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Review Article

Neuropeptide receptors as potential antiepileptic drug targets: focus on the ghrelin axis

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Epilepsy is a very serious neurological disorder which is often underrepresented. Around 50 million individuals worldwide have active epilepsy with recurrent seizures and in spite of the medical advances over the years, 30% of these patients remain as drug resistant (Pati and Alexopoulos, 2010). Even after several years of research, there is still a lack of good understanding on the pathophysiology of seizure disorders (Perucca, 2011). Investigators in this field believe that there is a great need for novel antiepileptic drugs (AEDs) that act differently than the drugs available on the market. The majority of AEDs act by blocking sodium channels (phenytoin, carbamazepine) or by the augment of GABAergic transmission (phenobarbital, valproic acid). A newer generation of AEDs has expanded therapeutic options, however these are not superior to the older drugs (Hitiris and Brodie, 2006). Patients with mesial temporal lobe epilepsy (mTLE) are among the most pharmacoresistant to these medications (Pati and Alexopoulos, 2010). In order to attempt the rectification of this dilemma, the neuropharmacologist needs to not only try and find AEDs with new mechanisms of action, but to also keep in mind what information is currently available on the pathophysiology of epilepsy. It is clear that during the complicated process of epileptogenesis, several different mechanisms are taking place, thus one should ideally identify new compounds that are capable of targeting different pathways simultaneously. The focus of epilepsy researchers is to identify compounds that are not only capable of attenuating seizures (anticonvulsant), but are also antiepileptogenic (can prevent epilepsy) or disease-modifying (halting its progression).

A lot of interest is being shown towards neuropeptides as a way to suppress epileptic seizures. A number of neuropeptides have been extensively studied in the pathogenesis of epilepsy, such as neuropeptide Y, galanin and somatostatin. However the poor ability of these neuropeptides to penetrate the blood brain barrier (BBB) serves as the main obstacle for brain drug development (Robertson et al., 2011). Neuropeptide-based treatments present a number of advantages in comparison to treatments that target classical neurotransmitter systems and ion channels, both in efficacy as well as safety (Hokfelt et al., 2003; Portelli et al., 2012a). Since neuropeptides are normally released from neurons in the presence of high frequency firing or pathological conditions, a likely advantage is that the clinical effects of neuropeptide receptor antagonists will only become evident under epileptic conditions where high frequency firing is involved, which in turn reduces the risk of adverse effects. Ghrelin, a pleiotropic peptide that has excited the scientific community generally since its discovery in 1999, has recently been introduced in the field of epilepsy.

Ghrelin is produced both centrally and peripherally. Ghrelin requires modification on the serine-3 by O-acylation with octanoate in order to bind to the G-protein coupled receptor (GPCR) growth hormone secretagogue receptor type 1a (GHSR1a) (Kojima et al., 1999). The ghrelin receptor gene encodes two types of GHSR mRNA, known as 1a and 1b, resulting in

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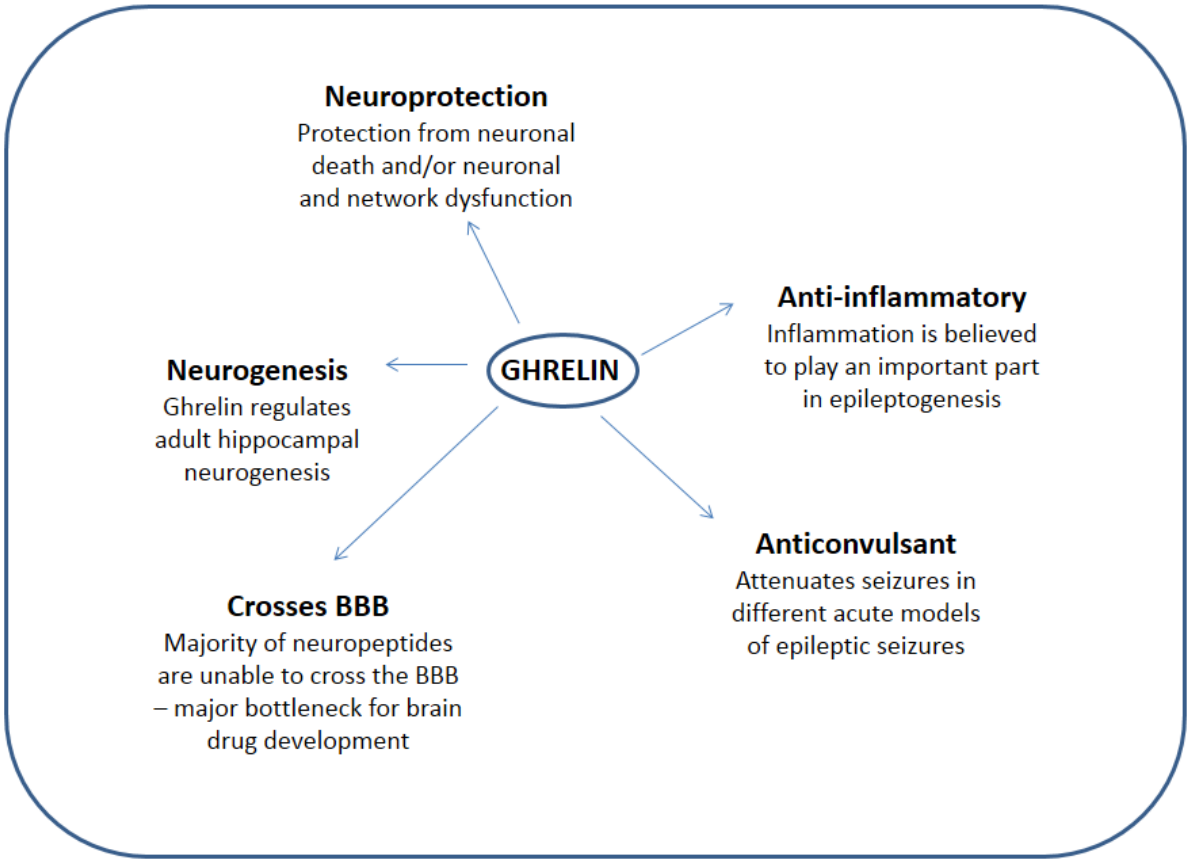

Figure 1: Some of the known effects of ghrelin, which makes this neuropeptide system a promising target for antiepileptic and antiepileptogenic treatments.

two isoforms: GHSR1a and GHSR1b (Camina, 2006). GHSR1a is given more importance, since GHSR1b is unable to bind or be activated by ghrelin. GHSR1a, as with ghrelin, is widely expressed both centrally and peripherally, including in seizure-prone regions such as the hippocampus. For a more detailed account of the cell biology of the ghrelin receptor, please refer to the review by Camina (2006).

In the past six years the ghrelin system has been reported to have anticonvulsant properties. Ghrelin has been shown to have an inhibitory effect on seizures induced by pentylenetetrazole (Obay et al., 2007; Obay et al., 2008), penicillin (Aslan et al., 2009), and kainic acid (Lee et al., 2010), but its anticonvulsant mechanism of action has remained elusive. Recently we have attempted to unravel ghrelin's anticonvulsant mechanism of action using the in vivo rat model for pilocarpine-induced limbic seizures, the mouse pilocarpine tail infusion model, transgenic mice with a GHSR deletion, electrophysiology in hippocampal slices, EEG recording in freely moving rats, and HEK293 cells expressing the human GHSR, to determine inverse agonism, activation, desensitization, internalization and resensitization (Portelli et al., 2012b). Ghrelin and the ghrelin-mimetic capromorelin attenuated pilocarpine-induced seizures in rats and mice. Experiments with transgenic mice established that ghrelin requires the GHSR for its anticonvulsant effect. Interestingly we found that $GHSR^{-/-}$ mice had a higher seizure threshold than $GHSR^{+/+}$ mice when administered the muscarinic agonist pilocarpine. This prompted us to look further into pharmacological modulation of the receptor, where we discovered that abolishing the constitutive activity of GHSR by inverse agonism results in the attenuation of seizures and epileptiform activity. We verified that ghrelin's potential to rapidly desensitize the GHSR is followed by internalization of the receptor and a slower resensitization process. This, together with our present novel findings that different ghrelin fragments possess similar agonistic potencies but different desensitization characteristics on the GHSR, led us to elucidate that ghrelin probably attenuated limbic seizures in rodents and epileptiform activity in hippocampal slices due to the desensitizing effect on the GHSR (Portelli et al., 2012b). This in turn constitutes a novel mechanism of anticonvulsant action whereby an endogenous agonist reduces the activity of a constitutively active receptor.

Ghrelin presents a number of advantages when compared to other well-established anticonvulsant neuropeptides. This neuropeptide can easily cross the BBB, and the ghrelin system has been attributed to affect a number of physiological processes, ranging from its potent anti-inflammatory and neuroprotective properties to its ability to protect the BBB and induce hippocampal neurogenesis (Moon et al., 2009; Portelli et al., 2012a) (Fig 1). Studies have shown that during the process of epileptogenesis, all previously mentioned physiological processes are negatively affected (Ravizza et al., 2006; Boer et al., 2008; Choi and Koh, 2008; Ravizza et al., 2008; Vezzani et al., 2011; Zlokovic, 2008; Coremans et al., 2010; Marchi et al., 2012; Parent and Kron, 2012; Vezzani et al., 2013). Recently, a clinical phase III trial for the ghrelin receptor agonist JMV1843 (Macimorelin), indicated for growth hormone deficiency in humans, has been completed and the drug was found to be well tolerated. It is now currently being developed by the company Æterna-Zentaris.

Our knowledge on the role of the ghrelin axis in the pathogenesis of epilepsy is still in its infancy. Targeting the ghrelin receptor has been shown to attenuate acute seizures in different models (Obay et al., 2007; Obay et al., 2008; Xu et al., 2009; Lee et al., 2010; Portelli et al., 2012a; Portelli et al., 2012b), and from what is already known with regard to this system's properties in view of inflammatory cascades, neuroprotection, neurogenesis, and BBB protection, it is promising that the ghrelin axis could play a role in the process of epileptogenesis.

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References

Aslan A., Yildirim M., Ayyildiz M., Guven A., Agar E. (2009). The role of nitric oxide in the inhibitory effect of ghrelin against penicillin-induced epileptiform activity in rat. *Neuropeptides*, 43, 295–302.

Boer K., Jansen F., Nellist M., Redeker S., Van den Ouweland A., Spliet W., van Nieuwenhuizen O., D. T., Crino P., Aronica E. (2008). Inflammatory processes in cortical tubers and subependymal giant cell tumors of tuberous sclerosis complex. *Epilepsy Res.*, 78, 7–21.

Camina J. (2006). Cell biology of the ghrelin receptor. *J. Neuroendocrinol.*, 18, 65–76.

Choi J., Koh S. (2008). Role of brain inflammation in epileptogenesis. *Yonsei. Med.*, 49, 1–18.

Coremans V., Ahmed T., Balschun D., D'Hooge R., DeVriese A., Cremer J., Antonucci F., Moons M., Baekelandt V., Reumers V., Cremer H., Eisch A., Lagace D., Janssens T., Bozzi Y., Caleo M., Conway E. (2010). Impaired neurogenesis, learning and memory and low seizure threshold associated with loss of neural precursor cell survivin. *BMC Neurosci.*, 11(2).

Hitiris N., Brodie M. (2006). Modern antiepileptic drugs: guidelines and beyond. *Curr. Opin. Neurol.*, 19, 175–180.

Hokfelt T., Bartfai T., Bloom F. (2003). Neuropeptides: oppurtunities for drug discovery. *Lancet Neurol.*, 2, 463–472.

Kojima M., Hosoda H., Date Y., Nakazato M., Matsuo H., Kangawa K. (1999). Ghrelin is a growth-hormone-releasing acylated peptide from the stomach. *Nature*, 402, 656–660.

Lee J., Lim E., Kim Y., Li E., Park S. (2010). Ghrelin attenuates kainic acid-induced neuronal cell death in the mouse hippocampus. *J. Endocrinol.*, 205, 263–270.

Marchi N., Granata T., Ghosh C., Janigro D. (2012). Blood-brain barrier dysfunction and epilepsy: pathophysiologic role and therapeutic approaches. *Epilepsia*, 53, 1877–1886.

Moon M., Kim S., Hwang L., Park S. (2009). Ghrelin regulates hippocampal neurogenesis in adult mice. *Endocr. J.*, 56, 525–531.

Obay B., Tasdemir E., Tumer C., Bilgin H., Atmaca M. (2008). Dose dependent effects of ghrelin on pentylenetetrazole-induced oxidative stress in a rat seizure model. *Peptides*, 29, 448–455.

Obay B., Tasdemir E., Tumer C., Bilgin H., Sermet A. (2007). Antiepileptic effects of ghrelin on pentylenetetrazole-induced seizures in rats. *Peptides*, 28, 1214–1219.

Parent J., Kron M. (2012). *Jasper's Basic Mechanisms of the Epilepsies - Neurogenesis and Epilepsy*. Bethesda (MD): National Center for Biotechnology Information (US), 4th edition.

Pati S., Alexopoulos A. (2010). Pharmacoresistant epilepsy: from pathogenesis to current and emerging therapies. *Cleve. Clin. J. Med.*, 77, 457–467.

Perucca E. (2011). The pharmacology of new antiepileptic drugs: does a novel mechanism of action really matter? *CNS Drugs*, 25, 907–912.

Portelli J., Michotte Y., Smolders I. (2012a). Ghrelin: an emerging new anticonvulsant neuropeptide. *Epilepsia*, 53, 585–595.

Portelli J., Thielemans L., Ver Donck L., Loyens E., Coppens J., Aourz N., Aerssens J., Vermoesen K., Clinckers R., Schaillier A., Michotte Y., Moechars D., Collingridge G., Bortolotto Z., Smolders I. (2012b). Inactivation of the constitutively active

ghrelin receptor attenuates limbic seizure activity in rodents. *Neurotherapeutics*, 9, 658–672.

Ravizza T., Boer K., Redeker S., Spliet W., van Rijen P., Troost D., Vezzani A., Aronica E. (2006). The IL-1beta system in epilepsy-associated malformations of cortical developement. *Neurobiol. Dis.*, 24, 128–143.

Ravizza T., Gagliardi B., Noe F., Boer K., Aronica E., Vezzani A. (2008). Innate and adaptive immunity during epileptogenesis and spontaneous seizures: evidence from experimental models and human temporal lobe epilepsy. *Neurobiol Dis.*, 29, 142–160.

Robertson C., Flynn S., White H., Bulaj G. (2011). Anticonvulsant neuropeptides as drug leads for neurological diseases. *Nat. Prod. Rep.*, 28, 741–762.

Vezzani A., French J., Bartfai T., Baram T. (2011). The role of inflammation in epilepsy. *Nat. Rev. Neurol.*, 7, 31–40.

Vezzani A., Friedman A., Dingledine R. (2013). The role of inflammation in epileptogenesis. *Neuropharmacology*, 69, 16–24.

Xu J., Wang S., Lin Y., Cao L., Wang R., Chi Z. (2009). Ghrelin protects against cell death of hippocampal neurons in pilocarpine-induced seizures in rats. *Neurosci. Lett.*, 453, 58–61.

Zlokovic B. (2008). The blood-brain barrier in health and chronic neurodegenerative disorders. *Neuron*, 57, 178–201.