Available online at www.sciencedirect.com
SCIENCE DIRECT
Brain Research 1066 (2005) 187 – 195
BRAIN RESEARCH
www.elsevier.com/locate/brainres

Research Report

Estrogen in the parabrachial nucleus attenuates the sympathoexcitation following MCAO in male rats

Tarek M. Saleh[a,b,*], Barry J. Connell[a,c], Alastair E. Cribb[a,b,c]

[a]Department of Biomedical Sciences, Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, P.E.I., Canada C1A 4P3
[b]Prince Edward Island Health Research Institute, University of Prince Edward Island, Charlottetown, P.E.I., Canada C1A 4P3
[c]Laboratory of Comparative Pharmacogenetics, University of Prince Edward Island, Charlottetown, P.E.I., Canada C1A 4P3

Accepted 21 October 2005
Available online 1 December 2005

Abstract

Recent investigations have provided evidence to suggest systemic estrogen administration prevented or reversed the sympathoexcitation observed following middle cerebral artery occlusion (MCAO) in male rats. The present investigation sought to determine the role of estrogen injected directly into the parabrachial nucleus (PBN) on the MCAO-induced sympathoexcitation as well as the role of the rostral ventrolateral medulla (RVLM) in mediating the sympathoexcitatory response. Male Sprague–Dawley rats were anesthetized with sodium thiobutabarbitol (100 mg/kg) and were instrumented to continuously record blood pressure, heart rate and renal sympathetic nerve activity (RSNA). Following occlusion of the middle cerebral artery, there was a significant increase in RSNA (from 3.8 ± 0.4 to 8.3 ± 0.6 μV/s; $P < 0.05$) which was significantly attenuated by the prior bilateral injection of estrogen (0.5 μM in 200 nl) into the PBN. Pre-injection of lidocaine (5% in 200 nl) directly into the RVLM resulted in only a slight reduction in the magnitude of the MCAO-induced sympathoexcitation ($P > 0.05$). Extracellular electrophysiological recordings from RVLM neurons demonstrated that MCAO did not produce any significant change in neuronal activity over the experimental time course ($P > 0.05$). Also, bilateral injection of estrogen into the PBN prior to MCAO or sham conditions did not result in any significant change in RVLM neuronal activity. These results indicate that estrogen receptors in the PBN play a major role in modulating the sympathoexcitatory response from ischemic forebrain nuclei, and that the pathway from the PBN to sympathetic preganglionic nuclei may not involve a synapse in the RVLM.

Theme: Endocrine and autonomic regulation
Topic: Cardiovascular regulation

Keywords: Autonomic tone; Rostral ventrolateral medulla; Stroke; Electrophysiology

1. Introduction

The majority of stroke patients in which significant autonomic and cardiovascular changes occur are usually the result of an occlusion of the middle cerebral artery (MCAO). In the clinical scenario, the autonomic dysfunction observed within 1–2 h of stroke is in the form of sympathoexcitation [13,14] and this has been replicated in an animal model following permanent MCAO [7,33]. In both humans and rats, occlusion of the right MCAO produces an ischemic lesion which includes the insular cortex [13,14], a forebrain nucleus involved in autonomic and cardiovascular regulation [16]. Thus, the therapeutic potential of drugs to recover or prevent autonomic dysfunction following MCAO is of considerable interest. Estrogen is well established as a cardioprotective and neuroprotective hormone [1,4,8,34], particularly as it relates to the central modulation of autonomic nervous system function [17,18,22,23]. In general, experiments in both male and female rats have demonstrated that estrogen administered either systemically or directly into various central

* Corresponding author. Department of Biomedical Sciences, Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, P.E.I., Canada C1A 4P3. Fax: +1 902 566 0832.
E-mail address: tsaleh@upei.ca (T.M. Saleh).

0006-8993/$ - see front matter © 2005 Elsevier B.V. All rights reserved.
doi:10.1016/j.brainres.2005.10.062

autonomic nuclei, resulted in significantly decreased blood pressure and basal sympathetic tone and/or enhanced parasympathetic tone within 30 min of injection ([3,9,17–23]). In addition to this ability of estrogen to decrease basal sympathetic tone, our lab showed that estrogen was also able to reduce the approximately 3 fold increase in renal sympathetic nerve activity (RSNA) observed following MCAO in male rats, and that this estrogen-mediated effect was via an action within the CNS [24,25].

Of the central autonomic nuclei involved in cardiovascular regulation, the parabrachial nucleus (PBN) in particular has been demonstrated to be an important synapse in the modulation of neurotransmission between the insular cortex and subcortical autonomic nuclei [10]. The PBN has a direct, reciprocal connection with the insula [30,32] and has been shown to be involved in relaying visceral afferent information to this region [2] as well as modulate changes in autonomic tone evoked by insular stimulation and/or lesion [5,7,14].

Therefore, to determine the role of estrogen receptors in the PBN in modulating the sympathoexcitatory response to MCAO, direct injection of the potent and selective estrogen receptor antagonist, ICI 182,780 into the PBN was used, resulting in a significant attenuation in the ability of systemically administered estrogen to attenuate the MCAO-induced sympathoexcitation [25]. Further, extracellular levels of estrogen within the PBN of male rats have been shown to increase immediately following MCAO [28]. Also, estrogen has been shown to almost completely block the increase in extracellular activity of PBN neurons observed following MCAO [28]. Although these investigation indirectly suggest that estrogen plays an important role in modulating descending sympathetic control, no study to date has determined the effect of estrogen injection directly into the PBN in modulating the sympathoexcitatory response to MCAO.

The PBN in turn projects to various medullary autonomic regulatory nuclei, including the rostral ventrolateral medulla (RVLM) [35], as well as directly to sympathetic preganglionic neurons in the intermediolateral cell column of the spinal cord [6,31]. Changes in RSNA as well as extracellular neuronal activity within the RVLM have been shown to be reciprocally related to alterations in baroreceptor activation indicating the importance of the RVLM in mediating acute changes in both blood pressure and sympathetic tone [11]. This, as well as numerous other neuroanatomical and neurophysiological evidence has given the RVLM the reputation of being a “presympathetic” nucleus [11], it is unknown if the RVLM is involved in mediating the sympathoexcitatory response of the ischemic insular cortex induced following MCAO.

Therefore, the current investigation was undertaken to determine the role of estrogen in the PBN in modulating the MCAO-induced increase in RSNA as well as the role of the RVLM in mediating the sympathoexcitatory response following MCAO. These results are necessary to increase our understanding of the role of estrogen in CNS in preventing the cardiovascular consequences of MCAO as well as the neuroanatomical pathway involved in relaying the autonomic dysfunction observed following MCAO.

2. Results

Prior to any central drug administration, baseline mean arterial pressure (MAP) and heart rate (HR) were measured (109 ± 11 mm Hg and 367 ± 33 beats/min, respectively) and no significant differences were found between any group of animals ($P > 0.05$; $n = 36$) for either parameter. MAP and HR were measured at 5 min post-drug injection (immediately prior to MCAO ($n = 28$) or sham ($n = 7$)). The MAP and HR values were not statistically different from pre-drug values ($P > 0.05$). In addition, over the time course of the experiment, no significant changes in either parameter were observed ($P > 0.05$; data not shown) in sham or MCAO groups except the group in which lidocaine was injected into the RVLM. Lidocaine in the RVLM resulted in a decrease in MAP and HR of 34 ± 11 mm Hg and 47 ± 23 beats/min, respectively. The average duration for this decrease in MAP and HR was 44 ± 11 min. Also in these animals, when baroreceptors were unloaded using sodium nitroprusside, the decrease in MAP (22 mm Hg) and reflex tachycardia (9 beats/min) were sustained for an average duration of 47 min. For all animals, rectal temperatures were 36 ± 1.0 °C throughout the time course of the study.

2.1. Effect of estrogen injection in the PBN on RSNA

At all time points measured following MCAO and the prior bilateral injection of saline into the PBN, renal sympathetic nerve activity (RSNA) was significantly elevated from a baseline value of 3.8 ± 0.5 μV/s to a peak value of 8.1 ± 0.7 μV/s at 90 min post-MCAO (Figs. 1A and C; $P < 0.05$). Prior to the termination of the experiment 4 h post-MCAO, RSNA remained significantly elevated (6.5 ± 0.6 μV/s; $P < 0.05$; Fig. 1C).

In contrast, following MCAO and the prior bilateral injection of estrogen into the PBN resulted in no significant change in RSNA at any time point measured post-MCAO ($P > 0.05$; Figs. 1B and C). The specificity of this estrogen-mediated effect was supported by the co-injection of estrogen with the estrogen receptor antagonist ICI 182,780 into the PBN. In these animals, the RSNA was significantly elevated from the baseline value of 3.4 ± 0.5 μV/s to a peak value of 8.3 ± 0.7 μV/s at 90 min post-MCAO ($P < 0.05$; Fig. 1C). As observed in saline pretreated animals, the RSNA remained significantly elevated at the termination of the experiment (240 min post-MCAO (6.9 ± 06 μV/s; $P < 0.05$; Fig. 1C).

2.2. Effect of lidocaine injection in the RVLM on RSNA

At all time points measured following sham surgery with prior bilateral saline injections into the RVLM, RSNA was