European Journal of Pharmacology 748 (2015) 68–75

Contents lists available at ScienceDirect

European Journal of Pharmacology

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

Cardiovascular pharmacology

Activation of serotonin 5-HT$_7$ receptor induces coronary flow increase in isolated rat heart

Ching-Chia Chang Chien [a], Ling-Wei Hsin [b], Ming-Jai Su [a,*]

[a] Institute of Pharmacology, College of Medicine, National Taiwan University, 11F No.1 s.1, Ren-ai Rd., Taipei 10051 Taiwan
[b] School of Pharmacy, Molecular Probes Development Core, Molecular Imaging Center, and Center for Innovative Therapeutics Discovery, National Taiwan University, 17, Xuzhou Road, Room 936, Taipei 10055, Taiwan

ARTICLE INFO

Article history:
Received 22 January 2014
Received in revised form
14 August 2014
Accepted 21 August 2014
Available online 6 September 2014

Keywords:
Serotonin
5-HT$_7$
5-HT$_{2A}$
Coronary flow
Nitric oxide

ABSTRACT

Serotonin (5-Hydroxytryptamine, 5-HT) can elicit both vasoconstrictive and relaxant responses on rat coronary artery. The constrictive response has been well discussed, but the mechanism of relaxant response is less studied. In the present study, we found serotonin (0.3 and 1 μM) increased coronary flow on isolated rat hearts, and treatment of nitric oxide synthase inhibitor NG-nitro-L-arginine methyl ester (L-NAME) 300 μM reduced but not totally blocked this coronary flow increasing effect. In L-NAME 10 μM treated heart, treatment of selective serotonin 5-HT$_7$ receptor antagonist SB269970 0.1 μM blocked serotonin induced coronary flow increasing response, and in the presence of 1 μM SB269970, serotonin turned into reducing coronary flow. Treatment of TCW295 (8-(2,4-Dimethoxyphenyl)-6-methoxy-2-phenethyl-1,2,3,4-tetrahydroisoquinolin-7-ol hydrochloride), a novel serotonin 5-HT$_{2A/7}$ receptor antagonist, inhibited both serotonin induced coronary flow increasing and decreasing effects. In conclusion, we found serotonin increases coronary flow of isolated rat heart by activating serotonin 5-HT$_7$ receptor activation, and this effect can be, at least partially, resistant to L-NAME.

© 2014 Elsevier B.V. All rights reserved.

1. Introduction

Serotonin (5-Hydroxytryptamine, 5-HT) has at least 14 subtypes of receptors, which are classified into 7 subgroups. Serotonin receptors are G protein coupled receptors except for serotonin 5-HT$_3$ receptor, which is a Na^+/K^+ channel (Cote et al., 2004; Pytliak et al., 2011). Serotonin 5-HT$_7$ receptor, which is coupled to Gs protein, is the latest identified serotonin receptor (Eglen et al., 1997). Activation of serotonin 5-HT$_7$ receptor causes relaxation on several vascular tissues (Cushing et al., 1996; Leung et al., 1996; Terron, 1996), and peripheral injection of serotonin induces prolonged hypotensive response via serotonin 5-HT$_7$ receptor activation (Centurion et al., 2004; Terron, 1997).

Serotonin has opposite effects, both constrictive and dilating effects, on human coronary artery. In patients with normal coronary arteries, intracoronary administration of serotonin causes vasodilation dose-dependently at lower doses, and at higher doses serotonin causes coronary artery constriction (Golino et al., 1991; McFadden et al., 1991). The vasoconstrictive effect is mainly mediated by serotonin 5-HT$_{2A}$ receptor, which is a Gq protein coupled receptor; to lesser extent, activation of serotonin 5-HT$_{1B}$ receptor, which is Gi coupled, also causes coronary artery constriction (Borton et al., 1990; Nilsson et al., 1999). Similar vasoconstrictive effects of serotonin on coronary artery have also been reported in bovine (Foy et al., 1992), porcine (Cushing and Cohen, 1992; Takenaka, 1959), canine (Brazenor and Angus, 1982; Cushing and Cohen, 1992; Lynch et al., 2009), monkey (Toda and Okamura, 1990), rabbit (Ellwood and Curtis, 1997), and rat (Lai et al., 1991).

The coronary dilating effect is endothelium-dependent in human (Golino et al., 1991). Serotonin 5-HT$_{1B}$ and 5-HT$_{2B}$ receptors are expressed on cultured human coronary artery endothelial cells and mediate serotonin induced nitric oxide (NO) release (Ishida et al., 1998; Ullmer et al., 1995). Endothelium-dependent and NO-dependent relaxing effects of serotonin on coronary arteries have also been reported in porcine (Cocks and Angus, 1983; Schoeffter and Hoyer, 1990), dog (Fujita et al., 2004), and guinea pig (Ellwood and Curtis, 1996). Besides, activation of serotonin 5-HT$_7$ receptor can relax dog coronary artery in the absence of endothelium (Cushing et al., 1996; Toda and Okamura, 1990).

In rat, serotonin induces NO release from endothelium and increases coronary flow (Bouchard et al., 2000; Joyeux et al., 2000); however, in our unpublished observation, nitric oxide synthase inhibitor Nω-Nitro-L-arginine methyl ester hydrochloride (L-NAME) cannot fully block serotonin induced

Abbreviations: 5-HT, 5-hydroxytryptamine; NO, nitric oxide; NOS, nitric oxide synthase; L-NAME, Nω-Nitro-L-arginine methyl ester hydrochloride

* Corresponding author. Tel.: +886 2 23123456x88317.
E-mail addresses: d95443008@ntu.edu.tw (C.-C. Chang Chien), lwhsin@ntu.edu.tw (L.-W. Hsin), mingja@ntu.edu.tw (M.-J. Su).

http://dx.doi.org/10.1016/j.ejphar.2014.08.027
0014-2999/© 2014 Elsevier B.V. All rights reserved.

coronary flow increase in isolated rat heart. Besides, to the best of our knowledge, role of serotonin 5-HT_7 receptor in serotonin induced coronary responses has not been examined in rat.

In the present study, we tested effect of L-NAME on serotonin induced coronary responses on isolated rat heart, and we also examined effect of 5-HT_7 antagonist SB269970 and TCW295 (8-(2,4-Dimethoxyphenyl)-6-methoxy-2-phenethyl-1,2,3,4-tetrahydroisoquinolin-7-ol hydrochloride), a novel 5-$HT_{2A/7}$ antagonist (US Patent: US 8552028 B2), on serotonin induced coronary flow responses.

2. Materials and methods

2.1. Animals

Adult male Sprague Dawley rats aged 2–3 months (275–350 g) were obtained from BioLasco Co. (Yilan, Taiwan) and kept in Laboratory Animal Center of National Taiwan University (Taipei, Taiwan). Rats were given *ad libitum* access to water and food. All animal procedures were performed according to the *Guide for the Care and Use of Laboratory Animals* of the National Institutes of Health, as well as the guidelines of the Animal Welfare Act. The animal studies were approved with a certificate number 20110073 by the Institutional Animal Care and Use Committee of the College of Medicine, National Taiwan University (Taipei, Taiwan).

2.2. Chemicals and solutions

Serotonin (5-Hydroxytryptamine, 5-HT) and Nω-Nitro-L-arginine methyl ester hydrochloride (L-NAME) were purchased from Sigma-Aldrich (Missouri, USA). SB269970 was purchased from Tocris Bioscience (Bristol, United Kingdom). TCW295 was provided by Prof. Xin's lab (School of Pharmacy, Molecular Probes Development Core, Molecular Imaging Center, and Center for Innovative Therapeutics Discovery, National Taiwan University). Chemical structure of TCW295 is demonstrated in Fig. 1, and binding affinities of TCW295 (US Patent: US 8552028 B2) to subtypes of serotonin receptor are listed in Table 1.

Perfusion solution used in this report was modified Tyrode's solution, of which composition was (in mM): 119.7 NaCl, 23.8 $NaHCO_3$, 5.6 Glucose, 1.2 $CaCl_2$, 1.1 $MgCl_2$, 0.3 NaH_2PO_4, and 5.0 KCl. Composition of Tyrode's solution used in isolated cardiac tissue was (in mM): 137 NaCl, 12 $NaHCO_3$, 5.5 glucose, 1.8 mM $CaCl_2$, 1 $MgCl_2$, 0.2 Na_2HPO_4, and 2.7 KCl. In some experiments, L-NAME was added in perfusion solution as needed at indicated doses.

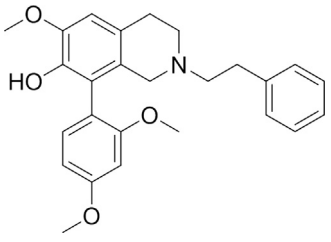

Fig. 1. Chemical structure of TCW295.

2.3. Langendorff apparatus

Coronary responses to serotonin were evaluated on Langendorff apparatus. Perfusion pressure was detected with physiological pressure transducer (ADInstruments, Castle Hill, Australia) and coronary flow was evaluated automatically by computer with software LabChart program (ADInstruments, Castle Hill, Australia), which can detect and convert speed of perfusion pump (ADInstruments, Castle Hill, Australia) into perfusion rate (ml/min); relationship between speed of pump and perfusion rate was calibrated each time before experiment. Perfusion at constant pressure was performed with the aid of STH pump controller (ADInstruments, Castle Hill, Australia), which finely adjusted speed of perfusion pump automatically in real time manner to keep perfusion pressure constant.

2.4. Effects of L-NAME on serotonin induced coronary response in isolated perfused rat heart

2.4.1. Preparation of isolated perfused rat heart

Rat heart was isolated and perfused on Langendorff apparatus as previously described (Ho et al., 2013) with little modification. Briefly, rat was weighed and injected with pentobarbital 50 mg/kg and heparin 300 IU/kg intraperitoneally. 15 min later, rat was killed by cervical dislocation and then heart was removed and rapidly mounted on Langendorff apparatus. The isolated heart was perfused under constant pressure of 70 mmHg with 37 °C modified Tyrode's solution. The buffer solution was continuously gased with 95% O_2–5% CO_2 (pH 7.4). Heart was left for stabilization for 30 min, and then subjected to different interventions as described in the following sections. Two silver recording electrodes were placed on apex to record electric cardiogram (EKG) and stimulating electrodes were placed on right atrium. Heart was constantly paced at 280 beat per minute (b.p.m.) with stimulation length of 2 ms and interval of 214 ms generated by a stimulator (Grass Instruments Co., MA, USA). Perfusion pressure, prefusion rate, and EKG were monitored and recorded digitally on computer with LabChart program (ADInstruments, Castle Hill, Australia).

2.4.2. Evaluation of influence of L-NAME on serotonin induced coronary response

Experiment protocol used is shown in Fig. 2A. After 30 min stabilization, serotonin 0.3 and 1.0 μM were added into perfusion solution consecutively to test coronary flow response to serotonin. The heart was then perfused with L-NAME 300 μM for 30 min to block activity of nitric oxide synthase (NOS), and then coronary responses to serotonin 0.3 and 1.0 μM were evaluated again in the presence of L-NAME 300 μM. For comparison, coronary response to adenosine was evaluated in the absence and presence of L-NAME 300 μM as mentioned above. Adenosine induced coronary artery dilation NO-dependently at low concentration (Hinschen et al., 2001).

2.5. Repeatability of serotonin induced coronary response in the absence or presence of low doses of L-NAME

Heart was prepared as described above (Section 2.4.1), and experimental protocol is shown in Fig. 2B. Hearts were subscribed

Table 1
Ki values of TCW295 to subtypes of serotonin receptor (in nM). "N.S." no significant binding.

	5-HT_{1A}	5-HT_{1B}	5-HT_{1D}	5-HT_{1E}	5-HT_{2A}	5-HT_{2B}	5-HT_{2C}	5-HT_3	5-HT_4	5-HT_5	5-HT_6	5-HT_7
TCW295	3590	$>10^4$	–	–	16	725	453	N.S.	N.S.	2970	1430	7.82

Download English Version:

https://daneshyari.com/en/article/2531532

Download Persian Version:

https://daneshyari.com/article/2531532

Daneshyari.com