Freezing as Flight Preparation: Evidence from Electrodermal, Cardiovascular and Gaze Recordings

Lara Rösler* & Matthias Gamer

Department of Psychology, Julius Maximilians University of Würzburg, Würzburg, Germany

*Corresponding author:

lara.roesler@uni-wuerzburg.de

Abstract

When confronted with threatening stimuli, animals typically respond with freezing behavior characterized by reduced movement and heart rate deceleration. Freezing-like responses during threat anticipation have also been observed in humans and are associated with anxiety. Recent evidence yet suggests that freezing does not necessarily reflect helpless immobility but can also aid the preparation of a threat escape. To investigate whether preparatory freezing can also be discerned in gaze behavior, we presented participants with aversive stimuli that sometimes could be avoided while measuring gaze, cardiovascular and electrodermal activity. In trials in which the threat could be escaped, participants displayed reduced heart rate, increased electrodermal activity and reduced visual exploration. Furthermore, heart rate deceleration and restricted visual exploration predicted the speed of flight responses. These results provide first evidence for freezing behavior in measures of visual exploration and suggest that such responding is adaptive in preparing the subsequent escape of approaching threats.

Keywords: freezing, flight, bradycardia, eye-tracking, threat, immobility

Introduction

Freezing is commonly observed in rodents as a reaction to relatively remote threat (R. J. Blanchard, Blanchard, Rodgers, & Weiss, 1990; Fanselow, 1994; Moscarello & LeDoux, 2013). While it is characterized by heart rate deceleration and reduced body motion, freezing does not always indicate defenseless immobility but can actively aid the animal in the preparation of an escape (D. C. Blanchard, Griebel, Pobbe, & Blanchard, 2011; Misslin, 2003). Freezing as compared to non-freezing rats show a heightened startle response, suggesting that freezing actively supports the detection of threat (Plappert, Pilz, & Schnitzler, 1993) and thus improves sympathetic fight-or-flight responses (Lang, Bradley, & Cuthbert, 1997). It therefore needs to be distinguished from tonic immobility which rats exhibit as a last resort to an unavoidable threat (Kozłowska, Walker, McLean, & Carrive, 2015). In humans, tonic immobility has been observed as an immediate response to traumatizing events and has therefore been suggested to be involved in the emergence of post-traumatic stress disorder (PTSD) (Bovin, Jager-Hyman, Gold, Marx, & Sloan, 2008; Rocha-Rego et al., 2009). Early-life stress is also linked to increased freezing-like behavior in adolescents strengthening the association between freezing and anxious attachment styles (e.g. Niermann et al., 2015; Roelofs, Hagenaars, & Stins, 2010). This finding is corroborated by various studies that show a relationship between reduced body sway and a history of anxiety or trauma in humans. However, different experimental designs have also demonstrated the benefits of postural freezing in humans as it potentially facilitates a subsequent threat avoidance (Azevedo et al., 2005; Gladwin, Hashemi, van Ast, & Roelofs, 2016; Stins & Beek, 2007). Supporting the notion of preparatory freezing mechanisms, decreases in heart rate have been linked to better discrimination of low spatial frequency information (Lojowska, Gladwin, Hermans, & Roelofs, 2015). The improved acuity of low spatial frequency vision occurs at the cost of high frequency recognition and might therefore facilitate a fast but sometimes unnecessary escape (Lojowska et al., 2015). With regard to the accumulating evidence of a role of freezing that surpasses mere immobility, it has recently been suggested that freezing represents a necessary mechanism for action preparation (Roelofs, 2017).

Research on the influence of threat on measures of visual exploration have primarily focused on the attentional capture of threatening stimuli. Accordingly, the use of dot-probe paradigms revealed shorter reaction times to threatening or painful stimuli (e.g. Khatibi, Dehghani, Sharpe, Asmundson, & Pouremad, 2009) and difficulties disengaging from them (Belopolsky, Devue, & Theeuwes, 2011; Mogg, Bradley, De Bono, & Painter, 1997). A meta-analysis on attentional biases towards pain confirmed that healthy participants and chronic pain patients show heightened attention towards impending pain signals but this bias was affected by the type of pain information and its timing (Crombez, Van Ryckeghem, Eccleston, & Van Damme, 2013). In line with the idea that pain captures attention, painful stimulation was recently seen to trigger attentional shifts toward the stimulation site (Schmidt, Gamer,

Forkmann, & Bingel, 2018). However, as participants were not able to escape the painful shock it is impossible to draw inferences about attentional processes during flight and freezing as a means to flight preparation.

The aim of the current study was therefore to investigate whether gaze patterns differ between trials in which a painful stimulus will certainly be administered and trials in which this threat can be actively avoided. We presented simplistic natural images to participants while the color of the preceding fixation cross indicated whether an unavoidable, an avoidable or no shock would be administered upon image disappearance. Throughout the entire experiment, we measured gaze, skin conductance and heart rate to investigate whether previously reported autonomic indicators of freezing would be accompanied by changes in visual exploration patterns. In line with the freezing for action hypothesis, we expected that flight trials would be accompanied by heart rate deceleration and a more focused visual exploration pattern characterized by a stronger central bias with fewer and longer fixations.

Methods

Participants

We invited 59 participants (11 males, age: $M = 23.79$ years, $SD = 5.32$ years) via a university-sponsored website targeted at participant recruitment to take part in our experiment. The study was conducted in line with the Declaration of Helsinki (version 2008) and approved by the local Ethics committee. Each participant provided written informed consent prior to the experiment and received extra course credit or monetary compensation. All participants had normal or corrected-to-normal vision and reported no history of pain-related diseases. Because of problems with the experimentation sequences, 3 participants had to be excluded. Another 3 participants were excluded because of a large proportion of invalid eye-tracking trials ($> 30\%$) and 3 participants were excluded because they did not follow task instructions on the day of testing. The final sample thus consisted of 50 participants (see Table 1), who were characterized regarding general anxiety, anxiety sensitivity and depression using established self-report questionnaires (see experimental procedure for further details). With the current sample size, medium to large effects (as reported in Löw, Weymar, & Hamm, 2015) in repeated measures analyses of variance (ANOVAs, $f = 0.25$) could be detected with a power of .95 at a significance level of $\alpha = .05$ when assuming a correlation of $r = .50$ between factor levels in the ANOVA (Faul, Erdfelder, Lang, & Buchner, 2007; see Supplemental Material). The experiment was not formally preregistered but de-identified data and analysis scripts can be found on the Open Science Framework (<https://osf.io/n7vua/9>).

Table 1. Demographics of participants included in the final sample ($n = 50$).

	<i>M</i>	<i>SD</i>
Age (years)	23.37	5.16
Sex (male/female)	(10/40)	
Handedness (right/left/both)	(45/3/2)	
STAI	34.88	8.22
ASI	33.74	11.67
BDI	4.81	5.00

Note. STAI = State-Trait Anxiety Inventory, trait version, ASI = Anxiety Sensitivity Index, BDI = Beck-Depression-Inventory.

Experimental Paradigm and Stimulation

Participants observed naturalistic images on a PC screen while their heart rate, skin conductance and eye movements were recorded. The stimulus set consisted of 60 naturalistic photographs (768×576 pixels, visual angle of $24.00^\circ \times 18.11^\circ$ at the current viewing distance of 50 cm) which were taken from the McGill Calibrated Colour Image Database (Olmos & Kingdom, 2004). As we also examined attentional biases during threat anticipation, we used the same images as Schmidt and colleagues (Schmidt et al., 2018) which were chosen for their uniform appearance (see Figure 1 for an example). The experiment was programmed using Presentation® (Neurobehavioral Systems Inc., Version 18.1.) and presented on a grey background of a 24" Asus VG248QE display (53.136 x 29.889 cm, 1920 x 1080 pixels, refresh rate 60 Hz). Pictures were presented pseudo-randomly, either in original or horizontally flipped orientation to account for effects of basic visual features on attentional orienting.

Visual stimulation consisted of an initially presented white fixation cross (6.5-8.5 s duration), a subsequent colored fixation cross (2 s duration), the naturalistic image (8 s duration) and a final grey screen which was either blank or contained a response prompt in the flight condition (1 s duration, see Figure 1). The color of the second fixation cross indicated whether a painful electrical stimulation would be either certainly (red; shock-condition), possibly (yellow; flight-condition) or certainly not (green; safety-condition) presented after stimulus offset. To simulate a possible escape to the threat, participants could avoid the shock by quickly hitting the space bar with their dominant hand upon stimulus offset in flight trials. During the first five of these flight trials, participants received a shock if they took longer than 240 ms to hit the space bar. In subsequent flight trials, the median reaction time of the first five flight trials was used as an individual threshold for each participant to ensure that flight attempts would sometimes but not always result in shocks (Lów et al., 2015). Each trial type appeared 20 times in pseudo-randomized

order, warranting that no more than three trials of the same type appeared after one another, resulting in a total of 60 trials.

Figure 1. Illustration of the trial structure. Participants accomplished three different trial types (shock, flight and safety) that were announced by a color cue. Note: Size of fixation cross and color cue are not drawn to scale.

Painful electrical stimulation was applied using a Digitimer DS7A constant current stimulator (Hertfordshire, United Kingdom) and a surface electrode attached to the dominant hand of the participant. Shock segments consisted of a train of three 2 ms square-wave pulses (separated by 50 ms) with alternating polarity. Shock intensity was individually adjusted before the experiment. This procedure involved repeatedly delivering electrical shocks starting at 0 mA and increasing in intensity in 0.1 mA increments. Each shock had to be rated verbally using a visual analog scale from 0 to 10 (VAS; 0 = “not painful at all” and 10 = “unbearably painful”). Participants were explicitly told that a VAS level of 4 should describe a clearly unpleasant yet bearable feeling. Whenever a shock was perceived as having an intensity of 4, the amperage was turned down to the minimum and the increasing electrical stimulation was rated anew. After three iterations, the mean amperage of the stimulations perceived as a VAS level 4 was calculated and, subsequently, an additional 50% of the respective amperage was added to determine the final stimulation used as a shock throughout the experiment. Across all participants, we applied an average amperage of 1.00 mA ($SD = 1.14$ mA).

Experimental Procedure

After signing the consent form, participants received task instructions and learned which fixation cross color was associated with which trial type. Subsequently, we attached heart rate and skin conductance electrodes to the chest and the non-dominant hand of participants respectively. The surface electrode for delivering electrical stimulation was attached to the dominant hand. Participants were then seated in a dimly lit sound-proof cabin and instructed to rest their heads on a chin rest to ensure minimal movement during the later eye-tracking recordings. After adjusting the shock intensity, the eye-tracker was calibrated and the calibration was validated using a nine-point grid. The experiment was then started and participants completed all 60 trials. Upon experiment completion, participants filled in several questionnaires assessing anxiety and depression (see Table 1). These consisted of the German versions of the Beck-Depression-Inventory (BDI; Kühner, Bürger, Keller, & Hautzinger, 2007), the trait version of the state-trait anxiety inventory (STAI; Laux, Glanzmann, Schaffner, & Spielberger, 1981) and the anxiety sensitivity index (ASI; Kemper, Ziegler, & Taylor, 2009).

Data recording and analysis

Heart Rate

Heart rate (HR) was measured using a BIOPAC MP160 device (BIOPAC Systems, Inc.) with a sampling rate of 500 Hz. Electrocardiogram (ECG) electrodes were placed on the manubrium sterni and the left lower rib cage. The reference electrode was placed at the right lower rib cage. In order to derive heart rate from the ECG signal, the data were first filtered with a 2 Hz high-pass filter to remove slow signal drifts. R-Waves were then detected from the ECG data using a semiautomatic method and R-R-intervals were converted to HR (in beats per minute). Then, a second-by-second sampling was applied (Velden & Wölk, 1987) for an interval of -1 to 15 s relative to the color cue preceding image onset. The HR in the last second prior to cue onset served as baseline and the mean heart rate change was derived by subtracting the baseline from the HR score of each postcue second.

Electrodermal Activity

Electrodermal activity (EDA) was measured with the same BIOPAC device at the thenar and hypothenar eminences of the participants' non-dominant hand by a constant voltage system (0.5 V) using a bipolar recording with two Hellige Ag/AgCl electrodes filled with 0.05 M NaCl electrolyte. EDA data were downsampled to 20 Hz and averaged in bins of 1 s between -1 and 15 s relative to the color cue preceding image onset. The mean skin conductance in the last second prior to cue onset served as baseline and was subtracted from the values of each postcue second.

Eye-Tracking

We tracked the participants' right eye using a mounted EyeLink 1000 Plus system with a sampling rate of 1000 Hz (SR Research Ltd., Ottawa, Canada). Eye-tracking data were initially processed using EyeLink's standard parser configuration which defined saccades as eye movements exceeding a velocity threshold of 30°/s or an acceleration threshold of 8,000°/s². Time periods with stable gaze between saccades were defined as fixations. To warrant baseline stability in all trials entering the analysis, we considered the last 300 ms before stimulus onset as fixation baseline. We subsequently examined baseline stability for each participant by conducting an iterative outlier removal procedure separately for x- and y-baseline coordinates (for a similar procedure see End & Gamer, 2017). Specifically, the smallest and the largest values were temporarily removed from the distribution and if one or both of these values were more than three standard deviations away from the mean baseline position of the remaining data, they were permanently excluded from the analysis. This procedure was repeated until no further exclusion had to be performed. Fixation x and y coordinates were then corrected for gaze drift by subtracting the baseline from the actual x and y coordinate values. Trials with an invalid baseline (6.9% of all trials) were excluded from all subsequent analyses of physiological responses and visual exploration behavior.

We looked at three different metrics to evaluate the dynamics of visual exploration during threat anticipation. To account for and investigate potential changes in gaze throughout the trial, we separated fixations in eight bins of 1s each as a first step. We then calculated average fixation durations, number of fixations and average distance of all fixations from center (in pixels) within these bins to investigate whether freezing behavior could be observed in eye movements by proxy of fewer and longer fixations which occurred closer to the center.

Statistical analysis

All of these three metrics were statistically evaluated using a repeated measures analyses of variance (ANOVA). These analyses included the within-subject factors trial-type (flight, shock or safety) and bin (15 levels for HR and EDA and 8 levels for measures of visual exploration). Degrees of freedom were adjusted according to the Greenhouse-Geisser method to compensate for potential violations of the sphericity assumption. Effect sizes are reported as generalized η^2 (Bakeman, 2005). Post-hoc tests were carried out to specifically compare the responses in flight trials to the other two conditions. Therefore, we calculated t-tests for each time bin and adjusted the *p*-values for multiple comparisons using the false discovery rate (FDR, Benjamini & Hochberg, 1995).

In order to exploratively examine the influence of physiological and eye-tracking indices of freezing on behavioral responses, we calculated a generalized linear mixed model (GLMM) on response times in

flight trials using the lme4 package (version 1.1-15; Bates, Mächler, Bolker, & Walker, 2014) for R. Trials where participants did not respond within 1s were excluded from the analysis (3.7%). Based on the results of the repeated measures ANOVAs (see below) as well as previous research using a comparable experimental paradigm (Löw et al., 2015), we concentrated on the responses in the second half of the stimulus presentation and therefore averaged time bins 5 to 8 for skin conductance, heart rate, center bias, fixation duration and fixation number and included these variables as predictors in the GLMM. Participant ID was included as random intercept. Model estimates were chosen to optimize the restricted maximum likelihood (REML) criterion and p -values for each predictor were obtained based on Satterthwaite's approximation of degrees of freedom using the lmerTest package (version 2.0-25; Kuznetsova, Brockhoff, & Christensen, 2017). All data preprocessing and analyses were carried out in R (Version 3.3.3, Core Team, 2018) on an α level of .05.

Results

Physiological Responses

The repeated-measures ANOVA using time bin and trial type (shock vs. flight vs. safety) on skin conductance changes yielded a main effect of bin ($F_{(14,686)} = 40.46$, $\epsilon = .12$, $p < .001$, $\eta^2 = .162$), a main effect of trial type ($F_{(2,98)} = 34.02$, $\epsilon = .71$, $p < .001$, $\eta^2 = .159$) and a significant interaction between bin and trial type ($F_{(28,1372)} = 38.72$, $\epsilon = .08$, $p < .001$, $\eta^2 = .215$). Similarly, the ANOVA on heart rate changes revealed a significant main effect of bin ($F_{(14,686)} = 22.69$, $\epsilon = .28$, $p < .001$, $\eta^2 = .095$) and a significant interaction between bin and trial type ($F_{(28,1372)} = 14.06$, $\epsilon = .22$, $p < .001$, $\eta^2 = .070$). There was, however, no statistically significant main effect of trial type ($F_{(2,98)} = 1.61$, $\epsilon = .91$, $p = .205$, $\eta^2 = .011$) suggesting that trial types do not per se elicit different cardiovascular responses but differ in the progress of these responses over time. As can be seen in Figure 2, flight trials were associated with a marked increase in skin conductance that started shortly after picture onset and was maintained until (potential) shock delivery. By contrast, a pronounced heart rate deceleration in flight trials only occurred during the second half of the picture viewing period and turned into an acceleration after picture offset.

Visual exploration

Measures of visual exploration were analyzed in a similar fashion as the physiological data but since these measures can only be meaningfully interpreted during the picture viewing period, these analyses were restricted to eight time bins. The repeated measures ANOVA on the average distance of fixations from the screen's center yielded a significant main effect of bin ($F_{(7,343)} = 39.04$, $\epsilon = .39$, $p < .001$, $\eta^2 = .098$), a significant main effect of trial type ($F_{(2,98)} = 82.06$, $\epsilon = .77$, $p < .001$, $\eta^2 = .143$) and a significant interaction of bin and trial type ($F_{(14,686)} = 12.90$, $\epsilon = .58$, $p < .001$, $\eta^2 = .021$). Similarly, we found a

significant main effect of bin ($F_{(7,343)} = 37.60$, $\varepsilon = .33$, $p < .001$, $\eta^2 = .137$), a significant main effect of trial type ($F_{(2,98)} = 6.04$, $\varepsilon = .74$, $p = .003$, $\eta^2 = .016$) and a significant main interaction of bin and trial type ($F_{(14,686)} = 10.15$, $\varepsilon = .45$, $p < .001$, $\eta^2 = .028$) for fixation durations. Also for the number of fixations we detected a significant main effect of bin ($F_{(7,343)} = 96.40$, $\varepsilon = .35$, $p < .001$, $\eta^2 = .205$), a significant main effect of trial type ($F_{(2,98)} = 9.25$, $\varepsilon = .74$, $p < .001$, $\eta^2 = .014$) and a significant main interaction of bin and trial type ($F_{(14,686)} = 10.28$, $\varepsilon = .53$, $p < .001$, $\eta^2 = .019$). Figure 2 indicates a stronger center bias in flight trials throughout the whole viewing period. By contrast, fixation durations and numbers in flight trials only differed from the respective values in shock and safety trials at the end of the picture viewing period. The observed increase in fixation durations and the related decrease in fixation numbers indicate a general reduction in visual exploration in anticipation of the flight response (for a further illustration of this reduced visual exploration see fixation density maps in Figure S1 of the supplemental material).

Prediction of flight behavior

Participants' mean response time in flight trials was 298.45 ms ($SD = 71.44$ ms). The GLMM revealed heart rate responses as well as center bias to be significantly related to response times in flight trials (Table 2). In detail, stronger heart rate decelerations as well as an increased center bias (i.e., a reduced distance of fixations to the screen's center) predicted faster response. The other predictors were not significantly related to response times in a combined model, which might be partly due to intercorrelations between eye-tracking measures (see Table S1 in supplementary material).

Table 2. Parameters of a generalized linear mixed model predicting response times in flight trials.

Predictor	β	SE	t	df	p
(Intercept)	265.958	57.210	4.65	660.2	< .001
Skin conductance	-0.410	1.160	-0.35	652.5	.724
Heart rate	0.500	0.208	2.40	656.6	.017
Center bias	0.084	0.022	3.83	615.1	< .001
Fixation duration	-0.005	0.016	-0.34	662.5	.736
Fixation number	1.059	3.002	0.35	663.6	.724

Note. p -values for each predictor were obtained based on Satterthwaite's approximation of degrees of freedom.

Figure 2. Mean changes in skin conductance level (A) and heart rate (B) relative to the last second prior to cue onset and average distance from center (C), fixation durations (D) and number of fixations (E) during picture presentation. Shaded areas denote standard errors of the mean. Time points with significant differences between the flight condition and the other two conditions (after false discovery rate correction) are displayed at the top of each figure as colored lines.

Discussion

The current study investigated autonomic and gaze responses to a painful shock that was either unavoidable or could be escaped via a button press. We observed fewer and longer fixations, which were also closer to the center of the screen, in trials in which a threat escape was possible. This freezing response at the level of visual exploration was accompanied by a stark heart rate deceleration and an overall increase of electrodermal activity. Additionally, reductions in heart rate and visual exploration of the stimulus predicted the speed of the escape response. Our results therefore provide evidence for freezing behavior in humans which seems to play an important role in action preparation.

In trials in which an escape of the threat was possible, participants exhibited clearly distinct gaze patterns which consisted of a reduced average distance to the center of the screen and fewer but longer fixations. These freezing-like eye movements that were demonstrated for the first time in the current study possibly aid a faster escape of the subsequent threat. Indeed, a GLMM analysis revealed that heart rate deceleration and reduced visual exploration of the presented stimuli were significant predictors of flight reaction times. Reduced scanning of the visual field and bradycardia might contribute to a sharpened perception of the environment which, in turn, enables a faster escape. Further support for a link between freezing measures and improved perception comes from a study which linked decreases in heart rate to better discrimination of low spatial frequency stimuli (Lojowska et al., 2015), potentially enabling crude but faster flight responses to threatening stimuli. Similarly, the expectation of pain at a specific location heightened multisensory processing as reflected by faster target detection at the pain-associated site (Vanden Bulcke, Crombez, Durnez, & Van Damme, 2015; Vanden Bulcke, Van Damme, Durnez, & Crombez, 2013). Again, reduced scanning of the environment might have served as a preparatory mechanism enabling a subsequent harm prevention.

The opportunity to actively escape the painful stimulation also led to clearly recognizable changes in the physiological reactions of our participants. Previously, Löw and colleagues had shown that attentive freezing, as indicated by fear bradycardia, exclusively took place when the threat could not be avoided (Löw et al., 2015). In contrast, we observed the most prominent heart rate deceleration when the painful stimulation following picture offset could be escaped via a button press. Our finding is supported by a study of Gladwin and colleagues which showed that fear bradycardia was strongly related to the preparatory state but only when a virtual gunshot resulting in painful electrical stimulation could be avoided (Gladwin et al., 2016). Which factors can explain these differential findings regarding the role of attentive freezing? The most striking difference between our experimental design and the study of Löw and colleagues is that our anticipatory state did not contain changing, dynamic stimuli but instead consisted of an 8 second viewing period of a static image. To observe changes with regard to threat

proximity, Löw and colleagues used stimuli that increased in size to inform participants about threat imminence. One possible interpretation is that the dynamically approaching stimuli led to differential autonomic responses potentially by precisely informing participants about shock proximity. A closer examination of Löw and colleagues' data yet shows that the active avoidance of threat was indeed associated with heart rate deceleration but not significantly more than the active button press in safe trials. This, in turn, suggests that the anticipation of a required button press, independent of whether a shock will follow or not, involves heart rate deceleration as a preparation for action. In the current study, we decided not to include a safe condition requiring fast responses since adaptive flight behavior is associated with the presence of threat. We can therefore not compare the anticipation of a button press in safe and unsafe trials. Overall, however, the strong fear bradycardia observed in flight trials which also predicted the speed of flight responses supports the action preparation function of freezing.

At first glance the notion of freezing as an adaptive preparatory flight mechanism is at odds with various animal and human studies which previously pointed to a relationship between anxiety and increased freezing behavior. Accordingly, rodents with high trait anxiety displayed more freezing behavior than their less anxious peers (Caldji et al., 1998; Frank et al., 2006). Similarly, humans who reported higher state anxiety responded with more pronounced reductions in heart rate and body sway to angry versus neutral or happy faces (Roelofs et al., 2010) and reduced postural sway when presented with a neutral target (Wada, Sunaga, & Nagai, 2001). This increased freezing response is not limited to humans with comparatively high state anxiety but extends to patients with a history of panic disorder (Lopes et al., 2009) and aversive life events (Hagenaars, Stins, & Roelofs, 2011). None of these experimental designs, however, allowed participants to escape the threatening situation. When we think of these freezing responses in terms of tonic immobility – a resigned acceptance of the approaching unavoidable threat – it becomes clear that these associations might point to an entirely different phenomenon. While highly anxious individuals might exhibit increased freezing as a sign of helplessness or resignation, freezing for action preparation, as measured in our study, might not be at all related to anxiety. Indeed, Gladwin and colleagues who also investigated human freezing responses when an escape of the threat was possible also failed to observe significant correlations between anxiety and freezing behavior (Gladwin et al., 2016). The present results therefore emphasize the necessity to distinguish between different types of freezing responses: 1) tonic immobility which is linked to anxiety and 2) freezing as a preparatory flight mechanism which is an adaptive response to a threat encounter.

The current investigation of cardiovascular, electrodermal and gaze responses to the anticipation of an electric shock that was either avoidable or unavoidable revealed that participants display increased freezing behavior when given the chance to escape the threat. This heightened freezing response was reflected in a stark heart rate deceleration, a simultaneous increase of electrodermal activity and reduced

visual exploration of the presented stimuli. In addition to primarily demonstrating freezing-like behavior in measures of visual exploration, the observed correlation between heart rate deceleration and reduced scanning of the presented stimuli with the speed of the flight response, furthermore suggests that freezing is an adaptive mechanism which serves as preparation for a quick escape of the imminent threat.

Author Contributions

M. Gamer developed the study concept. All authors contributed to the study design. Testing and data collection was performed by L. Rösler. L. Rösler performed the data analysis and interpretation under the supervision of M. Gamer.. L.Rösler drafted the manuscript, and M. Gamer provided critical revisions. All authors approved the final version of the manuscript for submission.

Acknowledgments

The authors would like to thank Julia Jochim and Tim Jelken for their help with participant recruitment and data acquisition.

Bibliography

- Azevedo, T. M., Volchan, E., Imbiriba, L. A., Rodrigues, E. C., Oliveira, J. M., Oliveira, L. F., ... Vargas, C. D. (2005). A freezing-like posture to pictures of mutilation. *Psychophysiology*, 42(3), 255–260. <http://doi.org/10.1111/j.1469-8986.2005.00287.x>
- Bakeman, R. (2005). Recommended effect size statistics for repeated measures designs. *Behavior Research Methods*, 37(3), 379–384. <http://doi.org/10.3758/BF03192707>
- Bates, D., Mächler, M., Bolker, B., & Walker, S. (2014). Fitting Linear Mixed-Effects Models using lme4. <http://doi.org/10.18637/jss.v067.i01>
- Belopolsky, A. V., Devue, C., & Theeuwes, J. (2011). Angry faces hold the eyes. *Visual Cognition*, 19(1), 27–36. <http://doi.org/10.1080/13506285.2010.536186>
- Benjamini, Y., & Hochberg, Y. (1995). Controlling the False Discovery Rate: A Practical and Powerful Approach to Multiple Testing. *Journal of the Royal Statistical Society: Series B*, 57(1), 289–300.
- Blanchard, D. C., Griebel, G., Pobbe, R., & Blanchard, R. J. (2011). Risk assessment as an evolved threat detection and analysis process. *Neuroscience & Biobehavioral Reviews*, 35(4), 991–998. <http://doi.org/10.1016/J.NEUBIOREV.2010.10.016>
- Blanchard, R. J., Blanchard, D. C., Rodgers, J., & Weiss, S. M. (1990). The characterization and modelling of antipredator defensive behavior. *Neuroscience & Biobehavioral Reviews*, 14(4), 463–

398 472. [http://doi.org/10.1016/S0149-7634\(05\)80069-7](http://doi.org/10.1016/S0149-7634(05)80069-7)

399 Bovin, M. J., Jager-Hyman, S., Gold, S. D., Marx, B. P., & Sloan, D. M. (2008). Tonic immobility
400 mediates the influence of peritraumatic fear and perceived inescapability on posttraumatic stress
401 symptom severity among sexual assault survivors. *Journal of Traumatic Stress*, 21(4), 402–409.
402 <http://doi.org/10.1002/jts.20354>

403 Caldji, C., Tannenbaum, B., Sharma, S., Francis, D., Plotsky, P. M., & Meaney, M. J. (1998). Maternal
404 care during infancy regulates the development of neural systems mediating the expression of
405 fearfulness in the rat. *Proceedings of the National Academy of Sciences*, 95(9), 5335–5340.

406 Crombez, G., Van Ryckeghem, D. M. L., Eccleston, C., & Van Damme, S. (2013). Attentional bias to
407 pain-related information: A meta-analysis. *PAIN*, 154(4), 497–510.
408 <http://doi.org/10.1016/J.PAIN.2012.11.013>

409 End, A., & Gamer, M. (2017). Preferential Processing of Social Features and Their Interplay with Physical
410 Saliency in Complex Naturalistic Scenes. *Frontiers in Psychology*, 8, 418.
411 <http://doi.org/10.3389/fpsyg.2017.00418>

412 Fanselow, M. S. (1994). Neural organization of the defensive behavior system responsible for fear.
413 *Psychonomic Bulletin & Review*, 1(4), 429–438. <http://doi.org/10.3758/BF03210947>

414 Faul, F., Erdfelder, E., Lang, A.-G., & Buchner, A. (2007). G*Power 3: A flexible statistical power
415 analysis program for the social, behavioral, and biomedical sciences. *Behavior Research Methods*,
416 39(2), 175–191. <http://doi.org/10.3758/BF03193146>

417 Frank, E., Salchner, P., Aldag, J. M., Salomé, N., Singewald, N., Landgraf, R., & Wigger, A. (2006).
418 Genetic predisposition to anxiety-related behavior determines coping style, neuroendocrine
419 responses, and neuronal activation during social defeat. *Behavioral Neuroscience*, 120(1), 60–71.
420 <http://doi.org/10.1037/0735-7044.120.1.60>

421 Gladwin, T. E., Hashemi, M. M., van Ast, V., & Roelofs, K. (2016). Ready and waiting: Freezing as
422 active action preparation under threat. *Neuroscience Letters*, 619, 182–188.
423 <http://doi.org/10.1016/J.NEULET.2016.03.027>

424 Hagenaars, M. A., Stins, J. F., & Roelofs, K. (2011). Aversive Life Events Enhance Human Freezing
425 Responses. *Journal of Experimental Psychology: General*, 141(1), 98.
426 <http://doi.org/10.1037/a0024211>

427 Kemper, C. J., Ziegler, M., & Taylor, S. (2009). Überprüfung der psychometrischen Qualität der

- 428 deutschen Version des Angstsensitivitätsindex-3. *Diagnostica*, 55(4), 223–233.
 429 <http://doi.org/10.1026/0012-1924.55.4.223>
- 430 Khatibi, A., Dehghani, M., Sharpe, L., Asmundson, G. J. G., & Pouretmad, H. (2009). Selective attention
 431 towards painful faces among chronic pain patients: Evidence from a modified version of the dot-
 432 probe. *Pain*, 142(1–2), 42–47. <http://doi.org/10.1016/J.PAIN.2008.11.020>
- 433 Kozłowska, K., Walker, P., McLean, L., & Carrive, P. (2015). Fear and the Defense Cascade: Clinical
 434 Implications and Management. *Harvard Review of Psychiatry*, 23(4), 263–87.
 435 <http://doi.org/10.1097/HRP.0000000000000065>
- 436 Kühner, C., Bürger, C., Keller, F., & Hautzinger, M. (2007). Reliabilität und Validität des revidierten
 437 Beck-Depressionsinventars (BDI-II). *Der Nervenarzt*, 78(6), 651–656.
 438 <http://doi.org/10.1007/s00115-006-2098-7>
- 439 Kuznetsova, A., Brockhoff, P. B., & Christensen, R. H. B. (2017). lmerTest Package: Tests in Linear
 440 Mixed Effects Models. *Journal of Statistical Software*, 82(13).
- 441 Lang, P. J., Bradley, M. M., & Cuthbert, B. N. (1997). Motivated Attention: Affect, Activation, and
 442 Action. In *Attention and Orienting: Sensory and Motivational Processes* (97th ed., p. 135).
- 443 Laux, L., Glanzmann, P., Schaffner, P., & Spielberger, C. D. (1981). Das State-Trait-Anxiety-Inventory
 444 (STAI). *Hogrefe, Göttingen*.
- 445 Lojowska, M., Gladwin, T. E., Hermans, E. J., & Roelofs, K. (2015). Freezing Promotes Perception of
 446 Coarse Visual Features. *Journal of Experimental Psychology: General*, 144(6), 1080.
 447 <http://doi.org/10.1037/xge0000117.supp>
- 448 Lopes, F. L., Azevedo, T. M., Imbiriba, L. A., Freire, R. C., Valença, A. M., Caldirola, D., ... Nardi, A. E.
 449 (2009). Freezing reaction in panic disorder patients associated with anticipatory anxiety. *Depression*
 450 *and Anxiety*, 26(10), 917–921. <http://doi.org/10.1002/da.20593>
- 451 Löw, A., Weymar, M., & Hamm, A. O. (2015). When Threat Is Near, Get Out of Here. *Psychological*
 452 *Science*, 26(11), 1706–1716. <http://doi.org/10.1177/0956797615597332>
- 453 Misslin, R. (2003). The defense system of fear: behavior and neurocircuitry. *Neurophysiologie*
 454 *Clinique/Clinical Neurophysiology*, 33(2), 55–66. [http://doi.org/10.1016/S0987-7053\(03\)00009-1](http://doi.org/10.1016/S0987-7053(03)00009-1)
- 455 Mogg, K., Bradley, B. P., De Bono, J. O., & Painter, M. (1997). Time Course of Attentional Bias for
 456 Threat Information in Non-Clinical Anxiety. *Behaviour research and therapy*, 35(4), 297–303.
- 457 Moscarello, J. M., & LeDoux, J. E. (2013). Active Avoidance Learning Requires Prefrontal Suppression

of Amygdala-Mediated Defensive Reactions. *Journal of Neuroscience*, 33(9), 3815–3823.
<http://doi.org/10.1523/jneurosci.2596-12.2013>

Niermann, H. C. M., Ly, V., Smeekens, S., Figner, B., Riksen-Walraven, J. M., & Roelofs, K. (2015). Infant attachment predicts bodily freezing in adolescence: evidence from a prospective longitudinal study. *Frontiers in Behavioral Neuroscience*, 9, 263. <http://doi.org/10.3389/fnbeh.2015.00263>

Plappert, C. F., Pilz, P. K. D., & Schnitzler, H.-U. (1993). Acoustic startle response and habituation in freezing and nonfreezing rats. *Behavioral Neuroscience*, 107(6), 981–987.
<http://doi.org/10.1037/0735-7044.107.6.981>

Rocha-Rego, V., Fiszman, A., Portugal, L. C., Garcia Pereira, M., de Oliveira, L., Mendlowicz, M. V., ... Volchan, E. (2009). Is tonic immobility the core sign among conventional peritraumatic signs and symptoms listed for PTSD? *Journal of Affective Disorders*, 115(1–2), 269–273.
<http://doi.org/10.1016/J.JAD.2008.09.005>

Roelofs, K. (2017). Freeze for action: Neurobiological mechanisms in animal and human freezing. *Philosophical Transactions of the Royal Society B: Biological Sciences*, 372(1718).
<http://doi.org/10.1098/rstb.2016.0206>

Roelofs, K., Hagenaaars, M. A., & Stins, J. (2010). Facing freeze: Social threat induces bodily freeze in humans. *Psychological Science*, 21(11), 1575–1581. <http://doi.org/10.1177/0956797610384746>

Schmidt, K., Gamer, M., Forkmann, K., & Bingel, U. (2018). Pain Affects Visual Orientation: an Eye-Tracking Study. *The Journal of Pain*, 19(2), 135–145. <http://doi.org/10.1016/J.JPAIN.2017.09.005>

Stins, J. F., & Beek, P. J. (2007). Effects of affective picture viewing on postural control. *BMC Neuroscience*, 8(1), 83. <http://doi.org/10.1186/1471-2202-8-83>

Vanden Bulcke, C., Crombez, G., Durnez, W., & Van Damme, S. (2015). Is attentional prioritization on a location where pain is expected modality-specific or multisensory? *Consciousness and Cognition*, 36, 246–255. <http://doi.org/10.1016/J.CONCOG.2015.07.003>

Vanden Bulcke, C., Van Damme, S., Durnez, W., & Crombez, G. (2013). The anticipation of pain at a specific location of the body prioritizes tactile stimuli at that location. *PAIN®*, 154(8), 1464–1468.
<http://doi.org/10.1016/J.PAIN.2013.05.009>

Velden, M., & Wölk, C. (1987). Depicting cardiac activity over real time: A proposal for standardization. *Journal of Psychophysiology*.

Wada, M., Sunaga, N., & Nagai, M. (2001). Anxiety affects the postural sway of the antero-posterior axis

488 in college students. *Neuroscience Letters*, 302(2–3), 157–159. <http://doi.org/10.1016/S0304->
489 3940(01)01662-7
490