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Improving Stimulus Realism: The effect of visual dimension on affective responding

Shannon Compton, *The University of Western Ontario*

Supervisor: Mitchell, Derek, *The University of Western Ontario*

A thesis submitted in partial fulfillment of the requirements for the Master of Science degree in Neuroscience

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Abstract

For decades researchers have used 2D stimuli under the assumption that they accurately represent real objects. This assumption has been challenged by recent vision and neuroeconomics research which has found that 2D images can evoke different neural and behavioural responses than real objects. The current study continues this line of research in the field of affective cognitive neuroscience; a field where small effect sizes are common and rapid habituation to affective stimuli used in the lab often occurs. The present study uses realistic 2D and 3D emotional images to determine the impact of visual dimension on affective responding. Subjective ratings revealed a perceptual advantage for 3D images which were rated more realistic and received some higher ratings of emotion than 2D images. Conversely, there were no differences in psychophysiological responding (i.e. skin conductance and electromyography) between 2D and 3D images. The implications of these results and future directions are discussed.

Keywords

2D/3D visual stimuli, emotion, affective responding, skin conductance, electromyography

Summary for Lay Audience

In order to generate an emotional reaction within a research setting, most psychology and neuroscience studies use emotional 2D images (e.g. kittens to induce pleasant emotions, spiders to induce fear). However, recent research suggests there are differences in the way the human brain respond to 2D images versus actual, physical objects. Real objects are better remembered, attended to, and are more highly valued than 2D images. This raises the question as to how well findings from studies which use 2D images can generalize to real world situations. In the laboratory, it is often difficult to mimic the impact of emotions in the real-world because emotional responses to images tends to weaken significantly when they are presented repeatedly. The present study aims to determine whether effects of emotion can be improved by using 3D images as they more closely resemble real objects. This study compared photorealistic 2D and 3D images of insects and arachnids of varying degrees of pleasantness (e.g. butterflies, scorpions). We predicted that 3D images would be perceived as more realistic and generate more intense emotional reactions compared to 2D images. To measure this, we explicitly asked participants to rate how realistic, pleasant, arousing, approachable, and dangerous they found each image. We also measured participants' bodily responses to the images as specific patterns of bodily responses are associated with different emotional reactions. The startle eye blink response is differentially affected by emotional images; positive images decrease the magnitude of the startle and negative images increase the magnitude of the startle. Skin conductance (SC) measures minute changes in the amount of sweat present on the skin. SC increases in response to emotionally arousing images, whether positive or negative. Our study found that 3D images showed greater subjective ratings for realism, arousal, and danger, but these same 3D images did not result in significant differences in visceral emotional reactions compared to 2D images. Before a definitive judgement can be made on whether there are differences in visceral reactions between 2D and 3D images, future research should compare these two image types using more arousing images, more bodily measures, and less repetitions.

Co-Authorship Statement

I, Shannon Compton, completed all experimental and written work for this thesis project. I designed the current study, recruited participants, completed all data collection, data processing, data analysis, and wrote the written work.

My supervisor, Dr. Derek Mitchell, contributed to all aspects of this thesis project including the formulation of the research question, consultation with task design, data analysis, interpretation, and editing of the written work.

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I would like to thank my advisory committee, including Dr. Jody Culham, Dr. Ingrid Johnsrude, and Dr. Ryan Stevenson, for their support and feedback throughout this project. I am especially grateful to Dr. Stevenson for his assistance in the editing process of this written work.

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List of Abbreviations

ACC	Anterior cingulate cortex
ANEW	Affective Norms for English Words
ANET	Affective Norms for English Text
ANOVA	Analysis of variance
ANS	Autonomic nervous system
CSEA	Center for the Study of Emotion and Attention
DmPFC	Dorsal medial prefrontal cortex
EMG	Electromyography
EmoPics	Emotional Picture System
FSQ	Fear of Spiders Questionnaire
GAPED	Geneva Affective Picture Database
IADS	International Affective Digitized Sound System
IAPS	International Affective Picture System
LGN	Lateral geniculate nucleus
NAPS	Nencki Affective Picture System
OASIS	Open Affective Standardized Image Set
OFC	Orbitofrontal cortex
PFC	Prefrontal cortex
SAM	Self Assessment Manikin
STAI	State Trait Anxiety Index
SCR	Skin Conductance Response
V1	Primary visual cortex
vIPFC	Ventral lateral prefrontal cortex
vmPFC	Ventral medial prefrontal cortex

CHAPTER 1

1 Introduction

Imagine you wake up on a typical Saturday morning with plans to meet your friend for breakfast at a local cafe. This everyday scenario is imbued with emotions that guide our behaviour. The happiness we feel when we walk outside to go to the cafe reinforces the health benefits associated with sun exposure and exercise. The fear we feel when exposed to threatening situations, like a car speeding past us as we cross the street, reminds us to be more cautious and aware of our surroundings when walking by the road in the future. The disgust we feel when we find mould on our bagel prevents us from consuming potentially harmful fungi. By recognizing our friend's emotional facial expression we are able to infer her internal emotional state and behave accordingly. For example, if our friend was frowning we would recognize that she was upset and our sense of empathy would compel us to ask what was bothering her. In this way, our ability to recognize emotions and the impact that ability has on our own emotional responses allows us to have successful social interactions and form fulfilling social connections. Not only do these emotions guide our behaviour but typical emotion processing is essential for us to function normally and successfully within the world around us.

1.1 Affective Cognitive Neuroscience

The field of affective cognitive neuroscience investigates the underlying neural mechanisms involved in the integration of emotional and cognitive systems. Research in this field identifies the brain areas responsible for emotional processing and the neural correlates behind emotional experiences. Information gained from these studies have revealed the ways emotions can impact the way we behave, learn, attend, remember, and interact socially.

Emotions have been described as “states elicited by rewards or punishers” where “a reward [is] something for which an animal (which includes humans) will work, and a punisher as something that an animal will work to escape from or avoid (Rolls, 2005, p. 1-2). This definition clearly indicates how emotions can influence our decision making; emotional psychophysiological responses and their neural correlates bias our behaviour towards rewards and away from

punishments (Damasio, Tranel, & Damasio, 1991). One example of how emotion can shape our behaviours is through fear conditioning. Fear conditioning is a form of associative learning encoded by the amygdala where a previously neutral stimulus (conditioned stimulus) is coupled with an innately fearful stimulus (unconditioned stimulus) until it too is able to produce a fear response (Phillips & LeDoux, 1992). This form of learning is biologically advantageous as it allows us to quickly recognize and respond to potentially dangerous situations. For example, if we burn ourselves by touching a hot pan we learn to associate the pan with pain and will be more cautious when handling it in the future. Emotion also biases our attention by enhancing the salience of biologically relevant stimuli. We preferentially attend to emotional stimuli; we are able to locate them within the visual field more quickly than neutral stimuli and we are less able to ignore them (Ohman, Flykt, & Esteves, 2001; Williams, Matthew, & MacLeod, 1996). For instance, it would be difficult to focus on driving if there was a wasp in the car with you, your attention would be drawn to the threatening stimulus. Just as with attention, emotional events are preferentially encoded into memory compared to neutral events. Flashbulb memories are the most compelling evidence of this; they are extremely vivid, detailed memories of surprising events that cause an extreme emotional reactions, usually associated with traumatic events such as the 9/11 terrorist attacks (Brown & Kulik, 1977). Finally, emotional processing is crucial for successful social interaction. By recognizing emotional facial expressions we are able to deduce what others are feeling and share in those feelings with them (Blair, 2003; Blair, 1995). Our ability to empathize with others facilitates our social interactions by ensuring that we conduct ourselves in ways that are deemed socially appropriate. As the breadth of these studies suggest, emotional processing is integral to normal functioning. In fact, it is so central to our wellbeing that abnormalities in emotional processing are present in almost every neuropsychiatric disorder (Vuilleumier, 2005). There are disruptions of mood in major depressive and bipolar disorder, fear and anxiety in anxiety disorders, aggression in antisocial personality disorder, and empathy in autism and conduct disorders to name a few. Affective cognitive research identifies how neural processing differs in these disorders allowing for the development of interventions and treatments which can then be tested and evaluated for effectiveness.

To perform this research, experimenters must elicit emotion within the controlled setting of a laboratory. The most common way experimenters evoke emotion is by using visual stimuli, usually 2D photographs. By using these 2D images experimenters can safely induce emotion and

are afforded a high level of experimental control; they can be sure that the stimuli appear consistent between participants.

1.2 Brain areas involved in emotion processing

While vision allows us to more easily complete practical tasks, it also allows us to admire paintings, regard our loved ones, or notice a spider crawling towards us. These types of stimuli elicit an emotional response; they influence internal states which reflect reward or punishment (Rolls, 1999). Emotion has such an impact on human cognition that it influences the way we process sensory information. Compared to neutral stimuli, emotional stimuli are more attention grabbing (Fenske & Raymond, 2006; Williams & Broadbent, 1986; Vuilleumier & Schwartz, 2001), more memorable (Cahill & McGaugh, 1995; Kensinger, 2009; Bradley, Greenwald, Petry, & Lang., 1992), and are able to prime reflexive actions (Bonnet, Bradley, Lang, & Requin, 1995; Both, Everaerd, & Laan, 2003). As such, the emotional pathways within the brain are widely connected and have strong bidirectional connections to the visual pathway (Amaral, Behniea, & Kelly, 2003; Price, 2003; Vuilleumier, 2005).

To better understand how emotion is able to accomplish this sensory modulation, we can examine the neural correlates of emotion. One region central to the neural processing of emotion and social behaviour is the amygdala, a bilateral structure within the medial temporal lobe (Aggleton, 2000). It is involved in almost every aspect of emotional processing including emotion regulation (Banks et al., 2007; LeDoux, 2007), fear conditioning (Davis & Whalen, 2001; Duvarci, Popa, & Pare, 2011), and emotion recognition (Yang et al., 2002; Garavan et al., 2001). The amygdala responds preferentially to emotional stimuli of both positive and negative valence (Yang et al., 2002; Hamann, Ely, Hoffman & Kilts, 2002; Sphors et al., 2018). Amygdala activation has been observed over a variety of emotionally evocative stimuli spanning different modalities and forms; this includes aversive natural stimuli (Krusemark & Li, 2011; Kensinger & Schacter, 2006), pleasant and negative events (Hamann, Ely, Grafton & Kilts, 1999), emotional films (Bride et al., 2014), aversive smells (Zald & Pardo, 1997), fearful vocal expressions (Phillips et al., 1998), and aversive tastes (Zald, Lee, Flugel, & Pardo, 1998). The amygdala sends more projections to the ventral visual pathway than it receives (Iwai & Yuki, 1987; Amaral, Behniea, & Kelly, 2003) suggesting that it modulates activity in these areas, particularly the inferior temporal cortex (area

TE; Iwai, Yukie, Suyama, & Shirakawa, 1987; Amaral & Price, 1984). This association between the amygdala and area TE is implicated in reward learning. A lesion study conducted by Spiegler & Mishkin (1981) found that macaques were only able to perform an object-reward association task when both area TE and the amygdala were intact; lesions to either of these areas impaired task performance. Since area TE is involved in object recognition, this suggests that the amygdala was responsible for linking the stimuli with the reward. The amygdala also modulates responding in the visual cortex in response to emotional facial expressions. Amygdalar lesions are associated with a reduction in fearful face recognition (Morris et al., 1998; Adolphs et al., 1999; Adolphs, Tranel, Damasio, & Damasio, 1995) and result in less activation in visual areas, such as the fusiform and occipital cortex, when viewing fearful faces compared to healthy controls (Vuilleumier et al., 2004). The amygdala's ability to increase activation in the visual cortex in response to emotional stimuli has also been observed in healthy adults when observing positive and negative emotional stimuli (Frank & Sabatinelli, 2014) and threatening stimuli, with activation in both the amygdala and visual cortex increasing with arousal (Bradley et al., 2003; Sabatinelli, Bradley, Fitzsimmons, & Lang, 2005).

While the amygdala is considered a central structure within the pathways of emotion due to its extensive connections throughout the brain (Amaral et al., 1992), the ventromedial prefrontal cortex (vmPFC) and orbitofrontal cortex (OFC) are also significantly involved in emotion processing. Both structures are within the prefrontal cortex (PFC) and have strong, bidirectional connections with the amygdala (Ghashghaei & Barbas, 2002; Amaral & Price, 1984; Barbas & De Olmos, 1990). Lesion studies suggest that both structures are involved in the emotional processes which underlie personality. A study by Barrash, Tranel, & Anderson (2000) found that bilateral damage to the vmPFC impaired emotional expression, emotional affect, interest, social behaviour, and insight. Likewise, a study by Hornak and colleagues (2003) found that patients with bilateral OFC lesions had deficits in emotion recognition of vocal stimuli, emotional affect, and social behaviour. The deficits in social behaviour (which were judged by an informant) were caused mostly due to the patients' inability to recognize or express emotions or provide insight into another's state of mind. The OFC has also been associated with insight into self behaviour, deficits to which impair social behaviour (Beer, John, Scabini, & Knight, 2006).

Although these are exclusive, the amygdala, vmPFC, and OFC have been identified as playing a key role in emotional processing and their function in this context illustrates how extensive the connections are between brain regions responsible for emotion and other systems. These structures respond to appetitive and aversive stimuli from every modality and yet visual stimuli are used most often in research on emotion. It is important to evaluate the visual stimuli used in these studies to ensure that they are not only effective at generating an emotional response but also that they accurately represent the emotional stimuli encountered outside a research setting.

1.3 Psychophysiological measures of emotion

Along with neural and behavioural measures, emotional responses can also be studied through measures of psychophysiological arousal.

Electromyography (EMG) is a measure of the electrical activity produced by muscle stimulation. The startle eye blink response is a response of the sympathetic autonomic nervous system (ANS) that is triggered by an unexpected stimuli, such as a loud burst of white noise. EMG electrodes can be placed on the orbicularis oculi muscle surrounding the eye to measure the magnitude of the eye blink response. Emotional modulation of the startle response is bidirectional where the valence of the stimuli presented affects the direction of the modulation. Positive images inhibit the startle response and negative images enhance the response (Vrana, Spence, & Lang, 1988; Bradley, Codispoti, Cuthbert, & Lang, 2001). Cuthbert, Bradley, & Lang (1996) illustrated this modulation in a study where participants viewed pleasant, neutral, and unpleasant images while being exposed to a startling stimuli (a burst of white noise). They found a valence by arousal interaction where, compared to neutral images, startle eye blink magnitude was larger for negative images (further increasing the more arousing the images were) and smaller for positive images (further decreasing the more arousing the images were).

Skin conductance (SC) measures the electrical conductance of the skin which increases with the level of sweat and oils present. As such, SC is used as a measure of arousal of the sympathetic ANS which innervates the sweat glands in response to threatening or arousing stimuli. A skin conductance response (SCR) is a rapid, transient increase in SC which can occur in response to external or internal stimuli. Since the ANS and SC by extension respond to environmental threat,

it is often used in emotion research. Unlike the startle eye blink response, the SC response to emotional stimuli is unidirectional; SC increases in response to stimuli of both positive and negative valence (Simons, Detenber, Roedema, & Reiss, 1999; Khalfa, Isabelle, Jean-Pierre, & Manon, 2002). A study by Bradley, Codispoti, Cuthbert & Lang (2001) found that SC increases to emotional images with arousal; the more emotionally arousing the image, the greater the response, regardless of whether the images were of positive or negative valence.

Amygdala activation has been linked to changes in psychophysiological responding related to fear, including SC and the startle eye blink response (Davis & Whalen, 2001; Wood, Van Hoef & Knight, 2014; Laine, Spitler, Mosher, & Gothard, 2009). Like the amygdala, SC and the startle eye blink response habituate to emotional stimuli. Bradley, Lang & Cuthbert (1993) conducted a habituation experiment where participants viewed pleasant, neutral, and unpleasant images repeatedly over three blocks while SCR, heart rate (HR), and startle eye blink EMG were measured. All three psychophysiological measures showed the expected valence effects within the first block and showed a large reduction in responding across the three blocks. While largely reduced, the startle eye blink response still displayed significant valence effects by the end of the final block while the other two measures no longer showed an effect. This persistence of valence effects is why the startle eye blink response is the primary psychophysiological measure in the current study. SC and EMG are often recorded together in studies of affective autonomic signalling to get a more complete view of how the brain processes emotional information.

1.4 Emotion and decision making

Our emotions do not only impact our internal states, they also impact our behaviour. Emotions influence our behaviour in ways that increase the likelihood of rewarding outcomes and decrease the likelihood of punishing outcomes. The somatic marker hypothesis (Damasio, 1994) postulates that emotional bodily responses (or ‘somatic markers’) are responsible for guiding decision making behaviour. Activation in the vmPFC is correlated with autonomic responses to emotional stimuli (Damasio, Tranel, & Damasio, 1990) so it is unsurprising that it has been implicated in the somatic marker hypothesis. In a study using a gambling task, both controls and patients with damage to the vmPFC were able to generate SCRs in response to rewards and punishments but only controls were able to generate anticipatory SCRs before making a decision, with larger SCRs

preceding riskier decisions (Bechara, Tranel, Damasio, & Damasio, 1996). From these results the experimenters concluded that vmPFC patients were impaired in their ability to change their autonomic responses in response to anticipated negative consequences. This study illustrates that autonomic signals generated by the vmPFC may play a role in guiding our decisions. While the somatic marker hypothesis has since been questioned (Dunn, Dalgleish, & Lawrence, 2006; Caramazza, Deldin, Hauser, & Tomb, 2002; Maia & McClelland, 2004), other studies have replicated the finding that autonomic signals related to emotion and their neural correlates play a role in decision making related to rewards and punishments (Bechara, Damasio, Damasio, & Lee, 1999; Guillaume et al., 2009; Bechara, Tranel, & Damasio, 2000).

In addition to the vmPFC, the OFC and ventrolateral prefrontal cortex (vlPFC) are also implicated in emotional decision making. Activation in the OFC has been linked to representations of reward and punishment (Rolls, 1999) and reward evaluation and choice difficulty (Arana et al., 2003). The OFC is involved in stimulus-reinforcement associated learning where a rewarding stimulus (e.g. an appetitive food) is associated with a neutral stimulus (i.e. an image) until the neutral stimulus alone becomes associated with reward (Rolls, 2000; Kringelbach & Rolls, 2004). During stimulus-reinforcement learning tasks, activity in the medial OFC is correlated with reward, with greater activation for greater gains, while activity in the lateral anterior OFC is associated with punishment, with greater activation for greater losses (O'Doherty et al., 2001). The OFC also represents outcome expectations and modulates associations between a stimulus and an expected outcome in response to violations of these expectations (Mitchell, 2011). In this way, the OFC impacts our behaviour by devaluing stimuli that are no longer rewarding (Gotfried, O'Doherty, & Dolan, 2003). Similarly, the vlPFC is involved in changing behaviour in response to changes in context (Mitchell, 2011). Lesions to the vlPFC result in impaired performance during reversal learning tasks, where one stimulus is associated with a reward and another with a punishment before the associations are reversed (Fellows & Farrah, 2003). This suggests that the vlPFC is necessary for alterations in decision making based on new information related to the reward and punishment value of certain behaviours. Taken together, these studies illustrate how the PFC influences decision making by optimizing behaviours that will result in rewards and minimizing behaviours that will result in punishments.

There is a saying about how we should not let our emotions make our decisions but these studies show that this would not only be difficult to accomplish but also maladaptive. Emotions, presently felt or anticipated, guide our behaviour in part due to the autonomic signals we receive or will experience once we make a decision. As previously mentioned, these autonomic signals can then be used as a measure of emotional arousal and have been in many affective cognitive neuroscience studies (Bradley, Lang, & Cuthbert, 1993; Kimmel & Gardern, 1986; Vrana, Spence, & Lang, 1988; Codispoti, Ferrari, & Bradley, 2007). The present study will use two measures of autonomic signaling to evaluate the differences in affective responding between emotional stimuli of two different modalities.

1.5 Emotion and attention

Attention is a cognitive process where certain stimuli are selected for further processing while other stimuli are neglected (Blair & Mitchell, 2009). The biased competition model of attention postulates that stimuli within the environment compete for neural representation and cognitive processing (Desimone & Duncan, 1995). As the name suggests, this competition is biased, specifically by bottom up and top down processes. Bottom up processes prioritize stimuli based on their low level visual features. Bottom up systems would bias attention towards stimuli that are large, colourful, and have a high contrast ratio (Beck & Kastner, 2009). Top down processes prioritize stimuli based on their higher order cognitive relevance. Top down systems would bias attention towards task relevant stimuli, for example stimuli which appear in a particular location within the visual field where participants were instructed to attend (Kastner, Weerd, Desimone, & Ungerleider, 1998). In either case, the neurons representing these stimuli would become highly active (Blair & Mitchell, 2009). The representation that generates the most activation will win the competition for attention.

So far, the processes described here identify ways that mundane stimuli compete for attention but a particular advantage is given to emotional stimuli. The preferential processing or enhanced encoding of emotional stimuli is thought to be conferred via the bidirectional interaction between the ventral visual stream and the amygdala (Pessoa and Ungerleider, 2004). Pessoa & Adolphs (2010) proposed a model where emotional stimuli are recognized by the amygdala which amplifies the activity of neurons representing the stimuli within the visual cortex. This model is supported

by tracer studies and studies of functional connectivity which have shown the amygdala is highly connected to the ventral visual cortex (Amaral, Behnia, & Kelly, 2003; Vuilleumier, 2005) and that activity in this cortex is intensified in response to emotional stimuli (Morris et al., 1998; Pessoa, McKenna, Gutierrez, & Ungerleider, 2002). Just as the amygdala modulates activity in the visual cortex, activity in the amygdala can be modulated. The PFC can modulate amygdala activity to prevent emotional stimuli from reaching awareness (Amting et al., 2009). This modulation may be a result of the PFC's role in directing visual attention to emotionally salient information. A study by Wolf and colleagues (2014) found that patients with vmPFC lesions exhibited deficits in identifying emotional facial expressions. Eye tracking data revealed that this impairment was due to the fact that patients were not attending to areas which would provide emotional information (e.g. the eyes), regardless of which facial expression was being displayed.

Although these studies reveal that emotional stimuli are salient, there are still issues associated with their use which has a negative impact on affective cognitive research. These limitations will be discussed and addressed in the current study.

1.6 Difficulties with emotion research

Small effect sizes are expected in emotion research as brain areas responsible for emotional encoding rapidly habituate to emotive stimuli (Plichta et al., 2014; Fischer et al., 2003; Wright et al., 2000). Unsurprisingly, this includes the amygdala which not only responds to emotional stimuli but also rapidly habituates to them (Plichta et al., 2014; Buchel, Morris, Dolan, & Friston, 1998; Wedig, Rauch, Albert, & Wright 2005). A neuroimaging study conducted by Brieter and colleagues (1996) found that the amygdala responds preferentially to faces of both positive and negative valences and rapidly habituates to them. This pattern of activation and habituation within the amygdala has been replicated in many studies featuring negative and positive stimuli (Wright et al., 2001; Fischer et al, 2003). Habituation to emotional stimuli is a common phenomenon in brain areas related to emotion; a neuroimaging study by Denny and colleagues (2014) found that repeated presentation of aversive stimuli resulted in a decrease in activation in several brain areas implicated in emotion processing including the amygdala but also the ventral PFC. Likewise, Wright and colleagues (2001) found significant habituation effects with both happy and fearful faces in the right amygdala and left dorsolateral PFC.

Emotion research is further restrained by ethical limitations. One of the central reasons why ethical guidelines are used in research is to ensure the individuals participating in research are not harmed. Although activation in brain areas related to emotion have been found to increase with arousal (Garavan et al., 2001), there is a possibility that exposing participants to highly arousing negative stimuli featuring death, violence, and/or gore may distress or traumatize some participants and thus cause them harm. There is evidence to suggest that images alone are enough to negatively impact an individual's emotional state; studies on media exposure following natural disasters or terrorist attacks reveal that frequent exposure to distressing images can cause anxiety and increase the likelihood of developing post-traumatic stress disorder (Ahern et al., 2002; Yeung et al., 2018; Busso et al., 2014; Bodas, Siman-Tov, Peleg, & Solomon, 2015). As such, highly emotional negative images are uncommon in emotion research even though they would evoke the most compelling effects of emotion.

One possible solution to these issues would be to improve upon the emotional stimuli that can be ethically used in emotion research by improving their level of realism. Emotional responses may be more robust and sustained if participants' experiences within the laboratory more closely resemble an authentic, real life experience. One possible way to accomplish this would be to use virtual 3D environments which are becoming more popular commercially in video gaming for exactly this reason (LaViola, 2008; Tachi, 2013).

1.7 Visual processing in the brain

Vision allows us to observe, navigate, and easily interact with our environment (Barry, 1997; Ekstrom, 2015; Cronin & Douglas, 2014). While not essential to daily living, having the ability to see assists us in almost every aspect of our daily functioning from simple tasks like washing and feeding ourselves to complex functions like operating a motor vehicle as we navigate through the city on our way to work.

Visual processing begins with the eye when images are projected through the cornea and lens onto the photoreceptors in the retina at the back of the eye (Enoch, Bedell, & Tobey, 1981). These photoreceptors then synapse onto a series of cells in the optic nerve which in turn pass the visual information into the optic tract and then to the lateral geniculate nucleus (LGN;

Hubel, 1963; Sefton & Swinburn, 1964). The LGN is divided into 6 layers, half of which receive information from one eye and half receiving information from the other eye (Bishop, Kozak, Levick, & Vakkur, 1962). This information corresponds to one visual hemifield; the left LGN processes information from the right visual field while the right LGN processes information from the left (Glees & le Gros Clark, 1941; Connolly & Van Essen, 1984). From here, visual information finally moves on to the cortex for processing, the first stop being the primary visual cortex (V1) in the occipital lobe (Hubel & Wiesel, 1974; Tootell, Silverman, & De Valois, 1981).

V1 processes basic visual features. Cells within V1 are sensitive to orientation; they respond to lines pointed in a particular direction which allows them to detect edges and bars (Hubel & Wiesel, 1977). After V1, a two pathway theory of vision has been suggested; a ventral 'vision for perception' pathway for object recognition and identification and a dorsal 'vision for action' pathway for object location and action-guided behaviour (Goodale & Milner, 1992; Mishkin, Ungerleider, & Macko, 1983; Goodale & Humphrey, 1998). The ventral pathway projects from V1 to the temporal lobe and includes V2 (which also processes basic visual features), V4 (which is involved in colour processing), and the inferior temporal lobe (Kobatake & Tanaka, 1994; Rousset, Thorpe, & Fabre-Thorpe, 2004). The dorsal pathway projects from V1 to the occipital and parietal lobes and includes V2, V3A and V5/MT (which are involved in motion perception), and the posterior parietal cortex (McKeefry et al., 1997; Goodale, 2011).

While the LGN processes visual information from both eyes, the information is still segregated within different layers. The first time visual information from both eyes actually integrate is within the V1 (Hubel & Wiesel, 1959; Bridge & Cumming, 2001). Simple and complex cells within the V1 use this information for edge detection (Hubel & Wiesel, 1962) but other cells respond to the fact that the visual information they receive from each eye is slightly different (Pettigrew, Nikara, & Bishop, 1968; Heeger, Polonsky, Blake, & Braun, 2000). Since human eyes are horizontally separated by approximately 6 cm, objects fall on a different part of the retina of each eye. This difference between the images projected onto each retina is referred to as binocular disparity. Stereopsis is the process by which binocular disparity is used to perceive depth. The binocular neurons in V1 alone are not enough for the conscious perception of depth (Cumming & Parker, 1997; Cumming & Parker, 1999) but work in conjunction with higher order visual areas such as V2 (Poggio, Motter, Squatrito, & Trotter, 1985;

Durand, Zhu, Celebrini, & Trotter, 2002), V3A (Felleman & Van Essen, 1987; Tsao et al., 2003; Wang et al., 2016), and V5/MT (Cumming, DeAngelis, & Newsome, 1998; Wang et al., 2016).

While stereopsis provides the most compelling depth information, it is not the only way humans are able to perceive depth. Monocular depth cues allow us to estimate distance with the visual information from one eye. Some examples of monocular depth cues include shadows (Kim & Anstis, 2016), occlusion (objects in front of other objects are perceived to be closer to the observer; Palou & Salembier, 2013), relative size (smaller objects are perceived to be farther away than larger objects; Sousa, Brenner, & Smeets, 2011), and motion parallax (when an observer is moving, objects that move fast are perceived to be closer than objects that move slower; Gibson, Gibson, Smith, & Flock, 1959).

1.8 Databases of affective stimuli

To facilitate research on emotion, several databases of standardized emotion-provoking stimuli have been created for use worldwide. Visual stimuli are most commonly used in affective cognitive research due to their convenience, the level of experimental control they afford, and their ability to generate unique behavioural and psychophysiological responses corresponding to particular emotional states (Mauss et al., 2005; Bradley et al., 1993; Rosenberg & Ekman, 1994). Many databases of emotional visual stimuli have been created to aide in this research including the International Affective Picture System (IAPS; Lang, Bradley, & Cuthbert, 1997), Geneva Affective Picture Database (GAPED; Dan-Glauser & Scherer, 2011), the Nencki Affective Picture System (NAPS; Marchewka, Zurawski, Jednorog, & Grabowska, 2013), the Emotional Picture System (EmoPicS; Wessa et al., 2010), and the recently created and freely available Open Affective Standardized Image Set (OASIS; Kurdi, Lozano, & Banaji, 2016). These databases provide normative valence, arousal, and dominance ratings for thousands of emotional images which cover the full range of valence and arousal dimensions, from very positive (e.g. happy babies, dogs) to very negative (e.g. spiders, cemeteries) and low arousal (e.g. flowers, baskets) to high arousal (e.g. erotic nudes, mutilated bodies).

The IAPS database was developed over 30 years ago and is now the most widely used database of emotional visual stimuli (Lang & Greenwald, 1988). The IAPS was developed by the

Center for the Study of Emotion and Attention (CSEA) with the goal of creating a standard set of images which could be used universally in psychological and neuroscience research to allow direct comparisons to be made between experiments and exact replication studies to be conducted (Lang & Greenwald, 1988). Since its creation, new stimuli have been consistently added to the database which is now comprised of over 1000 images across several different categories including images that feature humans, animals, objects, and scenes (Lang, Bradley, & Cuthbert, 2008). The database has been validated in many populations; most frequently with healthy adults but also adolescents (Vasa et al., 2011; Truedsson et al., 2019), older adults (Gruhn & Scheibe, 2008; Rehmert & Kisley, 2013), and clinical populations such as individuals with schizophrenia (Pankow et al., 2013), fibromyalgia (Rhudy et al., 2013), and depression (Gollan et al., 2016). Subsets of the database have also been tested across cultures with studies proposing different normative ratings for certain cultures including Spain (Molto et al., 1999), Belgium (Verschuere, Crombez, & Koster, 2001), Brazil (Ribeiro et al., 2005), India (Lohani, Gupta, & Srinivasan, 2013), and China (Huang et al., 2015; Gong & Wang, 2016). Along with these cross-cultural validation studies, IAPS is further trusted due to its reliability. IAPS images have a high internal consistency for valence and arousal ratings with both dimensions having highly reliable split-half coefficients when presented on paper ($r_s = 0.94$ and $r_s = 0.94$, $p < 0.001$) or on a computer ($r_s = 0.94$ and $r_s = 0.93$; Lang, Bradley, & Cuthbert, 1997).

Despite these benefits, there are some limitations to using this database. IAPS images are not matched on low level visual features like colour composition, luminance, or contrast (Coan & Allen, 2007) which can lead to differences in affective and visual processing (Bradley, Hamby, Low, & Lang, 2007; Wiens, Sand, & Olofsson, 2011). In order to correct for these physical differences, experimenters would need to use photoediting tools or select images based on their low level visual features. A test of spatial frequency, such as a wavelet analysis (Delplanque et al., 2007), would then need to be conducted to confirm that the images were not significantly different on these features. Another issue is that, while there are an extensive number of images in the IAPS database, the number of stimuli in some specific categories have been found to be limited (Marchewka, Zurawski, Jednorog, & Grabowska, 2013; Wessa et al., 2010; Dan-Glauser & Scherer, 2011). This is an issue for experiment designs that require many trials such as fMRI, EEG, or repetition experiments where the number of stimuli must be large enough to prevent any unintentional repetition (Dan-Glauser & Scherer, 2011). Finally, the fact that the IAPS database is

so widely used may also be one of its determinants. Many participants in psychological research are university students who participate in many studies. Thus the more studies that use IAPS images, the more likely participants are to have seen these images before which would lower their emotional impact (Marchewka, Zurawski, Jednorog, & Grabowska, 2013).

Along with the IAPS, the CSEA has created several other emotion-provoking databases including the International Affective Digitized Sound system (IADs), the Affective Norms for English Words (ANEW), and the Affective Norms for English Text (ANET) which provide normative ratings of emotion for sounds, English words, and brief English texts respectively. These databases are also used in research on emotion although they are not as extensive or ubiquitous as the IAPS.

Despite the number of emotional databases that exist there is still a lack of variety regarding visual stimuli. While the visual databases do contain a vast variety of images of different emotional content, they currently only feature 2D photographic stimuli. Moving forward, these databases should take advantage of the recent advances in vision technologies and work to create a set of standardized 3D images.

1.9 Questioning the ecological validity of 2D stimuli

For decades researchers have been using 2D stimuli under the assumption that they accurately represent real objects. This assumption that has been challenged by recent studies which show differences in neural, cognitive, and behavioural responding between the two stimulus types. For example, a neuroimaging study conducted by Snow and colleagues (2011) investigated the differences in repetition effects when using 2D images and real objects. Repetition suppression is a phenomenon where there is a characteristic reduction in the hemodynamic response (a direct measure of blood oxygenation levels within the brain and an indirect measure of neural activation) when stimuli are presented repeatedly. The Snow study found that there was a change in activation for 2D stimuli where there was a reduction in activation in a condition where stimuli were presented twice compared to a condition where they were only presented once. Conversely, there was no reduction in response when using real objects in the condition where stimuli were repeated. So while repetition suppression was observed, as expected, with 2D images, it was absent when

participants viewed real objects suggesting an inherent difference in the way these stimuli are perceived and processed.

As for cognition, Snow, Skiba, Coleman & Berryhill (2014) conducted a study which compared memory effects between 2D images and real objects. There were three conditions where participants were shown either black-and-white line drawings, coloured photographs, or real household objects. Participants then completed a free-recall task, where they wrote down as many stimuli as they remembered, and a recognition task, where they just had to state whether they had seen the object or not. Participants in the real object condition performed significantly better on both tasks whereas performance did not differ in the colored photograph or line drawing conditions.

The differences in responding between these two stimulus types extend to behavioural responses as well. Bushong, King, Camerer, & Rangel (2010) conducted a neuroeconomics study which compared the differences in valuation judgements between real objects and 2D images of those objects. Participants gave real objects significantly higher valuations than 2D images, increasing their willingness to pay for the item by 40-60%.

Taken together, these studies show a fundamental difference in the way 2D images and real objects are processed. Therefore, they raise questions about whether the results gained from studies which use 2D stimuli can be generalized to make claims about real world experiences as they have been in the past.

Several studies have been conducted to delineate the processes behind these observed differences in responding between 2D images and real objects. These studies most commonly focus on whether these differences exist because real objects can be interacted with in a purposeful way whereas 2D images cannot. One such study was an action priming study by Squires, Macdonald, Culham & Snow (2016). Action priming refers to the observation that viewing an object before being asked to interact with that same object facilitates grasping (Valyear et al., 2011). Interestingly, this study found that there was no difference in action priming between 2D images of tools and real tools. Although this study showed no difference in behavioural responses due to the interactive nature of real objects, a study by Gomez, Skiba, & Snow (2018) showed that there were attentional differences. Real objects were found to be more distracting during a flanker

task (where participants respond to a central image which is flanked by two distractor images) compared to 2D or 3D images only when the objects were graspable. As soon as a barrier was placed between the object and the participant, the differences in attentional effects disappeared. Further, a study by Marini, Breeding & Snow (2019) found that real objects elicited greater activation in action-related brain areas and brain areas related to stereopsis compared to 2D images. Taken together with past research, these studies suggest that the differences observed between real objects and other visual stimuli may be due to differences in visual processing and the fact that real objects are interactive.

While these studies provide a compelling argument that the brain responds differently to 2D images and real objects, this does not mean that all experiments should attempt to use real stimuli. While real objects would provide the most ecologically valid results, they are not always feasible to use within a laboratory setting, especially with research related to emotion. Biologically significant visual stimuli such as threat related stimuli (e.g. spiders, wolves) and social stimuli (e.g. emotional facial expressions) would be difficult to present in a controlled and consistent manner across participants. Even the act of storing and presenting these stimuli would be difficult in studies with designs that require many stimuli or many trials. In cases where using real stimuli is not possible, a compromise may be made by using 3D images. Just as with real objects, there is evidence to suggest that there are differences in responding between 2D and 3D stimuli. A study conducted by Rooney, Benson, & Hennessy (2012) compared participants' perception of and psychophysiological arousal to scenes from films presented in either 2D or 3D. Participants in the 3D film condition gave higher ratings of realism and had a higher heart rate compared to participants in the 2D condition. This difference in subjective ratings was replicated in a study by Gaebler and colleagues (2014) where participants rated a 3D film more immersive compared to the 2D version of that film.

While these studies provide intriguing results, most studies on emotion still use static 2D stimuli. To date, we are aware of only two prior studies that compared 2D and 3D emotional images. These were two neuroimaging studies conducted by Dores and colleagues (2013, 2014) which investigated the difference between pleasant, unpleasant, and neutral scenes presented in 2D and anaglyph 3D. These studies discovered greater activation in the amygdala for unpleasant 3D scenes compared to their 2D counterparts and greater activation in the postcentral and middle

frontal gyrus for both pleasant and unpleasant 3D scenes. Despite these promising results, these studies alone are not enough to conclude that 2D and 3D images produce different effects of emotion. The emotional scenes created for these studies were computer generated, giving both 2D and 3D stimuli a cartoonish appearance instead of looking like actual photographs of real scenes. Given the evidence that photographic 2D images and real objects are already known to engage the visual system and related systems in fundamental different ways (Snow et al., 2011; Gomez, Skiba & Snow, 2018; Marini, Breeding & Snow, 2019), it cannot be assumed that computer generated images would engage these systems in the same way as either stimulus type. Also, these studies used an anaglyph technique to create the illusion of depth. Red-cyan anaglyph glasses affect the colour of the stimuli further reducing its appearance of realism. Additionally, ghosting (where images appeared doubled as an image designed for one eye bleeds through to the other) is common with anaglyph glasses which impairs the stereoscopic effect and reduces image quality by blurring the edges of objects in the foreground (Woods & Rourke, 2004). Most emotion research does not use computer-generated 2D images but 2D photographs of actual stimuli. So even if 3D computer-generated images do elicit greater effects of emotion compared to computer-generated 2D images, this does not indicate that either have greater effects than realistic 2D images. Additional research needs to be conducted comparing photorealistic 2D and 3D emotional images before conclusions about differences in processing can be drawn.

1.10 Thesis objectives and hypotheses

Even though humans perceive the world in three dimensions, almost all prior studies on vision and emotion have used 2D stimuli. It was believed that 2D images were able to elicit responses that closely resembled those that would be experienced in the real world but recent research challenges this belief by revealing differences in neural, cognitive, and behavioural responding between 2D images and real objects (Snow et al., 2011; Snow, Skiba, Coleman & Berryhill, 2014; Bushong et al., 2010; Gomez, Skiba, & Snow, 2018). While the differences between 2D images and real objects are being explored, there is very little research comparing 2D and 3D images in the field of affective cognitive neuroscience. Research in this area could be especially beneficial for this field where small effect sizes are expected as brain areas and psychophysiological responses rapidly habituate to emotional stimuli (Breiter et al., 1996; Plichta et al., 2014; Bradley, Lang, & Cuthbert, 1993). This habituation could be counteracted if, like real objects, 3D images are more

resistant to repetition effects (Snow et al., 2011). This leaves a gap in current knowledge, there are no studies we are aware of that explore whether there are differences in affective responding between photorealistic 2D and 3D emotional images.

The overall objective of this thesis is to determine if 3D images can be used to improve stimulus realism and be used in emotion research to achieve more reliable effects of emotion. We predict that, compared to 2D images, 3D images will elicit greater affective responses. Specifically, we predict that 3D images will:

1. Receive higher subjective ratings of realism and emotion;
2. Initially elicit increases in psychophysiological responding;
3. Be more resistant to habituation effects; and
4. Experience a greater recovery of response when novel emotional images are presented after habituation.

The difficulties associated with emotion research increase the likelihood of committing a Type I error; effects may be missed due to the small or rapidly diminishing effects of emotion. The aim of this study is to determine whether classic emotion research methods can be improved upon by using stimuli that more closely represent real world experiences to bolster effects of emotion to ensure they are not wrongly overlooked. Unlike previous research, the present study will use photorealistic stimuli presented with 3D shutter glasses to more directly compare classic, photographic 2D stimuli to realistic 3D stimuli. The results of this study could expand beyond emotion research and inform all studies which use visual stimuli on how to improve effect sizes and ecological validity.

CHAPTER 2

2 Methods

2.1 Participants

Fifty-three participants (32 female, 21 male; mean age, 22.4; range, 18-35 years) from Western University took part in the current study. Three participants were excluded for being unable to distinguish between 2D and 3D stimuli (less than a 1-point difference in mean visual dimensionality scores between the two stimulus types). Of the 50 remaining participants, EMG and SCR data were collected from 45 participants. Participants were recruited from flyers posted around campus. All participants were screened for vision and psychiatric disorders and had normal or corrected-to-normal vision. Even though only participants with normal or corrected-to-normal vision were recruited, the Randot Stereo Vision Test (2018) was employed to ensure that all participants displayed normal depth perception. Eligible participants had a visual acuity score of 50 arcseconds or better, one participant was excluded for having a stereoacuity score greater than 400 arcseconds. All participants provided written informed consent and received monetary compensation at a rate of \$15/hour. This study was approved by the University of Western Ontario Research Ethics Board.

2.2 Stimuli

A 3D stimulus set was created by photographing 39 plant and taxidermic entomological specimens. The stimulus set consisted of 6 plant specimens, 16 beetle specimens, 3 wasp specimen, 12 butterfly specimens, and 2 arachnid specimens. One 2D and one 3D image was created of each specimen to create a stimulus set of 78 images. To create the stimuli, the specimens were positioned on a rock in front of a white background. Foliage was added to the background to provide additional monocular depth cues. A digital camera was placed approximately 188 cm away from the specimen, although images were later cropped and resized to optimize three-dimensionality when the images were viewed at 30 cm. To create the 2D images, two pictures were taken of the specimen from an established center line. To create the 3D images, one picture of the specimen was taken 3 cm to the left of the center line and a second picture was taken 3 cm

to the right of a center line to mimic the distance between human eyes. StereoPhoto Maker (Version 5.10; 2015) was used to superimpose the two images and merge them into a single .JPS file. The physical properties of the images were matched as closely as possible. Even though the specimen varied in size, the rock and foliage were consistent throughout the stimulus set in size and position. The images were then matched for colour, luminance, and size using MATLAB (2018a). To adjust colour and luminance, the top left corner of each image (which did not feature any objects) was measured as the average background colour and brightness. The rest of the image was adjusted so the background had an RGB of [230 230 230]. All images were sized to 1582 x 1315 pixels, with the background consistent and target centered in each image. For 3D images there was a 30 pixel separation between the left and right images (each 15 pixels away from the center). These stimuli were displayed using NVIDIA 3D Vision Photo Viewer on an ASUS 24-inch 3D gaming monitor (MG248Q) using NVIDIA 3D Vision 2 Goggles.

A subset of 36 of the 78 images in our stimulus set were chosen to use in the main study based on participants' ratings in a pilot study (see Appendix A for mean ratings). Stimuli were first split into three groups based on pleasantness ratings from a pilot experiment (Fig. 1); the images with the top third of pleasantness ratings were considered 'pleasant' (mean ratings between 6.33 – 8.17), the middle third 'neutral' (mean ratings between 4.31 – 5.77), and the bottom third 'unpleasant' (mean ratings between 1.58 – 3.38). Six images were then chosen from each emotion category based on visual dimensionality ratings; images with the greatest disparity in visual dimensionality ratings between the 2D and 3D version of the images were chosen (mean disparity 4.13 points). These images were chosen as the main study examines the effect of visual dimension on affective responding; thus, the images with the greatest perceived difference in visual dimensionality would be best suited to reveal any differences in responding between 2D and 3D stimuli if such a difference exists. Six stimulus subsets were created from these 36 images so that there was one 2D and one 3D image from each emotion category in each set. Subsets were created by matching stimulus size as closely as possible.

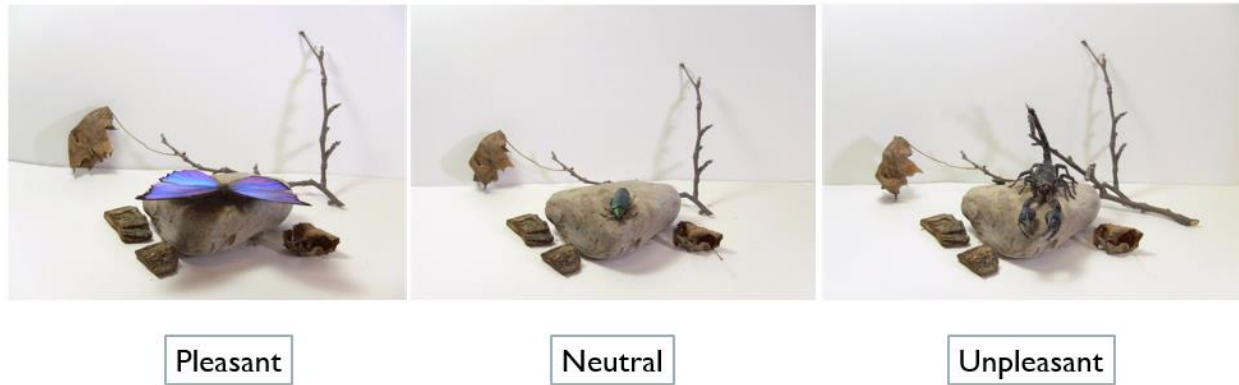


Figure 1. Examples of a pleasant, neutral, and unpleasant 2D stimuli in one stimulus subset.

An acoustic startle probe was created using Audacity (Version 2.1.2; Mazzoni, 2015). The probe was a 50ms white noise burst saved in a .wav file and presented at 104dB. The probe was administered through Sennheiser HD 25 Light DJ Headphones which were connected to a Behringer HA400 Stereo Headphone Amplifier.

2.3 Items

Trait anxiety was measured to determine if higher levels of anxiety resulted in increases in psychophysiological responding. The State Trait Anxiety Inventory (STAI; Spielberger et al., 1983) for Adults Form Y was administered to participants in order to evaluate their level of trait anxiety. The STAI is a self-evaluation questionnaire composed of two 20-item scales which measure state and trait anxiety respectively. The questionnaire is composed of statements related to how they are currently feeling or generally feel (i.e. *I feel nervous or restless*) and participants are asked to circle a number from 1-4 to indicate how much the statement applies to them (1 = almost never, 2 = sometimes, 3 = often, 4 = almost always). Half of the items are reverse-scored as they corresponded to statements associated with low anxiety (i.e. *I am “calm, cool, and collected”*). Participants received a score between 20 and 80 for each scale. All participants completed this questionnaire before they began the computer task. This measure was included to determine if higher levels of trait anxiety increased psychophysiological responding.

Additionally, individual scores of fear of insects and spiders were measured to determine if higher levels of fear resulted in increases in psychophysiological responding. The Fear of Spiders

Questionnaire (FSQ; Szymanski & O'Donohue, 1995) was adapted to assess participants' fear of insects as well as spiders. The adapted FSQ was administered to determine whether participants had a spider and/or insect phobia or to assess the level of fear towards spiders and/or insects in non-phobics. The FSQ is an 18-item self-evaluation questionnaire which asked participants how much they agreed or disagreed with statements relating to how they feel about or would respond to insects and/or spiders (i.e. *If I saw an insect/spider now, I would think it will harm me*) using a 7-point Likert scale (1 = strongly disagree, 7 = strongly agree, 4 = neither agree or disagree). After completing the adapted FSQ, participants received a score ranging from 18 to 126. Population means determine phobics score an average of 89.9 on the FSQ before undergoing treatment (Muris & Merckelbach, 1996), no participants in the present study scored above this threshold. The FSQ was chosen over the Spider Phobia Questionnaire because it more sensitive at measuring fear within the non-phobic range as it uses a 7-point scale as opposed to a dichotomous true or false scale (Muris & Merckelbach, 1996).

2.4 Subjective measures

Participants were asked to rate each image across six different dimensions. The six dimensions used were Arousal, Pleasantness, Approachability, Dangerousness, Realism, and Visual Dimensionality. Arousal and Pleasantness dimensions mirror the arousal and valence dimensions used in the Self-Assessment Manikin (SAM) scale used to assess IAPS images (Bradley & Lang, 1994). Approachability and Dangerousness dimensions were used instead of the dominance dimension used in the SAM scale to get a more complete picture of whether the image was being perceived as threatening. Realism was included as a direct measure of our prediction that 3D images would be perceived as more realistic than 2D images and Visual Dimensionality was included to ensure that participants were perceiving a difference between the 2D and 3D stimuli. For each image participants were asked to provide a rating on each dimension using a 9-point Likert scale (1 = min, 9 = max, 5 = neutral). Participants' starting position on the scale was highlighted in red; they used arrow keys to move between points on the scale and the enter key to confirm their selection. Participants had an unlimited amount of time to provide responses. The order that the dimensions appeared and the starting position on the scale was randomized. This was done to encourage participants to pay attention so they could rate the pictures independently

and to reduce the likelihood of response bias. Participants were provided with the definition of each dimension and information on how to use the scale before they completed the task.

2.5 Psychophysiological measures

The current study used two psychophysiological measures, EMG and SCR. Both are measures of the sympathetic ANS which are modulated by emotional stimuli. SCR shows a unidirectional response to emotional stimuli where the response increases with arousal. The EMG measure recorded the startle eye blink response to an acoustic startle probe. A startle response is a behaviour that occurs in response to an unexpected stimulus and can be modulated by emotional stimuli. Unlike skin conductance, the startle eye blink reflex shows a bidirectional response to emotional stimuli where positive stimuli inhibit the response and negative stimuli magnify the response.

Two EMG electrodes were placed on the orbicularis oculi of the left eye, one directly below the pupil and one near the outer corner of the eye (Fig. 2). EMG data were sampled at 2000 Hz with a pair of reusable 4mm Ag/AgCl shielded electrodes (Biopac model: EL254S) which were filled with an isotonic gel (Biopac model: GEL100). The raw signal was automatically filtered through a 30 Hz comb band stop filter during data collection to reduce noise. The signal was filtered offline using a 28-500 Hz band pass filter, the root mean square was derived, and 40 Hz low pass FIR filter in line with previously established guidelines (Blumenthal et al., 2005). The startle response was measured as the maximum amplitude of the EMG signal 20-200 ms after the presentation of the startle stimulus compared to a baseline which was determined as the average EMG activity 180 ms before stimulus presentation. EMG data were excluded if it contained excessive noise due to technical difficulties during recording or if participants were non-responders. Approximately 5-10% of healthy adults are startle non-responders (Blumenthal et al., 2005). For our EMG measure, non-responders were identified as participants who did not generate a startle eye blink response (EMG activity following startle probe was less than 2 standard deviations higher than baseline) but who clearly displayed voluntary eye blink responses. Data from three participants were excluded after they were identified as non-responders (6.1% of our sample) and data from two participants were excluded due to excessive noise.

This study measured the mean number of SCRs as opposed to the amplitude of the response as the number of SCRs was low overall. As with EMG, SCR data were gathered using the Biopac M160 Data Acquisition System and were processed and analyzed using the Acqknowledge 5 software. Data were sampled at 2000 Hz with a pair of disposable pre-gelled contact electrodes (Biopac model: EL507-10). The electrodes were placed on the distal phalanx of the first and middle fingers of the non-dominant hand (Fig. 2). SCR data were processed by removing movement manually and filtering the signal offline using a 1 Hz low pass FIR filter to eliminate noise. SCRs were identified and linked to stimulus events using the Acqknowledge software using a threshold of 0.01 microsiemens compared to a 2 second baseline. As with EMG, SCR data were excluded if participants were non-responders. Approximately 10% of the general population are SCR non-responders (Braithwaite, Watson, & Jones, 2015). For SCR, non-responders were identified as participants who displayed no event-related SCRs throughout the recording process (they did not display a 0.01 microsiemen increase in skin conductance level after stimulus onset). Data from five participants were excluded after they were identified as non-responders (10% of our sample).



Figure 2. SCR and EMG electrode placements.

2.6 Procedure: Pilot

In the pilot study participants completed a computer task where they provided subjective ratings across the six dimensions for all 78 images within the stimulus set. Participants viewed the stimuli on the 3D capable monitor with NVIDIA 3D Vision 2 Goggles while they provided ratings for the images on a laptop placed beside the 3D monitor. Stimuli were presented randomly and remained visible on the 3D monitor until participants completed rating the images on all six dimensions.

2.7 Procedure: Main study

Skin conductance electrodes were applied to a participant's non-dominant hand immediately after providing informed consent to ensure that the gel would have enough time to saturate the recording area before the computer task (saturation time >10 minutes). Participants completed the STAI Form-Y (Spielberger et al., 1983), the adapted FSQ (Szymanski & O'Donohue, 1995), and the Randot Stereovision Test (2018).

EMG electrodes were then placed under participants' left eye and SCR leads were affixed to the SCR electrodes. Both psychophysiological signals were tested for noise. For EMG, participants were asked to blink three times. The EMG signal was determined to be acceptable if the blinks were clearly distinguishable in the EMG signal. For SCR, participants were asked to take a deep breath. The SCR signal was determined to be acceptable if there was a corresponding increase in skin conductance level following the inhale. If either signal was unacceptable, the electrodes were replaced up to two more times in order to get a better signal.

Next, the participants completed the computer task which was displayed in MATLAB (2018a) using Psychtoolbox-3 (Kleiner et al., 2007). The computer task was broken down into three phases; a preparation phase, a habituation phase, and a novel phase (Fig. 3; Codispoti, Bradley & Ferrari, 2006; Codispoti, De Cesarei, Biondi & Ferrari, 2016). In the preparation phase, participants reviewed the task instructions, completed two test rating trials where they rated two test images (one 2D and one 3D image) across each subjective dimension, and were exposed to six acoustic startle probes to acclimate them to the noise. The habituation phase is broken down into three observation blocks where participants rated one set of six images repeatedly. During observation trials, participants placed their chin on a chin rest which was positioned 30 cm away

from the 3D monitor and asked not to move. There were 30 observation trials within each block where the images were presented quasi-randomly. Each observation block was broken down into five mini-blocks where each image was presented once. The same stimulus did not follow itself in either dimension (i.e. a negative 2D image would not be followed by a negative 2D or negative 3D image). Each image would follow all other images at nearly equivalent rates within one observation block. Since each image could follow four others without following itself in 2D or 3D, within the 30 trials of one block each image would follow every other image once with only four repeats. Of the 30 observation blocks, there was an acoustic startle on 3/5th of trials. A startle probe was not included on all trials to reduce participant expectancy. Startle probes were also not included on the first mini-block in block 1 or block 4 so participants could observe each image once undisturbed. Observation trials began with a fixation cross before the stimuli appeared. The stimuli was present on the screen for 8s. On startle trials, the acoustic startle probe occurred 7s after stimulus onset. The image was then followed by 15s of a fixation cross to avoid contamination of SCRs in the proceeding trial; SCRs can occur within 1 to 7s after stimulus onset and last for several seconds (Fig. 4). The acoustic startle was balanced across visual dimensionality and emotion category. Each observation block was preceded and proceeded by ratings trials where participants rate each image across the six dimensions. In the novel phase, participants viewed six new images in one block, again preceded and proceeded by rating trials. The novel phase was included to see if there is a recovery of response.

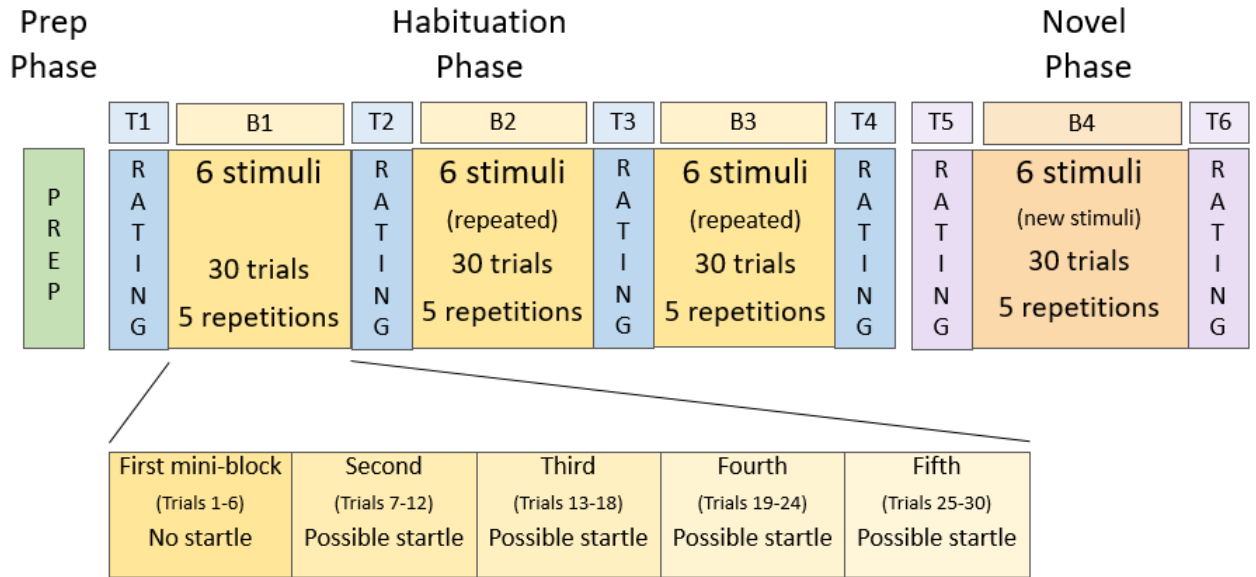


Figure 3. Computer task schematic.

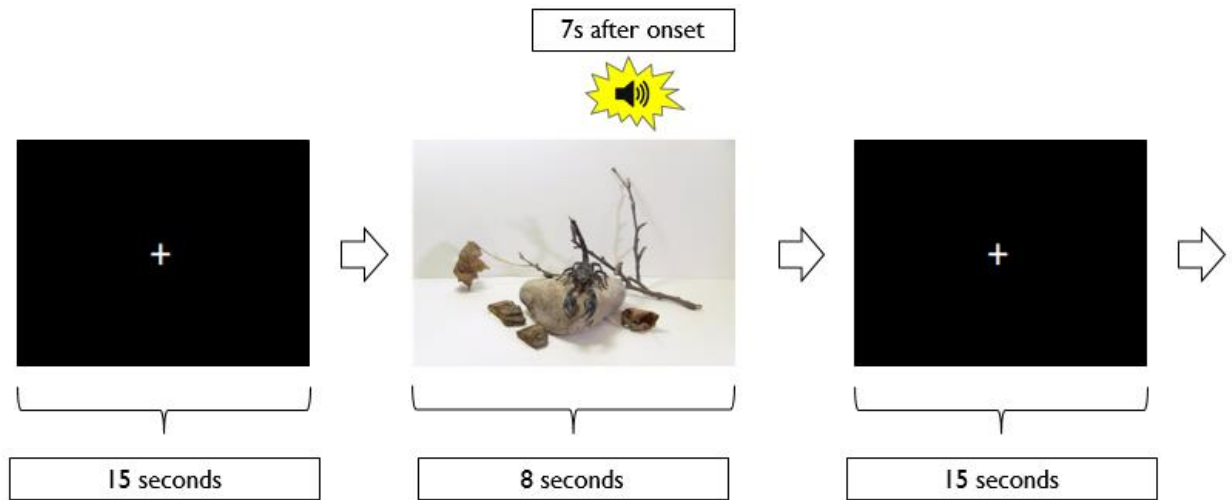


Figure 4. Example of observation trial. SCR data were collected over all 30 trials within an observation block; SCRs were considered event related if they began between 1 to 7s after stimulus onset. An acoustic startle probe was present on 3/5ths of observation trials thus EMG was collected on 18 trials within an observation block. EMG responses were measured 20-200ms after the startle probe.

2.7 Statistics

Statistical analyses were conducted using the IBM Statistical Package for Social Sciences version 25 (2017). Six 2 X 3 X 4 repeated measures ANOVAs were conducted to evaluate dimension (2D, 3D), emotion (pleasant, neutral, unpleasant), and time (1-4) for each of the six subjective ratings. Two 2 X 3 X 3 repeated measures ANOVAs were conducted to evaluate the effects of dimension (2D, 3D), emotion (pleasant, neutral, unpleasant), and block (1-3) for both psychophysiological measures. The threshold for significance was set at $p < 0.05$ for planned comparisons and post-hoc tests.

To evaluate if there was a difference in initial psychophysiological responding between the two stimulus types, 2 X 3 (Dimension, Emotion) repeated measures ANOVAs were conducted on the mean values (EMG maximum amplitude and number of SCRs) of the first block of the experiment comparing 2D and 3D images of each of the emotion categories.

To establish habituation effects in psychophysiological responding, difference scores were calculated by subtracting responses from the third block from responses in the first block to create a habituation index (where positive values represent amount of habituation). A 2 X 3 (Dimension, Emotion) repeated measures ANOVA was conducted to compare difference scores between 2D and 3D images of the same emotion category for both EMG and SCR measures.

To see if there was a recovery of response in psychophysiological responding, a recovery index was calculated by subtracting responses from the third block from responses from the fourth, novel block where participants viewed a new set of images (where positive values represent a recovery of response). Again, A 2 X 3 (Dimension, Emotion) repeated measures ANOVA was conducted to compare recovery scores between 2D and 3D images of the same emotion category for both EMG and SCR measures.

To determine if trait anxiety or fear of insects and/or spiders affected affective responding, trait anxiety and FSQ scores were correlated against mean EMG amplitude and mean number of SCRs.

CHAPTER 3

3 Results

3.1 Subjective Ratings

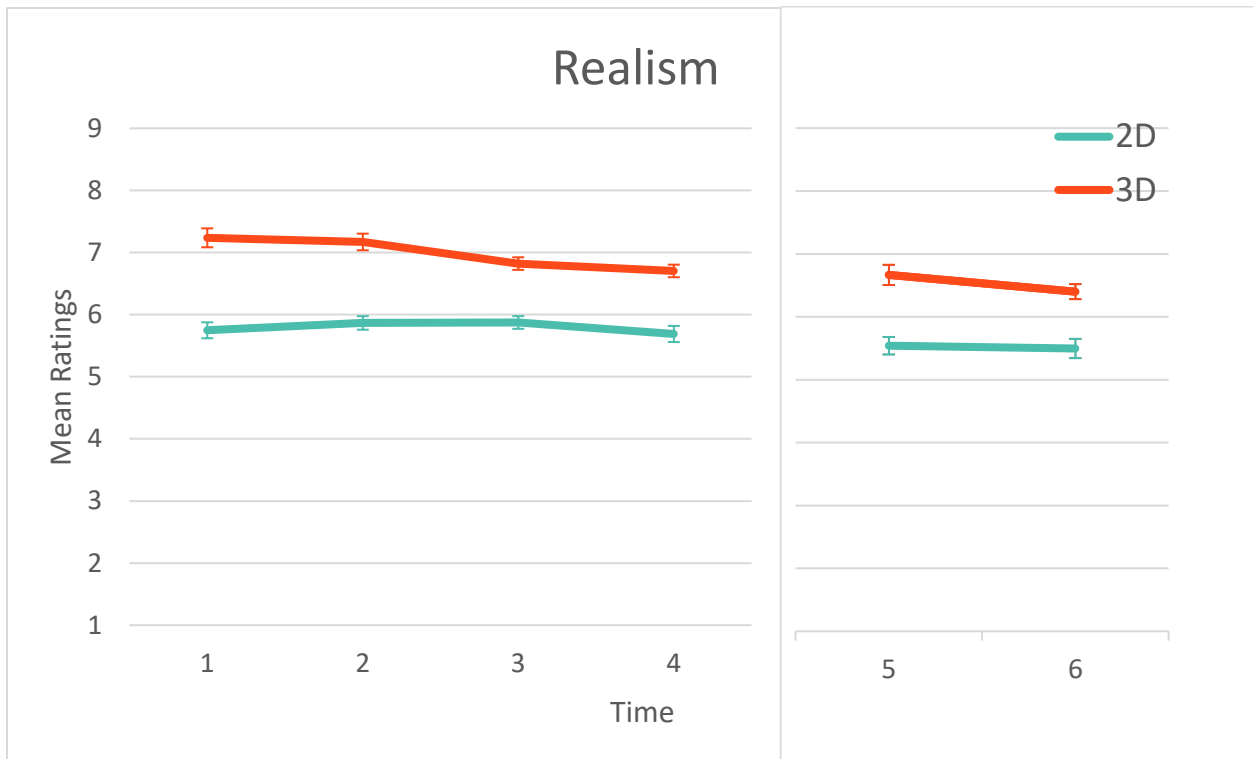
Six 2 (Dimension: 2D, 3D) X 3 (Emotion: Negative, Neutral, Positive) X 4 (Time) repeated measures ANOVAs were conducted on participants' subjective ratings of the stimuli, one for each of the six subjective dimensions (Realism, Arousal, Danger, Approachability, Pleasantness, and Visual Dimensionality).

3.1.1 Realism

The ANOVA for Realism ratings revealed a significant main effect of Dimension ($F = 51.49$, $df = 1$, $p < 0.001$, $\eta^2 = 0.505$), Emotion ($F = 4.53$, $df = 2$, $p = 0.013$, $\eta^2 = 0.093$) and Time ($F = 3.54$, $df = 2.31$, $p = 0.027$, $\eta^2 = 0.074$). 3D images were rated significantly more realistic than 2D images and negative images were rated more realistic than neutral or positive images. Realism ratings decreased over time for all stimulus types. There was a significant Dimension X Emotion interaction ($F = 3.30$, $df = 2$, $p = 0.042$, $\eta^2 = 0.070$) but no significant Dimension X Time ($F = 2.55$, $df = 2.19$, $p = 0.135$, $\eta^2 = 0.044$), Emotion X Time ($F = 0.318$, $df = 4.31$, $p = 0.878$, $\eta^2 = 0.007$), or three way interaction ($F = 0.455$, $df = 4.35$, $p = 0.784$, $\eta^2 = 0.010$).

A series of planned paired t-tests were conducted to uncover the nature of the Dimension X Emotion interaction. The t-tests revealed that 3D images were rated significantly more realistic than 2D images across all emotion categories (Negative: $t = 6.55$, $df = 44$, $p < 0.001$, $d = 0.893$; Neutral: $t = 4.02$, $df = 44$, $p < 0.001$, $d = 0.600$, Positive: $t = 3.57$, $df = 44$, $p < 0.001$, $d = 0.529$). In terms of the interaction, negative images were rated more realistic than neutral and positive images for 3D stimuli (Negative – Neutral: $t = 2.46$, $df = 44$, $p = 0.018$, $d = 0.363$; Negative – Positive: $t = 2.56$, $df = 44$, $p = 0.014$, $d = 0.381$; Neutral – Positive: $t = 2.46$, $df = 44$, $p = 0.018$, $d = 0.041$), while there were no significant differences in Realism ratings between the three emotion categories for 2D stimuli (Negative – Neutral: $t = 0.130$, $df = 44$, $p = 0.897$, $d = 0.022$; Negative – Positive: $t = 0.077$, $df = 44$, $p = 0.939$, $d = 0.015$; Neutral – Positive: $t = 0.070$, $df = 44$, $p = 0.944$, $d = 0.009$).

A)



B)

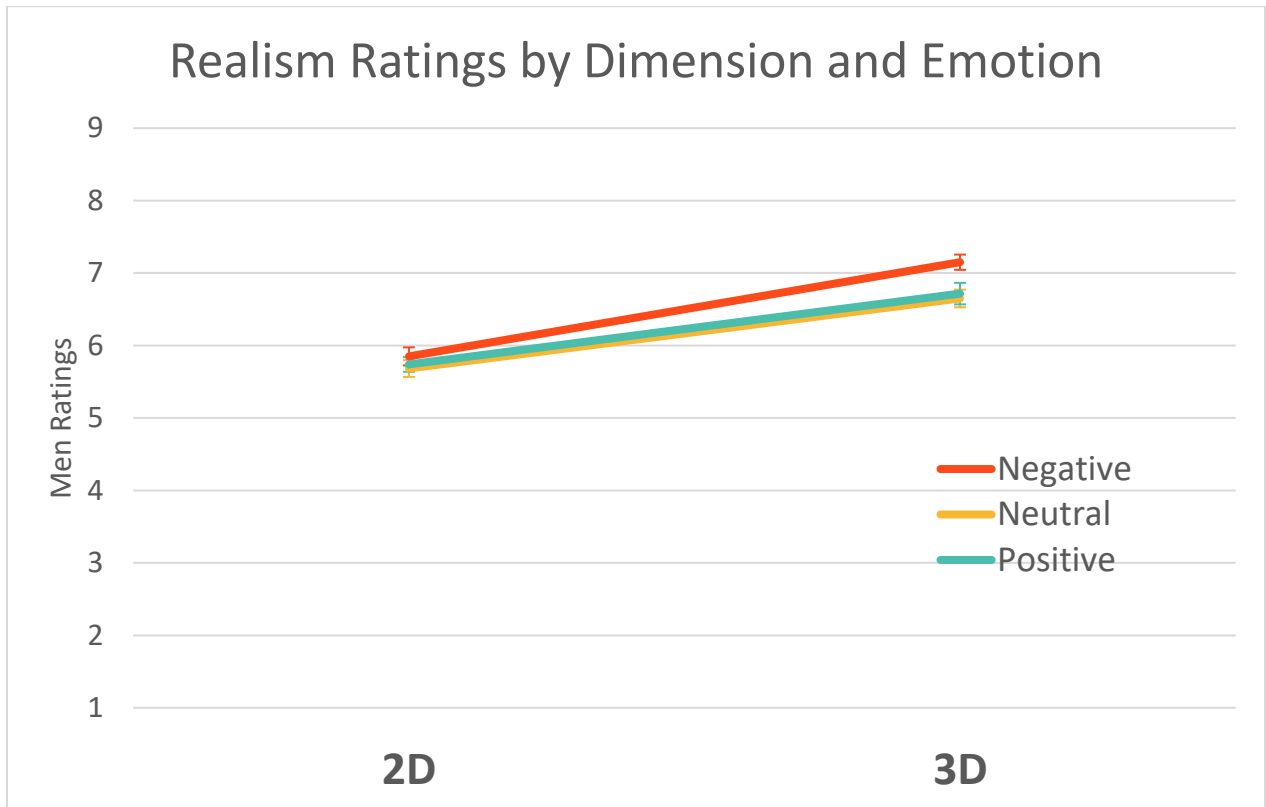


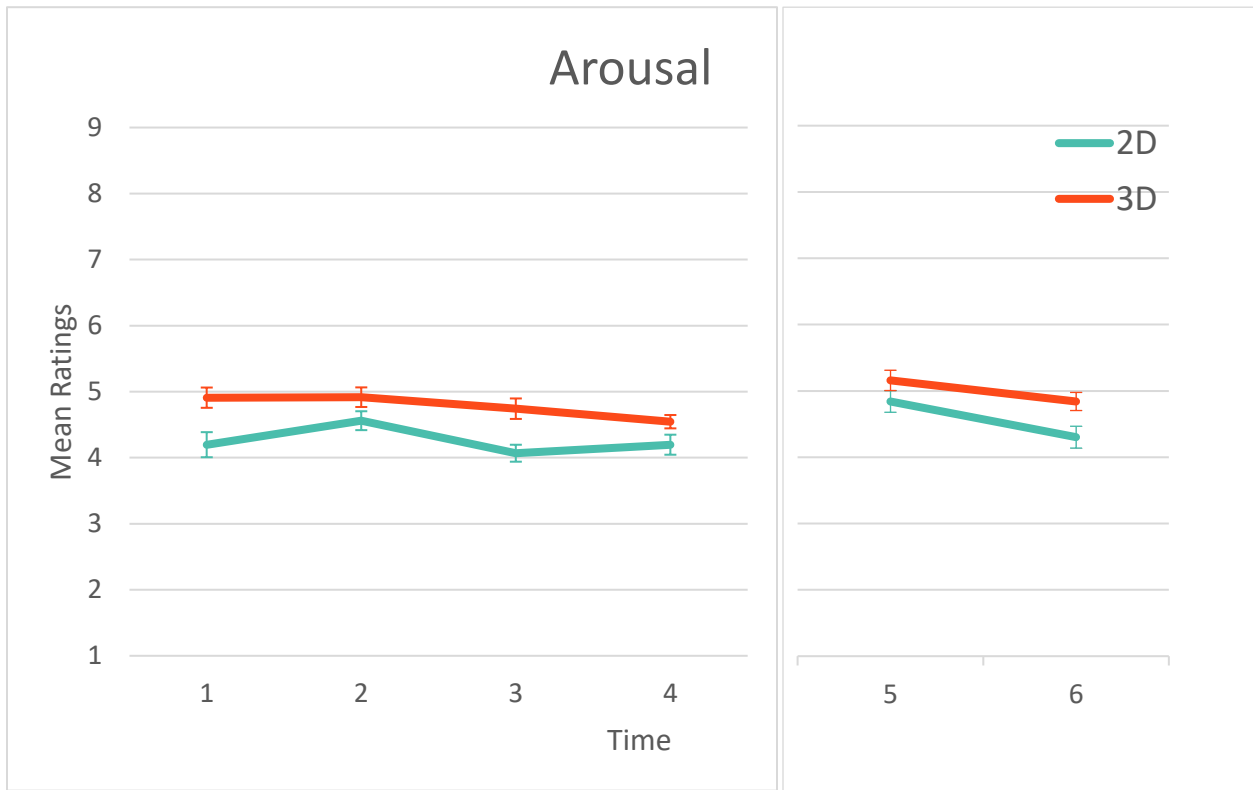
Figure 5. Mean subjective ratings of Realism; error bars represent standard error. A) Realism ratings for 2D and 3D images across the 6 points in time that participants were asked to rate images; one set of images was used for time points 1-4 and a novel set of images was used for time points 5-6. 3D images rated significantly more realistic than 2D images; Realism ratings decreased slightly for both stimulus types over time. B) There was a significant Dimension X Emotion interaction. 3D negative images were rated more realistic than neutral and positive 3D images while there was no difference in Realism ratings between emotion categories for 2D images.

3.1.2 Arousal

The ANOVA for Arousal ratings revealed a significant main effect of Dimension ($F = 22.3$, $df = 1$, $p < 0.001$, $\eta^2 = 0.317$), Emotion ($F = 3.88$, $df = 1.60$, $p = 0.033$, $\eta^2 = 0.075$) and Time ($F = 3.55$, $df = 3.00$, $p = 0.016$, $\eta^2 = 0.069$). 3D images were rated more arousing than 2D images. Negative and positive images were rated more arousing than neutral images. As with Realism, Arousal ratings decreased over time for all stimulus types. There were no significant interactions (Dimension X Emotion: $F = 1.18$, $df = 1.64$, $p = 0.306$, $\eta^2 = 0.024$; Dimension X Time: $F = 0.089$, $df = 2.54$, $p = 0.949$, $\eta^2 = 0.002$; Emotion X Time: $F = 0.648$, $df = 6.00$, $p = 0.692$, $\eta^2 = 0.013$; Dimension X Emotion X Time: $F = 0.742$, $df = 5.05$, $p = 0.616$, $\eta^2 = 0.015$).

The planned paired t-tests revealed that 3D images were rated significantly more arousing than 2D images across all emotion categories (Negative: $t = 3.42$, $df = 44$, $p < 0.001$, $d = 0.528$; Neutral: $t = 2.17$, $df = 44$, $p = 0.035$, $d = 0.374$; Positive: $t = 2.33$, $df = 44$, $p = 0.024$, $d = 0.245$). Negative and positive images were rated more arousing than neutral images for 3D stimuli (Negative – Neutral: $t = 2.27$, $df = 44$, $p < 0.001$, $d = 0.341$; Negative – Positive: $t = 0.639$, $df = 44$, $p = 0.562$, $d = 0.093$; Neutral – Positive: $t = 1.79$, $df = 44$, $p = 0.039$, $d = 0.274$) while there were no significant differences in arousal ratings between the three emotion categories for 2D stimuli (Negative – Neutral: $t = 1.16$, $df = 44$, $p = 0.250$, $d = 0.209$; Negative – Positive: $t = 0.250$, $df = 44$, $p = 0.804$, $d = 0.055$; Neutral – Positive: $t = 1.56$, $df = 44$, $p = 0.126$, $d = 0.305$).

A)



B)

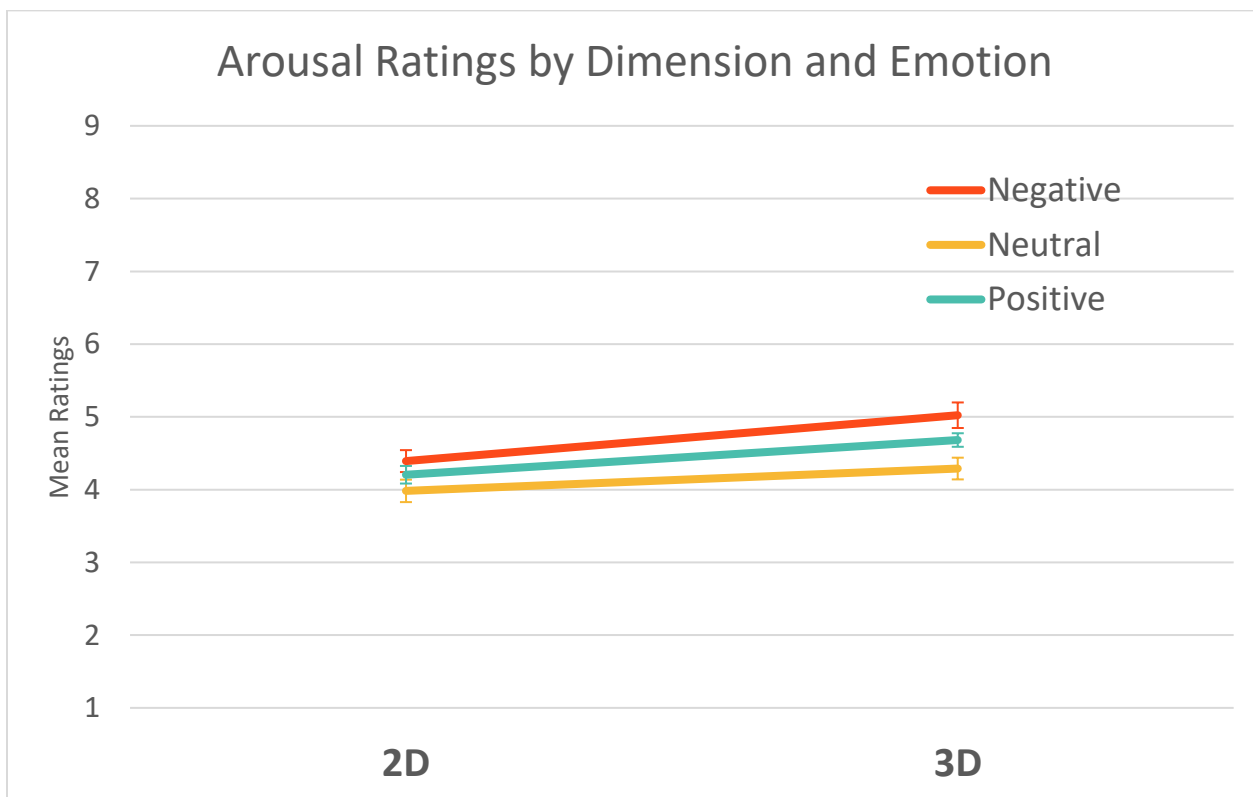


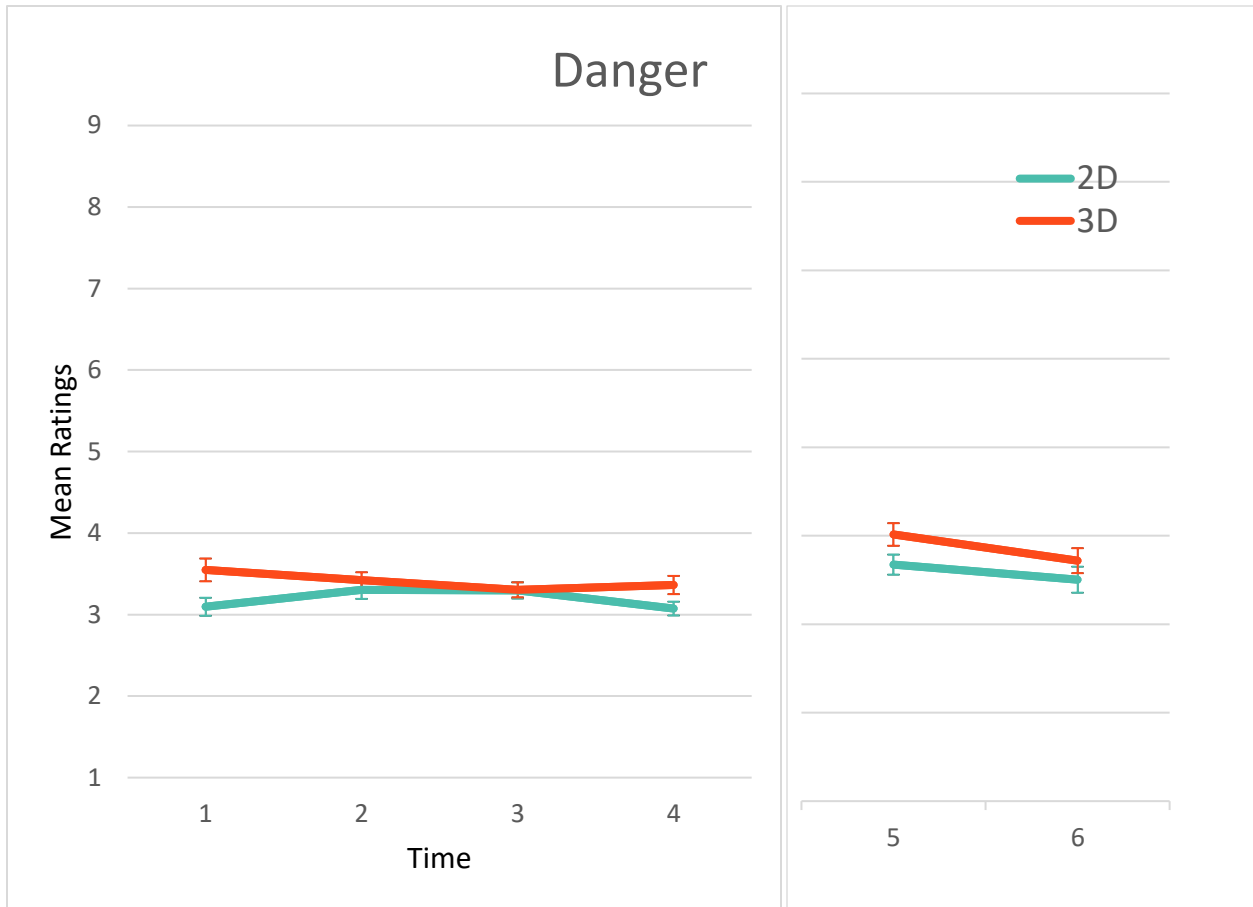
Figure 6. Mean subjective ratings of Arousal; error bars represent standard error. A) Arousal ratings for 2D and 3D images across the 6 points in time points. 3D images rated significantly more arousing than 2D images; arousal ratings decreased slightly for both stimulus types over time. B) Planned comparisons revealed 3D negative and positive images were rated more arousing than neutral images while there was no difference in arousal ratings between emotion categories for 2D images.

3.1.3 Danger

The ANOVA for Danger ratings revealed a significant main effect of Dimension ($F = 12.6$, $df = 1.00$, $p = 0.001$, $\eta^2 = 0.205$) and Emotion ($F = 76.5$, $df = 1.45$, $p < 0.001$, $\eta^2 = 0.609$) but no main effect for Time ($F = 0.837$, $df = 2.15$, $p = 0.476$, $\eta^2 = 0.071$). 3D images were rated significantly more dangerous than 2D images. As expected, negative images were rated significantly more dangerous than neutral images which were in turn rated more dangerous than positive images. There was a significant Dimension X Emotion interaction ($F = 3.74$, $df = 2.00$, $p = 0.027$, $\eta^2 = 0.071$) but no significant Dimension X Time ($F = 2.38$, $df = 2.55$, $p = 0.072$, $\eta^2 = 0.046$), Emotion X Time ($F = 0.887$, $df = 4.74$, $p = 0.505$, $\eta^2 = 0.018$), or three way interaction ($F = 0.187$, $df = 4.96$, $p = 0.967$, $\eta^2 = 0.004$).

The planned paired t-tests found that negative and neutral 3D images were rated more dangerous than negative and neutral 2D images (Negative: $t = 3.60$, $df = 44$, $p = 0.001$, $d = 0.388$; Neutral: $t = 2.74$, $df = 44$, $p = 0.008$, $d = 0.368$; Positive: $t = 0.101$, $df = 44$, $p = 0.920$, $d = 0.011$). Danger ratings for the emotion categories were as expected, with negative images being rated more dangerous than neutral images and neutral images rated more dangerous than positive images (**2D**: Negative – Neutral: $t = 7.07$, $df = 44$, $p < 0.001$, $d = 0.961$; Negative – Positive: $t = 10.4$, $df = 44$, $p < 0.001$, $d = 1.45$; Neutral – Positive: $t = 4.95$, $df = 44$, $p < 0.001$, $d = 0.502$; **3D**: Negative – Neutral: $t = 6.87$, $df = 44$, $p < 0.001$, $d = 0.905$; Negative – Positive: $t = 11.5$, $df = 44$, $p < 0.001$, $d = 1.74$; Neutral – Positive: $t = 6.82$, $df = 44$, $p < 0.001$, $d = 1.74$).

A)



B)

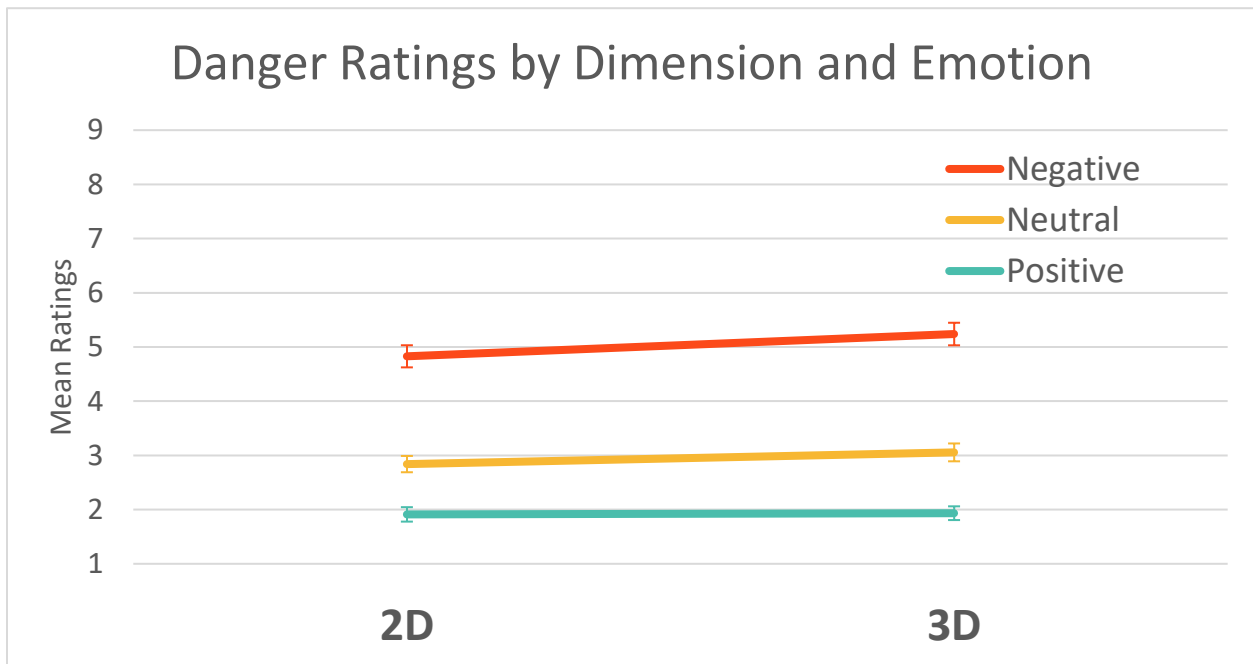


Figure 7. Mean subjective ratings of Danger; error bars represent standard error. A) Danger ratings for 2D and 3D images across the 6 points in time points. 3D images rated significantly more dangerous than 2D images. B) There was a main effect of emotion in the expected direction (Negative > Neutral > Positive) for both stimulus types. There was also a significant Dimension by Emotion interaction. 3D negative and neutral images were rated more dangerous than their 2D counterparts but there was no difference in danger ratings for positive images.

3.1.4 Approachability

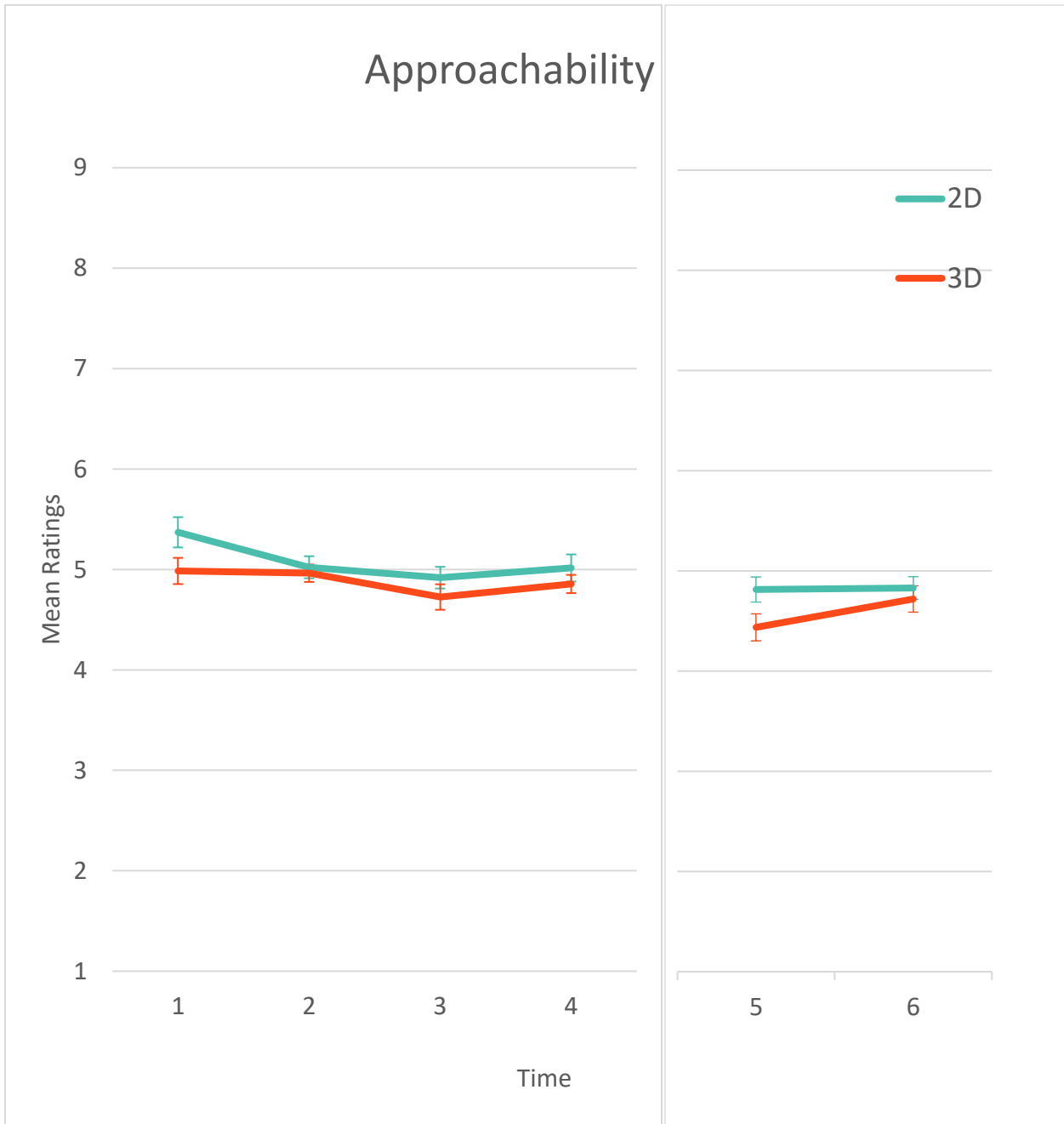
The ANOVA for Approachability ratings revealed a significant main effect of Dimension ($F = 10.28$, $df = 1.00$, $p = 0.003$, $\eta^2 = 0.189$) and Emotion ($F = 122.03$, $df = 2.00$, $p < 0.001$, $\eta^2 = 0.735$) but no main effect of Time ($F = 2.20$, $df = 2.09$, $p = 0.115$, $\eta^2 = 0.048$). 2D images were rated significantly more approachable than 3D images. As expected, positive images were rated significantly more approachable than neutral images which in turn were rated more approachable than negative images. There was a significant Emotion X Time interaction ($F = 2.39$, $df = 4.88$, $p = 0.041$, $\eta^2 = 0.051$) and a Dimension X Emotion X Time interaction ($F = 2.16$, $df = 6.00$, $p = 0.048$, $\eta^2 = 0.047$) but no Dimension X Emotion ($F = 2.76$, $df = 2.00$, $p = 0.069$, $\eta^2 = 0.059$) or Dimension X Time interaction ($F = 1.22$, $df = 3.00$, $p = 0.305$, $\eta^2 = 0.027$).

Paired t-tests were conducted to investigate the Emotion X Time interaction. Approachability ratings for positive images decreased over time ($t = 3.81$, $df = 44$, $p = 0.003$, $d = 0.373$) but there was no difference in approachability ratings for negative or neutral images over time (Negative: $t = 1.05$, $df = 44$, $p = 0.394$, $d = 0.059$; Neutral: $t = 0.822$, $df = 44$, $p = 0.311$, $d = 0.093$).

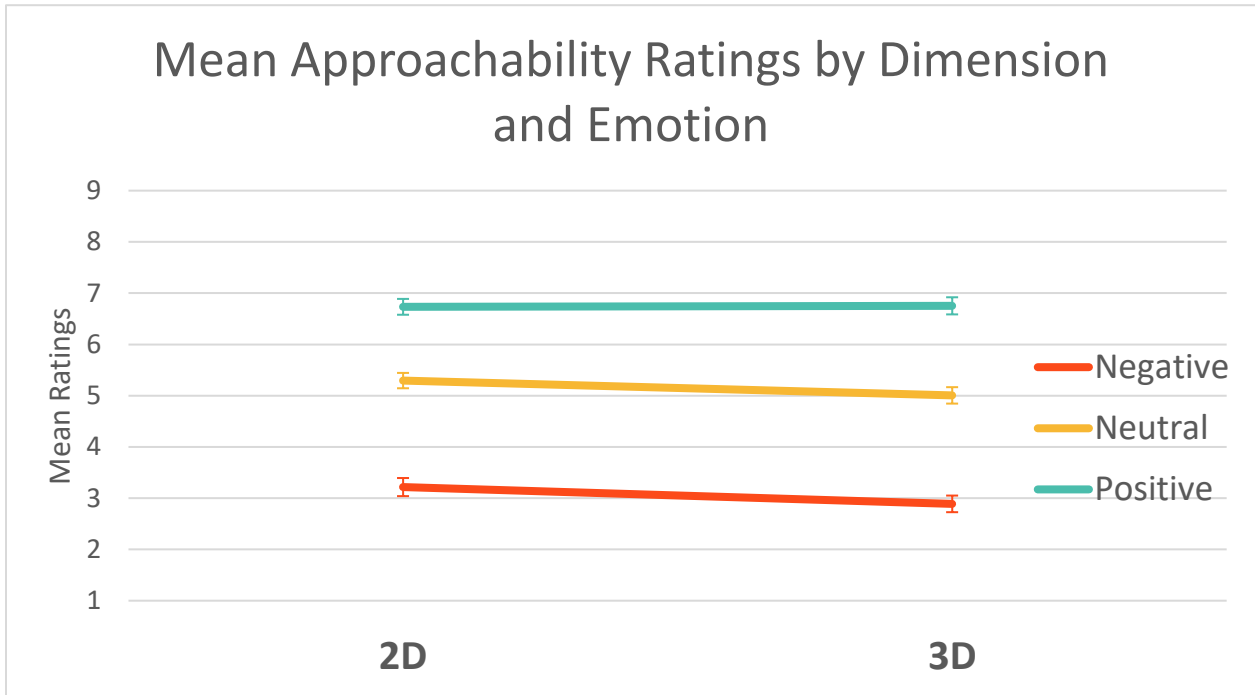
To begin to delineate the three-way interaction, three 2 X 4 (Dimension, Time) ANOVAs were conducted, one for each emotion category. The ANOVA for the negative images revealed a main effect of Dimension ($F = 10.3$, $df = 1.00$, $p = 0.003$, $\eta^2 = 0.189$) but not Time ($F = 0.338$, $df = 2.45$, $p = 0.757$, $\eta^2 = 0.008$) and no interaction ($F = 0.808$, $df = 3.00$, $p = 0.492$, $\eta^2 = 0.018$). The ANOVA for neutral images also revealed a main effect of Dimension ($F = 5.83$, $df = 1.00$, $p = 0.020$, $\eta^2 = 0.117$) but not Time ($F = 2.02$, $df = 2.23$, $p = 0.135$, $\eta^2 = 0.044$) and no interaction ($F = 0.882$, $df = 3.00$, $p = 0.452$, $\eta^2 = 0.020$). The ANOVA for positive images revealed a main

effect of Time ($F = 5.37$, $df = 2.29$, $p = 0.004$, $\eta_p^2 = 0.109$) but not Dimension ($F = 0.03$, $df = 1.00$, $p = 0.865$, $\eta_p^2 = 0.001$) and no interaction ($F = 3.89$, $df = 3.00$, $p = 0.011$, $\eta_p^2 = 0.081$). To further define the nature of the three-way interaction, a series of paired t-tests were conducted comparing 2D and 3D negative, neutral, and positive images at each time point. These t-tests revealed that the difference between 2D and 3D negative images was greatest in time point 4 ($t = 2.61$, $df = 44$, $p = 0.012$, $d = 0.394$; 2D > 3D) but the difference between 2D and 3D positive and neutral images was not significant during time point 4 (Positive: $t = 0.83$, $df = 44$, $p = 0.411$, $d = 0.111$; Neutral: $t = 0.35$, $df = 44$, $p = 0.730$, $d = 0.039$). The difference between 2D and 3D positive images was significant during time point 1 ($t = 2.78$, $df = 44$, $p = 0.008$, $d = 0.399$) while the difference between 2D and 3D negative and neutral images was not significant during this time point (Negative: $t = 1.60$, $df = 44$, $p = 0.118$, $d = 0.260$; Neutral: $t = 1.11$, $df = 44$, $p = 0.274$, $d = 0.154$). Finally, the difference between 2D and 3D neutral images was greatest for time point 3 ($t = 2.07$, $df = 44$, $p = 0.044$, $d = 0.346$) while the differences between 2D and 3D positive and negative images was not significant during this time point (Positive: $t = 1.30$, $df = 44$, $p = 0.202$, $d = 0.119$; Negative: $t = 0.780$, $df = 44$, $p = 0.437$, $d = 0.173$).

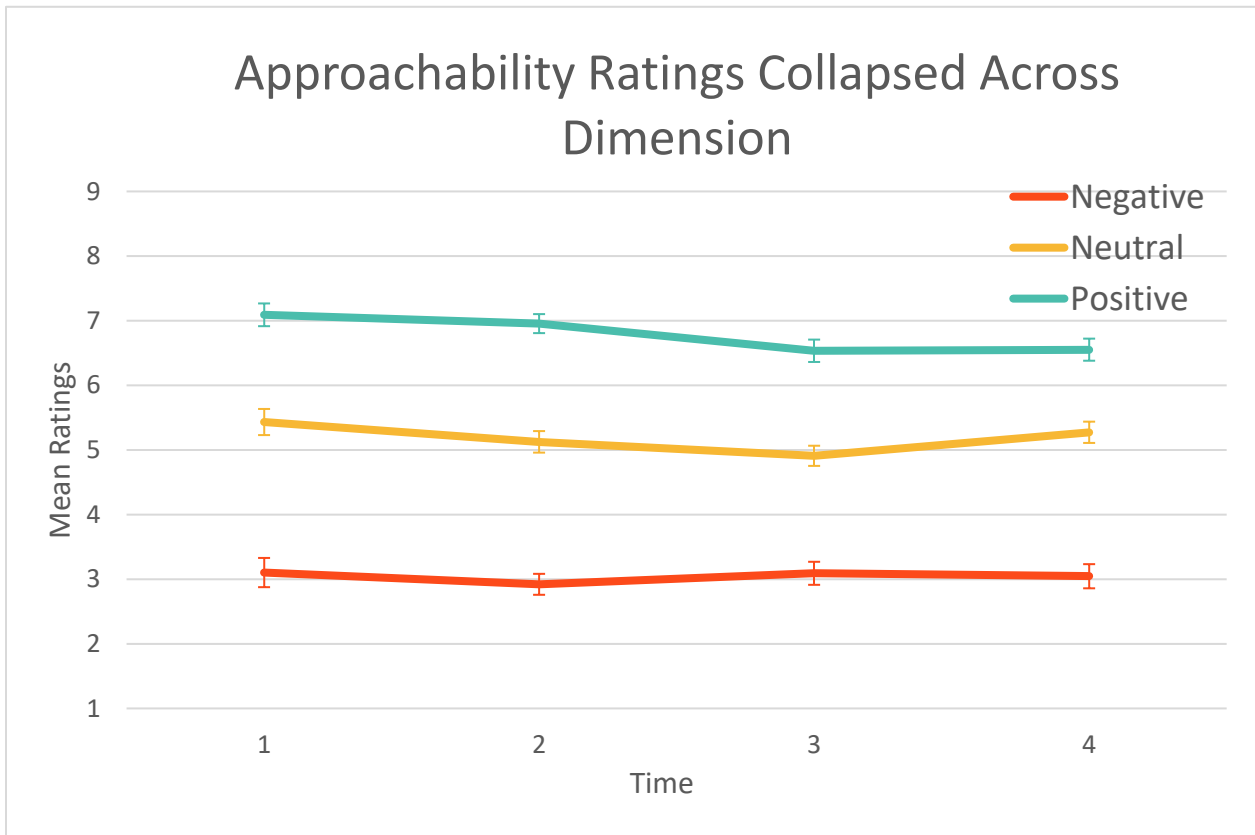
A)



B)



C)



D)

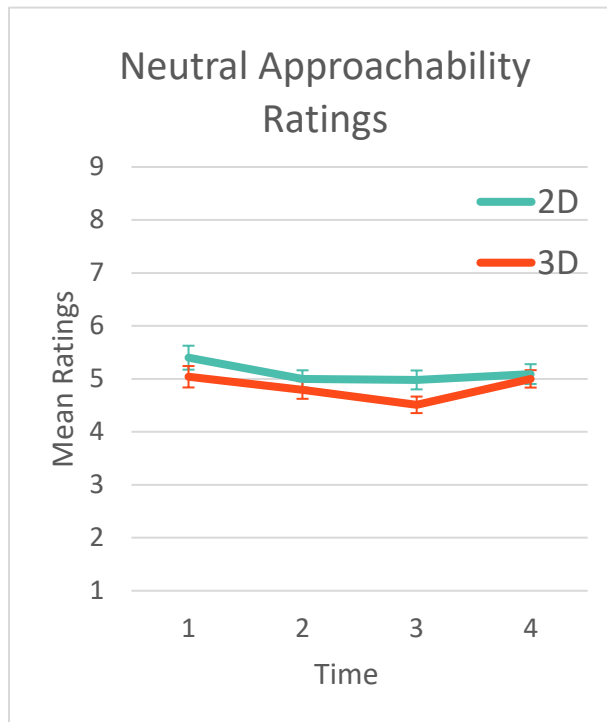
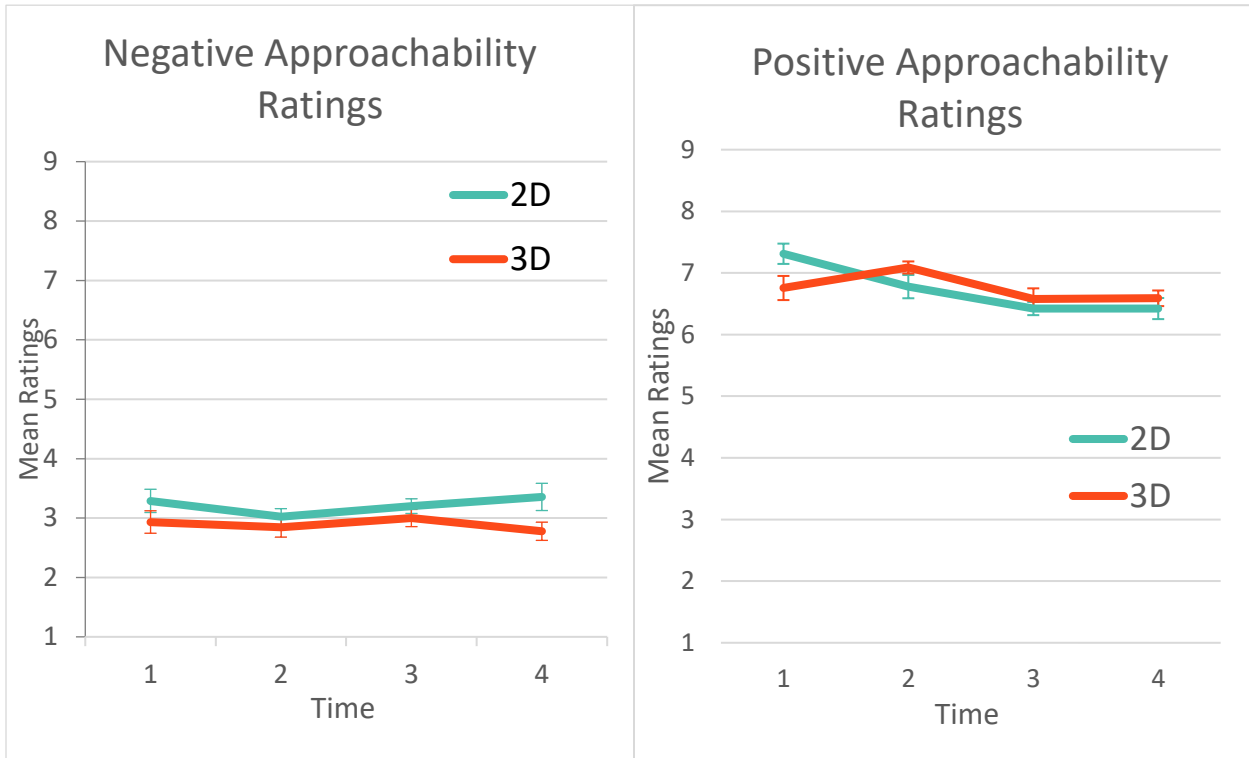
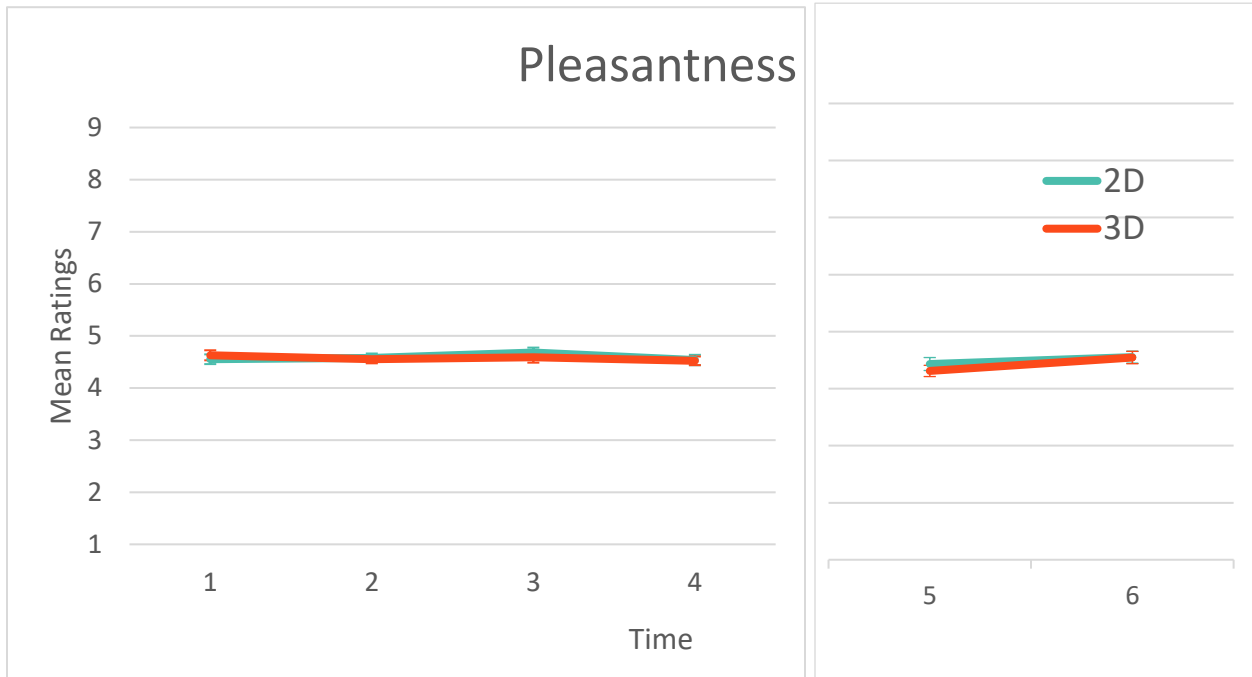


Figure 8. Mean subjective ratings of Approachability; error bars represent standard error. A) Approachability ratings for 2D and 3D images across the 6 points in time points. B) Approachability ratings showing main effect of emotion (Positive > Neutral > Negative). There was a significant Dimension by Emotion interaction. 3D negative and neutral images were rated less approachable than 2D images. C) Emotion by Time interaction. D) There was a significant three-way interaction. Between 2D and 3D images, negative images were significantly different in time point 4, positive images in time point 1, and neutral images in time point 3.

3.1.5 Pleasantness

The ANOVA for Pleasantness ratings revealed a significant main effect of Emotion ($F = 106.5$, $df = 2.00$, $p < 0.001$, $\eta_p^2 = 0.712$) but no main effect of Dimension ($F = 0.049$, $df = 1.00$, $p = 0.826$, $\eta_p^2 = 0.001$) or Time ($F = 0.454$, $df = 2.32$, $p = 0.665$, $\eta_p^2 = 0.010$). There were no significant interactions (Dimension X Emotion: $F = 2.27$, $df = 1.67$, $p = 0.119$, $\eta_p^2 = 0.050$; Dimension X Time: $F = 0.409$, $df = 3.00$, $p = 0.747$, $\eta_p^2 = 0.009$; Emotion X Time: $F = 1.59$, $df = 6.00$, $p = 0.150$, $\eta_p^2 = 0.036$; Dimension X Emotion X Time: $F = 1.56$, $df = 6.00$, $p = 0.160$, $\eta_p^2 = 0.035$). The Pleasantness ratings were as expected for the emotional categories with positive images being rated more pleasant than neutral images which were in turn rated more pleasant than negative images for both stimulus types (Negative – Neutral: $t = 6.77$, $df = 44$, $p < 0.001$, $d = 1.02$; Negative – Positive: $t = 14.0$, $df = 44$, $p < 0.001$, $d = 2.11$; Neutral – Positive: $t = 8.13$, $df = 44$, $p < 0.001$, $d = 1.22$).

A)



B)

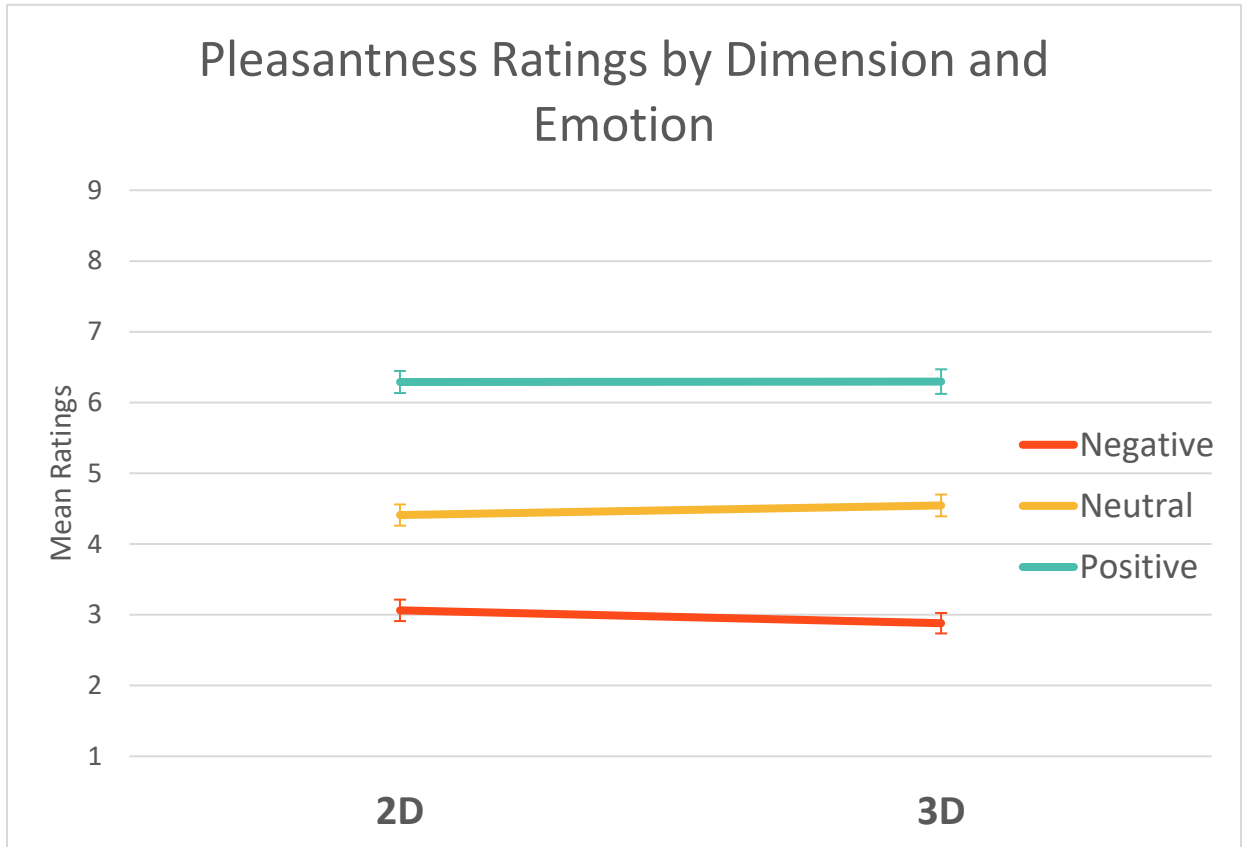
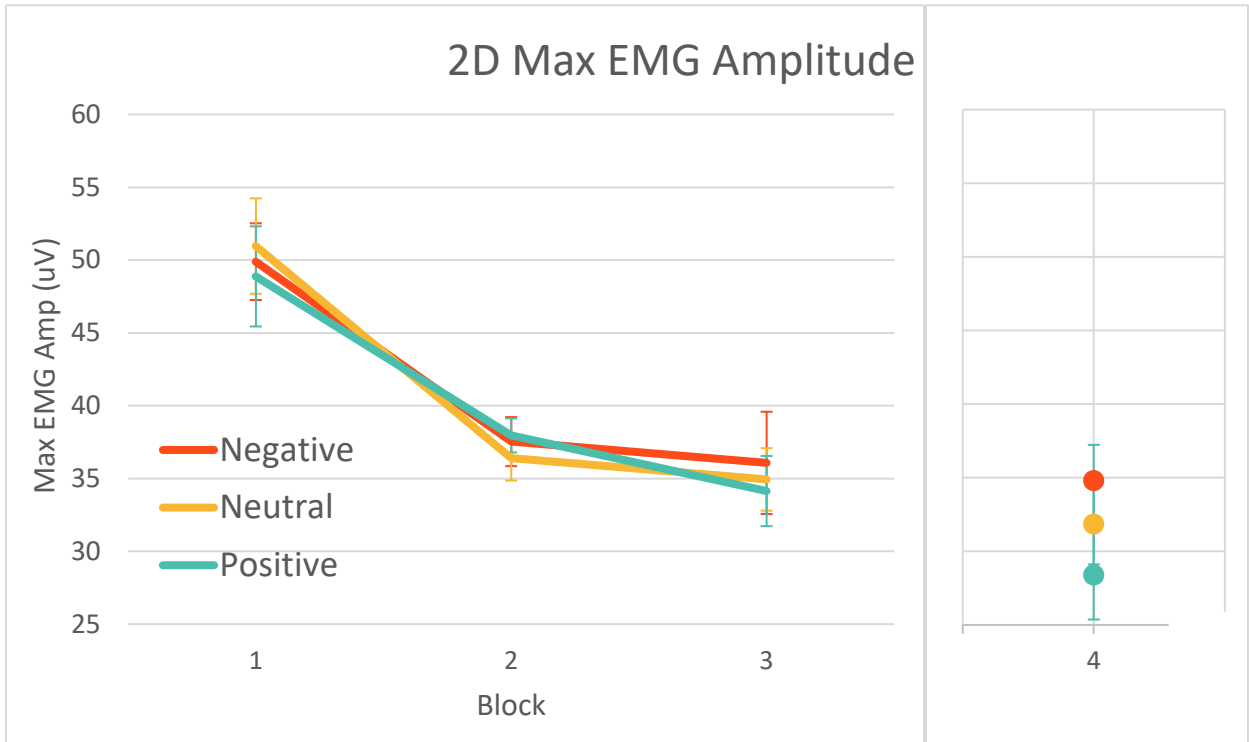


Figure 9. Mean subjective ratings of Pleasantness; error bars represent standard error. A) Pleasantness ratings for 2D and 3D images across the 6 points in time points. There were no significant differences between pleasantness ratings across dimension or time. B) There was a main effect of Emotion with pleasantness ratings in the expected direction (Positive > Neutral > Negative) for both stimulus types.

3.2 Electromyography results

As with the analysis of subjective ratings, a 2 X 3 X 3 (Dimension, Emotion, Block) repeated measures ANOVA was conducted on the mean EMG amplitude scores. A Greenhouse-Geisser correction was used for all main effects and interactions that had a significant Mauchly's Test of Sphericity ($p > 0.05$). A main effect of Block was uncovered ($F = 21.78$, $df = 1.14$, $p < 0.001$, $\eta_p^2 = 0.331$) but there was no main effect of Dimension ($F = 0.71$, $df = 1.00$, $p = 0.404$, $\eta_p^2 = 0.016$) or Emotion ($F = 0.12$, $df = 2.00$, $p = 0.884$, $\eta_p^2 = 0.003$). No significant interactions were discovered (Dimension X Emotion: $F = 0.048$, $df = 1.44$, $p = 0.905$, $\eta_p^2 = 0.001$; Dimension X Time: $F = 0.918$, $df = 2.00$, $p = 0.403$, $\eta_p^2 = 0.020$; Emotion X Time: $F = 1.11$, $df = 4.00$, $p = 0.355$, $\eta_p^2 = 0.025$; Dimension X Emotion X Time: $F = 0.453$, $df = 2.49$, $p = 0.680$, $\eta_p^2 = 0.010$).

A)



B)

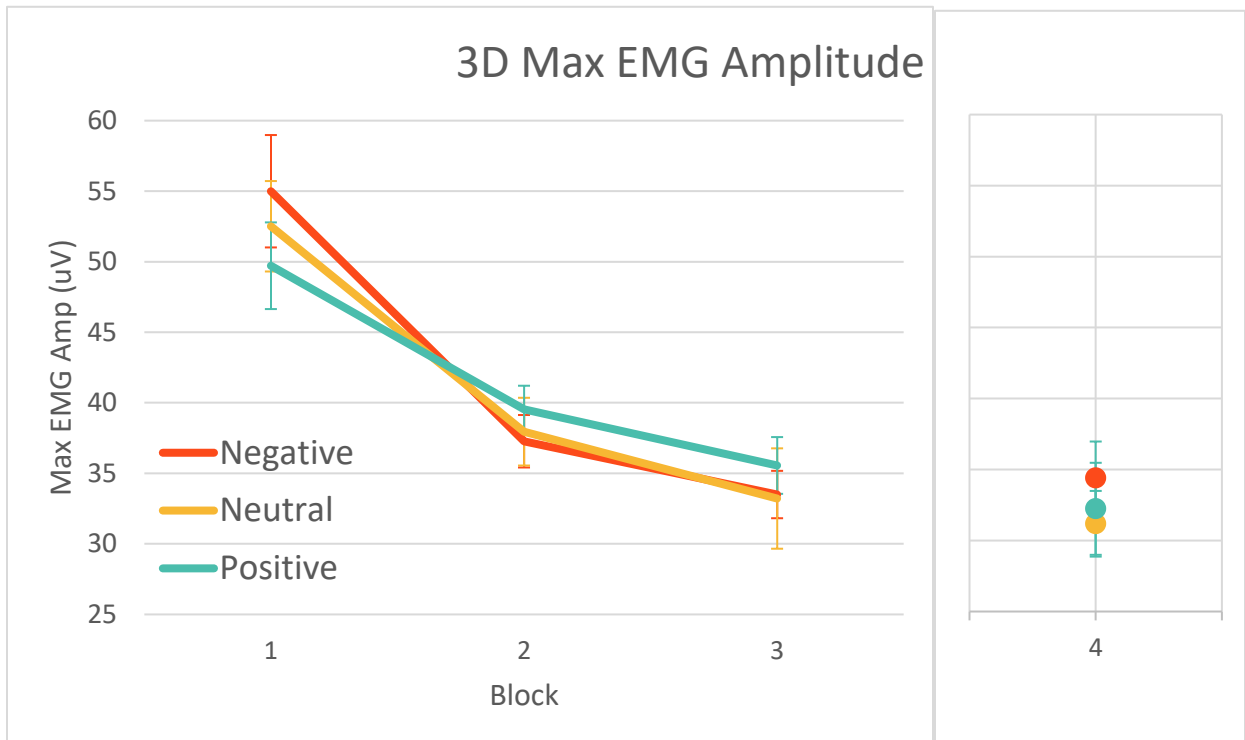


Figure 10. Mean maximum EMG amplitude after startle probe compared to a baseline; error bars represent standard error. A) EMG response to 2D images across blocks for each of the emotion categories. Participants observed one set of images for blocks 1-3 and a novel set of images for block 4. There was a significant decrease in EMG response over time for all emotion categories. B) EMG response to 3D images over time for each emotion category. As with 2D images, there was a significant reduction in EMG responding over time.

3.2.1. EMG first block

The first block was investigated more closely to determine if there were any differences in initial EMG responding between the two stimulus types. A Dimension X Emotion repeated measures ANOVA was conducted; there was not a main effect of Dimension ($F = 1.59$, $df = 1.00$, $p = 0.214$, $\eta_p^2 = 0.035$) or Emotion ($F = 1.19$, $df = 2.00$, $p = 0.930$, $\eta_p^2 = 0.026$) and the interaction was not significant ($F = 0.360$, $df = 1.63$, $p = 0.65$, $\eta_p^2 = 0.008$).

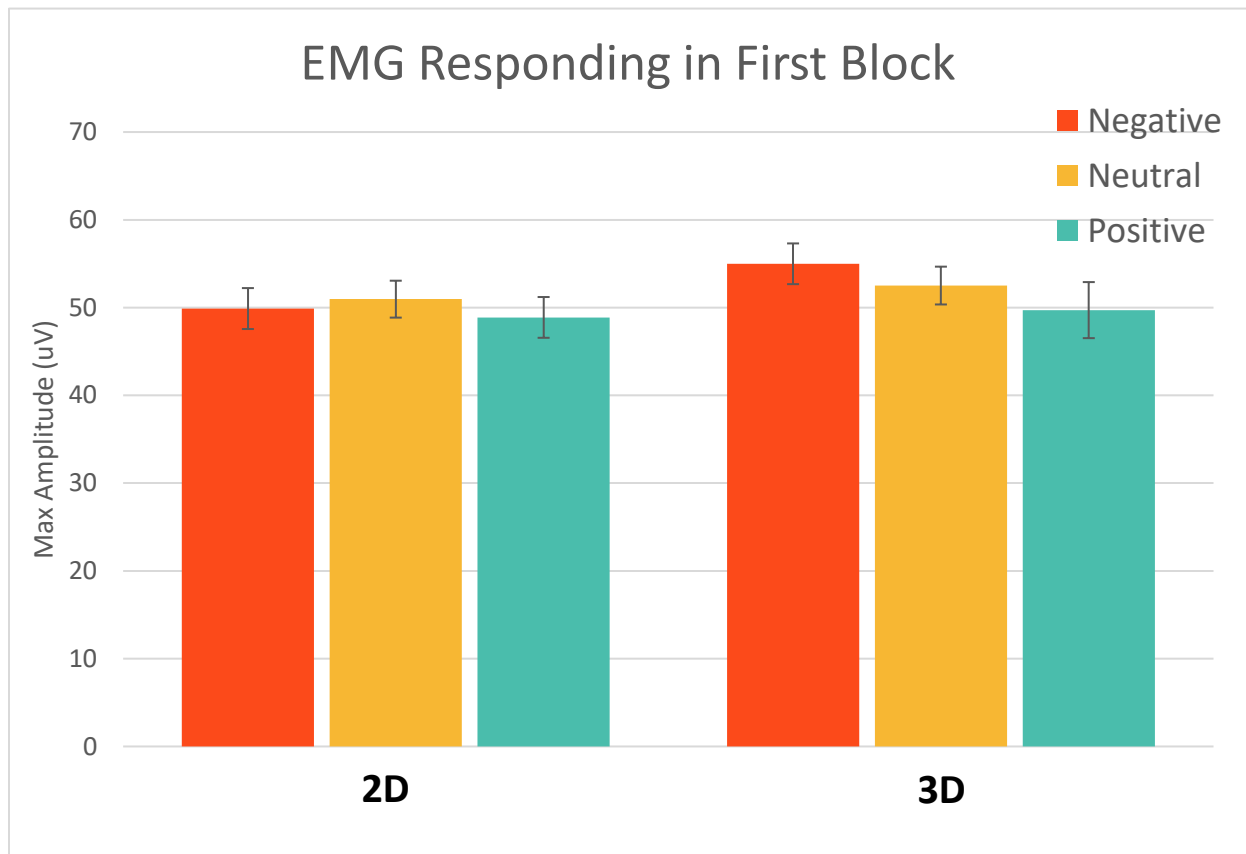


Figure 11. Mean maximum EMG amplitude in the first block of the experiment; error bars represent standard error. There were no significant differences in initial responding between 2D and 3D images. There was also no significant emotional modulation of the eye blink startle response.

3.2.2. EMG habituation

A habituation index was created by calculating difference score between the third and first block (Block 3 – Block 1) of the experiment. The third block was used as this is the last time participants observe the first set of images (a new set of images is presented in the fourth block). Positive scores on the habituation index indicate greater habituation. A Dimension X Emotion repeated measures ANOVA was conducted. There were no significant differences in habituation effects based on Dimension ($F = 1.75$, $df = 1.00$, $p = 0.192$, $\eta_p^2 = 0.038$) or Emotion ($F = 1.10$, $df = 2.00$, $p = 0.338$, $\eta_p^2 = 0.024$) and no significant interaction ($F = 0.619$, $df = 1.71$, $p = 0.517$, $\eta_p^2 = 0.014$).

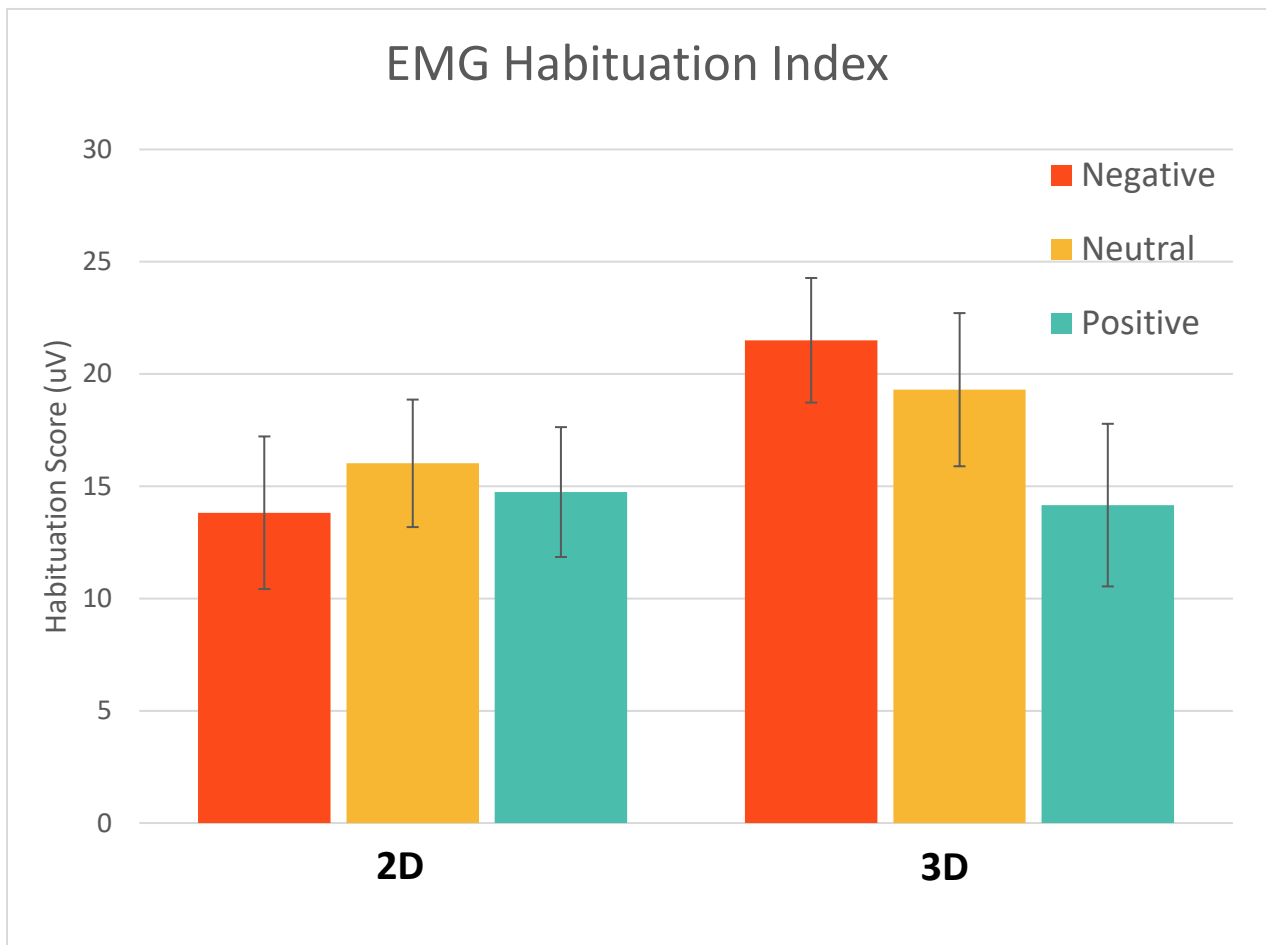


Figure 12. EMG Habituation Index; error bars represent standard error. There was no significant difference in habituation effects between the two dimensions.

3.2.3 EMG recovery

A Recovery Index was created by subtracting EMG response from the fourth, novel block from the third block (Block 4 – Block 3). Positive scores on the recovery index would indicate a recovery of response. A Dimension X Emotion repeated measures ANOVA was conducted. There were no significant differences in response recovery based on Dimension ($F = 0.654$, $df = 1.00$, $p = 0.423$, $\eta_p^2 = 0.015$) or Emotion ($F = 1.46$, $df = 1.67$, $p = 0.238$, $\eta_p^2 = 0.032$) and no significant interaction ($F = 0.020$, $df = 1.72$, $p = 0.969$, $\eta_p^2 = 0.00$).

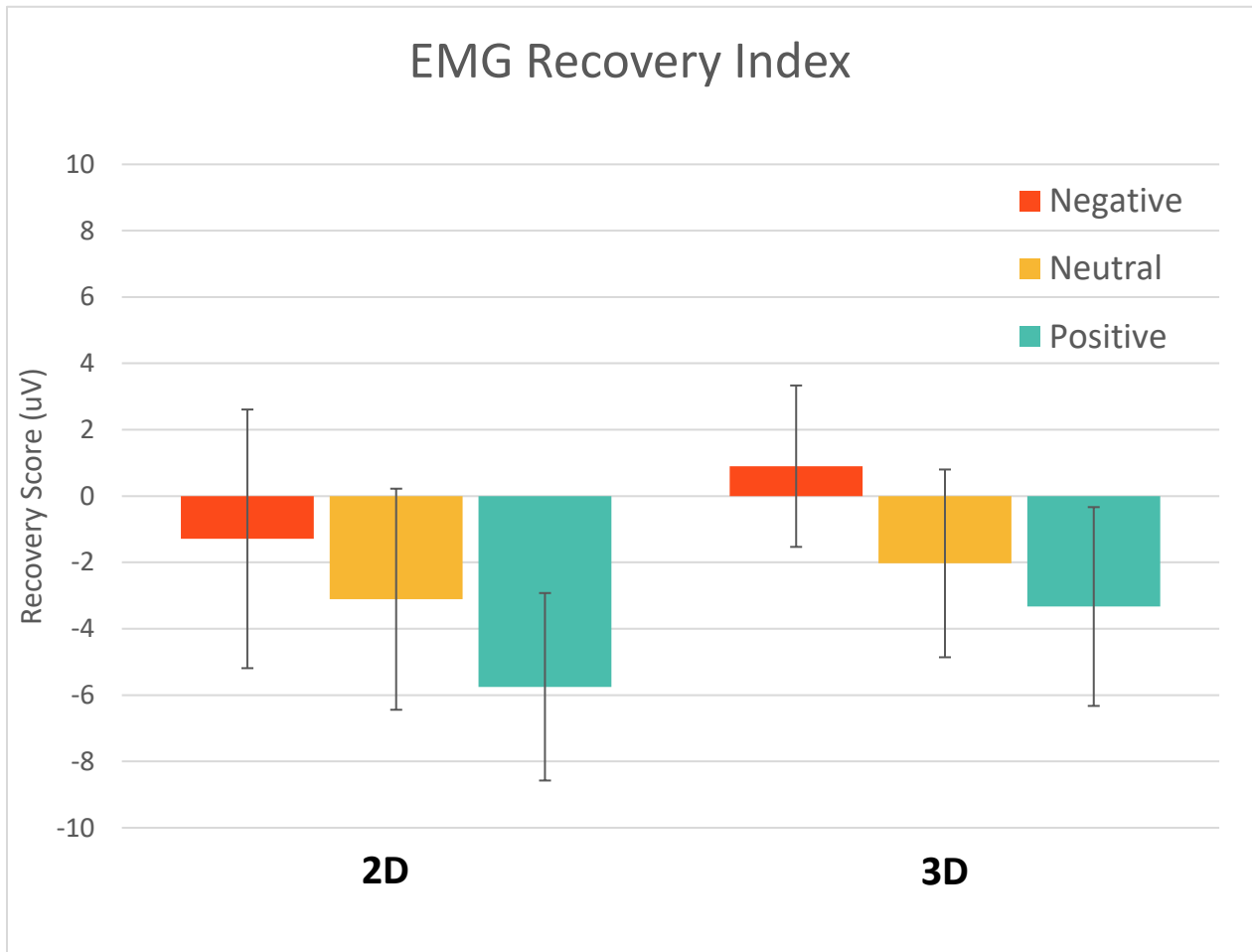


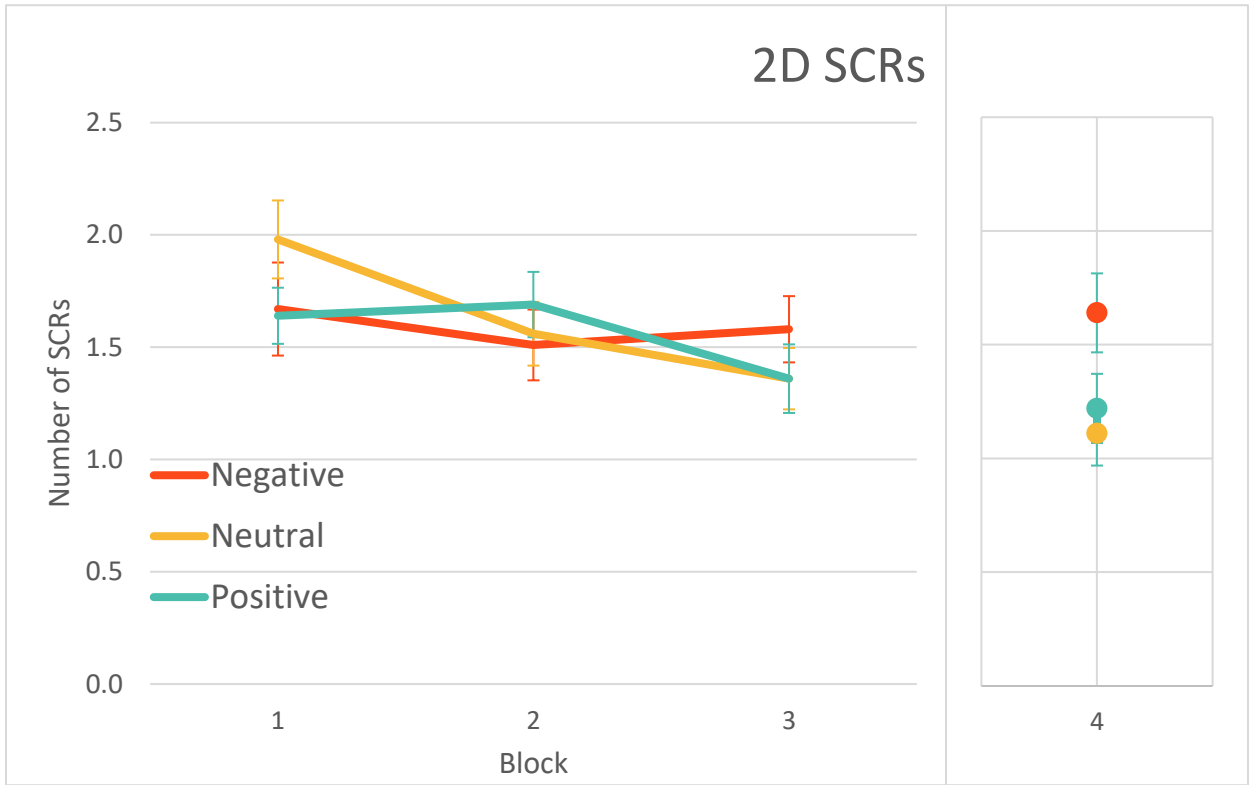
Figure 13. EMG Recovery Index; error bars represent standard error. There was no significant difference in recovery effects between the two dimensions.

3.3 Skin conductance response results

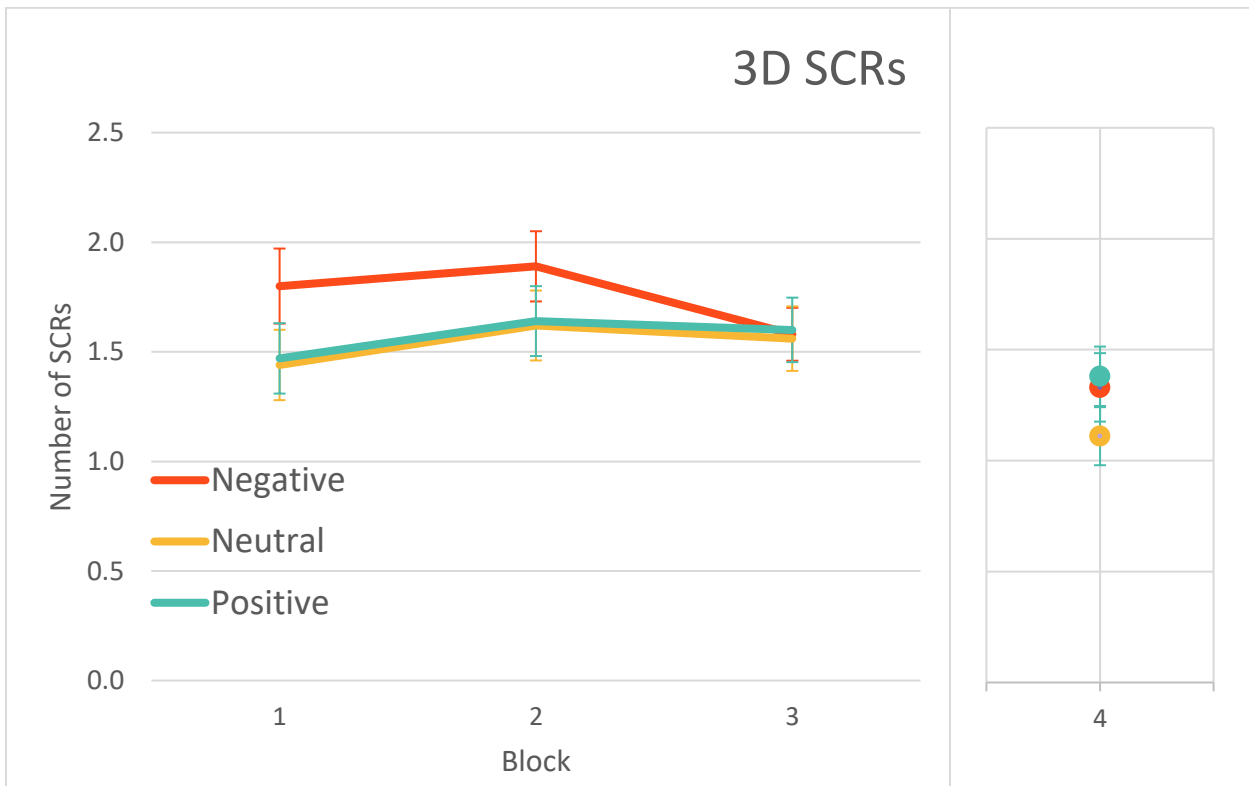
A 2 X 3 X 3 (Dimension, Emotion, Block) repeated measures ANOVA was conducted on the mean number of SCRs. A Greenhouse-Geisser correction was used as Mauchly's test for Sphericity was significant for all main effects and interactions ($p > 0.05$). No significant main effects were uncovered (Dimension: $F = 0.231$, $df = 1.00$, $p = 0.633$, $\eta_p^2 = 0.005$; Emotion: $F = 0.837$, $df = 2.00$, $p = 0.436$, $\eta_p^2 = 0.019$; Block: $F = 0.906$, $df = 2.00$, $p = 0.408$, $\eta_p^2 = 0.020$) but there was a significant Dimension by Block interaction ($F = 3.94$, $df = 2.00$, $p = 0.023$, $\eta_p^2 = 0.082$). No other interactions were significant (Dimension X Emotion: $F = 1.37$, $df = 2.00$, $p = 0.259$, $\eta_p^2 = 0.020$; Emotion X Time: $F = 0.415$, $df = 4.00$, $p = 0.789$, $\eta_p^2 = 0.009$; Dimension X Emotion X Time: $F = 1.61$, $df = .400$, $p = 0.175$, $\eta_p^2 = 0.035$).

Collapsing across emotion, a 2 X 3 repeated measures ANOVA was conducted to delineate the Dimension X Block interaction. There were no main effects of Dimension ($F = 0.231$, $df = 1$, $p = 0.633$, $\eta_p^2 = 0.005$) or Block ($F = 0.906$, $df = 2$, $p = 0.408$, $\eta_p^2 = 0.020$) but a Dimension X Block interaction was discovered ($F = 3.94$, $df = 2$, $p = 0.023$, $\eta_p^2 = 0.082$). Paired t-tests revealed this interaction was driven by the difference in response to 2D images in the first block and the third block where there was significantly more response in the first block compared to the third block ($t = 2.16$, $df = 44$, $p = 0.036$, $d = 0.320$). There was no significant difference in the number of SCRs for 3D images across blocks ($t = -0.046$, $df = 44$, $p = 0.964$, $d = 0.009$).

A)



B)



C)

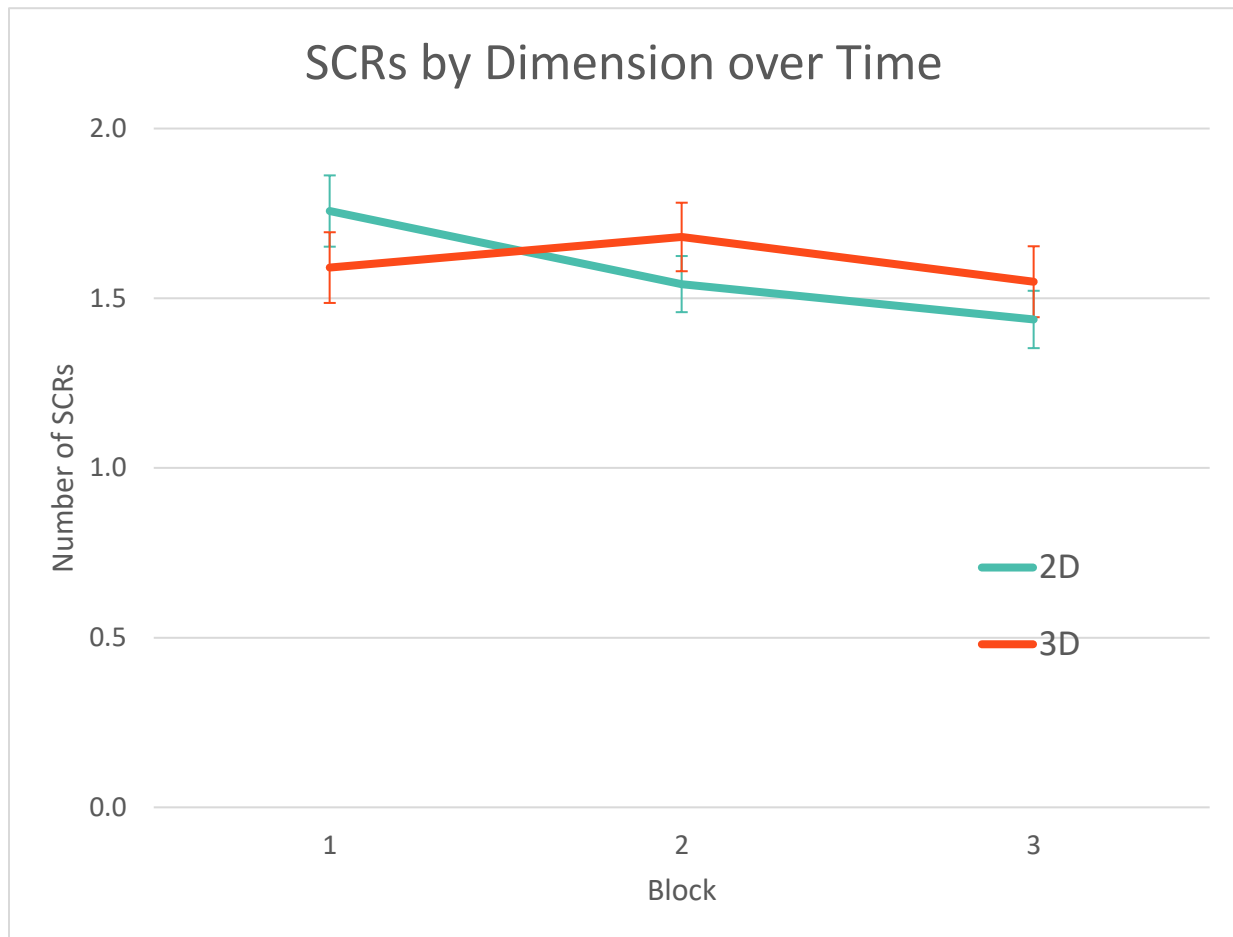


Figure 14. Mean amount of SCRs; error bars represent standard error. A) Number of SCRs to 2D images across blocks for each emotion category. B) Number of SCRs to 3D images across blocks for each emotion category. C) Number of SCRs by Dimension across time. The number of SCRs in response to 2D images decreased across blocks while there was no difference in SCRs to 3D images across blocks.

3.3.1 SCR first block

The first block was investigated more closely to determine if there were any differences in initial SCR responding between the two stimulus types. A Dimension X Emotion repeated measures ANOVA was conducted; no main effect of Dimension ($F = 1.19$, $df = 1.00$, $p = 0.276$, $\eta_p^2 = 0.004$) or Emotion ($F = 0.402$, $df = 2.00$, $p = 0.670$, $\eta_p^2 = 0.003$) was found. The interaction was not found to be significant ($F = 1.19$, $df = 2$, $p = 0.305$, $\eta_p^2 = 0.009$).

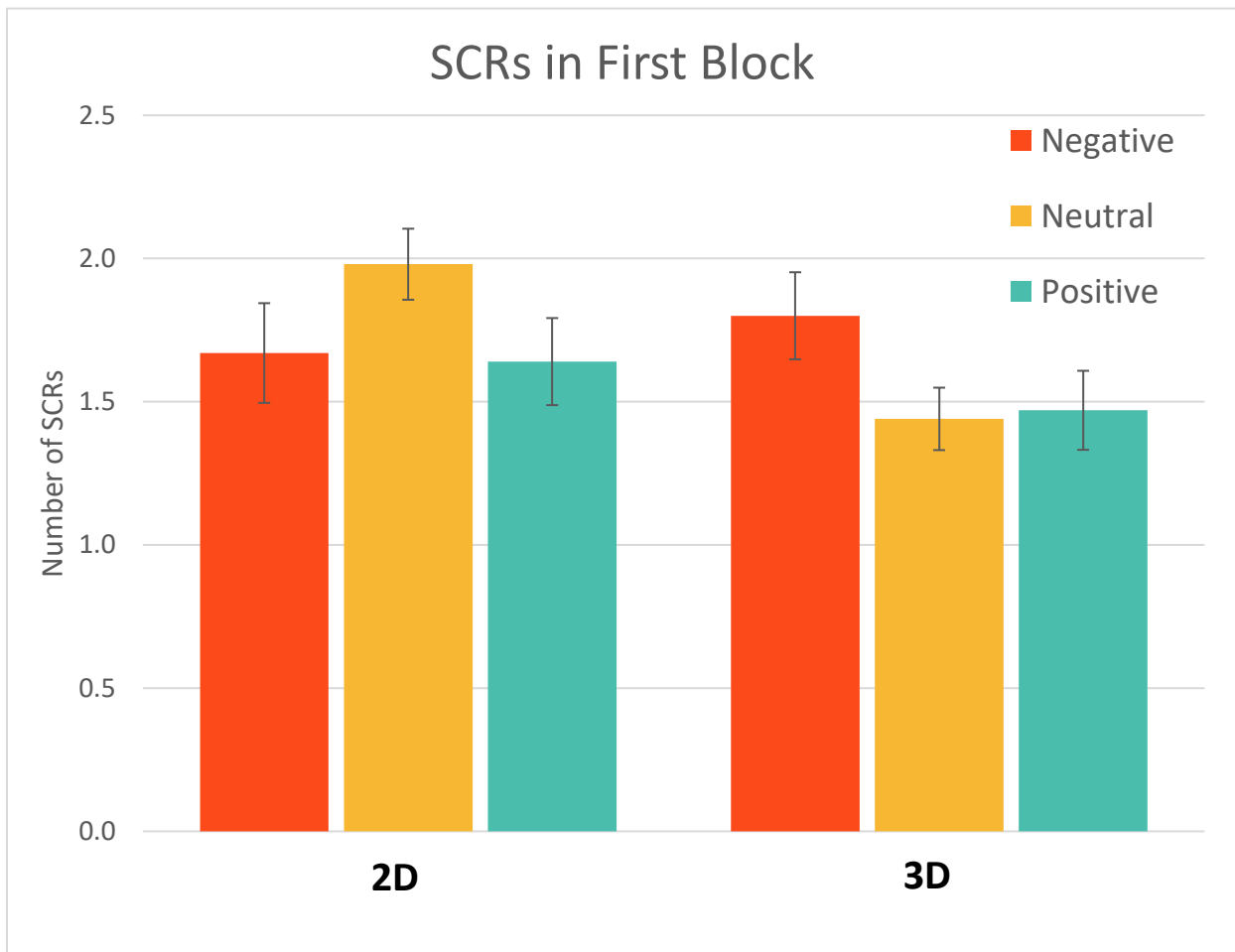


Figure 15. Mean number of SCRs in the first block of the experiment; error bars represent standard error. There was a significant difference in initial responding between 2D and 3D neutral images where 2D elicited more SCRs but no difference between negative or positive images between the two dimensions. Neutral 2D images elicited more SCRs than positive 2D images but there was no other significant difference between images of different emotion categories within a dimension.

3.3.2 SCR Habituation

A Habituation Index was created for SCRs, again difference scores were calculated by subtracting SCRs from the third block from the first block to create a habituation score (where higher scores represent more habituation). A Dimension X Emotion repeated measures ANOVA revealed a main effect of Dimension ($F = 7.36$, $df = 1.00$, $p = 0.009$, $\eta_p^2 = 0.143$) but not of Emotion ($F = 0.412$, $df = 2.00$, $p = 0.663$, $\eta_p^2 = 0.009$). 2D images experienced greater habituation compared to 3D images. The Dimension X Emotion interaction was found to be significant ($F = 3.22$, $df = 2.00$, $p = 0.045$, $\eta_p^2 = 0.068$).

Paired t-tests were conducted to delineate the Dimension X Emotion interaction. There was greater habituation for neutral 2D images compared to 3D images ($t = 3.39$, $df = 44$, $p = 0.001$, $d = 0.504$), there was no significant difference in habituation effects for negative or positive images ($t = 0.584$, $df = 44$, $p = 0.562$, $d = 0.087$; $t = -1.62$, $df = 44$, $p = 0.113$, $d = 0.238$).

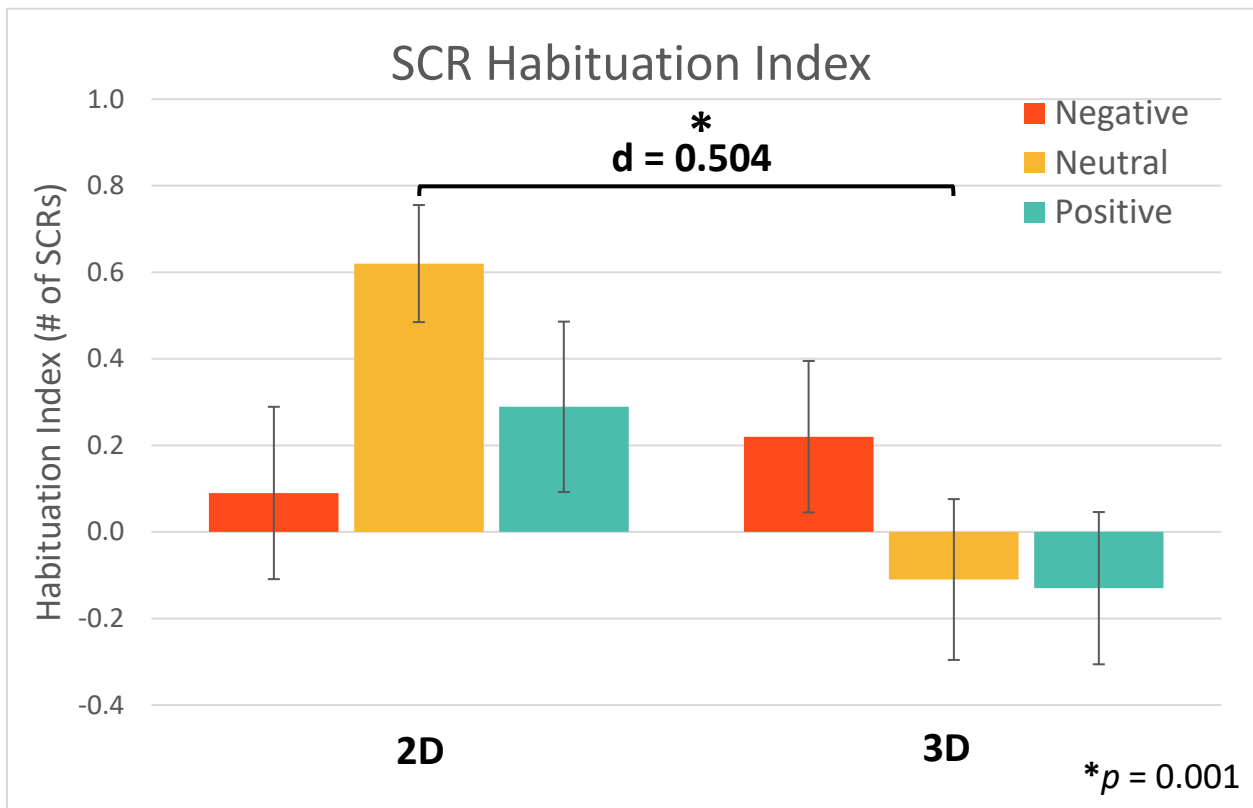


Figure 16. SCR Habituation Index; error bars represent standard error. There was no significant difference in habituation effects between the two dimensions.

3.3.3 SCR Recovery

A Recovery Index was created by subtracting SCRS from the fourth, novel block from EMG responses from the third block. A Dimension X Emotion ANOVA revealed there was no significant differences in response recovery based on Dimension ($F = 1.44$, $df = 1.00$, $p = 0.231$, $\eta_p^2 = 0.005$) or Emotion ($F = 0.81$, $df = 2.00$, $p = 0.446$, $\eta_p^2 = 0.006$). The interaction was also found not to be significant ($F = 0.15$, $df = 2.00$, $p = 0.862$, $\eta_p^2 = 0.001$).

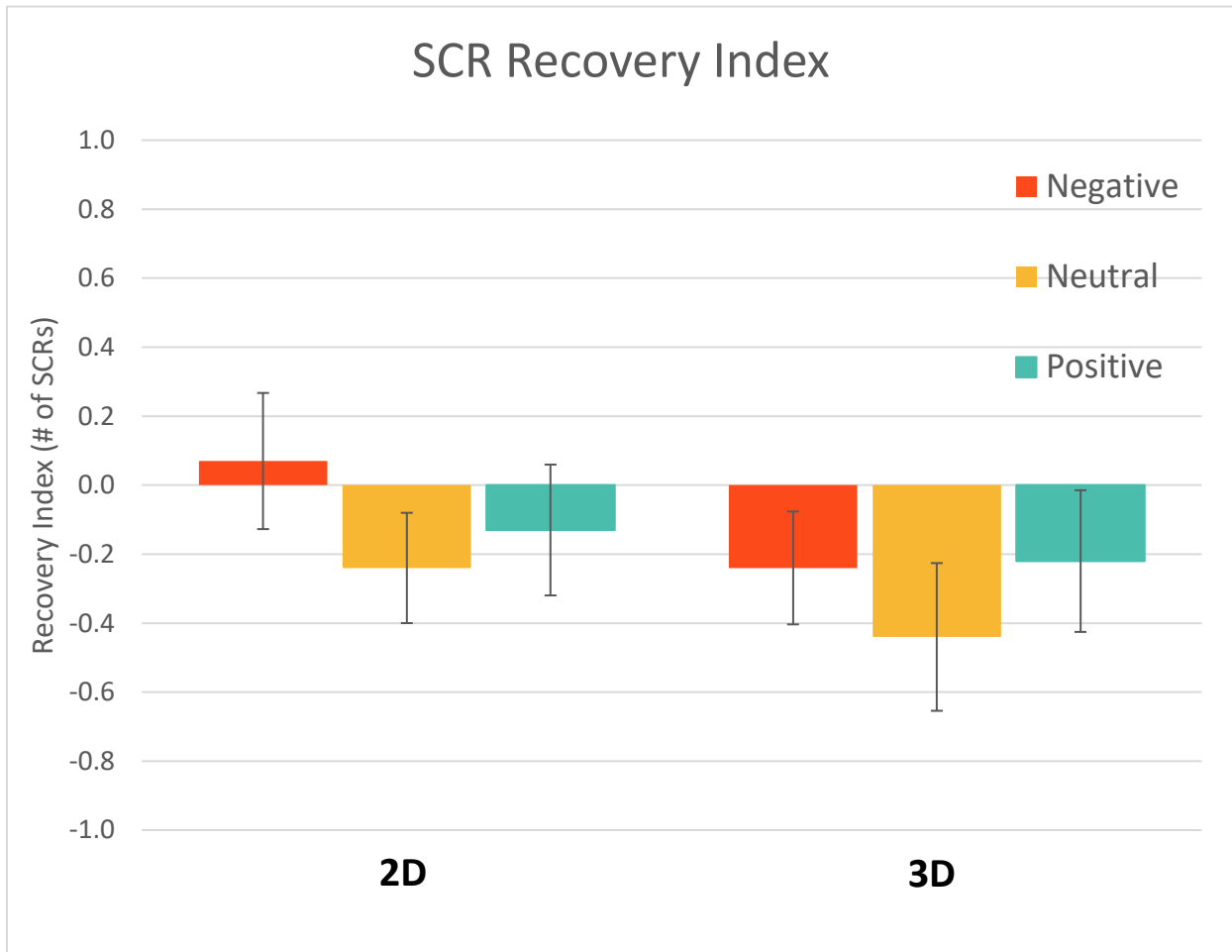


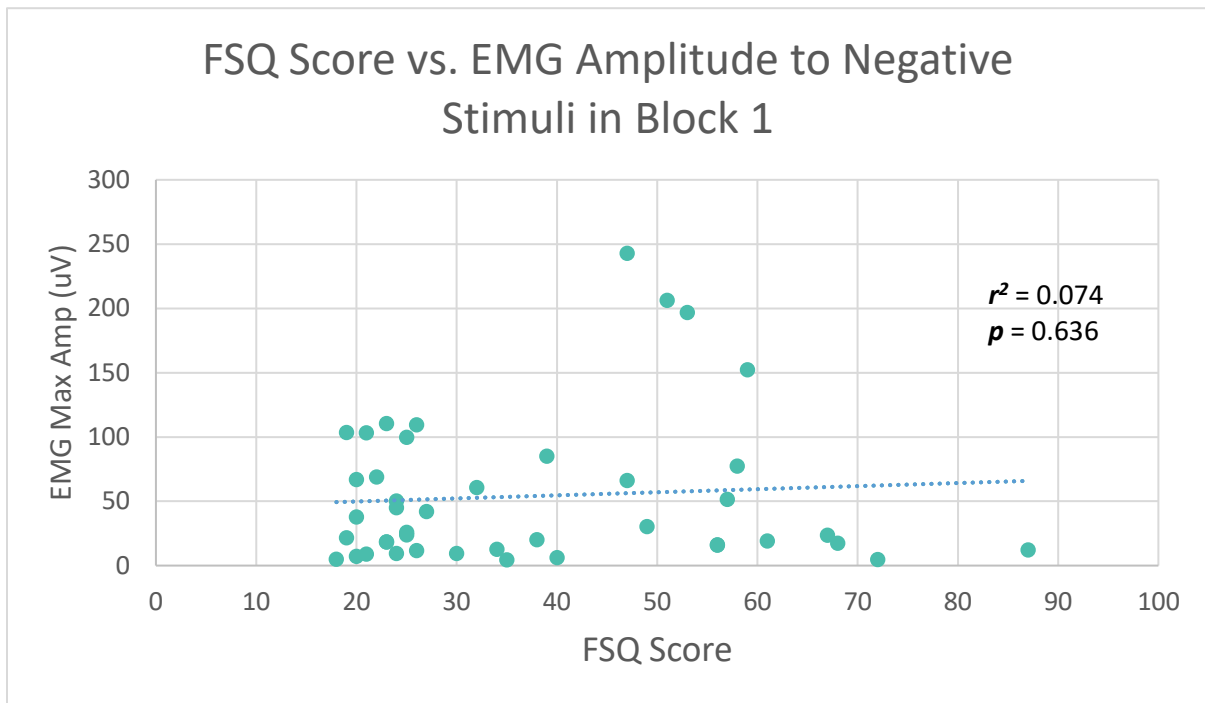
Figure 17. SCR Recovery Index; error bars represent standard error. There was no significant difference in recovery effects between the two dimensions.

3.4 Adapted Fear of Spiders Questionnaire Correlations

To determine if fear of insects and/or spiders had an effect on the startle eye blink response, scores on the adapted FSQ were correlated with maximum EMG amplitude to negative images in the first block. Only negative images were considered because fear would only have an impact on aversive stimuli. A bivariate correlation revealed that fear of insect and spiders did not have an impact on startle eye blink amplitude ($r^2 = 0.074$, $df = 43$, $p = 0.636$).

Likewise, to determine if fear of insects and/or spiders had an effect on skin conductance, scores on the adapted FSQ were correlated with the number of SCRs to negative images in the first block. A bivariate correlation revealed that fear of insect and spiders did not have a significant impact on number of SCRs ($r^2 = -0.127$, $df = 43$, $p = 0.418$).

A)



B)

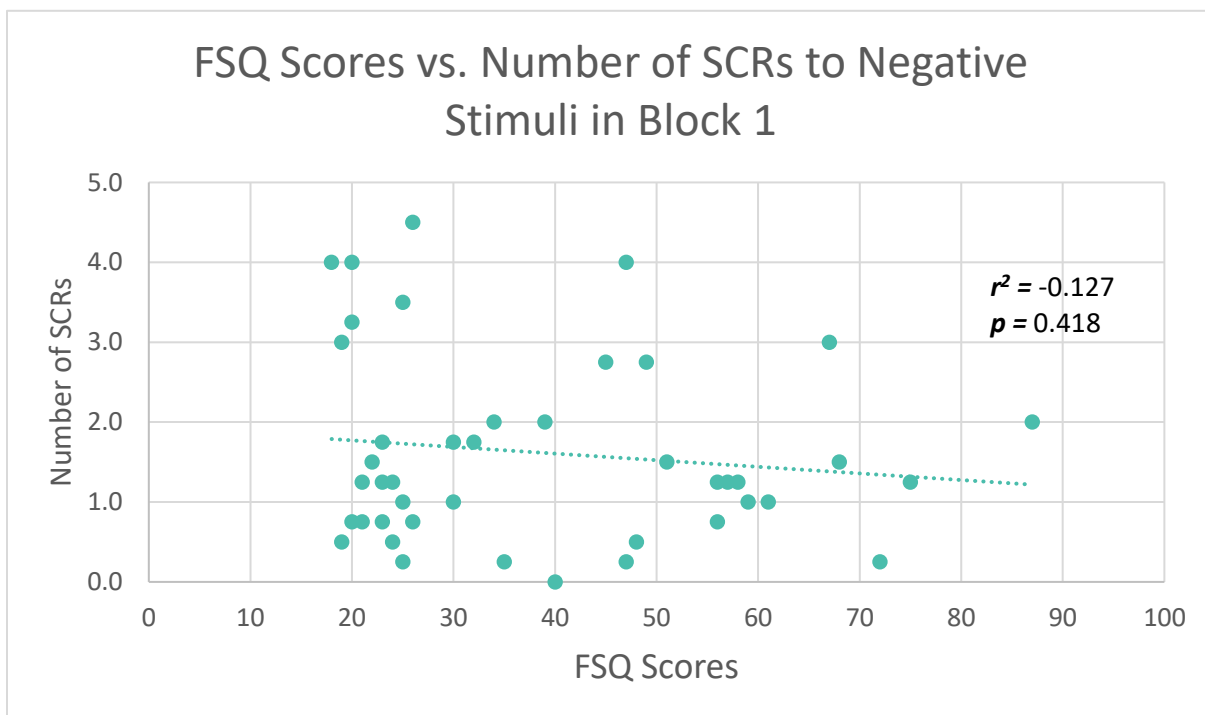


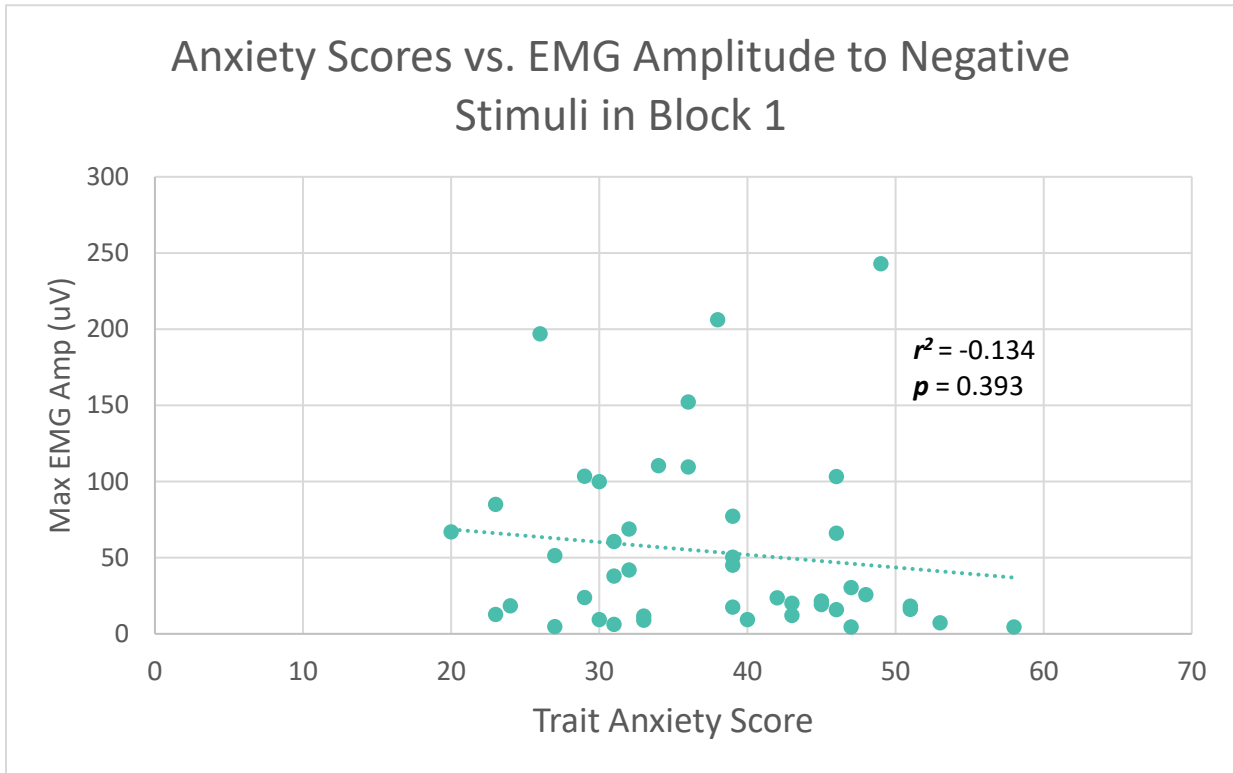
Figure 18. Impact of fear of insects and spiders on affective responding. A) No correlation found between FSQ scores and startle eye blink magnitude. B) No correlation found between FSQ scores and number of SCRs.

3.5 Trait Anxiety Score Correlations

To determine if trait anxiety had an effect on the startle eye blink response, trait anxiety scores from the STAI were correlated with maximum EMG amplitude to negative images in the first block. A bivariate correlation revealed that trait anxiety did not have an impact on startle eye blink amplitude ($r^2 = -0.134$, $df = 43$, $p = 0.393$).

Likewise, to determine if trait anxiety had an effect on skin conductance, trait anxiety scores from the STAI were correlated with the number of SCRs to negative images in the first block. A bivariate correlation revealed that trait anxiety did not have an impact on number of SCRs ($r^2 = -0.258$, $df = 43$, $p = 0.094$).

A)



B)

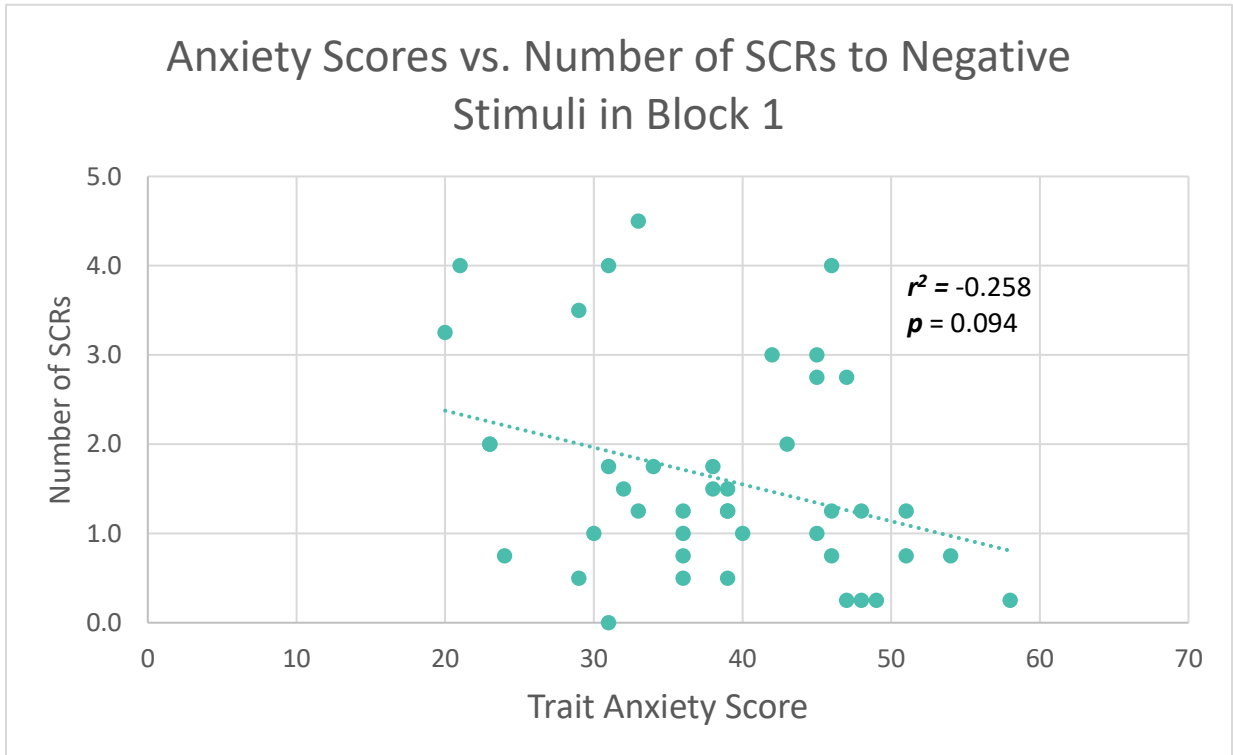


Figure 19. Impact of trait anxiety on affective responding. A) No correlation found between trait anxiety scores and startle eye blink magnitude. B) No correlation found between trait anxiety scores and number of SCRs.

CHAPTER 4

4 Discussion

4.1 Study Results

Our investigation into whether 3D images could improve stimulus realism and improve effects of emotion demonstrated a difference in how 2D and 3D emotional images are subjectively perceived, but no significant difference in psychophysical responding between the two stimulus types.

Differences in subjective perception between 2D and 3D images were discovered; 3D images were rated more realistic, arousing, and dangerous than 2D images whereas 2D images were rated more approachable than 3D images. In line with previous research (Tellegen, 1985; Lang, Greenwald, Bradley & Hamm, 1993; Codispoti, Ferrari, Bradley, 2007), negative and positive images were rated more arousing than neutral images. Negative images were also rated more realistic than neutral or positive images. There were a couple notable Dimension by Emotion interactions; 3D negative images were rated more realistic than 3D neutral and positive images but there was no difference in ratings for 2D images and 3D negative and neutral images were rated more dangerous than negative and neutral 2D images. These results demonstrate a subjective advantage to 3D images, particularly with negative images.

Contrary to our predictions, there was no significant difference in the EMG startle response magnitude between the two dimensions. Surprisingly, there was also no main effect of emotion even though previous research typically reports that the startle response should be differentially modulated by stimuli of different valence (Bradley, Lang, Cuthbert, 1993). There was a significant decrease in EMG response over time but there were no other main effects or significant interactions. There were also no differences in initial responding, habituation effects, or response recovery between the two dimensions.

Likewise, there was no significant difference in the number of SCRs between the two dimensions. Again, contrary to past research, there was no main effect of emotion indicating that emotional images did not increase the number of SCRs as expected (Bradley, Lang, Cuthbert,

1993). There was a significant Dimension by Block interaction where there was a significant decrease in the number of SCRs to 2D images across block but no decrease in the number of SCRs to 3D images. A closer investigation of habituation effects uncovered a significant Dimension by Emotion interaction where there was greater habituation for neutral 2D images than neutral 3D images. There were no differences in initial responding or response recovery between the two stimulus types. These results suggest there was no psychophysiological advantage to using emotional 3D stimuli.

Finally, fear of insects and/or spiders did not significantly correlate with EMG startle response magnitude or the number of SCRs to negative stimuli within the first block of the experiment. There was also no correlation between trait anxiety and EMG startle response magnitude or number of SCRs to negative images. This suggests that neither fear nor trait anxiety had an impact on psychophysiological responding.

4.2 Study implications

A possible conclusion that can be drawn from this experiment is that while using 3D images do provide a perceptual advantage for visual stimuli, they do not provide an advantage for responding in the autonomic nervous system. Although these results are not in line with our predictions, there is some previous research on 3D films which supports the finding that there is a subjective but not a psychophysiological benefit to 3D stimuli. Gaebler and colleagues (2014) found a subjective advantage to using 3D stimuli where 3D films were rated more immersive than 2D films. Conversely, Bride and colleagues (2014) conducted a psychophysiological study to compare skin conductance level and cardiac measures between 2D and 3D emotional film clips from popular films. There were 20 comparisons between the five psychophysiological measures used during the four film clips and only one significant difference between 2D and 3D films emerged. The thrilling 3D film clip elicited more SCRs than the 2D clip, otherwise, there was no increase in psychophysiological responding for 3D films. Rooney, Benson, & Hennessy (2012) compared both subjective and psychophysiological ratings in their study comparing 2D and 3D film scenes. This study revealed that 3D films received higher realism ratings and increased heart rate compared to 2D film but no significant difference in skin conductance level or skin temperature. They proposed that this dissociation between psychophysiological responses was due

to the fact that skin conductance level is controlled by the sympathetic nervous system whereas heart rate is influenced by both the sympathetic and parasympathetic nervous system. The sympathetic nervous system prepares the body for energy expenditure (the ‘fight or flight’ response) whereas the parasympathetic nervous system conserves energy (the ‘rest and digest’ response). The present study only included measures of the sympathetic nervous system (SCR and EMG) so it is unknown whether the 3D stimuli used in our study had an effect on psychophysiological responses controlled by the parasympathetic nervous system. As such, it is unknown whether there were differential psychophysiological responses associated with rest between our 2D and 3D stimuli. This is a limitation of the current study which should be addressed in future research.

A complication to the conclusion that there is a subjective but not an autonomic advantage for 3D images is that the present study did not observe the typical emotional modulation expected when using these psychophysiological measures. Previous research on the effect of emotional images on psychophysiological responses report effect sizes of emotion as $\eta^2 = 0.20$ for SCR (Codispoti & De Cesarei, 2007) and 0.25 for EMG (Anokhin & Golosheykin, 2010). In the current study, our effects of emotion were only $\eta^2 = 0.003$ and 0.019 for SCR and EMG respectively, a decrease in effect size from large effects to no effect and a small effect respectively. This may suggest that our stimuli were not as effective at eliciting an emotional response as those used in previous research. Of note, the current study used entomological stimuli for each of the stimulus categories as brain areas associated with emotion respond more strongly to threatening animate biological stimuli than to threatening inanimate stimuli (Coker-Appiah et al., 2013). However, the comparison studies used images of emotional faces, erotic couples, and mutilated bodies. Our stimuli may not have had enough variance between emotion categories and/or were not as arousing as the stimuli used in previous research.

To investigate this possibility, pleasantness and arousal ratings were examined more closely. While there was a significant effect of Emotion within pleasantness ratings, it is possible participants did not find the images emotionally distinct enough. While participants did rate the positive stimuli (which featured butterflies) more pleasant than the neutral stimuli (which featured beetles), it is possible that they did not find the pleasant stimuli objectively pleasant nor the neutral stimuli objectively neutral. Previous research has shown that participants tend to use

approximately 60% of a step-wise Likert scale like the one used in the current study (Matell & Jacoby, 1975). This raises the possibility that our results were a function of the participants' tendency to use the majority of the scale provided, even if they found our stimuli relatively benign. On a 9 point Likert scale, there was only a 3.31 point difference between average pleasantness ratings for positive and negative images, with positive images being rated only 1.29 points above neutral. This is a more limited range than is observed in studies comparing affective responding with IAPS images where a 4.5 point or greater difference between valence ratings is commonly observed (Codispoti, Ferrari, & Bradley, 2006; Codispoti & De Cesarei, 2007; Bradley, Lang, & Cuthbert, 1993; Sanchez-Navarro & Martinez-Selva, 2006; Bradley, Hamby, Low, & Lang, 2007; Codispoti, De Cesarei, Biondi, & Ferrari, 2016). While average pleasantness ratings did reflect positive, neutral, and negative scores for their respective emotion categories (6.29, 4.47, and 2.97), it is possible that these are relative ratings as only entomological stimuli were used; participants may not have actually considered the butterfly stimuli in the positive category pleasant but rated them pleasant compared to the beetle stimuli in the neutral category. Likewise, while there were main effects of Emotion and Dimension for arousal ratings, all average arousal ratings were under or at 5, the neutral point, on the 9 point scale. Since arousal is the driving force behind these psychophysiological measures (Bradley, Codispoti, Cuthbert & Lang, 2001; Cuthbert, Bradley, & Lang, 1996), it is possible that we did not see the expected differences in responding for emotion and the predicted differences in responding for dimension because the stimuli were not arousing enough.

Another possible explanation for our results is that there was a dissociation in the neural pathways associated with subjective ratings and psychophysiological responding. There is some evidence at the neural level that subjective emotional ratings and psychophysiological responses are governed by partially dissociable neurocognitive systems. When subjects are asked to provide emotional ratings for affective stimuli, brain areas responsible for emotional attention and interoception are activated, particularly the dorsomedial prefrontal cortex (dmPFC) and the anterior cingulate cortex (ACC; Taylor, Phan, Decker, & Liberzon., 2003; Buhle et al., 2014; Hariri, Bookheimer, & Mazziotta 2000; Lane et al., 1997; Northoff & Bermpohl, 2004; Hutcherson et al., 2005; Schienle, Wabnegger, Schoengassner, & Scharmuller, 2014). The amygdala is implicated in both the production of SCRs (Wood, Ver Hoef, & Knight, 2014) and startle eye blink modulation (Hitchcock & Davis, 1986; Angrilli et al., 1996). While the amygdala is not the only

structure involved in the generation of SCRs in response to emotional stimuli (Nagai et al., 2004; Critchley, Elliot, Mathais, & Dolan, 2000; Alvarez & Lahera, 2017) or startle eye blink modulation (Neuner et al., 2010), it is common to both types of responding. Previous research has shown a dissociation whereby neural regions associated with emotion were activated to fear-conditioned stimuli in the absence of differential autonomic responding (Tabbert, Stark, Kirsch, & Vaitl, 2006). Therefore it is possible that there was a dissociation between activity in areas of emotional attention that drove enhancement of subjective ratings of affect and realism for 3D stimuli, even though processes related to autonomic arousal did not differentiate between these stimuli. However, it should be noted that this explanation is highly speculative at this point; particularly given evidence that the brain areas responsible for subjective ratings and psychophysiological responding are often found to be highly overlapping. Providing emotional subjective ratings has been correlated with activation in the dmPFC and the ACC (Taylor, Phan, Decker, & Liberzon; 2003; Buhle et al., 2014) but also the middle PFC, amygdala (Phan et al., 2000), and middle temporal and fusiform gyri (Critchley et al., 2000). Psychophysiological responding has been correlated with activation in the amygdala (Wood, Ver Hoef, & Knight, 2014; Hitchcock & Davis, 1986) as well as the ACC (Tranel & Damasio, 1994) and the vmPFC (Damasio, Tranel, & Damasio, 1990). Also, activation in both the amygdala and mid-thalamic nuclei has been correlated with subjective rating conditions where SCRs were also present (Liberzon et al., 2000). So while the dmPFC and ACC has been associated with providing subjective emotional ratings and the amygdala has been associated with psychophysiological responding, the amount of overlap and brain areas involved in these processes complicates this interpretation of the results.

Another possible explanation for why effects of Dimension and Emotion were found for subjective and not psychophysiological ratings involves experimenter demand characteristics. Demand characteristics are changes in behaviour that occur when participants form a prediction about the expected results of an experiment and behave in a way to confirm those results. In the current study participants may have concluded that we were expecting a differences in subjective ratings between the two dimensions in favour of the 3D images and responded accordingly. Likewise, it is possible that participants did not actually experience an appreciable emotional reaction to our stimuli but knew conceptually that there should be an emotional difference between them. While our stimuli of spiders and butterflies did not produce the intended psychophysiological effects of emotion, other studies have shown that these reactions do exist to

these stimuli (Maltzman & Boyd, 1984; Wiemer, Gerdes, & Pauli, 2013; Stanley & Knight, 2004; Huijding & Jong, 2006; Anders et al., 2004; Neubert et al., 2017). So if participants did not experience a significant emotional reaction to our stimuli, it is possible they have had these reactions to these specimens in the past or know that other people in the population have these reactions so they reported experiencing differences between the stimuli that they did not actually feel.

Finally, it is also possible that the psychophysiological responses were diminished because the stimuli used in the current study may have generated an ambivalent response in the participants. Some findings suggest that activity in emotion related brain areas can be attenuated by ambiguous stimuli. A study by Wang and colleagues (2017) found a reduction in activation in the amygdala in response to ambiguous facial stimuli while a study by Kryklywy, Nates, & Mitchell (2009) found a reduction in insular activation in response to ambiguous emotional scenes. Activation in both of these areas has been found to be correlated with autonomic responding (Critchley, Elliot, Mathais, & Dolan, 2000; Neuner et al., 2010; Flynn, 1999). As the stimuli in the present study displayed a limited range of pleasantness ratings, it is possible there was limited activation in brain regions responsible for emotional encoding. This reduced response may explain why the expected emotional modulation of psychophysiological measures was not observed.

4.3 Limitations and future directions

While the current study provides a foundation for a line of research investigating the use of realistic 3D stimuli in affective cognitive neuroscience, it is not without limitations. As previously mentioned, the stimuli created may not have been emotionally distinct or arousing enough to produce psychophysiological effects of emotion. Additionally, while this study did use two measures of psychophysiological responding, they were both measures of the sympathetic nervous system. A measure of the parasympathetic nervous system, such as heart rate as was used successfully in a prior study comparing 2D and 3D scenes (see Rooney, Benson, & Hennessy, 2012), could have provided a more complete understanding of the effect of the stimuli on psychophysiological responding. Future studies should use stimuli with a greater amount of variance in pleasantness and arousal ratings as well as include measures of the

parasympathetic nervous system before a conclusive determination on whether visual dimension has an effect on psychophysiological responding can be made.

Since this study explored habituation effects, participants were exposed to a small number of stimuli repeatedly. It is possible that participants were less engaged with the stimuli and habituated more rapidly than expected because there was such a lack of variety in the images presented that they lost interest in the study faster than expected. Previous research comparing subjective and psychophysiological responding between emotional images vary in the number of stimuli used; whereas some use a small number as was done in the present study (Bradley, Lang, & Cuthbert, 1993; Codispoti, Ferrari, & Bradley, 2006; Codispoti, De Ceseari, Biondi, & Ferrari, 2016) others use dozens of different stimuli (Bradley, Hamby, Low, & Lang, 2007; Bradley, Codispoti, Cuthbert, & Lang, 2001; Sanchez-Navarro & Martinez-Selva, 2006). Future studies may consider increasing the number of stimuli used or look at habituation effects within stimulus category (e.g. positive-high arousal, negative-high arousal, etc.). Another limitation was that, in line with previous research (Codispoti, Ferrari, Bradley, 2006), participants were first exposed to the stimuli during a rating trial, when they were asked to rate the images on dimensions of emotion and realism. Psychophysiological responses were not being recorded during this time. Bradley and colleagues (1993) demonstrated a significant decrease in SCR magnitude after the first stimulus presentation so it is possible that emotional effects were present during these earlier presentations but not during the observation blocks when psychophysiological responses were being recorded. It is also notable that there was no recovery of response during the novel phase of the experiment. Including a novel block where new stimuli are presented has been shown to result in an increase of affective responding (Bradley, Lang, & Cuthbert, 1993; Codispoti, Ferrari, & Bradley, 2006), but this experiment did not find any significant recovery of response. This suggests that participants may have been fatigued by this point in the experiment which would reduce psychophysiological responding (Geldreich, 1939; Shiihara et al., 2000). Future studies comparing habituation effects between the two dimensions should address these limitations by using a greater number of stimuli and omitting the first rating trails to more conclusively determine whether using 3D emotional stimuli could result in different patterns of psychophysiological responding.

Future studies in this area should also investigate a greater variety of stimulus types. There is a growing body of research exploring the difference between 2D images and real objects but

future studies should compare all three stimulus types: 2D images, 3D images, and real objects. Just as the studies comparing 2D images and real objects have investigated the differences in memory effects (Snow, Skiba, Coleman, & Berryhill, 2014), attention (Gomez, Skiba, & Snow, 2018), behaviour (Breiter et al., 1996; Squires, MacDonald, Culham, & Snow, 2016), and repetition suppression (Snow et al., 2011), so too should studies comparing 2D, 3D, and real stimuli. Studies of this kind would elucidate the differences and similarities in behavioural, physiological, and neural responding between the stimulus types and allow researchers to make a more informed decision about what stimulus type would be most appropriate for their experiment.

While real objects would be the most ecologically valid, they would not be feasible for all experiment types, including some studies of emotion. For example, studies using stimuli related to a variety of threatening cues (i.e. guns, violent interactions, injuries, or snakes) or stimuli of emotional facial expressions would be difficult to operationalize with real objects. Although Nili, Goldberg, Weizman, & Dudai (2010) did conduct a study where a live snake was used in an fMRI experiment to measure brain regions associated with bravery. This study found that activation in the ACC and the right temporal pole was positively correlated with overcoming fear (i.e. when participants chose to bring the snake closer to them despite indicating that they were scared). As far as we are aware, this study is unique in its use of live stimuli. Another option is to deceive participants with videos of live stimuli as Mobbs and colleagues (2010) did in an fMRI experiment where participants believed a live tarantula was being placed at various distances away from their feet. Their study aimed to determine the neural correlates associated with the absolute proximity and approach and retreat movements of a phylogenetic threat. While habituation was observed in most brain areas, activity in the midbrain periaqueductal gray (an area associated with fear and panic; Nashold, Wilson, & Slaughter, 1969) was sustained throughout the experiment for participants who scored high on the FSQ. While robust effects of emotion were found in both of these studies, it is difficult to say if or how results would differ if more conventional dynamic stimuli were used (i.e. where participants knew they were observing film clips). Although, a similar issue arises with both of these studies as participants observed the real or believed to be real stimuli indirectly, which eliminates some of the benefits associated with observing real objects directly (e.g. three-dimensionality and the possibility of interaction). While these studies illustrate how real stimuli can be incorporated into emotion research, most likely 2D or 3D stimuli would be employed in studies of this kind. As such, it would be beneficial to know the differences in

neural processing between live, perceived to be live, and known to be pre-recorded stimuli. A manipulation allowing for these comparisons would allow researchers to determine whether there are benefits to using real stimuli which outweigh the costs, whether the deception of reality is sufficient to achieve these benefits, and whether deception is even necessary to achieve an effect.

Stimulus realism could also be improved upon by using dynamic 3D stimuli. In real life, emotion provoking stimuli rarely exist in static states. Research suggests that, compared to static stimuli, dynamic emotional stimuli elicit greater activation in brain areas associated with emotion (Trautmann, Fehr, & Herrmann, 2009), higher ratings of emotional intensity (Atkinson, Dittrich, Gemmel, & Young, 2003), and greater psychophysiological responses (Courtney et al., 2010). As the current study showed that participants perceived 3D images as subjectively more realistic and arousing than 2D images, dynamic 3D stimuli could further improve these effects and may then extend to psychophysiological responding. Another venue for future emotion research would be to explore emotion in virtual reality. Previous research has shown behavioural differences between real objects and 2D and 3D images due to the fact that real objects allow for subject interaction (Gomez, Skiba & Snow, 2018), a difference which could be eliminated in studies which use virtual reality.

Finally, our results revealed a dissociation between subjective and psychophysiological responding which may be explained by the fact that these processes have some divergent neural correlates. A neuroimaging study should be conducted to determine the pattern of brain activation associated with affective picture processing of 2D and 3D images. A study of this nature would not only clarify the results observed in the present study but would also provide a more complete understanding of the processes involved in processing emotional 2D and 3D stimuli.

4.4 Conclusions

This study investigated whether 3D images could be used in affective cognitive research to improve stimulus realism and achieve more reliable effects of emotion. 3D images were rated more realistic than 2D images, with a particular advantage observed for negative 3D images. They also received higher subjective ratings of arousal and danger than 2D images, indicating that 3D images did have an advantage in some emotion categories as well. Contrary to predictions, no difference

in psychophysiological responding was observed between emotional 2D and 3D images initially or across blocks. While 3D objects have been found to be resistant to repetition effects (Snow et al., 2011), this study did not find that emotional 3D images were more resistant to habituation effects than 2D images nor did they experience a greater recovery of response compared to 2D images. While these results appear to show a dissociation between the subjective experience of emotion and psychophysiological responding, the lack of differential psychophysiological responses may be attributed to potential methodological issues. Specifically, due to the effects of pre-exposure to the stimuli before psychophysiological recording, the reliance solely on sympathetic autonomic measurements, and the use of stimuli which may have only generated an ambivalent response. Further research should address these issues to explore the potential utility of 3D versus 2D stimuli in the field of affective cognitive neuroscience.

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Appendix A

1 Pilot Study

Fifty participants (31 female, 19 male; mean age, 20.7; range, 18-31 years) completed the pilot study. Six participants were excluded from data analysis based on their visual dimensionality ratings which revealed they were not rating 2D and 3D images differently (less than 1 standard deviation between mean ratings).

Five 2 (Dimension: 2D, 3D) X 3 (Negative, Neutral, Positive) way repeated measures ANOVAs were conducted on participants' subjective ratings of stimuli, one for each subjective dimension (Realism, Arousal, Danger, Approachability, and Pleasantness).

1.1 Realism

The ANOVA for Realism revealed a significant main effect of Dimension ($F = 6.74$, $df = 1$, $p = 0.013$, $\eta_p^2 = 0.144$) but not of Emotion ($F = 2.86$, $df = 1.50$, $p = 0.079$, $\eta_p^2 = 0.067$) and no Dimension X Emotion interaction ($F = 3.10$, $df = 2$, $p = 0.050$, $\eta_p^2 = 0.072$).

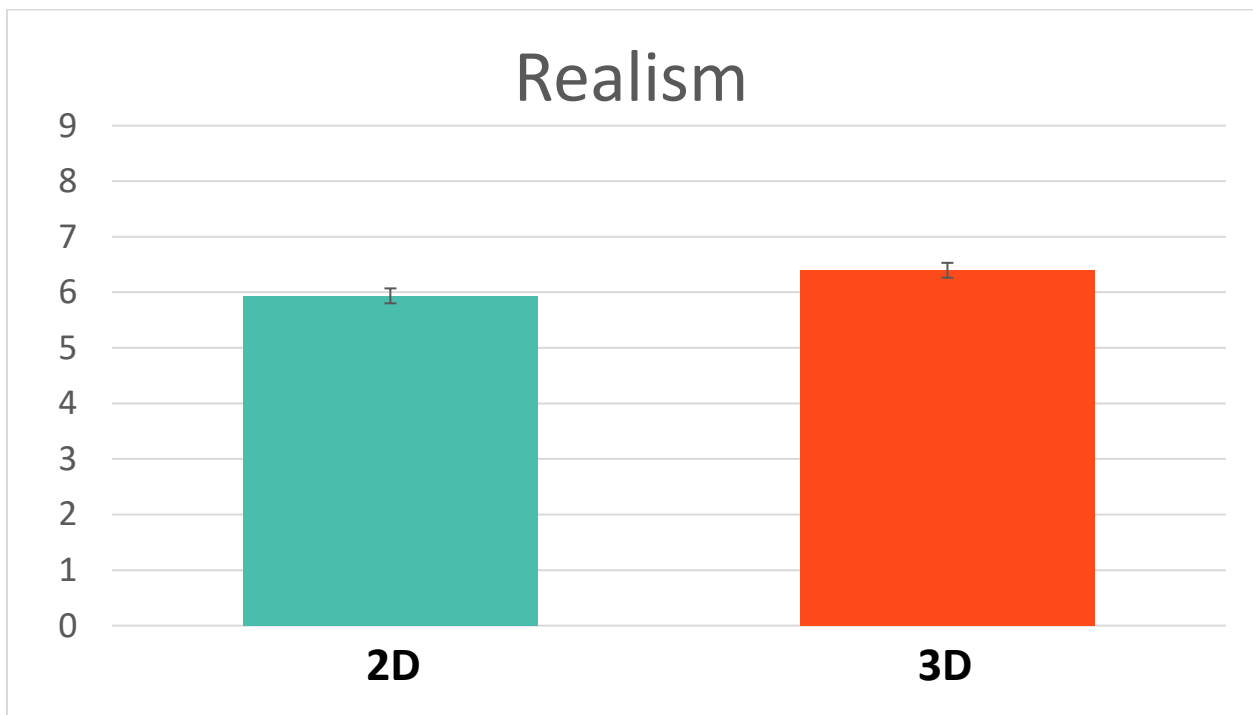


Figure 1. Mean realism ratings for the pilot study; error bars represent standard error. 3D images were rated more realistic than 2D images.

1.2 Arousal

The ANOVA for Realism revealed a significant main effect of Dimension ($F = 19.9$, $df = 1$, $p < 0.001$, $\eta^2 = 0.332$) but not of Emotion ($F = 0.450$, $df = 1.29$, $p = 0.642$, $\eta^2 = 0.011$) and no Dimension X Emotion interaction ($F = 0.910$, $df = 2$, $p = 0.408$, $\eta^2 = 0.022$).

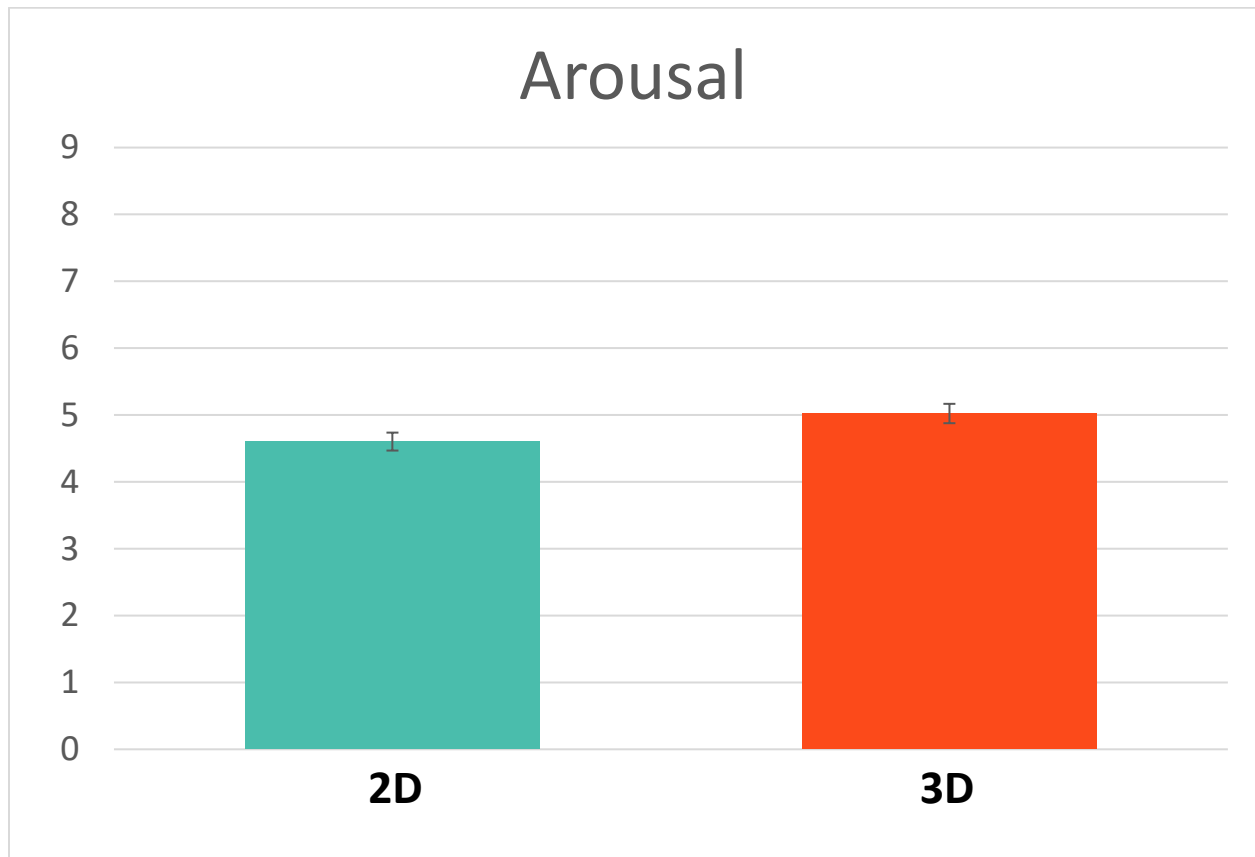


Figure 2. Mean arousal ratings for the pilot study; error bars represent standard error. 3D images were rated more arousing than 2D images.

1.3 Danger

The ANOVA for Realism revealed a significant main effect of Dimension ($F = 18.7$, $df = 1$, $p < 0.001$, $\eta_p^2 = 0.318$) and Emotion ($F = 169.8$, $df = 1.37$, $p < 0.001$, $\eta_p^2 = 0.809$) but no Dimension X Emotion interaction ($F = 1.64$, $df = 2$, $p = 0.201$, $\eta_p^2 = 0.039$).

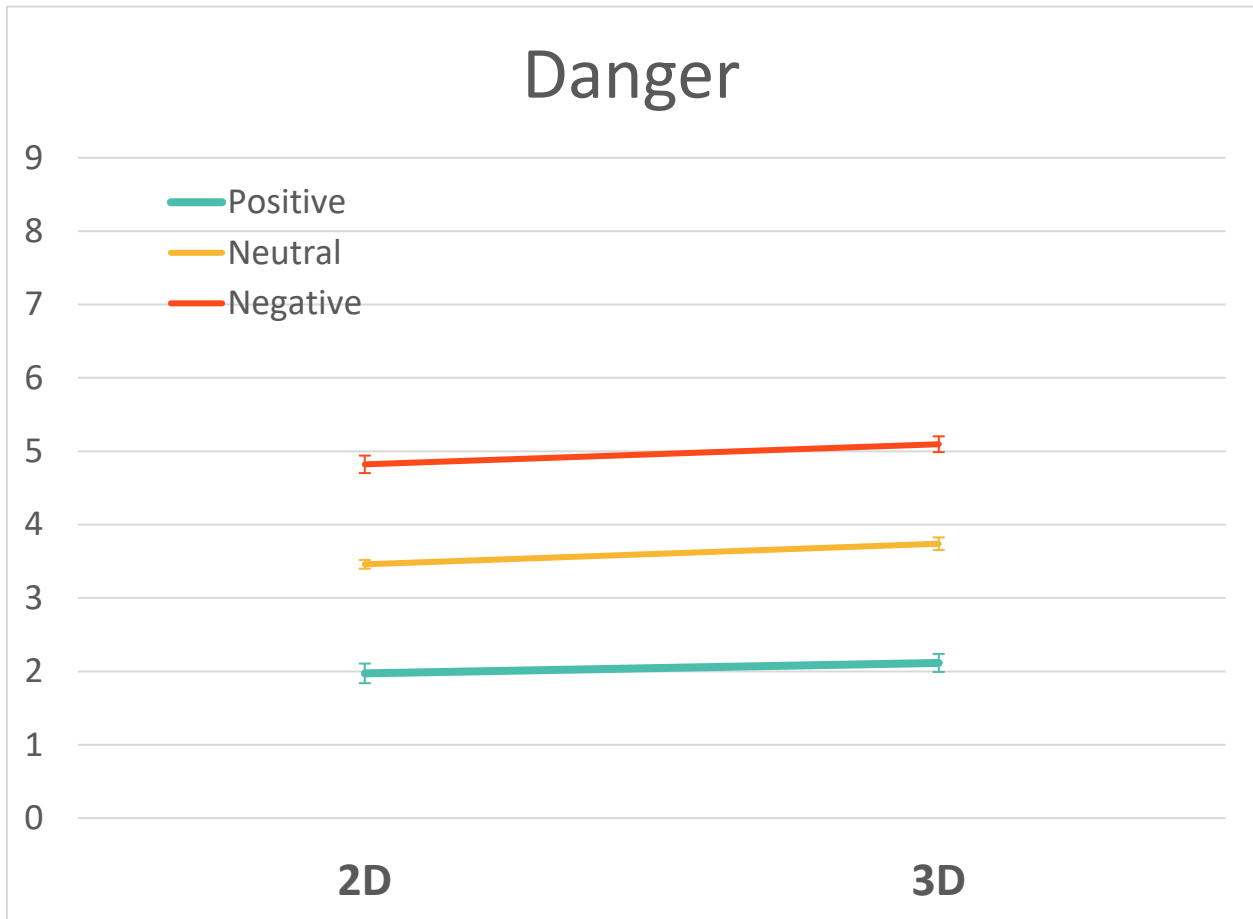


Figure 3. Mean Danger ratings for the pilot study; error bars represent standard error. 3D images were rated more arousing than 2D images. Effects of emotion were as expected (Negative > Neutral > Positive).

1.4 Approachability

The ANOVA for Realism revealed a significant main effect of Emotion ($F = 159.0$, $df = 1.48$, $p < 0.001$, $\eta_p^2 = 0.799$) but not Dimension ($F = 0.543$, $df = 1$, $p = 0.466$, $\eta_p^2 = 0.013$) and no Dimension X Emotion interaction ($F = 0.398$, $df = 2$, $p = 0.673$, $\eta_p^2 = 0.010$).

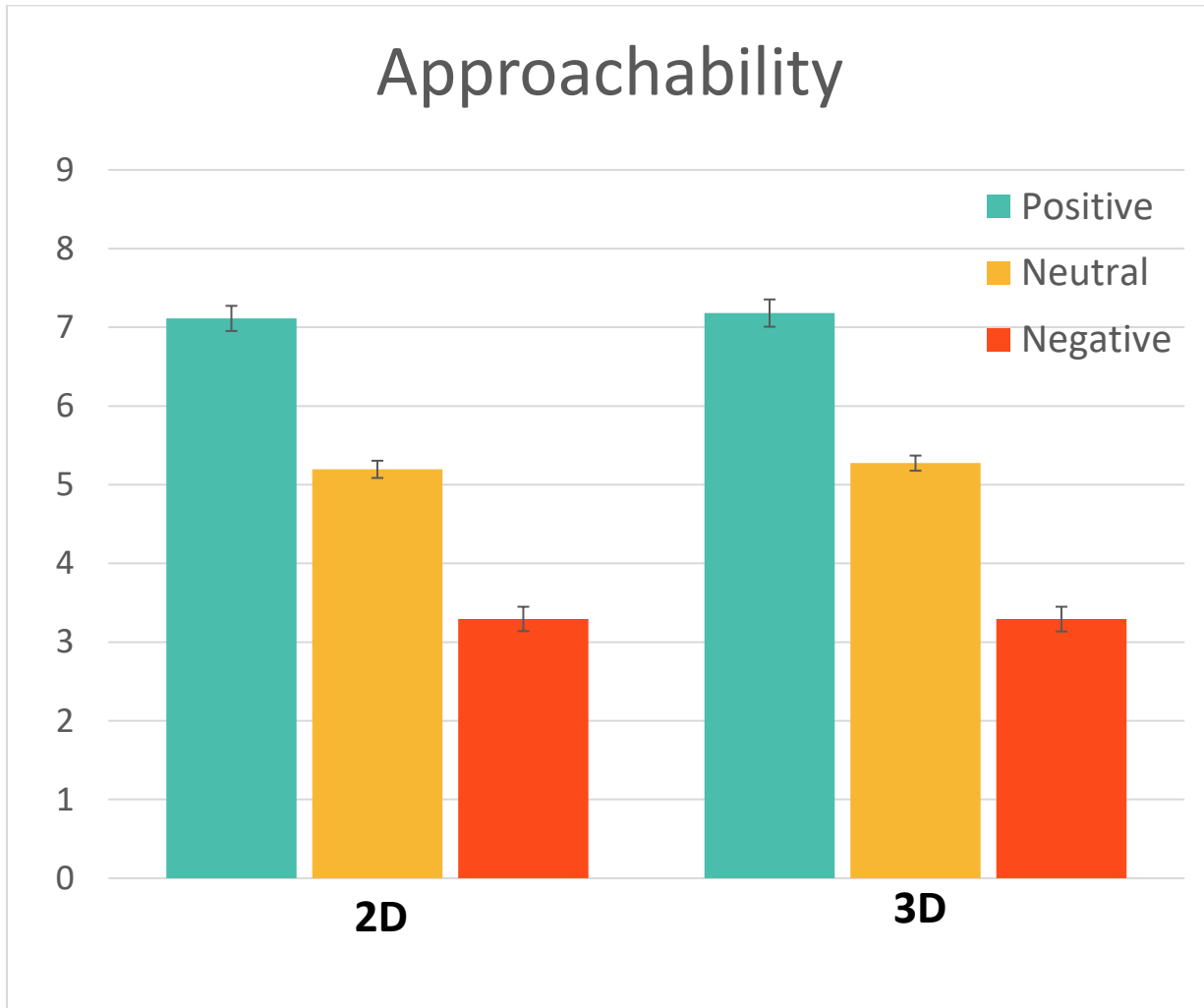


Figure 4. Mean Approachability ratings for the pilot study; error bars represent standard error. Effects of emotion were as expected (Positive > Neutral > Negative).

1.5 Pleasantness

The ANOVA for Realism revealed a significant main effect of Emotion ($F = 230.7$, $df = 1.63$, $p < 0.001$, $\eta_p^2 = 0.852$) but not Dimension ($F = 2.84$, $df = 1$, $p = 0.099$, $\eta_p^2 = 0.066$) and no Dimension X Emotion interaction ($F = 4.00$, $df = 2$, $p = 0.022$, $\eta_p^2 = 0.091$).

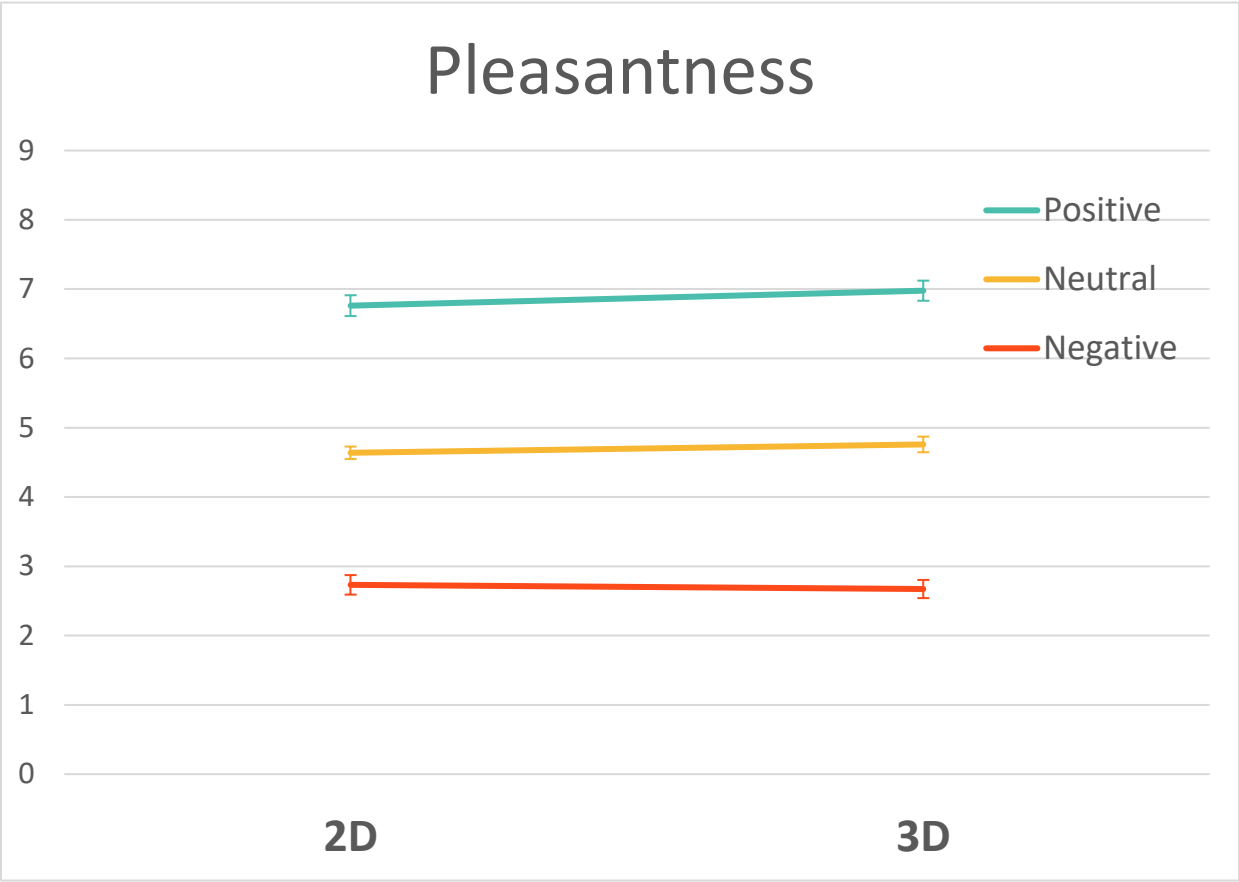


Figure 5. Mean Pleasantness ratings for the pilot study; error bars represent standard error. Effects of emotion were as expected (Positive > Neutral > Negative).

Appendix B



Date: 25 April 2019

To: Dr. Derek Mitchell

Project ID: 106132

Study Title: The impact of fear on cognition and social behaviour

Application Type: HSREB Amendment Form

Review Type: Delegated

Meeting Date / Full Board Reporting Date: 07/May/2019

Date Approval Issued: 25/Apr/2019

REB Approval Expiry Date: 23/Feb/2020

Dear Dr. Derek Mitchell ,

The Western University Health Sciences Research Ethics Board (HSREB) has reviewed and approved the WREM application form for the amendment, as of the date noted above.

Documents Approved:

Document Name	Document Type	Document Date	Document Version
Amended protocol add Experiment 6 April 23 2019 SUBMITTED	Protocol	23/Apr/2019	
Debriefing and re consent_April 23 2019 SUBMITTED	Consent Form	23/Apr/2019	Exp6 re consent clean
Letter of information and consent_EXPERIMENT 6 April 23 2019 SUBMITTED	Consent Form	23/Apr/2019	Exp 6 consent clean
Task and sample stimuli used	Other Data Collection Instruments	11/Apr/2019	Exp 6 stimuli

Documents Acknowledged:

Document Name	Document Type	Document Date
Summary of Changes	Summary of Changes	11/Apr/2019

REB members involved in the research project do not participate in the review, discussion or decision.

The Western University HSREB operates in compliance with, and is constituted in accordance with, the requirements of the TriCouncil Policy Statement: Ethical Conduct for Research Involving Humans (TCPS 2); the International Conference on Harmonisation Good Clinical Practice Consolidated Guideline (ICH GCP); Part C, Division 5 of the Food and Drug Regulations; Part 4 of the Natural Health Products Regulations; Part 3 of the Medical Devices Regulations and the provisions of the Ontario Personal Health Information Protection Act (PHIPA 2004) and its applicable regulations. The HSREB is registered with the U.S. Department of Health & Human Services under the IRB registration number IRB 00000940.

Please do not hesitate to contact us if you have any questions.

Sincerely,

Patricia Sargeant, Ethics Officer (ext. 85990) on behalf of Dr. Joseph Gilbert, HSREB Chair

Note: This correspondence includes an electronic signature (validation and approval via an online system that is compliant with all regulations).

Curriculum Vitae

Shannon Compton

Education

Masters of Neuroscience **2017 - Present**
University of Western Ontario, London, ON

Bachelor of Science, Honours (Psychology Major, Biology Minor) **2015**
University of Waterloo, Waterloo, ON

- Dean's Honour List
 - Relevant courses: Systems Neuroscience, Cognition and Cognitive Neuropsychology, Human Molecular Genetics, Principles of Human Physiology, Introduction to Bioinformatics, Analytical Methods in Molecular Biology, Biochemistry and Molecular Biology, Basic Research Methods
-

Research Experience

Research Assistant, Mental Health Nurses Research Alliance **Jan 2017 - Sept 2017**
Lawson Health Research Institute, London, ON

- Conduct study enrollment activities, participant interviews, data collection, and data entry.
- Manage conference submissions including creating abstracts, registering, and preparing travel and accommodations.
- Conduct literature reviews for papers by navigating databases such as PsycNet, CINHAL, and PubMed.
- Complete large scale grant applications for new projects.
- Compose project reports detailing project background, results, implications, and future directions for stakeholders.

Volunteer Research Assistant, Diversity and Intergroup Relationships Lab **2013 - 2014**
University of Waterloo, Waterloo, ON

- Recruit and schedule participants for social psychology using the cloud-based subject pool software SONA.
- Preparing materials for studies and running experiments with single and dyadic participants.
- Data entry, collection, and management. Included cleaning and coding data.
- Conducted data analysis using SPSS to calculate averages and graph data.
- Create online surveys for psychology studies

Volunteer Research Assistant, Dr. Richard Eibach Lab **2015**

University of Waterloo, Waterloo, ON

- Recruit and schedule participants for social psychology using the cloud-based subject pool software SONA.
 - Preparing materials for studies and running experiments involving deception.
 - Coding qualitative participant data.
-

Work Experience

Proctor **2017 - Present**
Western University, London, ON

Administrative Coordinator of Professional Programs **2015 - 2016**
College of Opticians of Ontario, Toronto, ON

- Provide administrative support for Registration and Quality Assurance Committees including case preparation, organization, and coordination of meetings.
- Assess professional membership portfolios and monitor member completion
- Following up with members regarding their Quality Assurance compliance and portfolio submissions; tracking correspondence with members and monitoring timelines and due dates.
- Attendance at trade shows, national examinations, and continuing education education events.
- Maintain systems for manual and electronic member case files and applicant files, ensuring appropriate data is collected and accurately entered.
- Compile data and statistics; graph data for accessible knowledge dissemination

Teacher's Assistant for Introduction to Cellular Biology **2012**

University of Waterloo, Waterloo, ON

- Conducted weekly tutorials and answered student queries.
- Marked student oral presentations and essays.
- Proctored midterm and final examinations.

Mature Adult Day Program Coordinator **2011 - 2012**

Community Home Assistance to Seniors, Bradford, ON

- Developed and led programs to promote cognitive stimulation for seniors with dementia and Alzheimer's disease.
- Provided one-on-one support and patient centred care to clients.
- Researched new trends in programming for cognitively impaired seniors.

Volunteer Experience

Tree Planting Volunteer **Summer 2017 - Present**

Reforest London, London, ON

- Planted trees in parks and naturalized areas to make London healthier and greener.

CT Department and Patient Registration Volunteer **2014 - 2015**

Grand River Hospital, Waterloo, ON

- Sort through hospital traffic to ensure patients are in the right department and prioritize patients based on appointment time and order of arrival.
- Transport paper work between departments.
- Provide customer service and patient interaction to those coming in for appointments.

Various Volunteer Positions **2003 - 2010**

Community Home Assistance to Seniors, Aurora, ON

- Assisted with various fundraising events including pancake breakfasts, awareness walks, and silent auctions.
- Delivered food to community-based seniors through the Meals-on-Wheels program.

Scholarship and Academic Honours

Dean's Honour List **2015**

Included five times on the University of Waterloo Science Dean Term Honour List

2011, 2013 - 2015

University of Waterloo President's Scholarship **2010**

Arts Alumni Entrance Scholarship **2010**

Bronze Governor General's Academic Medal **2010**

Youth Philanthropy Initiative Award Winner **2009**

Certification and Training

Western REM Training **2017**

WHMIS Training **2017**

Clinical Privacy and Confidentiality Module **2017**

AODA Accessibility Regulations Module **2017**

CPR/AED certified - Level C **2016**

Papers, Presentations, and Posters

Compton, S.A.H., Ritchie, M. B., Jones, I.G.R., Neufeld, R.W.J., & Mitchell, D.G.V. (2019, June). *The impact of violent gaming on social cognition as a function of trait anxiety*. Poster session presented at the Psychiatry Department of Western University's Academic Research Day, London, ON.

Compton, S.A.H., & Mitchell, D.G.V. (2019, May). *Improving stimulus realism: The effect of visual dimension on affective responding*. Poster session presented at the Western University Neuroscience Research Day, London, ON.

Compton, S.A.H., & Mitchell, D.G.V. (2019, May). *Improving stimulus realism: The effect of visual dimension on affective responding*. Poster session presented at the Western University Neuroscience Research Day, London, ON.

Compton, S.A.H., Ritchie, M. B., Jones, I.G.R., Neufeld, R.W.J., & Mitchell, D.G.V. (2018, June). *The impact of violent gaming on social cognition as a function of trait anxiety*. Poster session presented at Western University's London Joint Mental Health Research Day, London, ON.

Forchuk, C., Corring, D., **Compton, S.A.H.**, Harerimana, B., Marsella, O. (September, 2017). *Small steps for big outcomes: Building a smart technology strategy for people with mental illness*. MHEN Project.