Oxytocin's role on the cardiorespiratory activity of endotoxemic rats

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Abstract

Background: Recent findings concerning oxytocin indicate its anti-inflammatory, cardioprotective and parasympathetic modulating properties. In this study, we investigated the effects of systemically applied oxytocin on the cardiorespiratory activity in a rodent model of moderate endotoxemia.

Methods: Telemetrically recorded electrocardiogram (ECGs) from animals which received lipopolysac-charide (LPS); oxytocin (Ox); lipopolysaccharide + oxytocin (LPS + Ox), or vehicle (V) were analyzed using the ECG-derived respiration (EDR) technique to estimate the respiratory rate. The mean R-R interval and the spectral parameters of heart rate variability (HRV), such as the natural logarithm of the high frequency (lnHF) and low frequency (lnLF) components were also estimated up to 24 h after treatment.

Results: The endotoxemic animals (LPS) showed an elevated respiratory rate as well as a reduced mean R-R interval, lnHF and lnLF components compared to controls (V) from +5 to +12 h after the treatment. The administration of oxytocin significantly attenuated the hyperventilation produced by the LPS-induced endotoxemia (LPS + Ox) and restored the values of the mean R-R interval and such spectral parameters at different time points.

Conclusions: Our results support the existence of a link among the respiratory, cardiovascular, and immune systems in which oxytocin seems to act as a potential cardioprotective peptide by favoring cardiac cholinergic autonomic coupling. As a result, oxytocin diminished animal's endotoxemic tachyp-nea and restored the cardiorespiratory interactions, which was indicated by the spectral components of HRV.

Keywords: ECG-derived respiration; Anti-inflammatory cholinergic pathway HRV; Oxytocin; LPS

1. Introduction

The respiratory and cardiovascular systems are intimately linked, primarily via the autonomic nervous system (ANS) (Thayer et al., 2011). In fact, some authors have suggested the existence of a link among the respiratory, cardiovascular, and immune systems where the nucleus of the solitary tract (NTS) at the brain stem is the main site of neural integration (Thayer et al., 2011). Previous reports have shown that systemic inflammation may alter cardiac pacemaker dynamics. In fact, endotoxemia has been associated with the partial uncoupling of cardiac pacemaker cells from autonomic neural control, resulting in decreased HRV (Gholami et al., 2012). On the other hand, studies have indicated that some peptides like oxytocin possess cardioprotective prop-erties during ischemia-reperfusion injury (Faghihi et al., 2012). Oxytocin has been used to reduce myocardial infarction extension and ventricular arrhythmias. Also, it improves mean arterial pres-sure via nitric oxide production, protein kinase C activation, and reactive oxygen species balance (Faghihi et al., 2012). Actually, our previous study supports the findings of systemic cardioprotective and anti-inflammatory properties of oxytocin because a low dose of peripheral oxytocin reduced the signs of sickness behavior, such as lethargy and fever, and modified the heart rate variability (HRV) during moderated endotoxemia (Reyes-Lagos et al., 2016).

Within this framework, the aim of the present study was to analyze the effects of exogenous oxytocin after an immune chal-

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lenge induced by the administration of lipopolysaccharide (LPS) on the cardiorespiratory activity by using the ECG-derived res-piration (EDR) technique (Silva and Moody, 2014) in conjunction with the spectral analysis of HRV. Accordingly, the HRV reflects the associated physiological processes of the ANS linked to the respi-ratory and cardiovascular systems (Thayer et al., 2011), whilst the EDR allows to estimate the respiratory signal. We hypothesize that oxytocin plays a modulating role in the cardiorespiratory control during endotoxemia, mainly by favoring cardiac cholinergic auto-nomic coupling, resulting in a reduction of the respiratory rate and the restoration of spectral HRV parameters.

2. Methods

2.1. Electrocardiogram experimental data

Electrocardiogram (ECG) data from our previous study (Reyes-Lagos et al., 2016) were extracted from our database; these recordings were obtained from adult male Dark Agouti rats (DA/HanRj, 230–250 g) implanted with a telemetry transmitter (sampling frequency 2000 Hz) as described previously (Reyes-Lagos et al., 2016). These animals were maintained on a reversed 12:12 h light/dark cycle (lights off at 7:00 AM) and had ad libitum access to water and standard diet. The data were splitted into four different treatment groups: 1) vehicle (V; saline solution, n = 7); 2) oxytocin (Ox; 3 IU/kg administered subcutaneously, n = 8, 3) lipopolysaccharide (LPS; 0.1 mg/kg administered intraperitoneally, n = 8); 4) LPS + oxytocin (LPS + Ox; combined administration of 0.1 mg/kg LPS intraperitoneally and 3 IU/kg oxytocin subcuta-neously, n = 8). Baseline recordings started at 7:00 AM (time = –3 h) and drug administration was performed at 10:00 AM (time = 0 h) in each group. All experimental procedures were followed in accor-dance with the animal facilities of the University of Duisburg-Essen, the Animal Welfare Act (TierSchG) − Germany, the European Direc-tive 2010/63/EU, and with the National Institutes of Health − USA Animal Care guidelines. All procedures were approved by the Insti-tutional Animal Care and Use Committee (LANUV Düsseldorf, North Rhine-Westphalia, Germany).

2.2. Data analysis

Reliable and representative ECG segments of five minutes length were selected every hour (–3 h to +24 h), the ECG segments were analyzed for ECG-QRS detection and generation of R–R intervals using validated algorithms developed for MATLAB® (The Math-Works Inc., Massachusetts, U.S.A). The R–R intervals were analyzed using the Kubios software for HRV spectral analysis (University of Kuopio, Kuopio, Finland). We assessed the absolute power of the low frequency (LF) component, which is associated with the sympathetic and parasympathetic influences or even related to the baroreflex function, as well as the absolute power of the high fre-quency (HF) component, reflecting mainly the respiratory sinus arrhythmia (RSA), which is thereby considered as an indirect index of cardiac vagal control. The Kubios settings for the calculation of the spectral parameters were adjusted as follows: component LF (0.04–1.0 Hz) and HF (1.0–3.0 Hz), window of 256 points, 50% over-lap, interpolation rate of 8 Hz (Kuwahara et al., 1994). Additionally, we applied a signal processing technique for deriving respiratory signals from ECG's (EDR) based on the QRS area under curve. This technique is freely available in PhysioNet (www.physionet.org) and provides reliable respiratory-related data (Silva and Moody, 2014). The moving window was adjusted at 0.1 s. Respiratory derived signals were interpolated by a cubic spline fit at 100 Hz and dig-itally high-pass filtered using a cutoff frequency of 0.5 Hz. The EDR spectra peak (1.0–3.0 Hz) was calculated through Welch's power In Fig. 1 we show the estimated breathing rate by EDR, the mean R–R interval and the estimated spectral parameters from HRV (lnHF and lnLF) starting from −3 h to +24 h after treatment. A black/white bar at the bottom of each panel indicates, respec-tively, the dark/light cycle. Single administration of oxytocin by itself did not modify the respiratory rate dynamics in the long-term (V vs. Ox), whereas in the presence of endotoxemia oxytocin did induce significant changes (LPS vs. LPS + Ox; Fig. 1a at +6 h and +12 h, $F(16,459) = 6.487$; $p < 0.0001$). An increment of estimated respiratory rate, matched with a reduction of R–R interval at +5 h after treatment was also identified (Fig. 1a & b). These physiological changes continued until +12 h.

The spectral parameter lnHF was diminished from +8 to+12 h, the lowest value was detected at + 9 h (LPS vs. V; Fig. 1c, $F(16,459) = 2.071$; $p = 0.0083$). Similarly, a decrement of lnLF was found from +1 h to 12 h, with the lowest value identified at + 9 h (LPS vs. V; Fig. 1d, $F(16,459) = 4.450$; $p < 0.0001$). The administra-tion of oxytocin restored the lnHF and lnLF parameters +9 h after the treatment (LPS vs LPS + Ox; Fig. 1c & d, $F(16,459) = 2.071$; $p = 0.0083$ & $F(16,459) = 4.450$; $p < 0.0001$, respectively). Fisher's LSD post-hoc test showed significant differences among planned comparisons ($p < 0.05$).

Fig. 2 shows representative data of the estimated respiratory signals extracted by EDR technique. This figure illustrates a higher respiratory rate pattern during endotoxemia (LPS) compared to the data from the LPS + Ox group +6 h after the treatment.

4. Discussion

The reduction in the spectral parameter lnHF of HRV indicates a decreased respiratory sinus arrhythmia (RSA) and suggests partial uncoupling of cardiac pacemaker during LPS-induced endotox-emia at the long-term (LPS vs. V, Fig. 1c). In fact, a reduced RSA may decrease the gas exchange efficiency of the lung by failing to optimally synchronize the pulmonary perfusion to the alveo-lar ventilation through the ventilatory cycle (Yasuma and Hayano, 2004). Additionally, a reduced mean R–R interval (LPS vs. V, Fig. 1b) is related to either a vagal withdrawal or increased sympathetic activity. Noteworthy, the lowest value of the mean R–R interval (LPS vs. V; Fig. 1b at +9 h) is not in coincidence with the maximum value of respiratory rate (LPS vs. V; Fig. 1a at +6 h). Thus, these findings indicate that, despite a reduced RSA during LPS-induced endotoxemia (indicated by lower lnHF values), the estimated res-piratory rate presents a significant augmentation in conjunction with reduced mean R–R interval in response to a LPS challenge. In fact, the RSA may become reduced by an increased respiration rate (representative data in Fig. 2a) independently of the vagal nerve traffic (Eckberg and Eckberg, 1982). Yet the administration of oxytocin (LPS vs LPS + Ox) seems to revert at baseline levels the estimated respiratory rate (Fig. 1a), the mean R–R interval (Fig. 1b) and the spectral HRV parameters lnHF and lnLF (Fig. 1c & d). The decreased respiratory rate was found even +6 h and +12 h after oxytocin administration (LPS vs. LPS + Ox; Fig. 2b), although the increment in the HF power was only observed at +9 h of oxytocin administration (Fig. 1c). Thus, we speculate that, among different physiological mechanisms attributed to the observed changes in the spectral components lnHF and lnLF (Fig. 1c & d) and the esti-mated breathing rate in rats (Fig. 1a), the presence of oxytocin receptors in the heart (Faghihi et al., 2012) may have a direct effect on the cardiac pacemaker coupling. Additionally, oxytocin recep-tors in the lungs (Pequeux et al., 2005) as well as the activation of the cholinergic anti-inflammatory pathway should be both taken into consideration (Huston and Tracey, 2011)

5. Conclusion

In the present study, we found that spectral HRV parameters (lnHF and lnLF) are altered during moderate endotoxemia in rats. A depression in the HF component may indicate a reduced respiratory sinus arrhythmia and the changes in both HF and LF may indi-cate an uncoupling of the autonomic control during LPS-induced endotoxemia.

Yet our results also support the existence of a functional link among the respiratory, cardiovascular, and immune systems in which oxytocin seems to act as a potential cardioprotective peptide by favoring cardiac cholinergic autonomic coupling. Consequently, oxytocin diminished animal's tachypnea and restored the car-diorespiratory interactions indicated by the spectral components of HRV despite the administration of endotoxin. Finally, these findings confirm the suitability of the electrocardiogram derived respiration (EDR) technique to obtain the estimated respiration signal in rodent models.

Conflict of interest

The authors declare no conflict of interests.

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