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No effect of social exclusion on salivary cortisol secretion in women in a randomized controlled study

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Summary
Background: Lack of social support and social exclusion are associated with adverse effects for mental and physical health. Additionally, women appear to be more vulnerable to social triggers of health disturbances. Activity of the hypothalamus—pituitary—adrenal-axis (HPA-axis) might play a key role in this context as it has been shown both to be associated with psychosocial conditions and health outcomes and to respond differentially depending on gender. To test this hypothesis we thus investigated the effects of social exclusion on cortisol release in women. To experimentally vary social exclusion we employed a highly standardized paradigm (Cyberball) which already has been shown to affect subjective well-being and some physiological parameters.
Methods: Healthy women ($n = 89$) were randomly assigned to one of the three Cyberball conditions: social exclusion, inclusion and technical default, respectively. Salivary cortisol and subjective mood were assessed as primary outcome variables immediately prior to and every 15 min after Cyberball.
Results: Social exclusion had no effect on cortisol secretion though significant effects on mood were observed.
Conclusions: These results indicate that the Cyberball social exclusion paradigm alone though affecting psychological well-being does not affect HPA-axis functioning.

1. Introduction

Women are considered to be especially vulnerable to social triggers of health disturbances (Denton et al., 2004). One social trigger often analysed is the lack of social support and social exclusion (Hawthorne, 2008). Mechanisms mediating between the degree of social integration and physical and mental health are only partially understood. A potential mediator could be a dysregulation of HPA-axis functioning which is both known to be related to health disturbances and to social stress. Indeed, correlative research indicates that lack of social support is associated with an increased basal cortisol secretion (Cacioppo et al., 2000; Gunnar et al., 2003; Evolahti et al., 2006). Correlative studies are, however, ambiguous with respect to causal inferences. Thus, experimental research is needed to examine the effects of social exclusion on cortisol secretion.

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Experimental research on social exclusion, however, faces the problem of how to induce a standardized social exclusion condition. "Cyberball," a virtual ball game, represents one possibility of inducing social exclusion in a highly standardized manner (Williams et al., 2000). In this paradigm, participants are made to believe that they are playing with three other participants (which in fact are computer generated). During the game the degree of social inclusion (i.e., how often they receive the ball from the other participants) is manipulated: "included" participants receive the ball regularly throughout the game while "excluded" participants receive no further ball after the first throws. Previous research on this paradigm indicates that excluded participants not only perceive themselves as being excluded but also suffer from lower self-esteem compared to the included participants (Williams et al., 2000). Furthermore, fMRI studies have shown that the dorsal anterior cingulate cortex, a region also activated while experiencing physical pain, is activated during exclusion (Eisenberger et al., 2003). Effects of Cyberball on cortisol secretion have not been analysed, yet.

We therefore used the Cyberball paradigm to assess the effects of social exclusion on HPA functioning and mood in a randomized controlled study. We expected both an increase of cortisol secretion after social exclusion and an increase of bad mood.

2. Materials and methods

2.1. Participants and ethics

Participants were $n = 89$ healthy female students between 18 and 35 years, recruited by advertisement on the university campus and paid for participation. Exclusion criteria were acute or chronic infections, acute allergy, and diseases of the adrenal gland, regular use of any medication, gravidity, and actual or past mental illness. Detailed flows of participants and final samples according to the CONSORT criteria are displayed as supplemental information. All participants provided informed, written consent. The study was approved by the Local Ethics Committee and was found to conform to the guidelines of the World Health Organization (Declaration of Helsinki).

2.2. Experimental conditions

2.2.1. Independent variable: experimental variation of social exclusion

Experimental variation of social exclusion was done via the Cyberball paradigm (Williams et al., 2000). The participant is made to believe that he or she is connected to three other players (actually computer generated), one same sex, two opposite sex, whose photographs and names are displayed on the computer screen. Players are asked to throw a ball per mouse-click to each of the others. Every thrower is free to decide who receives the ball next. The ball is thrown 60 times. Three conditions were run: Social exclusion (after having received the ball three times, the participant does not receive it any more), technical default (control condition 1: game identical to social exclusion, but at the end of the game a pop-up window indicates that network problems precluded other players from throwing the ball to the participant), inclusion (control condition 2; the participant receives an average of every fourth ball).

To obtain the photographs for the three computer generated players, 40 students (20 of each sex) from other universities were photographed and rated by 10 male and 10 female students respectively, according to their attractiveness. For each sex two photographs with medium attractiveness ratings were selected and used for the experimental sessions.

Randomization and blinding: Subjects were randomly assigned to the experimental conditions and stratified with respect to oral contraceptive intake. An equal number of cards containing the respective condition were put in sealed opaque envelops prior to study onset. Envelopes were shuffled immediately prior to each experiment and a person not involved in data assessment and not in contact to participants drew an envelope and put the respective experimental settings at the Cyberball game. Experimenters in direct contact to the subjects were blinded until the end of an experimental session, when subjects were debriefed. To keep participants blind with respect to the hypotheses they were told the purpose of the study would be to examine effects of mental visualisation on the endocrine system (see also supplemental information).

2.2.2. Dependent variables

Cortisol response: To assess effects on cortisol saliva samples were taken every 15 min by means of Salivettes® (Sarstedt, Rommelsdorf, Germany) throughout the experiment and stored at $-20\ ^{\circ}C$ until analysis. After defreezing saliva samples were centrifuged at $1700 \times g$ for 3 min. All samples were visually untainted. A competitive luminescence immunoassay (IBL International®, Hamburg, Germany) was used to assess cortisol concentrations. The intra- and inter-assay CVs of this standard LIA Kit given by the manufacturer equal 6.08% and 7.76%, respectively. Within our laboratory CVs average below 2%. We thereby in larger studies do not run all samples in duplicate, but only some per plate. Within those samples approximately one out of hundred shows CVs above 10%. As primary outcome variable main effects of Cyberball on cortisol were analysed.

Psychological response: Subjective mood was assessed via Profile of Mood States (POMS) questionnaire (McNair et al., 1971) at baseline and after Cyberball. Anger and depression immediately after the Cyberball game were analysed as primary psychological outcome parameters.

2.2.3. Control variables

To prove whether groups were comparable with respect to potential psychological confounders the following variables were assessed by means of standardized tests (internal consistencies between $\alpha = 0.81$ and $\alpha = 0.98$) one week prior to the experiments: Perceived social support (Fydrich et al., 2007), Personality (NEO-PI-R; Costa and McCrae, 1992), Self-Esteem (Fleming and Courtney, 1984).

2.2.4. Procedure

A first appointment took place at least one week prior to the experiments. At that time, all subjects underwent an anamnestic interview in order to check for inclusion/exclusion criteria. Participants were informed about the details of the experiment and gave written consent. Additionally, they