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Threat and trait anxiety affect stability of gaze fixation

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ABSTRACT

Threat accelerates early visual information processing, as shown by shorter P100 latencies of pattern Visual Evoked Potentials in subjects with low trait anxiety, but the opposite is true for high anxious subjects. We sought to determine if, and how, threat and trait anxiety interact to affect stability of gaze fixation. We used video oculography to record gaze position in the presence and in the absence of a fixational stimulus, in a safe and a verbal threat condition in subjects characterised for their trait anxiety. Trait anxiety significantly predicted fixational instability in the threat condition. An extreme tertile analysis revealed that fixation was less stable in the high anxiety group, especially under threat or in the absence of a stimulus. The effects of anxiety extend to perceptual and sensorimotor processes. These results have implications for the understanding of individual differences in occulomotor planning and visually guided behavior.

1. Introduction

One of the primary roles of the gaze system is to bring and hold visual stimuli of interest onto the fovea, the central area of human retina showing the highest visual acuity, necessary for post-retinal processing. Even during fixation on a stationary stimulus/object (visually guided fixation), constant, small, involuntary "microsaccadic" eye movements prevent the naturally occurring adaptation of post-retinal cortical cellular mechanisms (Movshon and Lennie, 1979; Webster and De Valois, 1985), which would otherwise lead to depression of sensitivity/salience and even fading of a visual target following prolonged stabilisation of its image on the fovea (Troxler, 1804; Blakemore and Campbell, 1969). Apart from preventing retinal fading of a visual target, microsaccades also serve to bring the line of sight to visual details that are crucial for finely guided visuomotor tasks that require the highest level of spatial resolution (Steinman et al., 1973; Ko et al., 2010). Yet, in addition to improve spatial resolution, microsaccades are thought to transform the visual scene into a sequence of discrete views which, one at a time, are processed by attentional resources and guide decision making (Ballard et al., 1997; Ko et al., 2010; Kowler and Collewijn, 2010). Microsaccades may thus be prompted by cognition and reflect attentional processing and fine strategic visuomotor planning.

Initiation of visually triggered microsaccades involves occipital and parietal cortical inputs, including retinal input to "fixation neurons" in the superior colliculus, which then projects to the premotor circuit in the brain stem and cerebellum (Munoz and Istvan, 1998; Munoz and Wurtz, 1993a,b). Suppression of reflexive microsaccades is under the tonic control of frontal cortex and basal ganglia, which also project to the superior colliculus and brain stem premotor circuit (Hikosaka et al., 2000; Munoz et al., 2000; Schall, 1997 for review). Fixation in the absence of a visual stimulus (volitional fixation) is less accurate than stimulus-driven fixation (Smyrnis et al., 2003) and may be driven almost entirely by extra-retinal, prefrontal/basal ganglia input to the SC fixation neurons (Munoz and Wurtz, 1993a; Hikosaka et al., 2000; Munoz et al., 2000; Schall, 1997 for review). It has been hypothesized (Smyrnis et al., 2004) that these prefrontal regions maintain a "mental" representation of the fixation point and thus may be the same areas mediating spatial memory processing (Goldman-Rakic, 1988). Indeed, it is well-established that a particular region of the frontal lobe neocortex, the frontal eye field (FEF) is prominently involved in control of volitional eye movements, having a distinct sub-region for fixation. The dorsolateral prefrontal cortex in particular, acts as a ≪supervisory≫ area, inhibiting unwanted reflexive saccades when volitional maintenance of fixation is required (Gooding, 1999).

Suppression of microsaccades results in stable fixation such as that seen in trained athletes e.g. elite shooters and has been related to their superiority in selective and sustained attention (Di Russo et al., 2003). On the other hand, deficient fixational stability is

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seen in conditions characterised by attentional and strategic planning deficits due to fronto-striatal pathology, such as schizophrenia (Burton et al., 2008) and attention-deficit hyperactivity disorder (ADHD) (Munoz et al., 2003). Trait anxiety is associated with impoverished recruitment of attentional control mechanisms (Fox, 1993; Eysenck and Calvo, 1992; Bishop, 2009) and for this reason it is expected to impact on reflexive and volitional saccadic control. Threat commands visual attention through activation of the amygdala and the bed nucleus of stria terminalis (Lang et al., 2000) and is thus preferentially detected in humans (Ohman et al., 2001); it adaptively enhances contrast perception (Phelps et al., 2006) and accelerates the early P100 wave of pattern Visual Evoked Potentials (Laretzaki et al., 2010). The latter effect however, was not observed in high trait anxiety subjects (Laretzaki et al., 2010). This was probably a result of a hyper-responsive pre-attentive, amygdala-centred threat-detection system (Mathews et al., 1997), associated with deficient recruitment of prefrontal cortical mechanisms that are critical in the top–down control of selective attention to threat (Bishop et al., 2004; Ohman, 2005).

In the present study, we sought to determine if, and how, threat and trait anxiety interact to affect fixation of gaze. To this goal, we studied the effects of verbal threat on stimulus driven (a non-emotional target) and volitional (no-target, empty screen) fixation performance, in healthy subjects characterised for their trait anxiety. Subjects were also tested in the absence of threat. Based on available evidence, we predicted generally better fixation performance in the stimulus driven compared to volitional fixation condition. We also predicted that threat would impair fixation performance especially in high trait anxious subjects and this impairment would be more pronounced under volitional fixation since the latter is more critically determined by prefrontal input in the SC. Because of inadequate data in the literature, we made no predictions regarding group differences (high vs. low trait anxious subjects) in fixation performance in the absence of threat.

2. Materials and methods

2.1. Subjects

This study was conducted in compliance with the declaration of Helsinki and followed a protocol approved by the Ethics Committee of the University of Crete, and all participants gave their written informed consent. Participants ($n = 44$, age: 26 ± 5 yrs) were randomly recruited by phone from a pool of 560 healthy male volunteers, characterised for trait anxiety (median trait anxiety score: 36, mean: 36.45, range: 20–68), with the widely accepted State-Trait Anxiety Inventory (STAI-T) (Spielberger, 1983). Care was taken that the present sample was representative of the large cohort (20 were above and 24 below the cohort's median) covering the entire range of the cohort's STAI-T values. These volunteers had previously undergone thorough psychiatric and medical assessment including drug screening and they were free from history or presence of head trauma, medical neurological and psychiatric conditions, including use of prescribed or recreational drugs. Additional criteria for inclusion in the present study were absence of ocular or corneal disease, normal binocular and colour vision and optical correction (if needed) with spectacles for the viewing distance. Recruited subjects underwent an ophthalmological examination and a new urine drug screening test. Participants were tested between 9:00 am and 4:00 pm in one session.

2.2. Eye tracking

Fixational gaze movements from both eyes were recorded simultaneously using video oculography (EyeLink II, SR Research Ltd., Canada). Eyelink II consists of two miniature head-mounted infrared cameras that record eye position using pupil or pupil-cornea tracking. A third camera monitors subjects' head position relative to four infrared markers mounted on the display screen. According to the manufacturer, Eyelink II has a spatial resolution higher than 0.01° for two-dimensional eye tracking. In this study horizontal and vertical eye positions were recorded using the pupil-cornea tracking mode at a sample rate of 250 Hz. All measurements were performed with subjects seated on a chair and with their head stabilised by means of a chin rest to minimise head movements. A system calibration/validation was performed every time prior to recording to correlate the output results.

2.3. Subjective measures

The subjects' moods and feelings were self-rated on a 16-item visual analogue scales (VAS – see Supplemental file) originally developed for measuring drug-induced changes in mood and alertness (Aitken, 1969; Norris, 1971). Subsequently, these scales were found to be very sensitive to momentary changes in psychological states caused by psychological manipulations such as verbal threat (Bitsios et al., 1996, 1998a,b). They are easy and much faster (<60 s) to score than the Spielberger's State anxiety scale, they measure short term changes in anxiety and they are able to distinguish between changes in arousal levels and the emotion of anxiety. This is important as it has been shown previously that arousal may increase without accompanying increases in anxiety (Bitsios et al., 2004). The raw values (mm) for each item and each subject were weighted by multiplication with their respective factor loading and the weighted values for each item and subject were then allocated to 'alertness' (9 items), 'discontentment' (5 items) and 'anxiety' (2 items) factors, based on a principal component analysis (Bond and Lader, 1974). The average of the weighted group values for each factor was entered in the statistical analysis.

2.4. Part 1: testing procedure and training

Subjects had been previously informed that they would participate in one session where their fixational stability would be tested under various psychological conditions relevant to anxiety research. On arrival to the lab, all participants rested for 5 min during which they self rated their anxiety, alertness and mood using Visual Analogue Scale (VAS) questionnaires. Eye dominancy was determined by looking through a central hole in an A4 card, held by the participant in both hands away from the body. Subsequently, eye position data were obtained binocularly for two viewing conditions which were counterbalanced between subjects: in the absence of a fixational stimulus (volitional fixation), in which the volunteer was asked to keep fixation at the centre of the screen, and in the presence of a fixational stimulus (stimulus-driven fixation).

The stimulus, presented on a 21″ Sony GDM F-520 CRT monitor, was generated using custom written software for a VSG 2/5 stimulus generator card (Cambridge Research Systems Ltd., UK). The monitor was viewed from a distance of 100 cm. The stimulus was a 'profiled' spot, having a radial symmetrical spatial configuration of a blurred disk with a 0.25° flat top and a raised cosinusoidal skirt of 0.5° in diameter. The stimulus was presented at a Weber contrast of 40% for a total period of 15 s. The surround had a luminance (L) of 30 cd/m^2 (chromatic co-ordinates: $x = 0.310$, $y = 0.316$).

Recordings in this part served as 'training' in order to reduce novelty related arousal and familiarise subjects with the experimental procedures and were thus discarded from further analysis. At the end of part 1, subjects rated themselves again with the VAS, which was considered to be a more reliable baseline, since it was not confounded by novelty related arousal. Following these training procedures in part 1, subjects were given detailed instructions (see below) for part 2. They were reminded that they did not have to participate any further, however, all subjects agreed to participate and signed new consent forms for part 2 of the session.

2.5. Part 2: main session

The two viewing conditions described above were repeated with the same within-subject order as in training, under two psychological periods: they were both identical to training in part 1 but one was under verbal threat ("threat" period) and the other was not ("safe" period). Our verbal threat protocol has been described in detail previously (Bitsios et al., 2002, 1996; Hourdaki et al., 2005; Laretzaki et al., 2010). Briefly here, verbal threat (of electrical shock) was induced throughout the "threat" periods, by the presence of a Grass stimulator (SD 9) connected to the skin overlying the median nerve of the left wrist through disposable silver surface electrodes. Before and after connection to wrist electrodes, subjects completed the VAS questionnaires (see Fig. 1), in order to test whether electrode connection, a powerful contextual threat stimulus (Baas et al., 2002), induced adequate levels of anxiety. Subjects had been instructed to anticipate a total of 1–3 electric shocks but they were not aware of the exact number and timing of the electric shock(s). These were described as painful stimuli inducing a short-lived localized unpleasant sensation on the wrist. As threat was the actual variable of interest, no electric shock was actually delivered.

Half of the subjects within each trait anxiety group started with the "safe" and the other half with the "threat" condition. Therefore, there were four conditions (safe/volitional fixation, safe/stimulus driven fixation, threat/volitional fixation and threat/stimulus driven fixation) within each subject, which were all counterbalanced between subjects.

2.6. Data reduction and analysis

Data analysis was performed offline using custom-made scripts written in computational software (Matlab vs. 7.6.0.324). Fixation performance was evaluated using the Bivariate Ellipse Contour Area (BCEA), a mathematical description of fixation stability (Steinman, 1965). If the measured gaze positions are assumed to have a bivariate normal distribution, the ellipse area (BCEA) can be calculated using Eq. (1), where σ_H and σ_V are the standard deviations of position over the horizontal (x)

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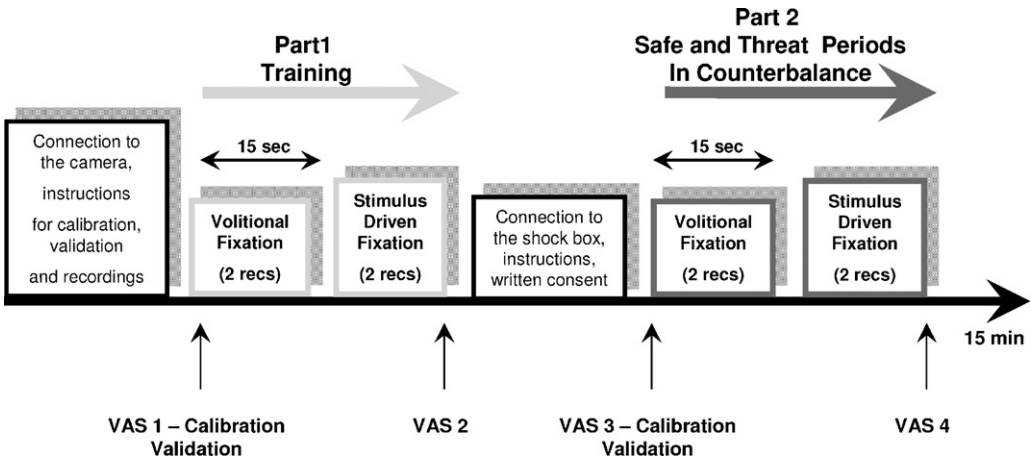

Fig. 1. The experimental session as followed for each volunteer, counterbalanced for safe and threat condition.

and the vertical (y) meridians, respectively, and ρ the product–moment correlation of the two position components. The value k establishes the confidence limit for the ellipse, i.e. the probability area (P) (Eq. (2)), where e is the base of the natural logarithm.

$$\text{BCEA} = 2k\pi\sigma_H\sigma_V(1-\rho^2)^{1/2} \quad (1)$$

$$P = 1 - e^{-k} \quad (2)$$

when $k = 1$, P describes fixation for 63.2% of the time (dispersion of the gaze about its mean position with ±1.0 standard deviation; Steinman, 1965). In the present study a value of $k = 1.14$ was used, with the calculated BCEAs corresponding to a P enclosing 68% of the recordings (see Fig. 3). Higher k values (i.e., 1.96) have also been applied but resulted in less repeatable BCEAs, due to inclusion of borderline position recordings. Fixation was expressed in visual angle area (arcmin^2) units. Blinks were identified, using an algorithm based on the horizontal and vertical position of the eye, and removed from further analysis, by discarding eye position data 0.1 s before the beginning and 0.1 s following the end of a blink.

The primary outcome variables of interest were BCEA in the four conditions (safe/volitional, safe/stimulus-driven, threat/volitional, threat/stimulus-driven) and the primary exposure of interest was trait anxiety. All scores entered the analyses as continuous variables. Multivariable linear regression models were performed to examine the association between trait anxiety and the outcomes of interest after adjusting for threat-induced state anxiety (defined as the Post–Pre electrode difference in VAS anxiety scores), age, education and smoking habit.

We also followed a dichotomous approach, comparing extreme tertile groups, based on trait anxiety scores. The cut-off scores for the lower and the higher tertile groups of the original cohort of 560 subjects were 33 and 40.0 respectively. Therefore, recruited subjects with a STAI-T score higher than 40 formed a high trait anxiety (HTA) group ($n = 14$) and those lower than 33 formed a low trait anxiety (LTA) group ($n = 14$). For this analysis, BCEA data were analysed using a mixed model repeated measures ANOVA with psychological period (safe, threat) and viewing condition (volitional fixation, stimulus driven fixation) as the within- and trait anxiety group as the between subjects factors. Pre- and post-electrode VAS scores for "alertness" "anxiety" and "discontentment" were compared with separate 2 × 2 (group by occasion) ANOVAs.

3. Results

Four subjects (STAI-T scores: 34, 35, 35 and 65) were excluded due to extremely poor fixation, which yielded empty cells in the SPSS or due to very large fixation areas and subsequent unreliable recordings. Therefore, 40 subjects were included in the regressions out of the 44 recruited in the study. The linear regressions revealed that trait anxiety was the only significant predictor of BCEA in the threat/volitional (beta: 0.598; $t = 4.5$; $p < 0.001$) and the threat/stimulus-driven conditions (beta: 0.384; $t = 2.4$; $p < 0.05$), explaining 43% and 15.8% of the BCEA variance respectively. Threat-induced, VAS-rated state anxiety, age, education and smoking habit did not predict BCEA in any of the two threat conditions (all $t < 1$ and all $p > 0.5$). No significant associations were found between BCEAs in the safe/volitional or safe/stimulus-driven conditions and trait or threat-induced state anxiety. The Pearson's r values between trait anxiety and BCEA were 0.630 ($p < 0.001$), 0.327 ($p < 0.02$), 0.262 ($p < 0.1$) and 0.098 ($p > 0.5$) for the threat/volitional, threat/stimulus-driven, safe/volitional and safe/stimulus-driven conditions respectively.

Table 1 shows the profile of the extreme tertile (LTA $n = 14$ and HTA $n = 13$) groups. Although the two groups differed significantly in trait anxiety, there were no group differences in demographic variables or in state (VAS) anxiety, discontentment and alertness at baseline part 1.

3.1. Subjective measures

Fig. 2 shows the group means of subjective mood and feelings obtained with the VAS on the day of testing. State anxiety increased in both groups after the application of electrodes [occasion main effect: $F(1,26) = 4.2$, $p < 0.05$] which was more anxiogenic

Table 1
Demographic characteristics for the two trait anxiety groups: low trait anxiety (LTA), high trait anxiety (HTA) (mean ± SD).

	LTA	HTA	t (or x^2)	P
Sample size	14	13		
Age (years)	26.9(3.6)	26.5(4.2)	<1	>0.7
Education (years)	18.1(2.7)	18.7(3.1)	<1	>0.5
Non-smokers/smokers[a]	8:6	7:6	<1	>0.9
Cigarettes/day	14.7(4.1)	19.5(13.2)	<1	>0.4
Baseline VAS anxiety, mm	12.0(16.2)	11.9(18.5)	<1	>0.9
Baseline VAS discontentment, mm	10.2(13.5)	8.3(12.1)	<1	>0.7
Baseline VAS alertness, mm	14.4(18.0)	19.0(23.2)	<1	>0.5
STAI-T score	29.0(2.7)	46.8(5.2)	11.3	<0.001

STAI-T: State Trait Anxiety Inventory-Trait questionnaire; VAS: visual analogue scales. For the t-tests df = 25.
[a] Chi square comparison.

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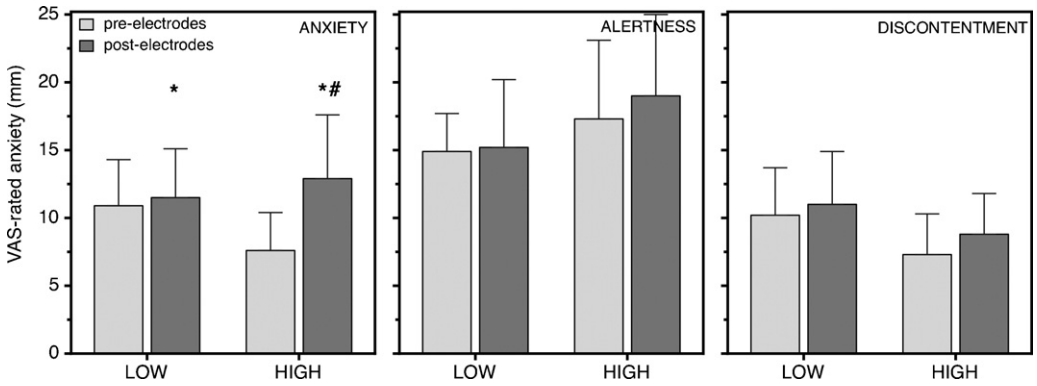

Fig. 2. Subjective anxiety, alertness and discontentment obtained before (light grey columns) and after (dark grey columns) threat induction via instructions and connection with shock electrodes. Columns represent group means and bars standard errors of the mean. *Significantly different from pre-electrode anxiety; #significantly different from post-electrode anxiety of the low trait anxiety group.

for the HTA group as evidenced by a significant group by occasion interaction [$F(1,26) = 4.9$, $p < 0.05$]. The group main effect was not significant ($F < 1$). No significant changes were observed in subjective alertness and discontentment (all F values <1.2).

3.2. Fixation stability

Eye position data under safe and threat psychological period for volitional and stimulus-drive fixation (through a 15 s eye tracking) are plotted in the scatter graphs of Fig. 3. Data from two subjects are presented, one with low and the other with high trait anxiety. The fitted Bivariate Contour Ellipse Areas (BCEAs) show that fixational performance is more stable under stimulus-driven fixation, for the safe condition and for the low trait anxiety participant.

Fig. 4 shows the ellipse areas of the two groups for the two psychological periods (safe and threat) and the two viewing conditions (volitional fixation, stimulus-driven fixation). It can be seen that compared to the LTA group, the HTA group had on average larger ellipse areas, especially for the "volitional fixation" condition in the threat period. These impressions were confirmed by the $2 \times 2 \times 2$ (psychological period $\times$ viewing condition $\times$ group) ANOVA. There were group [$F(1,26) = 6.8$, $p < 0.05$], psychological period [$F(1,26) = 13.8$, $p < 0.001$] and viewing condition [$F(1,26) = 17.9$, $p < 0.001$] main effects as well as group by psychological period [$F(1,26) = 8.5$, $p < 0.01$], group by viewing condition [$F(1,26) = 5.5$, $p = 0.05$] and psychological period by viewing condition [$F(1,26) = 8.2$, $p < 0.01$] interactions. The 3-way interaction reached a significant trend [$F(1,26) = 3.2$, $p = 0.083$]. Following an ANCOVA where state anxiety, measured as the post minus pre-electrode VAS scores (normalised after difference from baseline), was taken as the covariate, all the results above survived.

In order to improve presentation of the interactions between psychological and viewing conditions BCEA data was normalised for each subject. Fig. 5 depicts plots of fixation ratio (threat/baseline BCEA) for the two viewing conditions and the trait anxiety groups. Fixation ratio equal to 1.0 corresponds to no difference between the "threat" and "baseline" psychological conditions. Fixation ratio >1.0 corresponds to reduced fixation stability under the threat condition. It is evident that fixation ratio is increased in the HTA, compared to the LTA group, with the effect being more pronounced in the condition of volitional fixation.

4. Discussion

In this study, the ability of healthy males to maintain active fixation was tested in four different conditions: with or without the presence of a visual non-emotional fixation stimulus under a safe or a threatening context. We observed that fixation performance was generally more unstable in the volitional, compared to the stimulus-driven fixation condition replicating previous findings (Sansbury et al., 1973; Smyrnis et al., 2004) but our study is

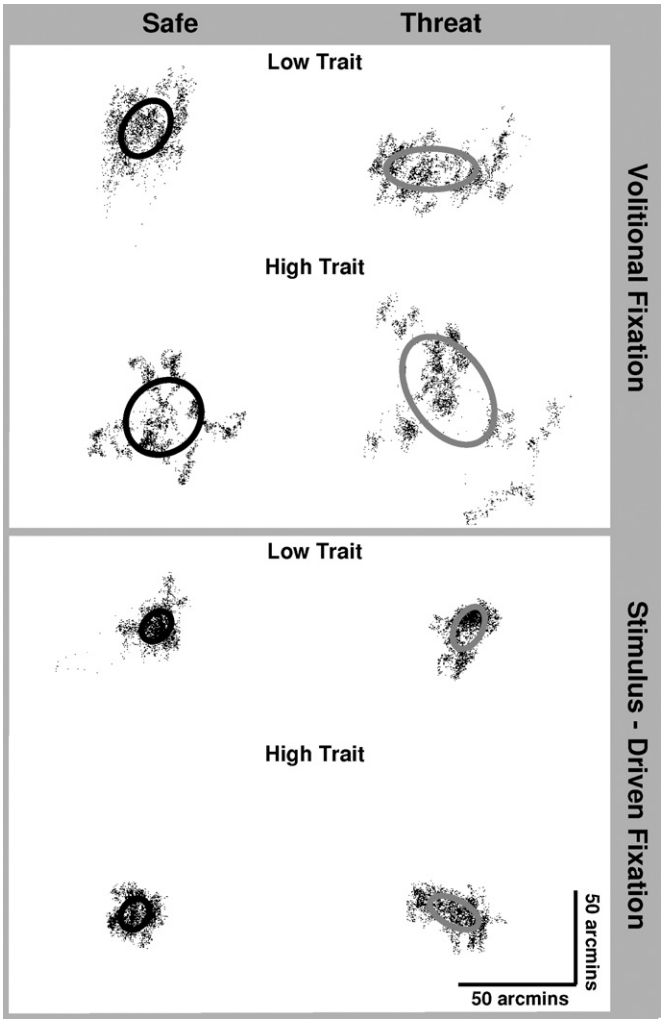

Fig. 3. Scatter graphs of fixation position and the fitted Bivariate Contour Ellipse Areas (BCEAs) under volitional fixation for two subject (one with low- and the other with high-trait anxiety) for the safe (left) and threat (right) psychological periods. Each BCEA contains 68% of fixation position through a 15 s eye tracking. The values at the bottom right corner of each scatter graph depict the area of the ellipses (fixation area) in arcmin^2.

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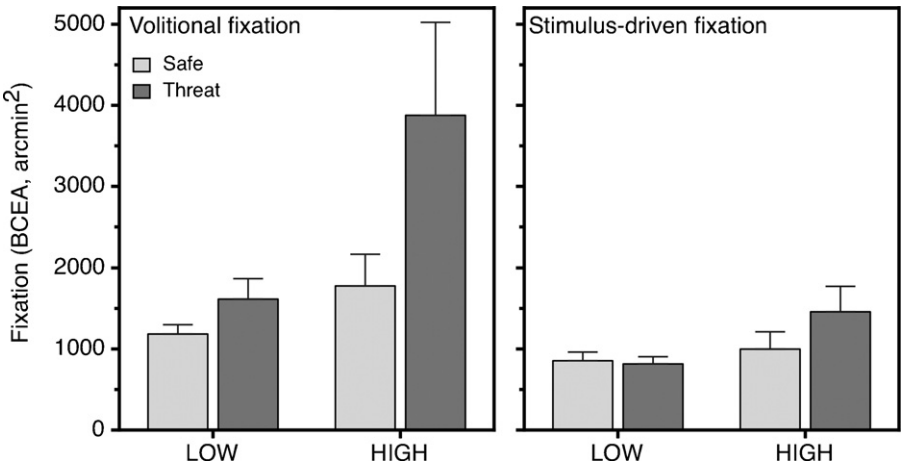

Fig. 4. Plots of fixation area (BCEA) during volitional (left) and stimulus-driven fixation (right) under the safe (light grey columns) and threat (dark grey columns) psychological periods for the high and low trait anxiety groups. The values represent group means and the bars the standard error of the mean.

the first to show that this was more pronounced in the threat condition. This is also the first study to demonstrate that trait anxiety affects fixation performance; while it is noticeable that the high trait anxious subjects had unstable fixation even in the absence of threat, their instability was more pronounced under threat or in the volitional fixation condition. These results suggest that threat alone is not sufficient to destabilise fixation, especially stimulus-driven fixation. Fixation instability is primarily associated with high trait anxiety with threat having an additive effect, which is particularly prominent in the absence of a fixational stimulus, when fixation is primarily mediated by extra-retinal cortical input.

Our results were consistent after continuous and dichotomous analyses and cannot be attributed to demographic differences or mood state at baseline (i.e. prior to the beginning of the experiment) as the two groups were similar in this respect. Our verbal threat procedures successfully elicited anxiety as evidenced by the significant occasion main effect in VAS-rated (state) anxiety, especially in the high trait anxiety group as evidenced by the significant group by occasion interaction. State anxiety reflects a combination of both trait anxiety and current environmental influences on mood state. The deficient fixation performance in the high trait anxiety group in the threat condition could therefore be due to a combination of higher trait and state anxiety in this condition. There are several reasons which, at first glance, argue against this possibility: (a) the regressions were adjusted for state anxiety and in the categorical analyses, the effects of trait anxiety on fixation survived after covarying for threat-elicited state anxiety (b) current anxious mood was previously found to have no effect on fixation performance (Smyrnis et al., 2004) and (c) the group main effect in the categorical analyses suggests less stable fixation of the high trait anxiety group also in the safe condition; this suggests that group differences in fixation performance reflect a deficit associated with vulnerability to anxiety rather than a symptomatic outcome of altered mood state in response to threat. However, it can be argued that a "safe" condition within an anxiety provoking context such as a threat-of-shock experiment may not always be assumed to be entirely devoid of increases in state anxiety. For this reason, we also checked fixation performance in the training part of the session, which took place prior to subjects' exposure to the anxiogenic verbal threat instructions. Analysis of these data revealed an identical pattern to that seen for the safe periods of part 2, i.e. greater fixation instability in the high trait anxious subjects under both viewing conditions, as well as greater instability in the volitional compared to stimulus-driven fixation condition (data not shown). Finally, it could be argued that these effects are not specific to trait anxiety or that a putative interaction of trait anxiety with other comorbid personality traits underlies our observations. Although this possibility cannot be entirely excluded, it is notable that schizotypy, one of the best characterised and measurable personality traits, which is frequently comorbid with anxiety, is not associated with fixation deficits in the presence or absence of a visual target (Thaker et al., 1996; Gooding et al., 2000; Smyrnis et al., 2004).

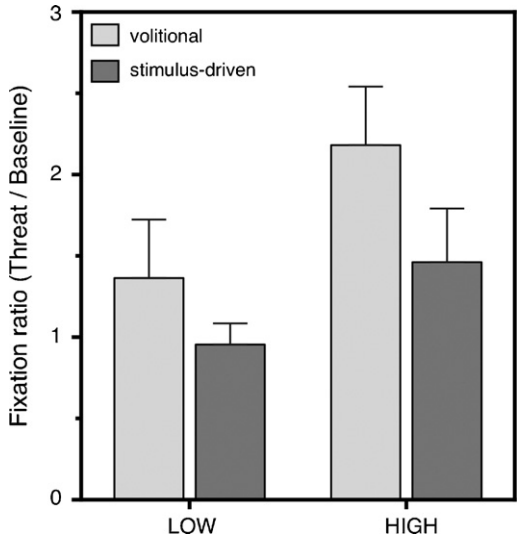

Fig. 5. Plots of fixation ratio (threat/safe fixation area) during volitional (light grey columns) and stimulus-driven (dark grey columns) fixation for the high and low trait anxiety groups. Fixation ratio equal to 1.0 corresponds to no difference between the threat and safe psychological conditions. The values represent group means and the bars the standard error of the mean.

It is interesting that fixation performance in the two fixation conditions, with and without a visual target, discriminated among the two trait anxiety groups of healthy subjects. It is striking that compared to low trait, the high trait anxious subjects had less stable fixation starting from the safe and visually guided condition and that their fixation instability was becoming more severe, presumably as a function of the recruitment of extra-retinal prefrontal areas. Indeed, BCEA in the safe/visually guided condition was smaller than in the threat/visually guided condition, which in turn was smaller than in the safe/volitional and finally the threat/volitional fixation condition. Volitional fixation is largely mediated by extra-retinal cortical input to the SC, probably of prefrontal origin (Munoz and Wurtz, 1993a; Gooding, 1999) and indeed, volitional fixation performance correlates with IQ (Smyrnis

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et al., 2004), a measure that heavily relies on higher cognitive mechanisms mediated by areas involved in executive functions such as the dorsolateral prefrontal cortex. The predominant theoretical stance has been that vulnerability to anxiety is associated with exaggerated responses of the threat-detection system, which is centred on the amygdala or related areas, such as the bed nucleus of the stria terminalis (Lang et al., 2000), but also with deficient active recruitment of prefrontal attentional mechanisms that are critical in the top–down control of selective attention to threat (Bishop et al., 2004; Ohman, 2005). Our results suggest that, beyond any potential abnormality in threat-related processing, trait anxiety is also characterised by inadequate recruitment of the PFC mechanisms that are implicated in the active control of attention in response to non-affective tasks, such as fixation performance in the absence of threat. This is in agreement with neuro-imaging evidence linking trait anxiety to impoverished recruitment of prefrontal attentional control mechanisms even in the absence of threat-related stimuli (Bishop, 2009), and with evidence showing that anxious individuals show deficits across a range of non-affective tasks that place demands on attentional or cognitive control (Mandler and Sarason, 1952; Fox, 1993; Eysenck and Calvo, 1992).

Eye movements were recorded under unnatural conditions in early research. For example, eye movements were recorded under monocular viewing or with the head immobilised on a biting board that ≪provided high stability of the head≫ (see Steinman, 2004 for a review). The results of the present study are based on binocular gaze recordings under more natural conditions, by avoiding the biting board and using a chin rest to only prevent large voluntary head movements. Therefore, our BCEA outcome variable may be confounded by small, involuntary and compensatory head movements and this may be seen as a limitation of our study. On the other hand, these conditions were more ecologically relevant, since they allowed for evaluation of the effect of trait anxiety and threat on the gaze rather than merely ocular stability. The observed fixation instability in high trait anxious subjects could be taken to mean decreased sensitivity to contrast, since fixation instability to a visual stimulus is thought to prevent sensory adaptation when sensitivity is reduced (Troxler, 1804; Blakemore and Campbell, 1969; Movshon and Lennie, 1979; Webster and De Valois, 1985). Although this hypothesis deserves testing with fixation targets of different contrasts and cannot be entirely ruled out, it does not accommodate these subjects' fixation instability in the volitional condition when no fixation target is present. It is more likely that our observations are in keeping with a role of microsaccades (unstable fixation) in attentional processing and strategic oculomotor planning (Ko et al., 2010; Kowler and Collewijn, 2010). This line of argument is open to two competing interpretations on the biological significance of our observations: (a) increased fixation instability in high trait anxious subjects represents a biologically adaptive search strategy i.e. more microsaccades to locate e perceived threat or (b) it represents a biologically maladaptive failure of attentional processing and strategic planning which would be more akin to loss of cognitive control and possibly (pertinent to our current paradigm) an internal state of panic. The second possibility finds some support from research in sport performance which suggests reliable anxiety-induced alterations in gaze performance with increased fixations in the periphery of the target (Moran et al., 2002; Savelsbergh et al., 2002), leading to inefficient and often ineffective search strategies (Janelle, 2002). Our results point to a primary "low level" effect of threat and trait anxiety on gaze fixation even in the absence of pictorial complexity such as that seen in visual search studies. It would be interesting to examine if perceptual training e.g. training to stabilize visually guided or volitional fixation under threat, could be beneficial in terms of reductions in subjective anxiety and its expression through its multiple physiological effector systems. This could have implications in sports performance or whenever performance under stress matters and also in the treatment of conditions associated with loss of cognitive control and strategic planning such as clinical anxiety. Better understanding of emotional and attentional interactions with early and basic visual processes might shed further light to the neurobiology of visually guided behavior, which may underlie a plethora of conditions from adaptive learning and conditioning processes to maladaptive anxiety states.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.biopsycho.2011.01.005.

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