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"The role of primary appraisal and perceived stress reactivity in healthy young women in subjective and physiological stress responses during a social evaluative stress task"

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#### **Abstract**

As stress has been deemed the "Health Epidemic of the 21st Century" by the World Health Organization (Fink, 2017) and severely impacts health negatively (DeLongis et al., 1988), it is important to determine the influence of psychological factors on the stress response. For that reason, this thesis aims to examine possible predictive effects of dispositional stress reactivity and primary appraisal on heart rate and subjectively perceived stress during a laboratory stress task, the Trier Social Stress Test (Kirschbaum et al., 1993). To check for possible predictive effects, linear regression analyses were conducted. Furthermore, a mediating effect of *primary* appraisal on the effect of stress reactivity on heart rate and subjectively perceived stress has been proposed. To check for this mediation effect, the bootstrapping method by Preacher and Hayes (2004) was applied. As a premise for this mediation model, a predictive effect of stress reactivity on primary appraisal has been proposed and checked for by using a linear regression approach. Except for the predictive effect of Reactivity to Failure on threat, none of the hypotheses could be confirmed neither when examining the entire available data set nor when utilizing an extreme case analysis approach.

# Abstract (deutsch)

Da Stress laut WHO eine der größten Gesundheitsgefahren des 21. Jahrhunderts darstellt (Fink, 2017) und die Gesundheit nachweislich negative beeinflusst (DeLongis, 1988), erscheint es sinnvoll, den Einfluss psychologischer Faktoren auf die Stressantwort zu untersuchen. Aus diesem Grund untersucht diese Arbeit die möglichen prädiktiven Effekte von dispositionaler stress reactivity und situationalem primary appraisal auf die Herzrate und subjektiv wahrgenommenen Stress während einem Stresstest im Labor, dem Trier Social Stress Test (Kirschbaum et al., 1993). Um diese Prädiktoreffekte festzustellen, wurden lineare Regressionsanalysen durchgeführt. Zudem wird überprüft, ob ein Mediationseffekt von Primary Appraisal auf den Effekt von stress reactivity auf Herzrate und subjektiv wahrgenommenen Stress besteht. Dies wurde mittels des Bootstrapping-Verfahrens nach Preacher und Hayes (2004) überprüft. Als Voraussetzung für dieses Mediationsmodel muss ein Prädiktoreffekt von stress reactivity hinsichtlich primary appraisal vorliegen. Dies wurde mittels linearer Regression überprüft. Abgesehen von dem Prädiktoreffekt von Reactivity to Failure hinsichtlich threat konnte keine der Hypothesen in der Stichprobe bestätigt werden. Auch im Extremfallvergleich konnte keine der Hypothesen bestätigt werden.

# 1.Theoretical Background

Stress is a widespread phenomenon nearly every individual experiences in the course of their day-to-day life (McEwen, 1998). In the past decades, due to globalization, social, and cultural changes, societal organization has transformed profoundly resulting in new challenges which cause significant stress, particularly in the psychosocial domain (Liu et al., 2017). Especially changes in demands in the working world pose such challenges: increasing requirements to work flexible long hours, constant availability via mail or phone, and deadline pressure (Kämpf, 2015; Hünefeld, 2019) make psychosocial stress an increasingly prevalent issue affecting a large proportion of the population.

More recently, restrictions in recreational activities and disruptions in social life due to Covid-19 regulations and lockdowns affected virtually every member of society: Social distancing, uncertainty, loss of personal space at home (if not living in a single household), isolation from family and friends, or, in the worst case, increases in domestic abuse and violence pose additional psychosocial stressors. This made the experience of psychosocial stress an even more common experience for individuals across all age groups since the beginning of the Covid-19 pandemic in Europe in Mach 2020 (Mohler-Kuo, 2020). Besides occupational and Covid-19 related stressors, other day-to-day psychosocial stressors may be acute life events, socioeconomic/financial strain, relationship stressors, and discrimination (Cuevas et al., 2019).

When confronted with a psychosocial stressor, a well-orchestrated response involving physiological and endocrine systems as well as psychological rections sets in (Andrews et al., 2013). Physiologically, the autonomous nervous system (ANS) plays a major role in the context of stress. Within milliseconds of stressor perception, the ANS responds and triggers the release of adrenaline and noradrenaline (Turner et al., 2019), which in turn initiates rapid changes in numerous autonomous bodily functions, such as an increase in *heart rate* (*HR*) (Chrousos, 2009). These rapid changes can be summarized to quickly provide brain and body with additional energy in order to prepare for "fight or flight" (Selye, 1936).

On the endocrine level, the hypothalamic-pituitary-adrenal (HPA) axis plays an important role in the stress response. The HPA axis becomes activated whenever threats and negative consequences are expected to set in, even before they actually occur (Herman et al., 2005). In such a situation, the HPA axis triggers the release of a cascade of hormones, the glucocorticoid cortisol being one of them (Andrews et al., 2013). Similar to the effects of the ANS response, cortisol release (and the release of other glucocorticoids) aims to increase available energy in order to cope with the stressful situation successfully.

Individuals also respond to stressful stimuli on the psychological level. The perception of a novel situation as stressful and threatening is the earliest critical component for ANS and HPA axis response activation (Andrews et al., 2013). If the situation is appraised as stressful, the individual also evaluates whether their coping resources are sufficient to deal with the situation successfully. These two appraisal processes can be referred to as the cognitive level of stress processing (Lazarus & Folkman, 1984; Lazarus, 1991). The physiological aspects of the stress response, triggered by ANS and HPA axis activation, are likely to be labeled with feelings of being "stressed" (Campbell & Ehlert, 2011). This process may be referred to as the subjective emotional stress response.

This well-tuned interplay of stress responsive systems has an adaptive function, but as psychosocial stress exposure increases in day-to-day life, long-term dysfunctions in these systems may establish - in turn leading to the occurrence of health problems (DeLongis et al., 1988).

In this regard, psychosocial stress has been shown to be linked to a plethora of negative mental and somatic health outcomes in a steadily growing number of studies (e.g., Cohen et al., 2007; Cohen et al., 2016; Kivimäki et al., 2006; Hammen, 2005; Mazure, 1998). Acute psychosocial stress may trigger allergic reactions (e.g., eczema, urticaria, asthma attacks), migraines, hypotensive or hypertensive attacks, gastrointestinal symptoms, different types of pain, as well as psychopathological events, such as psychotic episodes or panic attacks (Chrousos, 2009). Similarly, chronic psychosocial stress may also lead to negative physiological, neuropsychiatric, and behavioral outcomes, such as depression, anxiety, cognitive and/or executive dysfunctions, cardiovascular pathologies (e.g., hypertension),

metabolic disorders, neurovascular degenerative disease, osteoporosis, and sleep disorders (Chrousos, 2009).

Following conservative estimates, psychosocial stress accounts for up to 30% of the overall costs related to illnesses and accidents, summing up to 0.5 - 3.5% of the gross domestic product in Western nations (Nater et al., 2006). Because of the severe negative health impact of stress and its high monetary and societal costs, stress is considered to be the "Health Epidemic of the 21st Century" by the World Health Organization (Fink, 2017) and numerous research teams work on furthering our understanding of the stress process. Despite all efforts and a growing number of studies, the relationship between psychological factors and the individual stress response is still not understood completely (Kudielka et al., 2009). Determining the effects of psychological precursors on the stress reaction is thus needed to further our understanding of the processes linking stress to disease.

For that reason, two possible psychological determinants of the individual stress response, namely dispostional *stress reactivity* (*SR;* Schulz et al., 2005) and situational *primary appraisal* (*PA;* Lazarus & Folkman, 1984), and their shaping effects on psychological and physiological aspects of the stress response in a laboratory setting are examined and analyzed in the current project.

The majority of studies similar to the current project examined the influence of *SR* and *PA* on the HPA axis stress response, most commonly measured by assessing salivary cortisol concentration (Schulz et al., 2005; Schlotz et al., 2011a, 2011b). Research on the cumulative effects of both *SR* and *PA* on other aspects of the stress response is, to the author's knowledge of current literature, scarce. For that reason, this study aims to assess the influence of *SR* and *PA* on an aspect of the ANS stress response, namely *heart rate* (*HR*), and an aspect of the psychological stress response, measured by assessing *subjectively perceived stress* (*SubjStress*). In order to induce psychosocial stress in a laboratory setting, participants undergo a *Trier Social Stress Test* (*TSST*; Kirschbaum et al., 1993). The *TSST*, as well as *SR*, *PA*, *HR*, and *SubjStress* will be described and discussed in greater detail in the following sections.

#### 1.1 Theories of Stress

To this day, no unified standard definition for stress and its processes has been agreed upon (Fink, 2017). Generally, theoretical explanations of stress can be categorized into following types: stress as an external stimulus, stress as a response, and stress as a person-environment interaction, or as a person-environment transaction (Brough et al., 2009). Depending on the originating field of study of a stress concept, varying aspects of the stress process – physiological or psychological – are highlighted and examined in greater detail. A few of the most influential stress conceptualizations will be described in the following section:

# 1.1.1 Homeostasis (Cannon, 1932)

One of the earliest concepts relating to stress, homeostasis (Cannon, 1932), focuses on physiological processes affected in the context of stress. Homeostasis refers to an equilibrium of bodily functions, which is necessary for survival. Any internal or external effects challenging this equilibrium can be defined as a stressor (Chrousos & Gold, 1992). Stress can thus be defined as a response to an event in which homeostasis is either perceived to be threatened or actually threatened (Chrousos, 2009). In order to re-establish homeostasis, a complex adaptive response comprising behavioral and physiological changes sets in. In Cannon's homeostasis conceptualization (1932), this adaptive response was thought to be invariable across individuals and stressors.

In this context, Hans Selye (1950) posited the general adaptation syndrome: In response to stress, the body exhibits a defense reaction through the nervous and hormonal systems. As stress exposure repeats and intensifies, the body first shows an alarm reaction, then proceeds into a stage of resistance, and from there eventually into a state of exhaustion. Especially the third stage was conceptualized as a maladaptive process and thought to explain the negative effects of stress on health (Linnemann, 2016).

# 1.1.2. Allostatic Load (McEwen, 1998)

Similar to the concept of homeostasis, the allostatic load model (McEwen, 1998) describes how stress can have a negative influence on health. In this model, an organism seeks to re-establish homeostasis when faced with a stressor. In order to achieve this, adaptive systems, such as the HPA axis and the ANS, are activated. This adaptation process is called allostasis. Under- or overactivation of these allostatic systems can have detrimental effects on the body – it is thus thought that adaptation (or allostasis) comes with a "price", called the allostatic load. Allostatic load results from either overworked allostatic systems or when allostatic systems fail to shut down after stressor exposure has ceased. It may also result from occasions in which allostatic systems fail to respond to the stressor adequately. As allostatic load builds up, occurrence of detrimental physiological and psychological health outcomes becomes increasingly likely (Linnemann, 2016).

In contrast to the concept of homeostasis, the allostatic load model allows for individual differences in the stress response. These individual differences depend on two factors: Firstly, the condition of the body - McEwen (1998) suggests that individuals with conditions such as obesity or diabetes may be more vulnerable to stress; also, genetic makeup is considered to play a role in stress susceptibility. Secondly, individual differences in stress responses are dependent on how a person perceives and interprets the respective stress situation.

# 1.1.3. Transactional Stress Theory (Lazarus & Folkman, 1984)

In this context, one of the most influential stress theories originating from the field of psychology, the transactional stress theory (Lazarus & Folkman, 1984), needs to be mentioned. In contrast to homeostasis and allostasis approaches focusing mainly on physiological aspects in the context of stress, the transactional stress theory emphasizes the role of cognitions and appraisals of the stress situation. Following Lazarus' and Folkman's (1984) theory, an individual constantly appraises stimuli within their environment. Through these appraisal processes, an individual ascribes meaning to events and stimuli in the world around them (Boyd et al., 2009). These appraisal processes are affected by environmental factors, such as the novelty, unpredictability, uncertainty, and ambiguity of a situation (Mason, 1975),

and personal factors, including goals and self-efficacy (Biggs et al., 2017), as well as dispositional factors, such as *stress reactivity* (*SR;* Schulz et al., 2005). The variation in personal and dispositional factors as well as the complexity and ambiguity of environmental contexts explain the vast differences in appraisals that different individuals make of the same stimulus or event (Lazarus, 1991).

Two core types of appraisal processes are distinguished: *primary appraisal* (*PA*) and secondary appraisal. Through *primary appraisal* (*PA*), an individual evaluates the significance of an event regarding their well-being: the event can either be appraised as positive (exerting a positive effect on the individuals' well-being), as irrelevant, or as stressful (the event poses the risk of harm, loss, threat, or challenge). If a situation has been deemed stressful through *PA* processes, the individual assesses whether their resources are sufficient to successfully cope with the stressor. In the transactional stress theory, this process is called secondary appraisal. If a stimulus is appraised as stressful (through *PA*) and if coping resources have been evaluated as non-sufficient or barely sufficient (through secondary appraisal), a stress reaction of variable intensity sets in, comprising HPA axis and ANS activation as well as psychological and behavioral consequences.

Following the transactional perspective (Lazarus & Folkman, 1984), stress therefore can be defined as the person-environment transaction taking place when an individual is confronted with a stimulus that is appraised as either harmful, threatening, or challenging. Furthermore, the individual must evaluate their coping resources as insufficient (or barely sufficient) to overcome the stimulus (Biggs et al., 2017).

Even though it is unclear whether cognitions and appraisals are causal for emotional, biological, and behavioral aspects of the stress reaction (Phelps, 2006), they are considered to be an important link between stressor, the individual stress response, and subsequent negative health outcomes (Gaab, 2009).

#### 1.2. Trier Social Stress Test

As psychosocial stress has been shown to be a risk factor for subsequent disease (Chrousos, 2009), psychobiological stress research requires standardized and valid protocols to induce psychosocial stress under controlled conditions (Henze et al., 2017). The gold-standard of inducing psychosocial stress in a laboratory

setting is the *Trier Social Stress Test* (*TSST*; Kirschbaum et al., 1993). In search of a laboratory stress task that could induce reliable changes in endocrine, cardiovascular, and emotional stress parameters, the *TSST* was developed. Since its development, the *TSST* has been used in different protocol variations in healthy subjects as well as in clinical populations.

The *TSST* procedure, as applied in current project, runs as follows (Kudielka et al., 2007; Allen et al., 2013; Frisch et al., 2015): The stress task is preceded by a rest period of 30 minutes in order to minimize the impact of possible prior stressful events or other short-term effects possibly impacting the investigated outcome stress measures. The subsequent stress task consists of the following parts: a brief task introduction (2 minutes), a preparation period (3 minutes), a free speech task (5 minutes), and a mental arithmetic task (5 minutes). After the rest period is over, baseline measurements are assessed. Subsequently, the participant is guided to a neutrally furnished room by the experimenter, in which the stress task takes place. In this room a large table is located with two persons dressed in white lab coats seated at the table (the "selection committee", see Fig. 1).

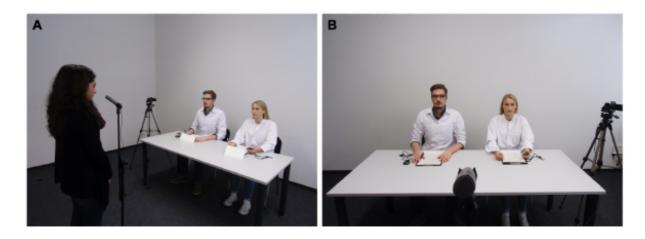


Figure 1. (A) Setup of the Trier Social Stress Test and (B) close-up of selection committee (before additional Covid-19 safety measures were applied).

The selection committee is comprised of one male and one female member that the participant is unacquainted with. The committee is equipped with writing materials and stop-watches. A video camera clearly visible to the participant is situated behind the selection committee as well as a microphone placed on the table

in front of the committee. The participant is asked to take a seat at a table equipped with paper and pens facing away from the committee.

The participant is then introduced to the task through a standardized instruction read out aloud to them by the experimenter. The participant is instructed to take over the role of an applicant who was invited to interview for their "dream-job". They are instructed that they should introduce themselves and convince the committee of their qualification in a free speech. They are further instructed to solely focus on personal attributes qualifying them for the job during the interview. Additionally, they are asked not to mention any prior work or study experiences qualifying them, as the selection committee has already received the participants curriculum vitae, university grades, and job references. After the job interview, the committee would present another task to the participant. The selection committee is introduced to be specifically trained in behavioral observation and that the participants performance is video- and audio-taped for subsequent analyses.

After ensuring that the participant understood the upcoming task, the participant is allowed to briefly prepare for the job interview by taking notes for 3 minutes, which they are not allowed to use during the speech task. After the preparation period, the participant is instructed to stand up from their preparation spot and stand in front of the committee table (in a distance of about 1.5 meters, see Fig. 1).

Then the 5-minute free speech task begins: The participant is asked to start their free speech by the male committee member. The committee is instructed to communicate with the participant in a distanced, reserved manner and to not respond with any verbal or facial feedback during the entire stress task. At the same time, the committee is trained to communicate with the participant in a way that does not elicit feelings of harassment or anger. If a subject finishes their speech in less than five minutes, the committee responds in a standardized manner: At first, they remain quiet for 20 sec in order to let an unpleasant silence occur. If the participant still remains silent after 20 sec, the committee tells the volunteer "You still have time left. Please continue!". As talking about one's own positive attributes for a period of five minutes tends to be difficult for most individuals, occurrence of unpleasant silences is very likely, inducing a sense of failure which in turn makes the free speech task an unpleasant and stressful experience.

Once the five minutes are over, the committee interrupts the free speech and introduces the 5-minute mental arithmetic task: Starting from 2043, the participant is asked to subtract the number 17 serially as fast and accurately as possible. Whenever the participant makes a mistake, they are instructed to start over at 2043. If a participant does well on the mental arithmetic task, they are interrupted by the committee by being asked to speak more loudly or to look up in the camera. This distracts the participant from the task and makes subsequent failure likely. After five minutes, the task is terminated.

The *TSST* comprises the necessary situational elements capable of eliciting a stress response (Dickerson & Kemeny, 2004): It contains elements of uncontrollability, forces situations of failure, in which participants cannot avoid negative consequences, and it poses social-evaluative threat, where task performance is negatively evaluated by others. By that, the *TSST* reliably induces physiological and psychological stress responses, such as an increase in salivary cortisol (Kirschbaum et al., 1993), an elevation in *HR* (Skoluda et al., 2015; Kirschbaum et al., 1993) and increased *SubjStress* (Hellhammer & Schubert; 2012). Furthermore, the *TSST* has been shown to induce stress in an ecologically valid manner (Henze et al., 2017): Compared to a naturalistic stressor (in this case a university oral exam), the *TSST* elicited similar cortisol and emotional stress responses.

As the *TSST* elicits a cardiovascular and emotional stress reaction reliably under standardized circumstances, it has been chosen as the stress task applied in current project.

# 1.3. Cardiovascular and Psychological Stress Response

When exposed to a *TSST*, the majority of individuals exert a stress reaction comprising endocrinological, cardiovascular, psychological, and behavioral elements. As most studies similar to current project investigated the cortisol stress reaction after *TSST* exposure, current project focuses on two aspects of the ANS and psychological stress response: *Heart rate* (*HR*) and *subjectively perceived stress* (*SubjStress*).

#### 1.3.1. Cardiovascular Stress Response: Heart Rate

A major stress reactive system significantly contributing to physiological aspects of the stress reaction is the ANS. ANS control of *HR* is essential to cardiovascular health. In periods of relative relaxation, *HR* is chronically inhibited by efferent parasympathetic nervous system (PNS) signals (Fisher & Newman, 2013). The extent of this inhibitory control is referred to as vagal tone (referencing the vagus nerve which innervates the viscera).

In periods of higher physiological demand, as well as psychosocial stress situations, the primary source of autonomic control of *HR* is exerted by increased sympathetic nervous system (SNS) activity (Hjemdahl et al., 1989; Trepel, 2004). The SNS is part of the sympatho-adreno-medullary (SAM) system, which mediates quick stress reactions through its neurotransmitters. These quick stress reactions are commonly referred to as "fight-or-flight" reactions (Rensing et al., 2006; Cannon, 1932). When the SNS is activated through stressor perception, it triggers the release of the catecholamines (noradrenaline and adrenaline) and transmits stress signals directly to relevant organ systems (Höch, 2019), resulting in an increase in in blood pressure, elevation of salivary alpha-amylase levels (Skoluda et al., 2017), as well as an increase in *HR* (Chrousos, 2009).

It has been proposed in numerous earlier studies (Light, 1981; Obrist, 1981) that high cardiovascular reactivity to acute psychosocial stressors, e.g., a large increase in *HR*, reflects a high likelihood of cardiovascular disease later in life. This is commonly referred to as the reactivity hypothesis (Turner et al., 2020). The reactivity hypothesis could be confirmed regarding numerous negative cardiovascular health outcomes, such as elevated systolic and diastolic blood pressure, risk of hypertension, early onset of hypertension, atherosclerosis, and clinical cardiac events (Chida & Steptoe, 2010; Turner et al., 2020). Also, high cardiovascular stress reactivity has been linked to anxiety and depressive disorders (Greaves-Lord et al., 2007; Kagan et al., 1987).

Although high cardiovascular stress reactivity is closely linked to subsequent cardiovascular disease and mental disorder, psychological determinants possibly influencing the cardiovascular stress response are still under-researched and thus

poorly understood. In order to close that gap in the literature, *HR* has been chosen as one of the outcome variables in current project.

# 1.3.2. Psychological Stress Response: Subjectively Perceived Stress

As mentioned above, the psychological stress response is associated with cognitive stress processing. This cognitive stress processing can be described through appraisal processes mentioned in the transactional stress theory (Lazarus & Folkman, 1984). The subjective emotional aspect of the psychological stress response may be described as the experience of *subjectively perceived stress* (*SubjStress*) and is closely tied to the cognitive aspect of stress processing (Gaab, 2009). The subjective stress experience is hard to objectify and heavily dependent on respective individual experiences as well as genetic predispositions regarding the acquisition and processing of experiences and emotions (Höch, 2019).

Generally, studies suggest that individuals feel more negatively when stressed: the general affect suffers, psychological and physiological symptoms are reported more frequently, and negative emotional tone increases (Baum et al., 1987). It has been suggested that the subjective emotional stress response can be measured most appropriately by the increase in distress that is elicited when an individual progresses from a "normal" situation into a stressful situation (Watson & Clark, 1984). For that reason, visual analogous scales (VAS) measuring *SubjStress* are applied in current project (for a more detailed description of the VAS and *SubjStress* measurement procedures, see down below).

Even though negative emotional stress reactivity has been linked to negative health outcomes (Denollet et al., 2000; Heponiemi, 2004), the link between possible psychological precursors and the subjective emotional stress response is still not understood completely. For that reason, *SubjStress* has been chosen as one of the outcome variables in current project.

#### 1.4. Psychological determinants of the stress reaction

Up until this point, numerous individual factors influencing the stress response have been determined, most often relating to the cortisol stress response: Two of the

most commonly reported influencing factors are age (e.g., Nater et al., 2013; Strahler et al., 2010) and sex (e.g., Kajantie & Phillips, 2007; Panagiotakopoulos & Neigh, 2014; Kirschbaum et al., 1999). A large proportion of reported sex differences in the stress reaction may be accounted for by menstrual cycle phase or hormonal contraceptive use in females (Kirschbaum et al., 1999; Childs et al., 2010). Also, lifestyle-related behaviors, such as caffeine, tobacco, alcohol, and cannabis use (Kudielka & Wüst, 2010; Kudielka et al., 2009;) have been shown to influence the stress response.

Concerning psychological factors influencing the stress response, studies have linked cognitive appraisal processes to the cortisol (Gaab et al, 2005) and the procoagulant stress response (Wirtz et al., 2006). Also, poor emotional regulation capacities have been shown to account for individual differences in stress responses (Campbell & Ehlert, 2012): Especially higher emotional suppression scores are linked to stronger cortisol stress reactivity (Lam et al., 2009).

In the superordinate study in which current project is embedded (for a more precise description, see down below), a plethora of psychological factors possibly influencing the stress response are assessed, e.g., chronic stress (Schulz & Schlotz, 1999), state and trait anxiety (Spielberger, 2010), Resilience (Rocalevent et al., 2015) and self-esteem (Rosenberg, 1965). Since examining the shaping effects of all psychological factors assessed in the superordinate study would greatly exceed the limits of current thesis, only two are examined in greater detail: *SR* (Schulz et al., 2005) and *PA* (Lazarus & Folkman, 1984).

#### 1.4.1. Stress reactivity (Schulz et al., 2005)

The concept of psychobiological *SR* refers to the tendency of an individual to respond to stressful stimuli with intense, immediate, and long-lasting stress reactions (Schulz et al., 2005). Psychobiological *SR* is considered to explain parts of the individual differences in the link between stress and disease (Lovallo & Gerin, 2003). As *SR* has been shown to be relatively stable over time (Burleson et al., 2003), *SR* can be conceptualized as a dispositional variable explaining interindividual stable differences in the subjective emotional stress experience and physiological responses when confronted with a stressor (Schulz et al., 2005). *SR* has also been

shown to be relatively consistent across response systems (Schlotz et al., 2008) and stressors (Hawkley et al., 2001).

*SR* can be conceptualized as specific or general: Whereas specific *SR* refers to the reactivity of a singular response system (e.g., endocrine, cardiovascular, or affective *SR*), general *SR* is reflected by aggregating the responses across domains and stressors (Schlotz, 2013). Concerning specific *SR* domains, high cardiovascular *SR* has been linked to adverse health outcomes numerous times (Light, 1981; Obrist, 1981; Chida & Steptoe, 2010; Turner et al., 2020; Greaves-Lord et al., 2007; Kagan et al., 1987). Endocrine and affective *SR* have been suggested to be risk factors for developing mental disorders, such as psychosis, anxiety disorders, and depression (Heim & Nemroff, 2001; Myin-Germeys & van Os, 2007).

In contrast to specific SR, general SR assumes a generalizability of stress responses across different response systems and stressors. It is based on the presumption that different aspects of the stress response have a common origin in cortical areas mediating HPA axis and ANS activation, as well as subjectiveemotional and behavioral responses (Schlotz, 2013). General SR can be measured in a laboratory setting or in everyday life. In order to measure stress responses ecologically validly, ambulatory assessment methods are used increasingly frequently (Schlotz, 2013). While ambulatory assessment methods present the opportunity to measure real-time SR is daily life, these methods are fraught with numerous practical problems: As general SR involves different response systems that need to be monitored across time and stressors, an aggregation of a plethora of measurement instances is needed in order to obtain a comprehensive and accurate assessment of SR (Schlotz et al., 2011b). As this is often too expensive and too difficult to conduct, many studies utilize self-report approaches asking individuals to report stress responses they typically show when confronted with different day-today stressors. These self-reported scores may be referred to as perceived SR (Schlotz et al., 2011a). Although ambulatory assessment and retrospective selfreport measures do not yield identical results, they can be expected to be correlated (Federenko et al., 2004). For that reason, a questionnaire measuring perceived SR, the Perceived Stress Reactivity Scale (PSRS; Schlotz et al., 2011a) is part of the questionnaires in the superordinate study.

Perceived SR is linked to a number of personality traits that are relevant to the stress process: Perceived SR has been shown to be negatively associated to selfefficacy (Bandura, 1997; Schlotz et al., 2011a). Individuals with low self-efficacy scores tend to react to stressful situations more intensely and appraise stressors as less manageable and more threatening (Bandura, 1998). This could be confirmed regarding the cortisol response: Individuals with low self-efficacy showed a higher cortisol reaction in response to a laboratory stressor (Kirschbaum et al., 1995). Also, perceived SR is strongly linked to neuroticism, defined as a tendency toward hostility, anxiety, depression, and self-consciousness (John & Srivastava, 1999). Although perceived SR and neuroticism concepts do overlap, SR is linked to stressful situations specifically, whereas neuroticism describes individual differences across multiple different situations (Schlotz et al., 2011a). It could be confirmed that individuals high in neuroticism or closely related constructs, such as trait anxiety, showed increased aggregated stress responses (Schwebel & Suls, 1999; Schlotz et al., 2006). Perceived SR is furthermore linked to negative health outcomes; this association is moderated by exposure to stress. Since stress has an adverse effect on sleep and mood, it was shown that high perceived SR is linked to poor sleep quality and depressive symptoms, especially when chronic stress was high (Schlotz et al., 2011a).

To the author's knowledge of current literature, studies addressing the link between perceived *SR* and physiological and psychological aspects of the acute stress response are extremely limited. Only two studies examined the link between perceived *SR* and the HPA axis stress response assessed through salivary cortisol concentration: Hammerfald et al. (2003) examined changes in salivary cortisol concentration in 41 healthy individuals after exposing them to a psychosocial laboratory stressor. Before being exposed to the laboratory stressor, perceived *SR* of the participants was assessed. Over the course of the experiment, eight saliva samples were collected in 10-minute intervals and the area under the cortisol reaction curve was calculated (Pruessner et al., 2003). The sample was then dichotomized into individuals with low or high values in perceived *SR*. The group high in perceived *SR* showed a significantly steeper increase in cortisol concentration. In another study by Schlotz et al. (2011b), 66 healthy participants underwent a similar procedure, but in this instance latent growth curve models

(Cudeck & Klebe, 2002) for change in salivary cortisol concentration were calculated. In this study, higher values in perceived *SR* were associated to steeper cortisol responses.

Concerning the association between perceived *SR* and changes in *HR* or *SubjStress*, research has yet to be conducted. Since *SR* is thought to be relatively stable across response systems (Schlotz et al., 2008) and because of its dispositional conceptualization, it can be assumed that *SR* predicts *HR* and *SubjStress* in a social evaluative stress situation.

# 1.4.2. Primary Appraisal (Lazarus & Folkman, 1984)

Besides individual differences in dispositional *SR*, stress responses may also be influenced through differences in cognitive appraisal processes (McEwen, 1998). In the transactional stress theory proposed by Lazarus and Folkman (1984), these cognitive appraisal processes are assumed to be highly important mediators of the stress process. As mentioned above, these cognitive processes may be subdivided into *PA* and secondary appraisal. Through *PA* processes the individual appraises whether the stressor is relevant to their well-being, i.e., threatening, or challenging, whereas through secondary appraisal processes one's own coping resources are evaluated. According to Lazarus and Folkman (1984), a stress reaction sets in if both cognitive appraisal processes occur, i.e., a threat or challenge is present and coping resources are inadequate. As *PA* and secondary appraisal processes always occur in relation to a specific event or situation, both appraisal processes are conceptualized as state variables (Gaab et al., 2005).

PA and secondary appraisal processes have been shown to be correlated with general personality factors relevant to the stress process: Similar to SR, PA has been negatively associated with self-efficacy (Gaab et al., 2005). Individuals scoring low on self-concept of own competence and control expectancy (both indices for low self-efficacy), appraised a psychosocial laboratory stressor as more threatening (Gaab et al., 2005). On the other hand, secondary appraisal has been positively linked to self-efficacy: Individuals with high values in self-efficacy tend to appraise their own coping resources as sufficient to cope with a laboratory stressor (Gaab et al., 2005). Also, neuroticism has been linked to PA processes: In a study by

Gallagher (1990), individuals high in neuroticism appraised future stressful academic events as more challenging than individuals scoring low in neuroticism.

Concerning the relationship of *PA* and secondary appraisal processes on the one hand and physiological and psychological aspects of the stress response on the other hand, the majority of studies focused on the cortisol stress response: In a study by Gaab et al. (2005), a significant association between *PA* and an increased salivary cortisol concentration in response to a laboratory stressor could be shown. Interestingly, this association could not be confirmed for secondary appraisal processes. This association pattern between *PA* and the cortisol response and the lack thereof regarding secondary appraisal could be replicated in a study by Schlotz et al. (2011b). In another study (Wirtz et al., 2006), the same pattern could be confirmed regarding a measure of the coagulant stress response (D-dimer): Whereas *PA* was correlated to the area under the D-dimer reaction curve, secondary appraisal did not show significant correlations. These findings suggest that *PA* processes are related more closely to the stress response than secondary appraisal processes. For that reason, only *PA* processes will be examined in current project.

Regarding the relationship between PA processes and the cardiovascular stress response, only a limited number of studies have been conducted, yielding inconclusive results: On the one hand, the predictive effect of appraising a laboratory stressor as threatening and challenging on HR increase was demonstrated in a series of experiments conducted by Tomaka et al. (1993). The same predictive effect of PA on HR increase could be replicated in a study conducted by Quigley et al. (2002). On the other hand, no significant correlations nor predictive effects of PA on HR increase were found in studies by Skoluda et al. (2011) and Maier et al. (2003). Even though data on the relationship between PA and HR is sparse and inconclusive, a positive association between PA and HR can be assumed: Since stress appraisal processes are thought to be the critical component in eliciting the ANS response resulting in an increase in HR (Andrews et al., 2013), a higher threat and challenge appraisal should therefore be related to a higher increase in HR. Following the transactional stress theory (Lazarus & Folkman, 1984), appraisal processes need to be involved in order to elicit a stress response comprising physiological, emotional, and behavioral aspects (i.e., an increase in HR). This

conceptualization implies a temporal precedence of *PA* before changes in *HR* occur. Following that reasoning, a predictive effect of *PA* on *HR* can be assumed.

Regarding the subjective emotional stress response and its relationship to *PA* processes, only a small number of studies have been conducted: In the aforementioned studies by Tomaka et al. (1993) and Maier et al. (2003), the predictive effects of *PA* on *SubjStress* could be shown. As appraisal processes are thought to elicit responses of the emotional stress system (Andrews et al., 2013) and are conceptualized to temporally precede changes in *SubjStress*, a predictive effect of *PA* on *SubjStress* can be assumed.

# 1.4.3. Stress Reactivity and Primary Appraisal

Cognitive appraisal processes are malleable through cognitive behavioral therapy (Gaab et al., 2003; Hammarfald et al., 2006) in order to modify physiological and psychological responses to stress, e.g., *HR* and *SubjStress*. In opposition, individual differences in *SR* are expected to be less susceptible to change due to their

origins in early developmental and genetic variability (Gunthert et al., 2007; Phillps, 2007). In order to generate implications for the conceptualization of future psychological stress interventions, a better understanding of the shaping roles of *PA* and *SR* on the cardiovascular and subjective emotional stress response appears useful.

A possible pathway of the influence of *SR* and *PA* on the stress response would be a model in which the association between *SR* and the stress response is mediated by *PA*: Because of the assumed temporal structure of the variables (i.e., *SR*, as a dispositional variable, temporally preceding *PA*, conceptualized as a situational state variable), an influencing effect of *SR* on *PA* can be assumed. This assumption taken together with the proposed predictive effects of *SR* and *PA* on *HR* and *SubjStress* speak for the plausibility of the mediation model. Additionally, in a study by Schlotz et al. (2011b), this mediation model could be confirmed using the cortisol stress response as outcome measure.

Given that reasoning, it can be assumed that the influence of *SR* on *HR* and *SubjStress* is mediated by *PA* (for a graphic depiction of proposed model, see Fig. 2).

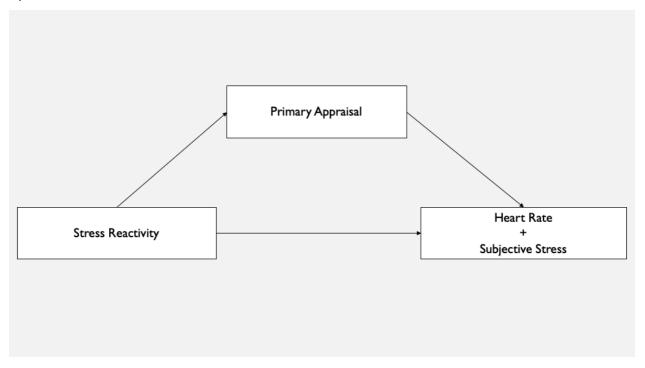


Figure 2. Proposed model testing for mediation of the effect of SR on HR and SubjStress by PA.

As a premise for proposed mediation model, *SR* and *PA* need to exhibit a significant association. As *SR* is defined as the disposition of an individual to respond to stressors with intense, immediate, and long-lasting stress reactions (Schulz et al., 2005) and also due to the temporal structure of *SR* and *PA*, a predictive effect of *SR* on *PA* can be assumed.

# 2. Research Question and Hypotheses

Following the reasoning above, following research question arises: What are the singular and cumulative effects of dispositional *stress reactivity* (*SR*) and *primary appraisal* (*PA*) on *heart rate* (*HR*) and *subjectively perceived stress* (*SubjStress*) in a social evaluative stress situation?

This research question yields following hypotheses:

- **H1)** *SR* significantly predicts *PA*.
- **H 2 + 3)** *SR* significantly predicts *HR* increase and increase in *SubjStress*.
- **H 4 + 5)** *PA* significantly predicts *HR* increase and increase in *SubjStress*.
- **H 6 + 7)** The effect of *SR* on *HR* and *SubjStress* is mediated by *PA*.

#### 3.Methods

For current research project, data gathered in a superordinate larger study called "Music, stress, and skin barrier recovery (MuSkiBa II)" conducted by Dr. Jasminka Majdandžić and Univ.-Prof. Dr. Urs Nater at the Institute for Clinical and Health Psychology at the Faculty of Psychology at the University of Vienna, Austria, was used for analysis. In this larger project, numerous factors possibly affecting the physiological and psychological stress response are assessed and examined. As the name of the greater study suggests, the effects of music or audiobook listening on the stress response and skin barrier recovery are the main focus of the future publication covering the entire data set of the study.

As opposed to the superordinate study, current project only focuses on data related to *stress reactivity* (*SR*) and *primary appraisal* (*PA*) scales and subscales, *subjectively perceived stress* (*SubjStress*) ratings and *heart rate* (*HR*) assessments only up to a certain point in the experimental procedure. For that reason, only steps of the procedure relevant for current project will be described in detail. In interests of completeness, all other steps will be briefly described.

#### 3.1. Participants

In the superordinate study, biological markers of HPA axis and ANS activity, including salivary cortisol and alpha-amylase concentration, are assessed. Since these two salivary stress markers and other biological markers of the stress reaction, such as cardiovascular activity, are highly susceptible to a plethora of confounding effects (Strahler et al., 2017), participants had to meet the requirements of an extensive catalogue of inclusion and exclusion criteria:

As secretion of both cortisol and alpha-amylase undergoes tremendous changes throughout the lifespan (Nater et al., 2013; Strahler et al., 2010), only individuals aged 18 to 35 years were eligible to participate in order to obtain comparable data without the need to control for age as a possible confounding variable. Since body weight, especially body mass considered to be overweight and/or obese, impacts HPA axis and ANS reactivity (Stadler et al., 2017; Phillips, 2011), study inclusion was only possible for participants with a body mass index (BMI) of 18 to 25 kg/m².

As gender socialization and biological sex differences affect the development of the HPA axis and thus the cortisol response to stress (Panagiotakopoulos & Neigh, 2014), only individuals with biological features considered female and who were assigned female at birth were eligible to participate. Since drastic changes in sex steroids occur during the female menstrual cycle, the respective menstrual cycle phases have an impact on HPA axis and ANS activity (Kirschbaum et al., 1999). Other profound sex steroid changes occur during pregnancy (Gisbrecht et al., 2013), or when hormonal contraceptives are used (Cornelisse et al., 2011), which in turn influence HPA axis responsivity. For that reason, participants had to have a regular menstrual cycle to be able to participate in the study. Participants could furthermore not use hormonal contraceptives or be pregnant. To control for possible influences of the menstrual cycle phase on the stress reaction, laboratory sessions with the participants were conducted only in the beginning of their respective follicle phases.

A wide variety of somatic health problems, including atopic conditions (e.g. asthma, dermatitis psoriasis), allergies or hypersensitivity reactions, pulmonary and respiratory diseases, cancer, chronic pain, diabetes mellitus, metabolic syndrome, hypertension and cardiovascular disease, neurological disorders, thyroid abnormalities, diseases of the digestive tract and infectious diseases (e.g. HIV, hepatitis) affect cortisol and alpha-amylase secretion (Strahler et al., 2017). Additionally, a variety of mental health problems, such as affective, anxiety and eating disorders also affect these systems (Knorr et al., 2010; Stadler et al., 2017; Morris et al., 2012). In order to control for possible influencing effects resulting from somatic illnesses and mental disorders, only individuals not suffering from any of the conditions named above were eligible to participate.

Also, individuals using medication possibly affecting HPA axis activity regularly, such as selective serotonin reuptake inhibitors, synthetic steroids, uterine-

active agents, diuretics and antidiuretics, sympathomimetic agents, phenothiazines, monoamine oxidase inhibitors or corticosteroids (Granger et al., 2009), or individuals using medication to treat an acute illness were excluded from study participation.

It has also been shown that habitual smoking (Kirschbaum et al., 1992), excessive alcohol use (Badrick et al., 2008) and recreational drug use (Fosnocht & Briand, 2016) have an impact on cortisol stress reactivity. For that reason, only non-habitually/completely non-smoking participants who do not drink alcohol excessively and have not taken recreational drugs in the past year (exception: cannabis in the past 14 days) were eligible for participation.

Further inclusion and exclusion criteria were the following: fluency in the German language, no non-correctable visual impairments, no (dental) surgery in the past 8 weeks, no tropical stay in the past 6 months, no previous experience with the social-evaluative stress task employed in the study, no personal relationships with study team members.

Participants were recruited through postings on social media (mainly Facebook groups related to the University of Vienna and/or platforms specifically for individuals interested in participating in scientific studies), to which a flyer with basic inclusion and exclusion criteria and a contact email address was attached. The flyer also advertised the expense allowance participants received after completing the study (45 €). Also, bulletins advertising for the study were posted around the University of Vienna.

Once prospective participants expressed interest via email, they were asked to provide a phone number and time periods in which an approximately 20-minute phone screening interview could take place. In that interview, aforementioned exclusion and inclusion criteria were specified and participation eligibility was determined. In order to control for possible confounding interviewer effects, only specifically trained members of the study team conducted the interview in a standardized manner.

Once participation eligibility was assessed, prospective participants were invited to participate in a laboratory session in the beginning of their respective follicle phases. Participants also received an email with a link to an extensive battery of online questionnaires they were asked to complete up to 24 hours before the

laboratory session.

Experimental assessments started in the beginning of February 2020 and were initially planned to be held out continuously until analyzable data of approximately 90 participants (30 participants for each condition, see below) would be available. Due to national lockdowns and restrictions resulting from the global Covid-19 pandemic, face-to-face laboratory sessions had to be suspended starting from March 2020. In December 2020, one single laboratory session was possible in between two nationwide lockdowns. Lockdown was alleviated in March 2021, which again allowed for face-to-face laboratory sessions. Due to the spread of the highly infections B1.1.7. variant of the SARS-CoV2 virus and resulting rising infection rates, laboratory sessions had to be suspended once again at the end of March 2021. All data gathered up to the end of March 2021 is considered in this analysis.

Because of the recurring restrictions and lockdowns, it was only possible to conduct the experiment with a very small number of participants. In total, data of 15 participants was available at the time in which statistical analyses were conducted. Due to missing values in *primary appraisal* (2 participants failed to fill out the *Primary Appraisal/Secondary Appraisal Scale*) or atypical stress reactions (decreased heart rate during social evaluative stress task compared to baseline levels in 2 participants), data of 4 participants had to be excluded.

After exclusion, data of 11 participants aged 21 to 32 (M = 25.55 years, SD = 3.48 years) was available for final analyses.

#### 3.2 Procedure

As stated above, current project is embedded in the MuSkiBa II study conducted at the University of Vienna. The MuSkiBa II study aims to examine the effects of music and/or audiobook listening on skin barrier recovery after exposure to psycho-social stress. In order to test for these effects, participants underwent a tape stripping procedure after exposure to the social-evaluative stress task. Subsequently, their trans-epidermal water loss (TEWL) was assessed in order to gain a measure for skin barrier function and recovery. Since both tape stripping procedure and TEWL measurements are not relevant for current project, they will be omitted from analysis and discussion. For further detail on both procedures the

reader is kindly referred to following literature (Rogiers, 2001; Gouin & Kiecolt-Glaser, 2011).

Two experimental and one control group were examined, yielding three conditions: music, audiobook, and silence (members of the silence group were given time to relax without listening to music or an audiobook, more detailed description further down below). Since all data relevant to current project was gathered before participants were exposed to either music or audiobook listening or the silence condition, it can be assumed that the respective experimental condition has not had an influence on relevant data. For that reason, experimental condition will be omitted from analysis and discussion.

Before each laboratory session, a wide range of psychological and socioeconomic variables was assessed through the aforementioned extensive online questionnaire battery. One of the questionnaires administered online is the *Perceived Stress Reactivity Scale (PSRS;* Schlotz et al., 2011a) measuring *stress reactivity (SR;* Schulz et al., 2005). The *PSRS* questionnaire and *SR* measurement will be discussed in detail further down below. Furthermore, participants tested after February 2020 were asked to provide a negative Covid-19 test or proof of vaccination at the beginning of the laboratory session to ensure safety of the test team.

Since cortisol levels in humans are subject to diurnal change patterns throughout the day (Adam et al., 2017), all participants were asked to arrive at the same time (approximately at 12:50) on the respective date of their laboratory session to control for possible influencing effects.

For the first 30 minutes, participants were given an acclimatization period in order to reduce their stress parameters to baseline levels. In this time period, participants were provided basic information about the study and asked to fill out an informed consent form. Furthermore, they were equipped with a *movisens* ECG sensor (EcgMove4, movisens GmbH, Karlsruhe, Germany) in order to measure *HR*. *HR* measurement, calculation of average *HR* and *HR* changes will be discussed in greater detail down below. Participants were also prepared for the tape stripping procedure and TEWL measurements and asked to choose a music playlist or an audiobook to listen to after being exposed to the social-evaluative stress task (for

participants in the silence condition, this part was skipped). During the acclimatization period participants also were able to practice collecting saliva samples. Due to the Covid-19 pandemic, participants tested after February 2020 were also asked to answer a short questionnaire assessing possible Covid-19 symptoms. Additionally, their temperature was measured twice to ensure safety of the test team.

After the acclimatization, participants were asked to set a marker on their movisens ECG sensor by double tapping it (M1) and to collect their first saliva sample. In order to do so, participants were asked to swallow one last time and then to collect their saliva for two minutes (without swallowing). In these two minutes, they were asked to fill out a number of questionnaires and rate their subjectively perceived stress (SubjStress<sub>Base</sub>). SubjStress measurement and calculations of increases in SubjStress will be discussed in greater detail down below. Subsequently, participants were asked to release their saliva through a straw into a polypropylene tube (SaliCap, IBL International, Hamburg, Germany) and to finish filling out the aforementioned questionnaires subsequently. The participants were then given the instructions for the *TSST* and were allowed to prepare for three minutes. After the preparation period, theparticipants were again asked to set a marker on their movisens ECG sensor (M2) and to start collecting their saliva for the second saliva sample. In that two-minute period, participants were again instructed to fill out a number of questionnaires and rate their SubjStress (SubjStress<sub>Pre</sub>). One of the questionnaires administered at that point is the *Primary Appraisal/Secondary* Appraisal Scales (PASA; Gaab et al., 2005) measuring primary appraisal (PA; Lazarus & Folkman, 1984). The PASA questionnaire and PA measurement will be discussed in detail further down below. After collecting their salive for two minutes, participants were again asked to release their saliva into a SaliCap and to finish filling out the questionnaires. Afterwards, the participants had to perform the 5minute free speech task and the 5-minute mental arithmetic task. The participants were then once again asked to double tap the *movisens* ECG sensor to set a marker (M3). They were also asked to again collect their saliva for two minutes and to then release it into a SaliCap. They were also again instructed to fill out a number of questionnaires and to rate their subjectively perceived stress (SubjStress<sub>Stress</sub>). All

steps described up to this point in the timeline of the study are the steps relevant for current project (for a graphic overview of steps described above, see Fig. 3).

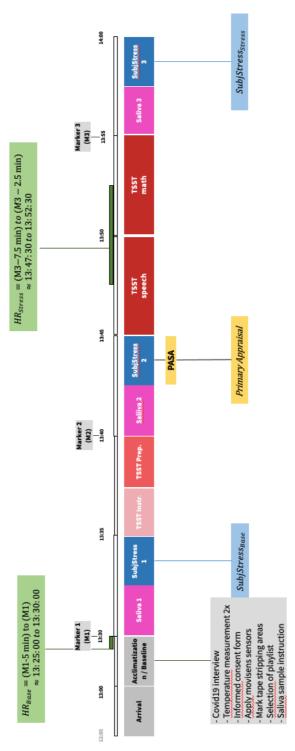


Figure 3. Detailed timeline of first part of study relevant for current thesis. Italics indicate measurements relevant for analyses of current project.

In interests of completeness, the entirety of the study procedure will be described briefly: The first TEWL measurement was conducted afterwards. The participants then underwent a tape stripping procedure and another TEWL measurement took place. After the TEWL measurement was completed, another marker was set on the ECG sensor, a saliva sample was collected, and participants filled out questionnaires. Then the music or audiobook listening session started. Participants listened to their previously chosen music playlist or audiobook while laying on a comfortable deckchair in a slightly darkened room (participants in the silence condition underwent the same procedure without listening to music or an audiobook). Afterwards, the measurement procedure described above was repeated (ECG-marker, saliva sample, TEWL measurement, questionnaires). This procedure was repeated three more times subsequently in 30-minute intervals. In between measurements, participants were given magazines to relax and pass time. After all measurements were completed, participants were debriefed and sent home (for a graphic overview of entire study timeline, see Fig. 4).

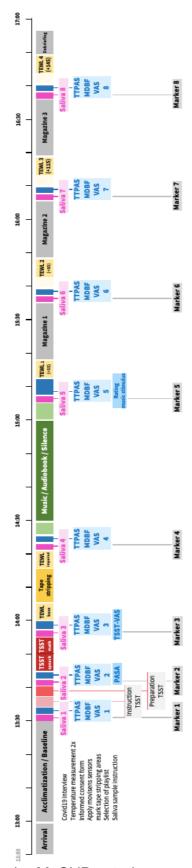


Figure 4. Detailed timeline of entire MuSkiBa study procedure.

#### 3.3. Measurements

# 3.3.1. Stress Reactivity

SR was assessed using the Perceived Stress Reactivity Scale (PSRS; Schlotz et al., 2011a). As mentioned above, the PSRS was part of the online questionnaire battery completed at least 24 hours before the respective laboratory session.

The questionnaire is the revision of an existing German scale assessing *SR* (Stress-Reaktivitäts-Skala, *SRS*; Schulz et al., 2005). The original *SRS* consists of 36 items covering 6 subscales. Each subscale represents a typically stressful area of everyday life (Reactivity to Work Overload, Reactivity to Social Conflicts, Reactivity to Social Stress, Reactivity to Failure, Anticipatory Stress Reactivity, Prolonged Stress Reactivity). Also, an overall stress reactivity index can be calculated by aggregating scores across all 6 subscales.

After revision and statistical reevaluation by Schlotz et al. (2011a) using data gathered across a large sample from the United Kingdom, the U.S., and Germany (N = 2040), 23 items and 5 subscales remained (Reactivity to Work Overload, Reactivity to Social Conflicts, Reactivity to Social Evaluation, Reactivity to Failure, Prolonged Stress Reactivity). The *PSRS* scales show high internal consistency (.62 <  $\alpha$  < .91) and high test-retest reliability (.62 <  $r_{\alpha}$ < .91).

To measure *SR*, items were generated that in the first part described typical stressful events covered by one of the subscales mentioned above. In the second part of each item, the participant is asked to rate their typical response to these potentially stressful events along a 3-step scale covering stress reactions of different intensities (e.g.: "When I make a mistake...": "(0) In general, I stay confident"; "(1) I sometimes feel unsure about my abilities"; "(2) I often have doubts in my abilities"). Higher scores in each scale indicate a higher stress reactivity in the respective domain.

Because the *TSST* is characterized by social-evaluative threat and since its tasks are designed to make failure very likely, only the subscales *Reactivity to Social Evaluation* (*ReacSocEval*) and *Reactivity to Failure* (*ReacFail*) were considered for analyses (analogous to Schlotz et al., 2011b).

# 3.3.2 Primary Appraisal

Primary Appraisal was assessed using the transactional stress questionnaire (Primary Appraisal Secondary Appraisal Scale, PASA; Gaab et al., 2005). Since the PASA measures anticipatory cognitive appraisal, the PASA is administered right after participants receive the instructions for the TSST, but before the TSST takes place.

The *PASA* is a questionnaire designed to cover each of the cognitive appraisal processes described in the transactional stress theory (Lazarus & Folkman, 1984): *Threat, Challenge*, Self-Concept of Own Abilities, and Control Expectancy. Each cognitive appraisal process is represented by a subscale of the *PASA*. Each subscale comprises 8 items, on which participants had to indicate to what extent each statement applied to them on a 6-step scale (e.g.: "I find this situation very unpleasant" (1) "Strongly disagree" up to (6) "Strongly agree"). Higher scores in each scale indicate a more stressful appraisal of the respective situation.

In order to compute a measure for *Primary Appraisal (PA)*, scores of the *Threat* and *Challenge* subscales are aggregated (for Secondary Appraisal, Self-Concept of Own Abilities and Control Expectancy scores are aggregated). Also, a general stress index can be calculated by aggregating *Primary Appraisal* and Secondary Appraisal scores. The *PASA* scales show high internal consistency (.63 <  $\alpha$  <.83).

Since the majority of studies found positive relations between *Threat* and *Challenge* appraisals (or *PA*, respectively) and hypothalamic-pituitary-adrenal and autonomous nervous system reactivity (e.g. Gaab et al., 2005; Wirtz et al., 2006; Denson et al., 2009), but not secondary appraisal, only *Threat, Challenge* and *PA* were considered for analyses (analogous to Schlotz et al., 2011b).

# 3.3.3. Subjectively Perceived Stress

Subjectively Perceived Stress (SubjStress) was assessed using visual analogous scales (VAS). A VAS is a line of 100 mm length on which participants indicate how stressed they feel by marking the line at the respective place. SubjStress was assessed at the time marks specified above. In order to obtain an

index of stress-induced change in *SubjStress*, delta scores were defined as the difference between the VAS administered right after the TSST and the VAS administered at baseline ( $\Delta_{SubjStress} = SubjStress_{Stress} - SubjStress_{Baseline}$ ; analogous to Skoluda et al., 2015).

#### 3.3.4. Heart Rate

Heart Rate (HR) was measured continuously in beats per minute (bpm) throughout each laboratory session using a movisens ECG sensor (EcgMove4, movisens GmbH, Karlsruhe, Germany). In order to obtain analyzable data, average HR of critical time periods relevant to the research question had to be specified in relation to the markers set at the time marks mentioned above. The length of the critical time window was set to 5 minutes.

As a baseline HR value, average HR of the 5-minute time period leading up to M1 ( $HR_{baseline} = \emptyset HR_{(M1-5\min)to\ (M1)}$ ) was computed using the *unisens Viewer* and *unisens Data Analyzer* software (movisens GmbH, Karlsruhe, Germany).

As a stress HR value, average HR of the 5-minute time period comprising the last 2,5 minutes of the TSST speech task and the first 2,5 minutes of the TSST arithmetic task was computed  $(HR_{stress} = \emptyset HR_{(M3-7.5 \min) to (M3-2.5 min)})$ .

In order to obtain an index of stress-induced change in HR, delta scores were defined as the difference between average HR during the TSST and average HR at baseline ( $\Delta_{HR} = HR_{stress} - HR_{baseline}$ ). For a graphic depiction of HR measurements and calculations, see Fig. 3.

#### 3.4. Statistical Analyses

Initially, it was planned to conduct statistical analyses on data gathered across a medium-sized sample tested continuously starting from February 2020. For that reason, analyses relying on regression models seemed feasible. As the Covid-19 pandemic reached Europe in the middle of February 2020 and Austria went into its first Lockdown on March 16<sup>th</sup> 2020, face-to-face laboratory sessions had to be suspended for extended time periods, which in turn drastically reduced the number of tested participants.

For hypotheses 1 through 5, linear regression analyses were planned initially. For hypotheses 6 and 7 it was planned to conduct mediation analyses by applying the bootstrapping method by Preacher and Hayes (2004). All statistical analyses were conducted using IBM SPSS Statistics (Version 26) predictive analytics software.

To estimate sample sizes allowing for significant results for the respective hypotheses, G\*Power analyses for bivariate and multivariate linear regression models were conducted (Faul et al., 2007; Faul et al., 2009) using effect size estimates established in a similar study (Schlotz et al, 2011b). In this study, the cumulative effects of *SR* and *PA* on the cortisol response in context of a social evaluative stress task were examined.

G\*Power analyses yielded sample size estimates greatly exceeding the number of participants available for current analyses (with sample size estimates for proposed hypotheses ranging from n = 44 up to n = 631), which made statistically significant findings highly improbable.

For that reason, results from initially planned statistical analyses will only be briefly discussed. Additionally, a descriptive approach, called extreme case analysis, examining the participants with either the highest or lowest value in the independent variables will be followed.

#### 4. Results

#### 4.1. Descriptive Statistics

Regarding scales relating to SR, mean value of ReacSocEval was M = 5.55 (SD = 2.54, Range 0 to 8) and the mean value of ReacFail was M = 5.09 (SD = 1.58, Range 0 to 10). Scales related to PA were threat (M = 13.0, SD = 4.63, Range 0 to 24), challenge (M = 18.73, SD = 3.32, Range 0 to 24), and PA (M = 15.36, SD = 2.89, Range 0 to 24). The mean increase in HR was M = 23.48 (SD = 12.32, Range 6.27 to 47.41). Mean increase in SubjStress, was M = 46.0 (SD = 30.16, Range 16.0 to 98.0).

#### 4.1.1. Manipulation check

To check if the TSST successfully induced stress, a series of one-sample t-test were conducted. To check if the TSST task introduction reliably induced stress, a dependent t-test between  $SubjStress_{Baseline}$  and  $SubjStress_{Pre}$  was conducted. As this test reached statistical significance ( $t(10) = -5.35 \ p < .001$ ) and because of the direction of the t-value, a stress inducing effect of the TSST task introduction can be assumed. To further check if TSST task exposure induced stress, a dependent t-test between  $SubjStress_{Baseline}$  and  $SubjStress_{Stress}$  as well as  $HR_{baseline}$  and  $HR_{stress}$  were conducted. Both t-tests for SubjStress (t(10) = -5.06, t < .001) and for t (t (10) = -6.32, t < .001) reached statistical significance implying that t t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t < t

# 4.2. Regression and mediation models

# 4.2.1. Predictive effects of SR on PA (H1)

With one exception, the predictive effect of either SR scale on any of the three scales related to PA could not be confirmed. Using ReacSocEval as the independent and threat as the dependent variable did not yield a statistically significant prediction model (F (1,9) = .43, p = .53). ReacSocEval did not contribute to the model significantly (B = .21, D = .53). This was also the case for Challenge (C (1,9) = .31, D = .59; C = -.19, D = .59), and C (C (1,9) = .04, C = .85; C = .07, C = .85) as dependent variables.

Using *ReacFail* as the independent variable yielded a significant prediction model when using *threat* (F (1,9) = 5.33, p = 0.05; B = .60, p = 0.05) as the outcome variable. In regard to *challenge* (F (1,9) = .39, p = .55; B = -.21, P = .55) and PA (F (1,9) = 1.38, P = .27, P = .37, P = .27) as outcome variables, no predictive effects of *ReacFail* could be found.

#### 4.2.2. Predictive effects of SR on HR and SubjStress (H2 + H3)

The predictive effect of either *SR* scale on *HR* could not be confirmed. Using *ReacSocEval* as the independent variable did not yield a statistically significant

model (F (1,9) = .51, p = .49); ReacSocEval consequently did not contribute to the model significantly ( $\mathcal{B}$  = .23, p = .49). ReacFail did also not predict HR in a statistically significant manner (F (1,9) = .14, p = .71;  $\mathcal{B}$  = .13, p = .71).

Both SR scales did also not predict SubjStress in the given sample: Using ReacSocEval (F (1,9) = .6, p = .46; B = -.25, p = .46) as well as using ReacFail (F (1,9) = .05, p = .83; B = .074, p = .83) did not yield a significant regression model.

# 4.2.3. Predictive effects of PA on HR and SubjStress (H4 + H5)

Neither the two *PA* subscales nor *PA* predicted *HR* significantly. Using *threat* as the independent variable did not yield a statistically significant result (F (1,9) = .01, p = .92); *threat* did consequently not contribute to the model significantly ( $\mathcal{B}$  = .03, p = .92). *Challenge* (F (1,9) = .1, p = .76;  $\mathcal{B}$  = .10, p = .76) and PA (F (1,9) = .07, p = .80;  $\mathcal{B}$  = .09, p = .80) did also not predict *HR*.

None of the three *PA*-related scales yielded significant prediction models concerning *SubjStress:* Regression models using *threat* (F (1,9) = 1.03, p = .34; B = .32, p = .34), *challenge* (F (1,9) = .79, p = .40; B = .28, D = .40), or D (D = .19, D = .20; D = .42, D = .20) as predictors did not reach statistical significance.

### 4.2.4. Mediation of effect of SR on HR by PA (H6)

As none of the three scales related to *PA* as well as all *SR* scales yielded any significant prediction models regarding *HR* and *SubjStress*, a moderating effect of *PA* on the effect of *SR* on the outcome measures is highly improbable. In interests of concision, the results of the analysis only using *PA* as mediator will be briefly described:

The relationship between *ReacSocEval* and *HR* was not mediated by *PA*. As Fig. 5 illustrates, the standardized regression coefficient between *ReacSocEval* and *PA* and between *PA* and *HR* did not reach statistical significance. The standardized indirect effect was (.065)(.071) = 0.046. Using bootstrapping methods (Preacher & Hayes, 2004), unstandardized indirect effects were calculated for 10,000 bootstrapped samples. Then the 95% confidence interval was computed by assessing the indirect effects at the 2.5<sup>th</sup> and 97.5<sup>th</sup> percentiles. The bootstrapped

unstandardized indirect effect was .0224 and the 95% confidence interval ranged from -1.49 to 1.84. Since this interval contains 0, the indirect effect was non-significant (Fig. 5).

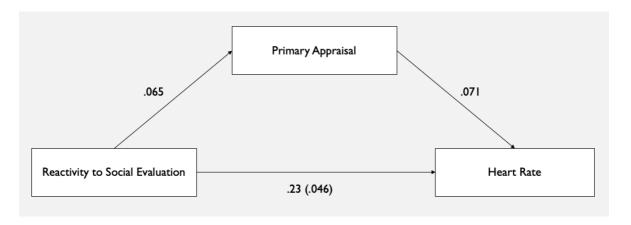


Figure 5. Mediation model with ReacSocEval as independent and HR as dependent variable. All paths p > .05.

The relationship between *ReacFail* and *HR* was also not mediated by *PA*. With both standardized coefficients between *ReacFail* and *PA* as well as *PA* and *HR* not being significant, the standardized indirect effect of the model was (.36)(.041) = 0.015. The bootstrapped unstandardized indirect effect was .13 with a 95% confidence interval ranging from -3.71 to 5.23. Thus, the indirect effect was not significant (Fig. 6).

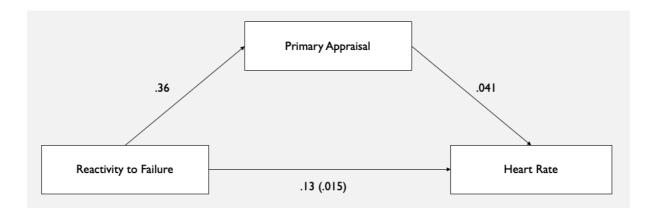


Figure 6. Mediation model with ReacFail as independent and HR as dependent variable. All paths p > .05.

## 4.2.4. Mediation of effect of SR on SubjStress by PA (H7)

The effect of *ReacSocEval* on *SubjStress* was not mediated by *PA*. The standardized regression coefficients between *ReacSocEval* and *PA* as well as *PA* and *SubjStress* were non-significant. The standardized indirect effect of the model was (.065) (.43) = .028. The bootstrapped unstandardized indirect effect was .34 with a 95% confidence interval ranging from -2.82 to 5.29. Thus, the indirect effect was non-significant (Fig. 7).

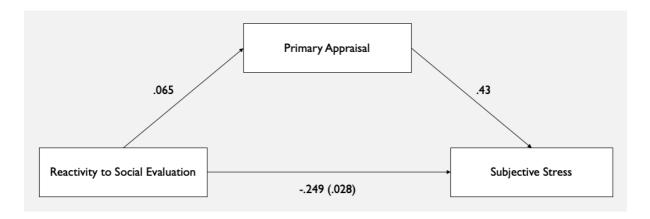


Figure 7. Mediation model with ReacSocEval as independent and SubjStress as dependent variable. All paths p > .05.

Futhermore, the relationship between *ReacFail* on *Subjstress* was not mediated by *PA*. Standardized regression coefficients between *ReacFail* and *PA* as well as *PA* and *SubjStress* were not significant. The standardized indirect effect was (.36) (.45) = .16. The bootstrapped unstandardized indirect effect was 3.15 with the 95% confidence interval ranging from -4.57 to 16.37. Thus, the indirect effect was non-significant (Fig. 8).

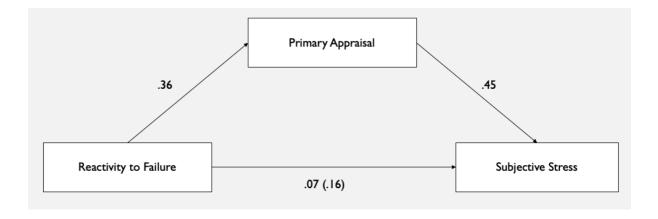


Figure 8. Mediation model with ReacFail as independent and SubjStress as dependent variable. All paths p > .05.

## 4.3. Extreme case analysis

As available sample size was far too small in order to realistically obtain statistically significant results using methods relying on regression, a descriptive approach outlining the participants with the most extreme values in the independent variables (*ReacSocEval* and *ReacFail*) will be followed.

As only two subscales of *SR*, namely *ReacSocEval* and *ReacFail*, were considered for analyses, both the participants scoring lowest and highest in *ReacSocEval* and *ReacFail* will be described: The participant scoring the lowest value in *ReacSocEval* = 2 also exhibited the lowest value in *ReacFail* = 3 (low *SR* participant, see Fig. 9). Similarly, the participant scoring highest in *ReacSocEval* = 9 also showed the largest value in *ReacFail* = 7 (high *SR* participant, see Fig. 9). This pattern could be expected, as moderate correlations of the *PSRS* subscales have already been demonstrated (Schulz et al., 2005).

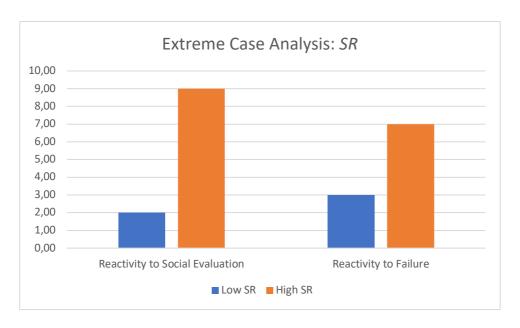


Figure 9. SR-Values of participants scoring highest and lowest in the independent variables (ReacSocEval and ReacFail).

As all paths of proposed mediation model (Fig. 2) implied a positive predictive effect of both SR subscales on PA and its related subscales, as well as HR, and SubjStress, it could be expected that the low SR participant exhibits considerably lower values in threat, challenge, PA, HR, and SubjStress compared to the high SR participant. Regarding PA and its related subscales, the low SR participant exhibited greater values in challenge = 22 and PA = 19 compared to the high SR participant (challenge = 15, PA = 15). Only in threat the high SR participant scored slightly higher than the low SR participant (low SR threat = 16 vs. high SR threat = 18). Considering the significant prediction model found when entering threat as the independent and threat as the dependent variable in a linear regression model, it was expected to find a similar pattern in the extreme case analysis approach. Interestingly, this was not the case.

Regarding HR, the low SR participant showed a lower increase in HR = 19.70 as compared to the high SR participant (HR = 26.05). Given the variance of HR in the complete sample, ranging from 6.27 up to 41.48, this difference in HR between the two examined participants appears negligible. Contrary to the prediction derived from proposed mediation model, the low SR participant showed a considerably larger increase in SubjStress compared to the high SR participant (low SR SubjStress = 75 vs. high SR SubjStress = 24). For a graphic overview of the two described participants, see Fig. 10 a-c.

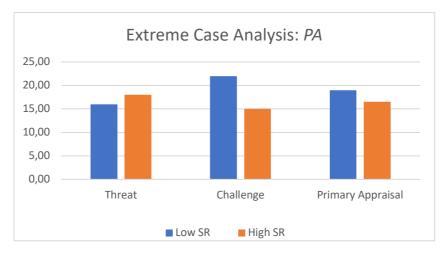


Figure 10a. Values of low SR and high SR participant in threat, challenge, and PA.

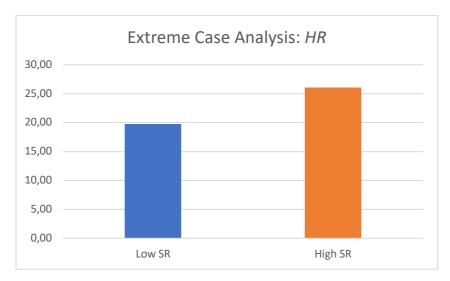


Figure 10b. Values of low SR and high SR participant in HR.

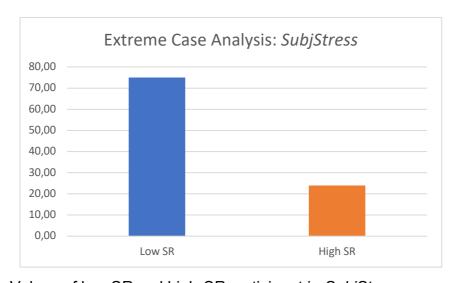


Figure 10c. Values of low SR and high SR participant in SubjStress.

#### 5. Discussion

The aim of this thesis was to examine possible predictive effects of dispositional *SR* and situational *PA* on *HR* and *SubjStress*. To determine the singular predictive effects of *SR* and *PA* on *HR* and *SubjStress*, linear regression analyses were conducted. To determine the cumulative predictive effect of *SR* and *PA*, a bootstrapping method (Preacher & Hayes, 2004) checking for the mediating effect of *PA* on the association between *SR* and the outcome measures was applied.

Using linear regression analyses, most of the proposed predictions regarding the predictive effects of *SR* on *PA* could not be confirmed. The only significant prediction model was obtained when entering *ReacFail* into the model as the independent and *threat* as the outcome variable – given the exceptionally small sample size, this significant finding implies a remarkably strong association between *ReacFail* and the *PA*-subscale *threat* in examined sample.

Regarding the predictive effects of *SR* on *HR* and *SubjStress*, no significant effects could be shown. Similarly, *PA* and its subscales also did not exhibit a predictive effect on *HR* and *SubjStress* in examined sample.

Also, the proposed mediation models (see Fig. 2) could not be confirmed: After conducting a mediation analysis using bootstrapping methods (Preacher & Hayes, 2004), no mediating effect of *PA* on the effect of *SR* on both *HR* and *SubjStress* could be proven. As *SR* did not exhibit a significant predictive effect on both outcome variables, the non-significant findings of the mediation analyses come to no surprise.

Available sample size was extremely small, leading to low statistical power of current thesis. This low statistical power decreases the probability that a statistically significant finding reflects a true effect (Button et al., 2013). For that reason, all findings described above have to be taken with caution – all interpretations and implications derived from available data do not stand on a firm empirical foundation and have to be appreciated with the highest precaution.

Because of that, a descriptive approach outlining both extreme cases in the independent variables (*ReacFail* and *ReacSocEval*) was conducted. All extreme case predictions derived from proposed mediation model could not be observed in the two described participants: The low *SR* and the high *SR* participant exhibited

similar values in all *PA* scales as well as *HR* increase. Surprisingly, the low *SR* participant reported a considerably larger increase in *SubjStress* compared to the high *SR* participant when both were exposed to the psychosocial stress-task as the hypotheses predicted the opposite case. As the extreme case approach reduces the already small data set even further, all findings derived from it also must be interpreted with high caution.

The significant predictive effect of *ReacFail* on *threat* found in the examined sample could not be observed in the participants outlined in the extreme case analysis.

Speaking for the whole examined sample, this finding implies a strong association between the two variables. As the extent of *threat* posed by the stress task was rated right after the *TSST* was introduced as an event posing a high likelihood of failure, it appears sensible that individuals reacting intensely to failure would rate a *TSST* situation as highly threatening. This finding is in line with a similar study (Schlotz et al., 2011b), in which a moderate correlation between *ReacSocEval* and *threat* was found. Concerning the findings in the extreme case analysis, a different pattern could be observed: While the high *SR* participant was expected to exhibit a greater value in *threat*, both the low *SR* and high *SR* showed reported similar values in *threat* after being introduced to the *TSST*. Measurement errors in form of self-report and recall bias in observed values in *threat* and *ReacFail* in the low *SR* and high *SR* participant may explain this pattern contradicting the hypotheses.

All other regression models concerning the predictive effects of *SR* on *PA* did not reach significance. Similarly, the findings derived from the extreme case analysis did not support the hypotheses. Several reasons for this lack of significant findings stand out: It appears sensible to examine the rating process of *SR* assessed through the *PSRS* (Schlotz et al., 2011a) and to evaluate possible biases that result from using self-report measures.

The *PSRS* was constructed to measure the typical stress response of an individual across a wide range of stressful situations. As this reflects a general tendency that is thought to be stable over time (Burleson et al., 2003), the construct measured by the *PSRS*, *SR*, can be viewed as a dispositional variable (Schulz et al., 2005). The individual filling out the questionnaire is asked to recall a number of

stressful situations of a similar kind (e.g. social-evaluative stressful situations or situation that pose a high risk of failure), evaluate their individual stress response to each one of these situations, and subsequently aggregate these individual responses into an abstract, self-perceived stress response representing the individuals typical stress response intensity. This recall process requires a high cognitive load as an aggregation of several past experienced instances is required. This makes an accurate recall of perceived *SR* difficult and suggests a high susceptibility to recall biases (Bound et al., 2001). Furthermore, Schlotz (2013) mentions that subjective self-report measures of *SR* may confound the actual individual *SR* with frequency of exposure to daily life stress – meaning, that individuals experiencing social-evaluation or failure more frequently tend to evaluate their *SR* to such exposures higher. This too might have led to inaccurate estimates of perceived *SR*, which in turn partially explains the lack of significant predictive effects of *SR* on *PA*.

Additionally, both the *PSRS* (Schlotz et al., 2011a) as well as the *PASA* (Gaab et al., 2005) utilize self-reports to measure *SR* and *PA*. Self-reported data typically bears the risk of potentially being biased (Donaldson & Grant-Vallone, 2002); the most prominent bias in this context would be social desirability: On the one hand, being able to manage stress successfully is a highly desirable trait in today's society. This might have influenced the self-reported *SR* and *PA* values: Reporting to be marginally stress reactive (i.e., scoring low in *SR*) and to seldomly consider situations to be stressful (i.e., scoring low in *PA*) reflect a high capability of managing stress successfully – meaning that participants might have reported a lower *SR* and *PA* score to fit this desirable trait. These ratings then would not reflect the actual *SR* and *PA* levels in these participants. On the other hand, leading a stressful life can be seen as an indicator of leading a successful professional life. This might have influenced the *SR* and *PA* ratings in the other direction.

This self-report bias caused by social desirability (Donaldson & Grant-Vallone, 2002), as well as the small sample size might explain the non-significant findings, even though a positive correlation and even a positive predictive effect of *SR* on *PA* is theoretically implied.

Regarding the predictive effects of *SR* on *HR* and *SubjStress*, none of the models derived from the hypotheses reached statistical significance. This could on

the one hand be due to the way the *PSRS* operationalizes *SR*: *SR* is defined as an individual's perceived typical response intensity to stressful situations in everyday life. Contrasting to that operationalization stands the *TSST*, being a highly standardized stress task in a laboratory setting. Even though it utilizes a jobinterview in an effort to mimic an everyday situation, it still is very unlikely for individuals to have encountered a comparable situation unprepared and under similar circumstances – being, that their job-interview performance is video- and audio-taped while having to stand in front of a selection committee dressed in white lab-coats. Because of these striking dissimilarities, participants might have rated their typical stress response intensity in respect to social-evaluative and failure-related stressful situations that were greatly different from the *TSST* situation. This might have led to perceived *SR* ratings that do not relate to situations similar to the *TSST*, which in turn would explain the lack of predictive value of *SR* ratings in examined sample.

Furthermore, both outcome stress measures assessed only reflect the stress response of exposure to a single stressor whereas stress reactivity assesses retrospective reports of aggregated stress responses, reflecting a personality trait. It has already been shown that correlations of singular stress responses and personality traits were rather small in comparison to the correlation of aggregated stress responses over repeated stress exposures and personality traits (Pruessner et al., 1997). Following Pruessner's (1997) findings, a significant relationship between the *PSRS* subscales and the outcome measures can be expected if participants are exposed to multiple *TSST* or other standardized stress tasks. Implementing this reasoning into future research might lead to more definite results.

The transactional stress theory (Lazarus & Folkman, 1984) suggests that if a situation is appraised as threatening and challenging a stress reaction comprising physiological as well as psychological aspects (in this case an increase in *HR* and *SubjStress*) sets in, implying a predictive effect of *PA* on the outcome measures. In current data set, none of the proposed predictions of scales related to *PA* on the outcome measures could be observed. The lack of predictive effects may once again be due to measurement errors in *PA* caused by social desirability. Furthermore, observed individual differences in *HR* and *SubjStress* might not only be attributable to individual differences in *PA* (and *SR*), but also to the *TSST* procedure itself: Even

though the *TSST* is a highly standardized procedure, and the committee members were all well-trained before experimental assessments took place, the *TSST* procedure leaves room for variability. As the committee did not always comprise the same two members of the study team, interactional nuances between the participant and the committee members of the respective testing date could not be fully controlled (Allen et al., 2017), which may have led to some degree of variability in the outcome measures. In a study by Buchanan et al. (2012), it could be shown that the committee members may show an empathic stress reaction towards the participants in a *TSST* procedure: In some instances, the committee members may contagiously "catch" the stress reaction of the participant and subconsciously provide mild social support, which modulates neurophysiological and psychological stress responses (Frisch et al., 2015). This poses a threat to the internal validity of the *TSST*s conducted in current project and might be an explanation for the lack of statistically significant predictive effects.

Also, the mediation model of the effect of *SR* on *HR* and *SubjStress* by *PA* did not reach significance. Even though bootstrapping methods have been argued to be a powerful tool to test the significance of mediation models in the field of psychology especially with small samples (Koopman et al., 2015), minimum sample size recommendations still comprise 20-80 cases (Preacher & Hayes, 2004; Shrout & Bolger, 2002). Due to recurring lockdowns starting from March 16<sup>th</sup> 2020 up until the point of data analysis in April 2021, even the minimum recommendation of 20 participants could not be reached. Additionally, as the *TSST* and other procedures related to the MuSkiBa-study are relatively labor-intensive processes requiring the cooperation of four study team members, the maximum number of participants tested per week was also restricted due to limited availabilities of the testing team. The exceptionally small sample size resulting from these restrictions poses the most impacting limitation of current project. As the main part of that limitation is due to the Covid-19 pandemic and is thus incontrollable, no recommendation can be made to improve this limitation in future studies similar to current project.

Another limitation of current project is the method of assessment of *SR*. In current project, *SR* was assessed by utilizing the *PSRS* (Schlotz et al., 2011a), a questionnaire asking for self-reported ratings of aggregated typical stress reactions

to day-to-day stressors. As already mentioned above, this assessment method may be confounded by self-report biases, such as social desirability, recall biases and insufficient introspective abilities (Donaldson & Grant-Vallone, 2002), as well as frequency of exposure to daily life stress (Schlotz, 2013). These confounds may have led to inaccurate assessments of SR yielding inconclusive results. An alternate, more accurate method to assess SR may be the utilization of ecological momentary assessment methods (EMA) (Shiffman et al, 2008): Ecological momentary assessment involves repeated measurements of a participants' current behaviors and experiences in real time in their natural environment. By doing so, EMA methods yiel high external validity when compared to global retrospective self-reports, such as the PSRS (Schlotz et al., 2011a). EMA methods are furthermore able to assess all response systems relevant to SR by utilizing technologies ranging from self-reports assessing momentary levels of perceived stress, over real-time saliva sampling, especially during or after stressor exposure, to physiological sensors measuring HR. The data gathered using EMA methods can then be aggregated by highly trained professionals, thus minimizing recall bias. This procedure would yield a comprehensive, accurate measure of SR. As EMA procedures are very costly and labor-intensive and require a high level of compliance of the participants, applying these methods in current project was not feasible and the more economical, but possibly less accurate self-report measure of SR was chosen instead. A direct comparison of SR as measured by the PSRS (Schlotz et al., 2011a) on the one hand and SR as measured by EMA methods may be a project of interest for further psychobiological stress research interested in the shaping effects of SR on the stress response. Furthermore, inserting these two different assessments of SR into the proposed mediation model using a larger sample size and comparing the predictive effects of both SR assessments might be another possible direction for further research.

In conclusion, the findings of current project did not provide results in line with the hypotheses: Apart from the predictive effects of *ReacFail* on *threat*, none of the other proposed predictive models of *SR* on *PA* reached statistical significance. Furthermore, neither *SR* nor *PA* predicted *HR* and *SubjStress*; a mediating effect of *PA* on the effect of *SR* on the outcome variables also could not be found. This lack of significant findings is primarily due to the exceptionally small sample size. Also, as

only self-report measures were used as independent variables, self-report biases, such as social desirability and insufficient introspective abilities of the participants (Donaldson & Grant-Vallone, 2002), could have led to the lack of significant findings. These possible confounds caused by self-report biases may also have led to the findings contradicting the expectations derived from the hypotheses in the extreme case analysis.

For further research, the possible confound caused by self-report biases could be accounted for by implying EMA methods (Shiffman et al., 2008) to measure *SR*. Furthermore, higher correlations between *SR* and the outcome measures could be expected when participants are exposed to multiple *TSST* and/or other standardized stress tasks. Lasty, as the limited sample size was mainly due to restrictions caused by the Covid-19 pandemic, the author wishes the MuSkiBa research team great success in conducting face-to-face experiments in these trying times in order to increase the volume of analyzable data.

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