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**ABSTRACT** 46 Negative affective information may be presented outside of awareness and change 47 physiological activity. By increasing peripheral physiological activity, subliminally presented 48 49 negative affective information may contribute to the development of disease. The current systematic review evaluated 65 studies in which negative affective stimuli were presented 50 51 subliminally to a healthy sample while cardiovascular, electrodermal, electromyographical, hormonal, or immunological activity was measured. Overall, 41% of the tested contrasts 52 indicated significant increases due to negative affective stimuli compared to control stimuli. 53 These effects were most pronounced in fear-conditioning studies measuring skin conductance 54 response amplitude and priming studies measuring systolic blood pressure. However, across 55 the included studies the methodology varied substantially and the number of contrasts per 56 physiological parameter was limited. Thus, although some evidence exists that subliminally 57 presented negative affective stimuli can induce adverse peripheral physiological changes, this 58 has not yet been addressed sufficiently. 59 Keywords: implicit processes, awareness, fear-conditioning, priming, cardiovascular 60 activity, electrodermal activity, electromyographical activity, peripheral physiology 61

Peripheral physiological responses to subliminally presented negative affective stimuli:

A systematic review

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Can information that occurs outside of awareness affect perception, motivation, decisions, and emotions? Research addressing this question is flourishing in various fields within psychology, including organizational (e.g., Uhlmann et al., 2012), emotion (e.g., Zajonc, 1980), clinical (e.g., Jones, Vilensky, Vasey, & Fazio, 2013), cognitive (Kihlstrom, 1987), and social psychology (e.g., Bargh & Chartrand, 1999; Fazio, 2001). Surprisingly, the potential role of unconscious processes in the relationship between negative affective information and health has remained understudied. In psychosomatic research, the limits of conscious awareness have long been of interest and explored (Lane, 2008). For example in the 1930s, a psychoanalytic approach was used to address unconscious emotional conflict in the etiology of hypertension (Alexander, 1939), but experimental tests of this particular method failed to provide supportive evidence (Lane, 2008). Notwithstanding, the possible adverse influence of negative affective information outside of awareness on physiological systems is consistent with current theoretical insights (Brosschot, 2010; Brosschot, Verkuil, & Thayer, 2010; Brown, 2004; Damasio, 1994; Lane, 2008). However, experimental evidence is still scarce. Given that several studies indeed showed that unconscious processes influence the experience of emotions (e.g., Dannlowski et al., 2006; Murphy & Zajonc, 1993) and behavior (e.g., Aarts, Custers, & Marien, 2008; Cohen, Moyal, Lichtenstein-Vidne, & Henik, 2016) it seems crucial to examine whether physiological parameters can be affected by negative affective stimuli when these are presented outside of awareness. In fact, the guest for evidence of this kind appears to have a long history. In the early days of psychological research, Jung (1907) and Peterson and Jung (1907) performed several studies regarding the effect of word-associations on galvanic skin responses (GSRs). In these

studies they would repeatedly read out a list of neutral words to participants that had to

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verbalize whatever associated word came to mind. The researchers observed that participants gave different verbal responses to some of the same words and, importantly, that the GSRs were larger than what they had seen before. Notably, this was one of the first psychophysiological experiments and not much was known about the electrodermal response at the time. An in-depth interview with the participants on these words revealed personal affective associations and that the changes in verbal responses had been unintentional. It was concluded that the GSR was able to detect affective associations with neutral words. The different verbal responses and GSRs together were assumed to be a new method to measure an attempt of the mind to prohibit further conscious processing of something that was considered harmful to the self and was referred to as the psycho-physical galvanic reflex. Although the authors faced considerable methodological restrictions using the electrodermal response, it seems that these findings are the first (published) displays of the physiological changes that involuntarily accompany an affective state. Later, McGinnies (1949) was able to display negative affective words below threshold of awareness using a tachistoscope at an interval of 10 ms. He found larger GSRs to the affective words compared to the neutral words, which was interpreted as evidence for *perceptual defense*: a distortion of perception to protect the individual from unpleasant experiences. Moreover, Lazarus and McCleary (1951) provided evidence that after a conditioning procedure individuals were able to discriminate between stimuli of different affective valence before conscious recognition as indicated with changes in GSR, which was referred to as *subception*. Notably, the results of these studies have been largely discussed in light of the repression hypothesis as they were believed to indicate that individuals tend to reject and keep something out of consciousness when it may negatively affect one's wellbeing. These experimental researchers were pioneers and gave way to find ostensibly more objective evidence of physiological effects of subliminal negative affective information. The research instigated fierce criticism from peers, who

performed what we would now call observational studies, and, as a result of the zeitgeist, may have been overlooked in their importance (for a historical discussion the reader is referred to MacKinnon and Dukes, 1962).

More recently, influential evidence of the effects of subliminally presented negative affective stimuli on physiology is offered by neuroscience studies that have found amygdala activation in response to fear-inducing stimuli that were presented below threshold of awareness (e.g., Critchley, Mathias, & Dolan, 2002; LeDoux, 2000; Pessoa, 2005). These findings suggest physiological arousal can be elicited using this type of stimulus presentation and support the earlier findings with GSR that differences in affective valence of stimuli can be determined even when these are presented outside of awareness. However, far less studies seem to have addressed peripheral physiological parameters, such as blood pressure or cortisol. Considering the potential relevance of unconscious processes in psychosomatic research, the aim of the current study was to provide a systematic review of the evidence for the physiological effects of subliminally presented negative affective stimuli from different fields within psychology.

This systematic review focused on studies that manipulated awareness of negative affective stimuli. In experimental designs, awareness is usually manipulated by presenting a stimulus below the threshold of awareness, i.e., subliminally, typically followed (and often preceded) by an irrelevant different stimulus, i.e., mask (e.g., Bargh & Chatrand, 2000; Marcel, 1983; Tamietto & De Gelder, 2010; Wiens & Öhman, 2007). Typically, this subliminal manipulation has been applied to two paradigms: priming with stimuli with an innate affective valence (e.g., Van den Bussche, Van den Noortgate, & Reynvoet, 2009b), from here on referred to as 'priming studies', and priming with fear-conditioned stimuli (e.g., Wiens & Öhman, 2007), from here on referred to as 'fear-conditioning studies'. The mechanism underlying the first paradigm, priming, is believed to be the activation of

cognitive representations of the prime content, which is reflected in a change in a variety of behavioral responses such as reaction times to targets (Fazio, 2001). In addition to behavioral responses, physiological responses have also been found to be influenced by subliminal affective primes (e.g., Hull, Slone, Meteyer, & Matthews, 2002). In fear-conditioning, an association between an unconditioned stimulus (US), such as a shock or a loud noise, that automatically elicits a response (i.e., unconditioned conditioned response, UCS) and a novel stimulus is formed. The result is a conditioned response (CR) to the now conditioned stimulus (CS+). In contrast, the stimuli that are not combined with a US are referred to as CS-. The participant is assumed to learn to differentiate between the CS+ and CS-. Presentation of the CS+ is expected to elicit a physiological response that is similar to presentation of the US alone, as if it was the negative experience itself (e.g., Öhman & Mineka, 2001). The advantage of fear-conditioning over priming is that it offers more control over the specific affective associations with the stimulus.

Theoretically, the subliminal presentation of negative affective stimuli in experimental

Theoretically, the subliminal presentation of negative affective stimuli in experimental paradigms activates unconscious negative affectivity and should result in measurable changes in physiological activity (Brosschot, 2010; Brosschot et al., 2010; Lane, 2008). Since the dysregulation of adaptive peripheral physiological activity is assumed to be the final step in the relation between psychological negative affect and adverse health outcomes (e.g., McEwen, 1998b), we only included studies using peripheral physiological parameters. Most of these parameters are believed to be more directly involved in increased somatic health risks than central nervous system parameters. For example stronger responses of systolic blood pressure (SBP), diastolic blood pressure (DBP), and heart rate variability (HRV) to mental stress were found to be predictive of cardiovascular (CV) disease risk and other health-related outcomes (e.g., Chida & Steptoe, 2010; Malik et al., 1996; Thayer, Yamamoto, & Brosschot, 2010). Furthermore, chronically elevated cortisol increases vulnerability for

disease states, for example through immunosuppression and numerous other pathophysiological effects (McEwen, 1998a). As described, results generally confirm that subliminally presented stimuli affect the brain (e.g., Critchley et al., 2002; LeDoux, 2000; Pessoa, 2005), but this central activity does not necessarily provide information on peripheral activity. Moreover, findings regarding central activity have already been substantially elaborated on elsewhere (e.g., Brooks et al., 2012; Gianaros & Wager, 2015). In contrast, results on peripheral activity have scarcely been addressed and the potential health risks have not been evaluated. Thus, we focused on the peripheral physiological parameters that indicate physiological changes within the organism: CV and electrodermal (EDA) parameters of autonomic activity, musculoskeletal, i.e., electromyographical (EMG), hormonal, and immunological parameters. Additionally, by including only studies that tested a healthy population we attempted to elucidate the more general mechanisms that theoretically precede physical illnesses.

Searching the literature for research on the main concepts of this study, i.e., 'unconscious' is considerably hindered by a lack of consensus on terminology, (see also Brosschot et al., 2010; Eriksen, 1960; Merikle, 1984). To overcome this issue we paid special attention to building a comprehensive keyword profile in an attempt to find all relevant studies. The complex method of building this profile is explained in detail in the method section. Basically, we systematically expanded an initial simple keyword profile with a large set of new keywords. Possible relevant keywords for 'unconscious' were for example alternatives such as 'subconscious' and 'without awareness'. A comprehensive and systematically built topic-specific profile increases the degree of certainty in finding all relevant articles. Moreover, it ensures replicability across databases and researchers while facilitating updates with exactly the same search profile over time.

Furthermore, we addressed two methodological issues regarding subliminal stimulus presentation. First, as pointed out by Eriksen (1960) and Merikle (1984), to obtain valid results regarding the effects of subliminally presented stimuli, a check of awareness of the presented stimuli is required to ensure that the stimuli are indeed not consciously perceived. Moreover, verbal report of awareness is subjective and objective measures of (non)awareness should be used (Merikle, 1984). However, when recognition is reported using an objective measure, it implies that a participant has also consciously perceived (or processed) the stimulus, which is not necessarily true (Merikle, Smilek, & Eastwood, 2001). To overcome this conundrum, we have extracted information on the type of awareness check without ascribing any value to the specific type of check. Second, changes in physiology after subliminal presentation of stimuli may be a consequence of the procedure itself, for example by seeing flashes on the screen or the use of masks that might have been arousing in some way. We addressed this by selecting studies with adequate control stimuli, i.e., stimuli that had no negative affective connotation, that were presented in the same way as the negative affective stimulus, either in between or within-group designs.

Taken together, the primary research question of this systematic review is whether subliminally presented negative affective stimuli increase peripheral physiological activity compared with control stimuli. By providing an overview of studies regarding the role of non-conscious processes and potentially pathophysiologic mechanisms, this systematic review may add significant overarching knowledge about the effect of negative affective information on somatic health.

213 Methods

## **Keyword profile**

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We composed an elaborative keyword profile using BOOLEAN logic to formulate and combine the three sets of keywords pertaining to the three concepts: "unconscious", "negative affect", and "physiology". We started with a basic keyword profile in which the sets were separated by 'AND': (unconscious\* OR subconscious\* OR nonconscious OR nonconscious OR preconscious OR pre-conscious OR sublimin\* OR implicit\*) AND (stress\* OR arousal\* OR (negative and (affect\* OR emot\*)) OR anxi\* OR anger OR angr\* OR fear OR threat\*) AND (cortis\* OR glucocort\* OR adren\* OR noradren\* OR SCL\* OR GSR\* OR blood\* OR blood-pressure OR systol\* OR diastol\* OR cardiac\* OR heart\* OR cardiovasc\* OR immun\*). Subsequently, for each set we aimed to gather an exhaustive list of alternative keywords through the help of a native English speaker, the Thesaurus of PsycINFO, the synonym list of MS Word 2010, and previously found articles. For example in the case of the set "unconscious" we came up with 64 different conceptualizations, such as "nonconscious", "proprioception", and "repressed", see Table 1. Some keywords were written differently across the articles and were thus formulated in all possible ways, for example "mindwandering", "mind-wandering", and "mind wandering". Instead of adding all keywords at once to the basic keyword profile each new keyword was added individually and its additional value was evaluated in terms of the number of new relevant articles found. This was established by searching the databases with a profile containing the new word and the two sets to which the word did not belong, while the set to which the new word did belong was "excluded" by using the NOT function of BOOLEAN logic. For instance in the case of the word "repressed" the evaluative profile would be: repressed AND (set keywords for "stress") AND (set keywords for "physiology") NOT (set keywords for "unconscious" without the new keyword). This profile would yield *only* the articles that the keyword

"repressed" added to the basic profile. When these articles were considered to be relevant, the keyword was added to its set in the basic profile. When the new keyword did not yield relevant articles it was not used anymore. The final profile that was build using this procedure is provided in Table 2.

**Table 1.** Keywords for "unconscious"

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absence of awareness latent inhibition repressed absent-minded less conscious represser access dissociation masked repressing
access dissociation masked repressing
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<b></b>
affective stimuli masked pictures routinized
affective valence masked stimuli stimulus awareness
automatic processing meta-consciousness subconscious
automatic emotional mind-wandering subliminal
aware non verbal suboptimal
awareness nonattended suppressed
conscious awareness nonconscious suppresser
daydreaming oblivious suppressing
degree of awareness outside of awareness train of thought
emotional awareness preattented unaware
first order mental states preattentive unawareness
habitual preconscious unconscious
implicit pre-cognition unknowing
interoceptive awareness precognitive unnoticed unwanted thoughts
intuition primary proces-level unpremeditated
intuitive prime unwitting
involuntary priming without attention
lack of attention proprioception
latent proprioceptive

**Table 2.** Keyword profiles as inserted into the databases

Database	Web of Science	PsycINFO
Search	Core Collection	Basic Search
details	Advanced Search	
Keyword profile	((TS=(unconscious* or subconscious* or nonconscious or non-conscious or preconscious or preconscious or preconscious or preconscious or preconscious or preconscious or sublimin* or implicit* or "automatic emotion" or "automatic affect" or "automatic affective" or unattend* or mindwandering or "emotional awareness" or "interoceptive awareness" or "degree of awareness" or "stimulus awareness" or "conscious awareness" or "involuntary stress" or "latent inhibition" or precogn* or pre-attent* or "automatic processing" or masked* or nonverbal or "non verbal communication") AND TS=(stress* or arousal* or (negative and (affect* or emot*)) or anxi* or anger or angr* or fear or threat* or ruminat* or worr* or "psychological tension" or shock* or "affective stimuli" or "priming" or "prime" or (emotional and (stimuli or circuit* or content* or state* or stimulation or expression))) AND TS= (cortis* or glucocort* or adren* or noradren* or SCL* or GSR* or blood* or bloodpressure or systol* or diastol* or cardiac* or heart* or cardiovasc* or immun* or "physiological arousal" or "physiological measures" or "physiological correlates" or "physiological activity" or "skin conductance" or autonomic* or EMG or (fac* AND (electromyography or muscle*))))) AND LANGUAGE: (English) AND DOCUMENT TYPES: (Article)	(unconscious* or subconscious* or nonconscious or non-conscious or preconscious or preconscious or preconscious or preconscious or sublimin* or implicit* or "automatic emotional" or "automatic emotion" or "automatic affect" or "automatic affective" or unattend* or mind-wandering or "emotional awareness" or "interoceptive awareness" or "degree of awareness" or "stimulus awareness" or "conscious awareness" or "involuntary stress" or "latent inhibition" or precogn* or preattent* or "automatic processing" or masked* or nonverbal or "non verbal communication") AND (stress* or arousal* or (negative and (affect* or emot*)) or anxi* or anger or angr* or fear or threat* or ruminat* or worr* or "psychological tension" or shock* or "affective stimuli" or "priming" or "prime" or (emotional and (stimuli or circuit* or content* or state* or stimulation or expression))) AND (cortis* or glucocort* or adren* or noradren* or SCL* or GSR* or blood* or blood-pressure or systol* or diastol* or cardiac* or heart* or cardiovasc* or immun* or "physiological arousal" or "physiological measures" or "physiological activity" or "skin conductance" or autonomic* or EMG or (fac* AND (electromyography or muscle*)))
	Indexes=SCI-EXPANDED, SSCI	Peer-reviewed

## **Search strategy**

The procedures described by the PRISMA (Preferred reporting Items for Systematic Reviews and Meta-Analyses) Statement (Moher et al., 2009) were applied, to the extent that they apply to experimental research, to the literature search, data collection, and reporting of the results. The final keyword profile was used in Web of Knowledge (Core collection; field: 'topic') and PsycINFO (field: 'all text') on June 16, 2015. In Web of Science the search was limited to 'Article' as document type and 'English' as language. The used indexes were 'SCI-

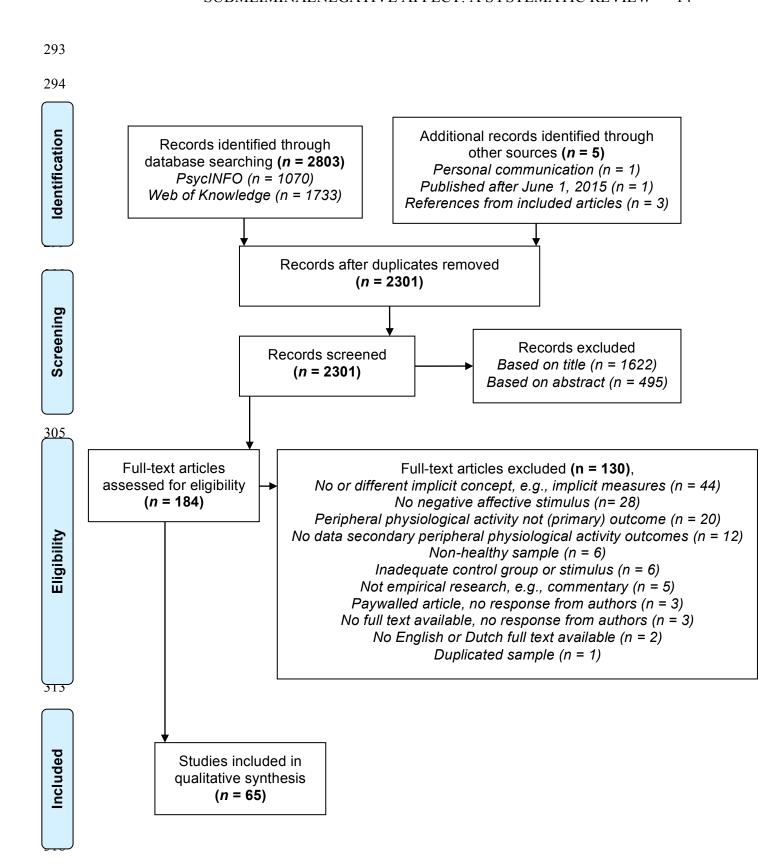
Expanded' and 'SSCI'. No limit to the time span was applied. In PsycINFO the limiters 'peer-reviewed' and 'human subjects' were applied. All duplicate publications were removed. For seven eligible articles the full-text could not be obtained through online methods; in one case we received the full-text version of the article from the authors, in two cases the authors were already deceased, and in the remaining four cases there was no response from the authors. The latter studies were discarded (Esteves, Dimberg, & Öhman, 1994; Esteves, Parra, Dimberg, & Öhman, 1994; Ohira, 1992, 1994). Finally, we checked all references of the final selection of articles, i.e., a snowballing procedure, for articles that might not have been picked up by the keyword-profile. This resulted in ten possible new inclusions, of which three were eligible for inclusion. The databases were checked again for new articles on 16 December 2015 and resulted in one additional relevant article. Finally, one eligible article was accepted for publication at time of the second search and was obtained through personal communication.

## Study selection and data collection

In total 2301 articles were evaluated for eligibility (See Figure 1). Articles were included when (1) subjects were healthy human adults, (2) an experimental design was used, (3) manipulation involved a negative affective stimulus, (4) the negative affective stimulus was manipulated out of the subject's awareness, i.e., processed without requiring conscious processing, (5) a control stimulus was used that was presented exactly like the negative affective stimulus for either between or within-group designs but was either of positive or neutral valence, (6) the dependent measure was a peripheral physiological outcome measure, (7) the article was peer-reviewed (e.g., no dissertations, conference proceedings, or editorials), (8) full-text was available in either English or Dutch.<sup>1</sup>

Eligibility was evaluated independently by two reviewers, the first and third author. A third reviewer, the second author, was consulted in case of disagreement. Articles that could not unanimously be excluded based on the information available at one step automatically were included in the next step to prevent invalid exclusion. The first round of exclusion was based on title; articles with titles that clearly implied an unrelated subject were discarded. After this round 679 articles were left. In the second round, exclusion was based on abstract and resulted in 184 potential eligible articles. Finally, in the third round the full-texts were evaluated which lead to the final inclusion of 54 articles. From articles that discussed multiple experiments studies that met the inclusion criteria were included as separate studies, resulting in a final selection of 65 studies.

The main features of the studies were extracted, as displayed in Table 3: Sample description, the nature of the negative affective stimulus, the key features of the design such as type of stimuli and presentation method, the type and data handling of the physiological parameters, awareness check, and the results. Data extraction was checked by at least one other author.



**Figure 1.** Flow chart of the selection process. Adapted from Moher et al. (2009).