

## The continuum of consciousness in cardiovascular stress research : an experimental expedition

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# Chapter 8

## **General Discussion**

The aim of this thesis was to examine the association of stress-related cognition outside of awareness with cardiovascular (CV) activity. As elaborated on in the Introduction, studies on the relationship between psychological stress and CV disease have not been able to fully explain how this negative psychological state may result in a prolonged adverse physiological state (8,9, 22-25). One possible explanation is the occurrence of stress-related cognitions outside of awareness, that is, unconscious stress, which may activate and even prolong physiological stress-responses (26-28). This thesis represents a first attempt to examine the role of processes outside of awareness in CV responses to stressors through a systematic review and a series of experiments, described in the previous six chapters. In this last Chapter these findings are first briefly summarized and then discussed in terms of relevant theoretical and methodological issues. The reader will also find a description of the strengths and limitations of the studies in this thesis, and the possible future of the unconscious stress hypothesis is portrayed.

#### **Main findings**

Unconscious processes and their relationship with physiological activity have not been described in the context of psychological stress. However, a vast amount of studies have been performed using methods to present stimuli outside of awareness of the participants while measuring peripheral physiological activity (e.g., 26,27). In order to attain accumulation of knowledge on this topic, a systematic review was performed (Chapter 2). This review included 65 experimental studies in which negative affective stimuli versus control stimuli were presented outside of awareness. In other words, the stimuli that were presented did not require deliberate processing. During this procedure peripheral physiological responses (i.e., CV, electrodermal, electromyographical, hormonal, and immunological parameters) were recorded. Mainly two methods have been used to reduce the chances that participants were aware of the presented stimuli: subliminal priming and subliminal presentation of fear conditioned stimuli. From this literature a lack of agreement on various methodological aspects was apparent as well as a large variety of outcome measures. Nevertheless, the findings seem to indicate that negative affective stimuli in subliminal priming studies increase systolic blood pressure (SBP) relative to the control stimuli. In the fear conditioning studies, fear conditioned stimuli that were very briefly presented increased skin conductance response (SCR) amplitude. Insufficient data were available for other physiological outcomes. Notably, none were performed with hormonal or immunological outcomes. Taken together, based on this overview I cannot conclude with certainty that unconscious stress, operationalized as negative affective stimuli presented outside of awareness, negatively influences health-relevant parameters, such as blood pressure (BP) and heart rate variability (HRV; 95). Not only do the findings call for more studies using these parameters, it also stresses the need for

more consensus in the field regarding adequate subliminal presentation, awareness checks, consistency in data reporting and interpretation, and execution of replication studies.

The three subsequent chapters described studies in which the unconscious stress hypothesis was tested using both methods to manipulate awareness of stimuli mentioned above: subliminal priming and subliminal presentation of fear conditioned stimuli.

In **Chapter 3**, subliminal priming was used to test the unconscious stress hypothesis. Healthy participants were presented either threatening or neutral words (the primes), while performing a categorization task. Although mean arterial pressure (MAP) and total peripheral resistance (TPR) were higher and heart rate variability (HRV) was lower in the threat condition as expected, statistical significance was only obtained for TPR. Interestingly, TPR is believed to be associated with perceived threat (99,100). In addition to this main finding, moderating effects of trait worry and baseline HRV, which are common predictors of adverse health outcomes, were not related to the CV reactivity to the different primes. Furthermore, using the Implicit Positive and Negative Affect Test (IPANAT, 84) as implicit measure of affect after the priming task, a small and statistically marginal significant positive relationship was found between implicit positive affect (IPA) and mean arterial pressure (MAP), but overall implicit affect was not related to CV reactivity to the primes. Thus, unconscious stress, operationalized as threat words presented outside of awareness, can elicit an increase in TPR, which can be interpreted as a physiological state reflecting perceived threat. However, the priming effects appear to have been too weak to sort effects on other affective and CV parameters.

One of the problems in the field is the lack of replication studies to validate previous findings (see Chapter 2). Therefore, **Chapter 4** describes a replication of studies of Hull et al. (2002, 62) and (partially) of Garfinkel et al. (2016, 61), that showed that repetitive subliminal presentation of the prime word 'angry', increased blood pressure (BP) and heart rate (HR) in comparison with the prime 'relax'. In the study in Chapter 4 none of these differential changes in BP or HR were replicated. Furthermore, in contrast to expectations, higher implicit negative affect (INA) was related to lower SBP and diastolic blood pressure (DBP) during the priming task. In line with the expectations, lower IPA was related to a higher TPR during priming as well as during the recovery phase. Thus, the primes did not seem to affect the physiological state, but affect measured with an implicit measure did relate to CV activity. This indicates that although the subliminal presentation of primes may not have been successful, implicit measures may provide additional information in the relationship between psychological stress and CV activity.

In **Chapter 5**, fear conditioning was used to create a stressor that was equal across participants. The stressor was then presented subliminally. Neutral stimuli

were paired with a shock (CS+) set at an intensity that was barely tolerable for the participants. Other neutral stimuli were not paired with the shock (CS-). The CS+ and CS- were presented above and below the threshold of awareness. In this study, skin conductance was also included as a physiological measure since Chapter 2 indicated that this is a widely used measure in the field, and would therefore provide a good comparison with previous studies. SCR magnitude was larger in response to the CS+, compared to the CS-, also when CS+ stimuli were presented subliminally. However, no such effects were found for the CV variables. Thus, only partial evidence for the unconscious stress hypothesis was obtained, which may indicate that fear conditioned stimuli outside of awareness may affect the physiological state, but not necessarily in a way that is detrimental to health.

A different approach to unconscious processes was to assess stress-related processes with so called *implicit measures* and relate these to CV activity during and after a laboratory stressor. In **Chapter 6**, psychological stress was induced using a mathematical task with and without anger harassment. The IPANAT was used as an implicit measure to capture implicit positive and negative affect, while a self-report measure of affect was used to capture explicit or 'conscious' assessment. Both were administered during the recovery phase. The addition of harassment to the mathematical task did not yield additional CV reactivity or recovery. Importantly, higher INA was related to higher SBP, and lower TPR and HRV during the task and to slower recovery of DBP, while lower IPA was related to slower recovery of SBP and DBP. Thus, the relationship of the IPANAT subscales with CV recovery after a stressor at least seems to indicate that CV activity might partially be explained by implicit affective processes, as predicted by the unconscious stress hypothesis.

In **Chapter 7**, an anger recall procedure was used to induce psychological stress, as a different laboratory stressor, which was compared to 'happy recall' as a control condition. This time, an affective Lexical Decision-making Task (LDT; 89) was used as implicit measure of unconscious stress, indicated by cognitive activation of information and expressed as automatic vigilance (81). Again, no differences in CV reactivity or recovery were found between the conditions, but a higher automatic vigilance index for negative information (AVI-N) was related to a higher DBP during the recovery and a lower automatic vigilance index for positive information (AVI-P) was related to slower recovery of SBP, HR, and TPR. Importantly, self-reported affect was not related to CV reactivity or recovery.

An overview of the main findings in this thesis is presented in Table 1. In short, we could not replicate the increased SBP found by the systematic review in Chapter 2 using subliminal presentation of negative affective stimuli versus control stimuli (Chapters 3, 4, & 5). However, we did find some support with respect to the TPR effects found in Chapter 2 using general threat words versus neutral words (Chapter 3). Furthermore, higher INA was related to decreased SBP and DBP in response to the

| Method  | Unconscious stress operationalization   | Main findings   |   |   |  |
|---|---|---|---|---|--|
|   |   | CV activity   |   | Other outcomes  |  |
| Systematic<br>review<br>(Chapter 2)                   | Negative affective<br>stimuli versus<br>nonnegative affective<br>stimuli presented<br>subliminally              | $\begin{array}{l} SBP (12) \uparrow \\ DBP (10) \leftrightarrow \\ HR (18) \leftrightarrow \\ PEP (8) \leftrightarrow \\ HRV (6) \leftrightarrow \end{array}$                                     | $\begin{array}{l} CO(1) \uparrow \\ VC(1) \leftrightarrow \\ TPR(1) \uparrow \\ RR(1) \leftrightarrow \end{array}$                      | SCR amplitude (27) $\uparrow$<br>SCR magnitude (9) $\leftrightarrow$<br>SCR rise time (1) $\uparrow$<br>SCR frequency (2) $\leftrightarrow$<br>SCR latency (1) $\leftrightarrow$<br>GSR (4) $\uparrow$<br>SCL (4) $\uparrow$  | Corrugator supercilii<br>(20) ↔<br>Orbicularis oculi muscles<br>(7) ↑<br>Zygomatic major (14) ↔<br>General EMG activity<br>(2) ↔ |
| Subliminal<br>priming<br>(Chapter 3)                  | Threat-related<br>versus neutral words<br>presented subliminally  | $\begin{array}{l} MAP \leftrightarrow \\ RMSSD \leftrightarrow \\ TPR \uparrow \end{array}$   |   | $\uparrow$ INA: MAP ↔, RMSSD ↔, TPR ↔<br>$\uparrow$ IPA: MAP ↔, RMSSD ↔, TPR ↔  |  |
| Subliminal<br>priming<br>(Chapter 4)                  | 'Angry' [woedend]<br>versus 'relax' [rustig]<br>presented repeatedly<br>and subliminally                        | $SBP \leftrightarrow$ $DBP \leftrightarrow$ $HR \leftrightarrow$  |   | ↓ INA: SBP $\uparrow$ , DBP $\uparrow$ , HR $\leftrightarrow$ , TPR $\leftrightarrow$<br>↓ IPA: SBP $\leftrightarrow$ , DBP $\leftrightarrow$ , HR $\leftrightarrow$ , TPR $\uparrow$   |  |
| Fear<br>conditioning<br>(Chapter 5)                   | Fear conditioned<br>images (CS+) versus<br>neutral images (CS-)<br>presented subliminally<br>and supraliminally | $SBP \leftrightarrow$ $DBP \leftrightarrow$ $HR \leftrightarrow$  |   | SC magnitude ↑  |  |
| Mathematics<br>and anger<br>harassment<br>(Chapter 6) | IPANAT as implicit<br>measure   | $\begin{array}{l} \textit{Reactivity:} \\ \textit{SBP} \uparrow \\ \textit{DBP} \uparrow \\ \textit{HR} \leftrightarrow \\ \textit{RMSSD} \leftrightarrow \\ \textit{TPR} \downarrow \end{array}$ | Recovery:<br>SBP $\leftrightarrow$<br>DBP $\leftrightarrow$<br>HR $\leftrightarrow$<br>RMSSD $\leftrightarrow$<br>TPR $\leftrightarrow$ | Reactivity:   ↑ INA: SBP ↑, DBP ↔, HR ↔, RMSSD ↓, TPR ↓   ↑ IPA: SBP ↔, DBP ↔, HR ↔, RMSSD ↔, TPR ↔ <i>Recovery</i> :   ↑ INA: SBP ↔, DBP ↑, HR ↔, RMSSD ↔, TPR ↔   ↓ IPA: SBP ↑, DBP ↑, HR ↔, RMSSD ↔, TPR ↔   |  |
| Emotional<br>recall<br>(Chapter 7)                    | LDT as implicit<br>measure after an<br>anger or happy recall<br>procedure                                       | Reactivity:SBP $\uparrow$ DBP $\uparrow$ HR $\leftrightarrow$ RMSSD $\leftrightarrow$ TPR $\downarrow$  | Recovery:<br>SBP $\leftrightarrow$<br>DBP $\leftrightarrow$<br>HR $\leftrightarrow$<br>RMSSD $\leftrightarrow$<br>TPR $\leftrightarrow$ | $\begin{array}{l} \textit{Reactivity:} \\ \uparrow \ \text{AVI-N: SBP} \leftrightarrow, \text{DBP} \uparrow, \text{HR} \leftrightarrow, \text{RMSSD} \leftrightarrow, \text{TPR} \leftrightarrow \\ \uparrow \ \text{AVI-P: SBP} \leftrightarrow, \text{DBP} \leftrightarrow, \text{HR} \leftrightarrow, \text{RMSSD} \leftrightarrow, \text{TPR} \leftrightarrow \\ \textit{Recovery:} \\ \uparrow \ \text{AVI-N: SBP} \leftrightarrow, \text{DBP} \leftrightarrow, \text{HR} \leftrightarrow, \text{RMSSD} \leftrightarrow, \text{TPR} \leftrightarrow \\ \uparrow \ \text{AVI-P: SBP} \leftrightarrow, \text{DBP} \downarrow, \text{HR} \downarrow, \text{RMSSD} \leftrightarrow, \text{TPR} \uparrow \end{array}$ |  |

#### TABLE 1 Summary of the main findings presented in this thesis

**Note.**  $\uparrow$  = Increase,  $\downarrow$  = Decrease,  $\leftrightarrow$  = No change. For Chapter 2 the number of studies on which the conclusion is based is indicated between brackets. Regarding recovery, higher values indicate slower recovery (Chapter 6) and lower values indicate faster recovery (Chapter 7). *Abbreviations*: CV = Cardiovascular, SBP = Systolic blood pressure, DBP = Diastolic blood pressure, HR = Heart rate, PEP = Pre-ejection period, HRV = Heart rate variability, CO = Cardiac output, VC = Ventricular contractions, TPR = Total peripheral resistance, RR = Respiration rate, SCR = Skin conductance response, GSR = Galvanic skin response, SCL = Skin conductance level, EMG = Electromyographical, MAP = Mean arterial pressure, RMSSD = Root mean squared successive differences, INA = Implicit negative affect, IPA = Implicit positive affect, CS = Conditional stimulus, IPANAT = Implicit Positive and Negative Affect Test, LDT = Lexical Decision-making Task, AVI-N = Automatic vigilance index for negative words, AVI-P = Automatic vigilance index for positive words

word 'angry' versus 'relax' (Chapter 4). In two studies with explicit stress-inducing situations, high INA was related to higher SBP and lower HRV and TPR during the stressor, but during recovery from the stressor low IPA was related to slower recovery of SBP and DBP (Chapter 6). Furthermore, high AVI-N was related to increased DBP during the stressor, but high AVI-P was related to faster recovery of DBP and TPR.\

#### Implications for the unconscious stress hypothesis

The findings provide tentative support for the unconscious stress hypothesis. Across studies, we found that subliminally presented stress-related stimuli increased TPR, but not SBP, DBP, and HR, and we found no decrease of HRV. Additionally, in the fear conditioning study in particular we found increased SC magnitude to the stress-related stimuli above and below the threshold of awareness. Furthermore, the implicit measures of psychological stress were related to CV activity during a stressor (Chapter 6), albeit sometimes in the opposite direction of what was expected, and after (Chapters 6 and 7) a stressor, where the explicit measure was not related to the CV activity. The unconscious stress hypothesis states that stress-related cognition may occur outside of awareness by which it negatively affects the physiological state, in addition to what is within the realm of awareness (26,27). Based on the findings, it seems evident that what is outside of awareness may influence the physiological state, but not on all health-relevant parameters, and not for all stressful stimuli and stressors used. Consequently, the unconscious stress hypothesis is only partially supported.

The crucial issue in addressing unconscious stress has been its operationalization. In line with the definition of psychological stress (16), we focused on the activation of affective representations of a stressor. In all studies we intended to activate or measure these representations and expected concurrent physiological responses. However, the different operationalizations of unconscious stress in the studies were inconsistently related to physiological changes. An explanation may be that we erroneously assumed that the various stressful stimuli and situations used would all have physiological effects. Possibly, the stimuli used were either too general or too mild. In contrast, previous studies that used specific stimuli related to the specific samples (e.g., negative stereotypes of ageing in elderly people; 63, social ties with real friends/acquaintances; 199) found clear CV effects.

The anger primes of Chapter 4 and the fear conditioned stimuli in Chapter 5 may have been too mild. As we also argue in the respective chapter, perhaps the word 'angry', at least the Dutch translation 'woedend', may not be sufficiently intense to influence CV activity. This may also hold for the fear conditioned stimuli: although the shock was perceived as uncomfortable, participants indicated that the intensity could have been higher in hindsight, which has been suggested to be a frequent methodological issue (269). In addition to the comparison of our findings with the replication of Garfinkel et al. (2016, 61) and Hull et al. (2002, 62) other factors mentioned in the systematic review in Chapter 2, such as the type of stimuli, may be of influence as well. The research group of Gendolla (e.g., 132, 144, 163) has repeatedly found effects of subliminally presenting depictions of facial expressions on CV activity. However, they measured CV activity during a (demanding) math task and focused on the effects of task difficulty in relation to the primes. Thus, perhaps it is another factor, for example adding additional cognitive demands, that intensifies the CV response patterns to subliminal stimuli. Still, presenting fearful faces subliminally has been shown to elicit amygdala activation associated with detection of salient stimuli (72). One could then argue that subliminal processing of emotional expressions is different (i.e., has stronger effects) from that of other images or words, as used in the current dissertation. Nevertheless, another study that reported changes in CV activity used religious images (172). The conclusion then seems inevitable that this matter needs further and more systematic research. All in all, it is still possible that in real life, outside the laboratory, stress-related information above or below threshold of awareness elicits much stronger affective responses, and that this information has considerable physiological effects. It might be difficult to model these effects in the laboratory, except when the stress-related stimuli used are individualized and meaningful.

Additionally, we assumed that affective representations of the stressor outside of awareness would be induced by subliminal presentation and measured indirectly with the implicit measures. However, although affective changes processes outside of awareness may exist, but we can never be certain that a change in affectivity occurred nor that induction and assessment occurs without conscious processing by the participants, not even when changes on the IPANAT and LDT were observed (144,194,195). Thus, the partial support provided here is fuel for more questions regarding unconscious processes in this line of research. The idea that psychological stress may also be unconscious is therefore not dismissed in this thesis, rather it shows that the existence of the phenomenon is probable and deserves further exploration. In other words, based on the current findings but also the literature, I think it is fair to state that we have only scratched the surface on the continuum of consciousness in cardiovascular stress research.

#### Theoretical considerations beyond unconscious stress

The expedition undertaken in this thesis ran into various alternative options and point of views that I feel should be explicated in this discussion if one is to build upon the knowledge gathered. First of all, from the systematic review (Chapter 2) it became apparent that there are various theoretical viewpoints on unconscious processes and physiological activation that influence study designs and conclusions, despite using similar methodologies. From these different viewpoints two are relevant in terms of the current findings. The "preparedness theory" (186,268) states that fear conditioning of subliminal findings can only be successful when the stimuli relate to evolution-based aversive images, such as snakes. However, I have argued that the fear conditioning itself, that is, combining a neutral image with an aversive stimulus, creates a stimulus that instigates threat in the individual, which then generates a physiological response in Chapter 5. Although we did not find an effect of the fear conditioning procedure on the CV variables, skin conductance measures did show this effect (Chapter 5), both supraliminally and subliminally. Thus, the preparedness theory did not hold in the current set-up.

Furthermore, it has been suggested that implicit affect can facilitate coping with challenges in the so-called implicit-affect-primes-effort model (IAPE model; e.g., 331-333). In other words, subliminal priming of the affective state can influence whether a task is considered easy (i.e., in case of activated happiness and anger) or difficult (i.e., in case of activated sadness and fear). The former case (i.e., easy) would be characterized by mainly higher cardiac responses (PEP and HR) to a difficult task, while the latter (i.e., difficult) would be characterized by more vascular responses (TPR). In the studies where we presented subliminal stimuli, we used the word 'angry' (which would induce perceived task easiness and dominantly PEP and HR), threat-related words (which would induce perceived difficulty and dominantly TPR), and fear conditioned images (also perceived difficulty). Notably, we used a simple categorization task (Chapters 3 and 4) and no task (Chapter 5) rather than a highly demanding cognitive task that is commonly used in studies testing the IAPE model. However, we found no effects on HR, but PEP was not used as an outcome measure. Moreover, only vascular activity (TPR) appears to have been affected, and only with threatening stimuli. So, regardless of the specific valence of the stimuli, there was no evidence for an effect of subliminal primes on effort-related cardiac responses.

Another related theoretical paradigm, that was not addressed in the systematic review, is the idea of the existence of a core affect, which is a neuropsychobiological state described in terms of the dimension pleasure-displeasure and that of activation-deactivation (310). It challenges the traditional 'basic emotions' view (e.g., 334) on emotions that suggests that an event leads to an emotion, which then triggers feelings, behavioral, and physiological responses (for a review see 335). In core affect, physiological responses are part of the concept itself that prepare the individual for action. In contrast, self-reported 'feelings' are considered to be an affective quality assigned to a specific situation or object, which does not necessarily relate to the core affect. The hypothesis of core affect seems theoretically related to the unconscious stress hypothesis, which also describes a nonspecific change in (negative) affectivity (26,27). Importantly, the findings from the current thesis confirm that processes beyond self-report are related to physiological changes, but also that the affective processes are likely to operate on a two-dimensional plane of pleasure/displeasure and activation/deactivation (310) rather than in specific categories of emotion, such

as fear or anger, referring to the findings regarding positive affectivity. Therefore, the role of core affect might be an interesting line of research in stress and health, encompassing emotion-theory and processes beyond awareness.

#### **Contributions to stress research**

Apart from offering partial support for the notion that stress-related physiological activity is to some extent caused by unconscious stress, that is, the unconscious stress hypothesis, this thesis potentially contributes to stress research in several additional ways.

First, and perhaps somewhat surprising, is the finding that TPR, and not the other CV variables, appeared to be most sensitive to unconscious stress. Specifically, although the SBP, DBP, and HR may not have been affected by subliminal priming and subliminally presented fear conditioned images, TPR was changed. It increased in response to subliminally presented threat-related words (as expected; Chapter 3). Furthermore, during a mental challenge and emotional task it decreased (not as expected; Chapters 6 and 7). Additionally, a higher TPR was found to be related to lower IPA during and after a priming task (as expected; Chapter 4) and to lower INA during a difficult mental task (not as expected; Chapter 6) as measured with the IPANAT, but also to a higher AVI-P after emotional recall as measured with the LDT (not as expected; Chapter 7). A vascular response pattern to psychological stress, as contrasted with a cardiac response pattern, may be particularly detrimental for health as it has been related to hypertension and hypertrophy (336-338) which may have severe consequences for health (97,98,339). Furthermore, TPR has been found to increase in response to worrying (340). Moreover, TPR has been related to psychological stress and is believed to be specifically related to threat appraisal (99,100,172,341), that is, when a situation is appraised as threatening, TPR increases. Additionally, the Cognitive Activation Theory of Stress (CATS; 16) describes that only after a stimulus is appraised as threatening, it is followed by a nonspecific stress response and cognitive processing. In other words, the observed changes in TPR in response to the stressrelated stimuli are in line with the psychobiosocial model of Blascovich (99) and our definition of psychological stress that is based on the CATS. Moreover, the findings regarding TPR may be a key element in the relationship explaining the relationship between psychological stress and adverse CV health outcomes. Based on the current findings, it could be argued that TPR is indicative of psychological stress, regardless of the level of awareness at which the stressor is processed. Thus, in light of the current findings, it appears that TPR may be a sensitive outcome measure that responds to stress inductions even outside of awareness.

Another implication of the findings is the evident additional explanatory value of the implicit measures in stress research. When implicit measures were taken into account, they were related to the physiological changes, over and above the self-report measures of psychological stress. Moreover, self-reported affectivity was not related to the CV activity. These findings not only call for the application, further development, and testing of additional *implicit* measures, but in general may also explain why the causal relationship between CV health and psychological stress remains unresolved. This supports the unconscious stress hypothesis and stipulates the additional value of these measures in explaining the relationship between psychological processes and health.

Finally, two unintended findings in this thesis challenge the use of some standard laboratory stressors without proper control manipulations. We found that anger harassment in addition to a neutral mathematical task and that anger recall versus happy recall did not result in differentiating CV changes. In stress research, there are several methods to induce stress that are widely used and accepted within the field, including anger harassment and anger recall procedures (92). However, these tasks are mostly used in a battery of multiple tasks and lack adequate control groups. Most studies merely compare healthy versus non-healthy participants in CV reactivity on these tasks (92,342). In contrast, in the current thesis we aimed to use control groups in our designs to ensure that the findings would be attributable to the negative affective component rather than other cognitive processes such as calculating or emotional recall. Surprisingly, the mental arithmetic and anger harassment elicited similar CV responses, as did the anger and happiness recall. This seems to call into question the role of angry affect as such, to elicit these physiological effects. Practically, these findings imply that certain widely used stress manipulations to induce CV responses in the literature have not been sufficiently controlled for. In other words, this is a setback in our understanding of the mechanisms underlying psychological stress and CV activity, at least in healthy individuals, which should encourage researchers to formulate statements on this relationship in non-healthy samples with conceptual caution.

#### **Strengths and limitations**

A strength of the current thesis is the overarching approach undertaken to address the unconscious stress hypothesis and to expand knowledge on the relationship between processes outside of awareness with negative affective associations and peripheral physiological responses. I have used several experimental designs that applied to the theoretical notions described in Chapter 1. The use of these various methodologies based on commonly executed psychological experiments, provides a new overview of the important issues regarding the different methods and findings in light of their relevance to the unconscious stress hypothesis. The combination of experiments provides a fruitful base for further research on this topic.

A second strength of this thesis is the careful localization and summary of earlier similar studies and their results. The new method of finding and systematically testing

all possible keywords in a single, changing keyword profile, enabled us, and will enable others, to point out much more precisely the state of the art, or 'what we know' and what we 'do not yet know'. It also indicates that building upon literature to which one is familiar is not sufficient and a much more thorough search following the proposed method is needed to find all relevant literature (Chapter 2). Moreover, these studies were evaluated on the quality of the reported information in terms of risks for bias. This is a common requirement in for example the medical sciences exemplified by the Risk of Bias Tool by the Cochrane collaboration (343), but such evaluations of quality have not been previously applied to psychological experimental research. The quality ratings in Chapter 2 disclosed the lack of replication studies and limitations in reporting and peer-review. This unnecessarily frustrates the progress of science as it could be easily addressed by for example pre-registration and open access publication of datasets. In line with our own recommendations, but limited by financial resources, we have published one chapter open access (Chapter 6) and the data from the respective papers of all the chapters have been made accessible online. By providing a systematic method for literature search, a tool to quantify the quality of psychological experiments, and making the data of this thesis available online (see section 'Publications'), I have strived to contribute to the academic landscape beyond the scope of this thesis.

A third strength of this thesis is that we aimed to conceptually replicate two studies (61,62) of which the outcomes provided substantial support for the unconscious stress hypothesis (Chapter 4), namely an increased CV response to the word 'angry' versus the word 'relax'. However, we were not able to obtain the same results. Failures to replicate may be attributed to differences between the current and the original study (e.g., 258), but effects of subliminal priming are rarely replicated (60), which seems to be applicable in this study. Moreover, this nonreplication once again shows the importance of verifying the results of previous studies before drawing firm conclusions based on the data. Having stated this, I would encourage replication of the findings in this thesis and the conclusions as I have stated them since they should also be sufficiently verified (or falsified).

Despite these strengths, the work also has several limitations that should be addressed. First, the thesis does not contain a systematic review and/or meta-analysis on the relationship between implicit measures and peripheral physiological responses. Therefore, although we have tried to be sufficiently comprehensive in the respective chapters, we cannot state with certainty that all available information has been used to determine the study designs and weigh the findings regarding the role of implicit measures in stress research. A second limitation is the lack of a baseline measurement of the implicit and self-report measures in Chapters 6 and 7, which implies that for these state measures we could not sufficiently check for pre-existing values or trait aspects of the measured constructs. However, we chose this design to prevent carry-over effects of the measurement onto the manipulation, CV activity, and the measurement of interest afterwards. Finally, we have attempted in the studies with subliminal presentations to ensure that participants were unaware of the stimuli and used awareness checks to verify the success of these attempts (121,122,256). However, neither subjective or objective measures of awareness will be fully able to capture what the participants actually saw (194-196).

With respect to the implicit measures, there are several more options to consider (e.g., 74,81). One option that we have considered was the Morphing Faces Task (MFT, 327), as described in Chapter 7, which we thought would represent a tendency to more readily detect faces displaying (negative) affectivity after a stressor. However, due to high correlations amongst the different expressions, the test could not be considered reliable and could not be used further to address the research question. Another task, the Approach Avoidance Task (344-347), is particularly interesting. In this task, participants have to avoid or approach a stimulus on the screen based on an irrelevant feature (e.g., color) with a joystick which lead to a decrease or increase in size of the stimulus. The stimuli are usually images with a relevant content of two categories, such as angry versus happy faces (e.g., 348), insects versus noninsects (349), or alcoholic beverages versus soft drinks (350). Although the AAT seems a promising implicit measure, in the application to stress research it has some disadvantages. Similar to the IAT, and as discussed in Chapter 1, it can only be used for two categories which is very limiting considering the nonspecific nature of psychological stress (CATS; 16,74). Furthermore, when looking at the outcome measure, it indicates a tendency to either avoid or to approach. However, in a stressful situation one may be inclined to do both, that is, one may want to approach the stressor and confront it, or one may want to avoid the stressor (e.g., 351,352), and both may be associated with an unconscious stress experience. This would complicate interpretation of the findings in this context. All in all, we have not further pursued the ideas around the AAT.

#### **Future directions**

Based on the current findings and relating literature, we see several future directions of studies on unconscious processes in stress research in addition to those already described in the previous chapters. There are several other methods that we have not applied, but that I think should be considered.

First, an idea that is not new, but simply has not been addressed here, is the role of personality traits or other more or less stable characteristics that relate to the inability to (verbally) address one's mental state, such as alexithymia or levels of emotional awareness (28). People with alexithymia may to some degree be less equipped to identify, express, and regulating their emotions, but also an increased self-reporting of negative affectivity (353-355) which has been related to all-cause mortality (356), and CV disease (357-359). This relationship has been explained by

autonomic dysregulation or adverse coping behavior originating from the characteristics of alexithymia (360,361). Furthermore, emotional awareness can be described in several levels that indicate the degree to which one is able to recognize and describe one's own emotions and that of others (362). Lower levels of emotional awareness have been related to for example hypertension (28). These constructs are two examples of explanations why some develop somatic diseases based on dysregulation of emotion processing. Other examples of this restricted reporting of emotions, which should not be confused with the absence of experiencing emotions (e.g., 353), are Type D personality, neuroticism, and defensiveness (as described in 28). Future studies could include these characteristics to relate to stress reactivity and recovery and development of (CV) disease over time in addition to self-report and implicit measures of mental stress.

Second, in this thesis we have mainly focused on response-based measures of psychological stress, self-report and implicit, to explain CV activity. The unconscious stress hypothesis is partially based on the absence of sufficient explanatory value of the self-report measures (25-27). However, recording other physiological or behavioral modalities that inherently do not require any inquiry from participants may further clarify CV responses to psychological stress. One can think of the use of pupil dilation, which has been shown to provide information about mental load (363), or eye movements, which have been found useful in distinguishing fearful from nonfearful subjects (364). Another example is the use of EMG measurements as described in Chapter 2. Facial muscle activity has been studied as a form of nonverbal behavior and emotional expression or experience (365,366), which could provide an additional source of information regarding the affective state and its relationship with health (367). Moreover, several methods of assessing and categorizing facial emotional expressions have been developed (e.g., 368-370) and e-health applications are currently evaluated (e.g., 371).

Third, I believe that the clarification of the relationship between psychological stress and CV health is hindered by a suboptimal use of the collected data in psychosomatic medicine and psychophysiological research. This could be overcome through the implementation of advances in statistical analytical techniques. One can think of using machine learning to predict participants' outcomes (e.g., 372), and quantum mechanical techniques to understand the sensation-perception dynamics (373), but also of the shift away from the interpretation of *p* values (e.g., 374-376). Perhaps further progress in psychological science can be achieved by building upon (and re-analyzing) existing data and taking this knowledge into the 21st century.

#### **Final conclusions**

Taking together all the findings and literature discussed here, I have found tentative verification of the unconscious stress hypothesis. It has also become clear that, from

what has turned out to be a first expedition into the continuum of consciousness in cardiovascular stress research, there is a remaining abundance of options available to assess and influence processes outside of awareness that can be applied in the context of psychological stress and health.

"There is great need to spell out explicitly the assumed characteristics of the unconscious and to search for explanations of so called unconscious phenomena in terms of more commonplace psychological variables. To do so may destroy the titillating mystery that the unconscious seems to hold but then that is the business of science." Eriksen (1960, p.120)