oocytes). (d) Co-application of glabridin (30 µM) leads to a leftward shift in the dose-response curve of GABA on α1β2γ2 receptors (n of 4 oocytes). Higher concentrations of glabridin lead to an activation of the $GABA_A$ receptor (n of 4 oocytes). (e) Representative voltage-clamp recording of a *Xenopus* oocyte exposed to increasing concentrations of GABA in the absence (left) and presence (right) of 30 µM glabridin.