1 **Bursts and variability of beta oscillations mediate the effect of anxiety on motor exploration and**
2 **motor learning.**

3 **Sebastian Sporn[1], Thomas P. Hein[2], María Herrojo Ruiz[2,3]***

4 [1] School of Psychology, University of Birmingham, Birmingham, UK

5 [2] Department of Psychology, Goldsmiths University of London, London, UK

6 [3] Neurology Department, Charité University of Medicine Berlin, Germany

7 ***Corresponding author**: María Herrojo Ruiz, email: M.Herrojo-Ruiz@gold.ac.uk. Address: Department of Psychology,
8 Goldsmiths, University of London. Lewisham Way, New Cross, London SE14 6NW (UK).

9 **Keywords: Anxiety, Motor learning, Variability, Beta Oscillations, Reward**

10

11

12

13

14

15

16

17

18

19

20

21

22

23

Abstract

Anxiety results in sub-optimal motor performance and learning through mechanisms still unknown. Here we addressed whether state anxiety impairs motor learning through changes in behavioral and neural variability. Participants completed a reward-based motor sequence learning paradigm, with separate phases for exploration (baseline) and learning. Anxiety was manipulated either during baseline or learning. We show that anxiety at baseline reduces motor variability, undermining subsequent reward-based learning. By contrast, unconstrained baseline exploration led to successful motor learning, even under the effect of anxiety. The behavioral changes were driven by changes in the variability of sensorimotor beta oscillations (13-30Hz, SBO). Moreover, bursts of SBO, a marker of physiological beta, lasted longer under the effect of anxiety, resembling recent findings of pathophysiological beta in movement disorders. Our findings suggest that changes in variability and burst duration in SBO represent a neural mechanism through which anxiety constrains movement variability, with detrimental consequences for motor learning.

Introduction

Anxiety involves anticipatory changes in physiological and psychological – cognitive, emotional, behavioral – responses to a potential and uncertain future threat[1-2]. Previous work on the neurobiology of anxiety established that trait anxiety interferes with prefrontal control of attention in perceptual tasks, whereas state anxiety modulates the amygdala during detection of threat-related stimuli[2-3]. In the area of motor control, research has shown that stress and anxiety have detrimental effects on performance[4-5]. These results have been partially interpreted as the interference of anxiety with information-processing resources[6]. However, the effects of anxiety on motor learning are often inconsistent and a mechanistic understanding is still lacking. Delineating mechanisms through which anxiety influences motor learning is important to ameliorate its impact in different settings, including in motor rehabilitation programmes.

Motor variability could be the primary component of motor learning that is affected by anxiety; it is defined as the variation of performance across repetitions[7], and is driven by various factors including sensory and neuromuscular noise[8]. As a form of action exploration, movement variability is increasingly recognized to benefit motor learning[9-11]. These findings are consistent with the vast amount of research on reward-based reinforcement learning demonstrating increased learning following initial exploration[12]. More recently movement variability was shown to benefit motor learning when it takes the form of 'intentional' exploration of the task space, not as motor noise[13]. Yet contextual factors can reduce variability. For instance, recent work on ritualistic behavior reveals that state anxiety leads to movement redundancy, repetition, and rigidity to regain a feeling of control[14]. This finding resembles the reduction in behavioral variability and exploration that manifests across animal species during the fight or flight response in stressful environments[15]. Based on these results we set out to test the hypothesis that state anxiety modulates motor learning through a reduction in motor variability and action exploration.

Additionally, we posited that changes in motor exploration are driven by neural variability in premotor and motor areas. Support for our hypothesis comes from recent data in animal studies demonstrating that variability in the primate premotor cortex tracks behavioral variability during motor planning[16]. Further evidence in rodents and primates supports that changes in variability in single-neuron activity in motor cortex drive motor exploration during initial learning, and reduce it following intensive training[17-18]. Also, the basal ganglia are crucial for modulating variability during learning and production, as shown in songbirds and, indirectly, in patients with Parkinson's disease[11, 19-20].

In the present study, we analyzed sensorimotor beta oscillations (SBO, 13-30Hz) as a candidate mechanism

2

66 driving motor exploration and variability. Beta oscillations have been linked to different aspects of
67 performance and motor learning[21-23], as well as reward-based learning[24]. Although amplitude or power
68 changes was traditionally the primary focus of research on oscillations, there is a renewed interest towards
69 assessing dynamic properties of oscillations, such as the presence of brief bursts[25], which are considered to
70 be a central feature of physiological beta in motor-premotor cortex and the basal ganglia[26-28]. The
71 assessment of variability and burst duration of SBO thus allows us to capture dynamic changes in neural
72 variability induced by anxiety and their link to behavioral effects.
73 To test our hypotheses, we recorded electroencephalography (EEG) in three groups of participants while
74 they completed a reward-based motor sequence learning paradigm, with separate phases for motor
75 exploration (baseline) and reward-based learning. Crucially, different sequences were used in each phase of
76 the task to exclude carry-over effects of learning from the baseline period. We manipulated anxiety by
77 informing participants about an upcoming public speaking task that would require them to describe an
78 unknown art object to a panel of experts[14]. Using a between-subject design, the anxiety manipulation
79 targeted either the baseline or the reward-based learning phase. Analysis of the EEG signals aimed to assess
80 anxiety-related changes in the variability and burst duration in SBO in relation to changes in behavioral
81 variability.
82 Our primary finding was that anxiety impairs reward-based learning by constraining motor variability and
83 action exploration during the baseline phase. Importantly, these effects were mediated by increased within-
84 trial variability and burst duration in SBO. A second experiment served to demonstrate that anxiety during
85 reward-based learning has an opposing effect on motor variability and learning rates depending on the
86 presence or absence of a preceding baseline exploration phase.
87
Results
88
89
90 Sixty participants completed our reward-based motor sequence learning task, consisting of three blocks of
91 100 trials each over two phases (**Figure 1**): a baseline motor exploration (block 1) and a reward-based
92 learning phase (blocks 2 and 3: termed training hereafter). Prior to the experimental task, we recorded in
93 each participant 3 min of EEG at rest with eyes open. Next, on a digital piano, participants played two
94 different sequences of seven and eight notes during the exploration and training phases respectively (**Figure
95 1A**). They were explicitly taught the tone sequences prior to the start of the experiment, yet precise
96 instructions about the timing or loudness (keystroke velocity) were not provided.
97   During the exploration phase, participants were informed they could freely change the rhythm and/or
98 the loudness of the performance of sequence1 every trial, and that no reward or feedback would be
99 provided. During training, however, participants received performance-based feedback in the form of a 0-
100 100 score at the end of each trial, and were informed that the overall average score would be translated
101 into monetary reward. They were directly instructed to explore the temporal or loudness dimension (or
102 both) and to use feedback scores to discover the unknown performance objective (which, unbeknownst to
103 them, was a specific rhythmic pattern). The task-related dimension was therefore timing, whereas keystroke
104 velocity (Kvel) was the non-task related dimension. Timing in our task referred to the pattern of inter-onset-
105 intervals between consecutive keystrokes (IOI, ms). The score increased when the difference between the
106 coefficient of variation of the performed and target rhythm patterns (IOIs) decreased (see *Materials and
107 Methods*).
108   Participants were pseudo-randomly allocated to either a control group or to one of two experimental
109 groups (**Figure 1B**): anxiety during exploration (anx1); anxiety during the first block of training (anx2). The
110 lack of anxiety manipulation during block3 thus allowed us to assess the dissociable effects of anxiety

3

111   during baseline exploration or training on the learning rates during the last training block. We measured
112   changes in heart-rate variability (HRV), heart-rate (HR) and state anxiety scores four times throughout the
113   experimental session: resting state (3 min, prior to performance blocks); block1; block2; block3. The HRV
114   significantly dropped during the targeted blocks relative to the initial resting phase in each group (**Figure 1 –**
115   **figure supplement 1**), confirming that the experimental manipulation succeeded in inducing physiological
116   responses consistent with an anxious state[29]. Statistical analysis of behavioral and neural measures focused
117   on the separate comparison between each experimental group and the control group (contrasts: anx1 –
118   controls, anx2 – controls). See *Materials and Methods*.
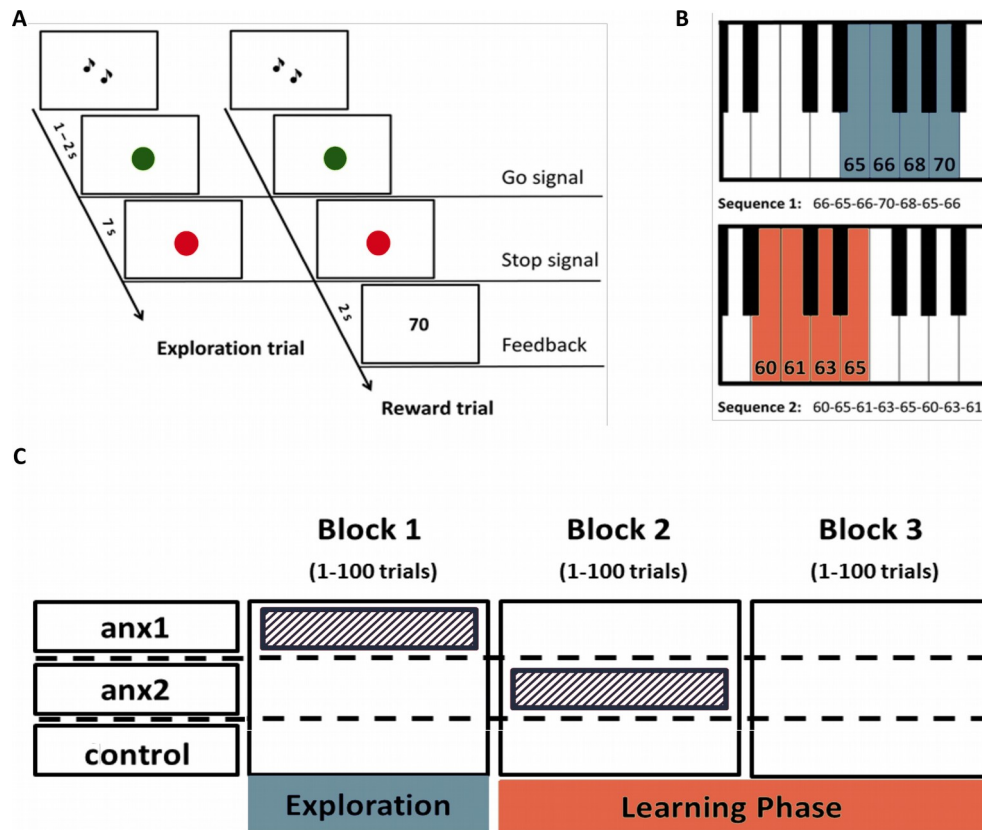
Figure 1. **A Novel Paradigm for Testing Exploration and Reward-Based Learning during Sequence Performance.** (A)
Schematic of the task. Participants played sequence1 during 100 exploration trials, followed by 200 trials of reward-
based learning performing sequence2. After each reward-based learning trial, participants received a performance-
related score between 0-100. (B) Pitch content of the sequences used in the exploration (sequence1) and reward-
based learning blocks (sequence2), respectively. (C) Schematic of the anxiety manipulation. The shaded area denotes
the phase in which anxiety was induced in each group, using the threat of an upcoming public speaking task, which
took place immediately after that block was completed.

**General Effects of Baseline Task-related Variability and Exploration on Reward-based Learning**

All groups of participants demonstrated significant improvement in the achieved scores during reward-

4

157   based learning, confirming they effectively used feedback to approach the hidden target performance
158   (**Figure 2**: $p < 0.05$, after control of the false discovery rate at level $q = 0.05$ due to multiple comparisons[30],
159   termed FDR-corrected thereafter; anx1: non-parametric effect size[31], $PS_{dep}= 0.80$; anx2: $PS_{dep}= 0.88$; controls:
160   $PS_{dep}= 0.90$). Detailed analysis of the trial-by-trial changes in scores and performance will be reported
161   elsewhere.
162   Assessment of motor variability was performed separately in the task-related temporal dimension and
163   the non-task-related keystroke velocity dimension. Temporal variability – and similarly for keystroke velocity
164   – was estimated using two different measures (**Figure 2B**): the within-trial and across-trials coefficient of
165   variation of IOI (cvIOI). The within-trial cvIOI provided a total of 100 values across each experimental block.
166   By contrast, the across-trials cvIOI provided one single value per experimental block. Because the score
167   obtained during reward-based learning was explicitly related to the within-trial cvIOI, we predicted that
168   higher values of this parameter at baseline would be associated with higher reward during the subsequent
169   training phases. Of note, higher within-trial cvIOI values denote a larger departure from an isochronous
170   performance of the sequence. However, we also hypothesized that a higher degree of exploration across
171   trials at baseline (that is, playing different temporal patterns in each trial), and therefore higher across-trials
172   cvIOI, would improve subsequent reward-based learning.

173

5
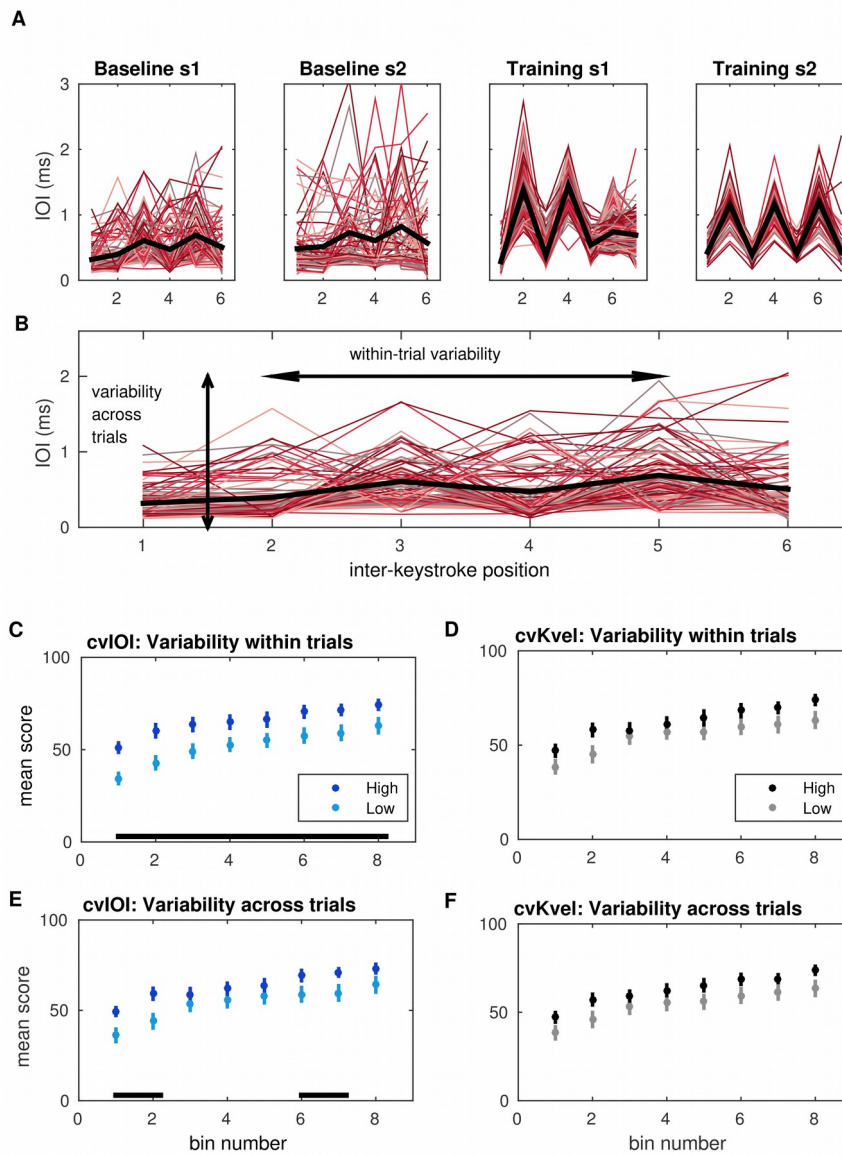
Figure 2. Temporal variability within and across trials at baseline contributes to subsequent reward-based learning. (A) Illustration of timing performance during baseline exploration (left panels) and training (right panels) blocks in two representative participants, s1 and s2. X-axis represents the position of the inter-keystroke interval (sequence1: 7 notes, corresponding to 6 inter-keystroke temporal intervals; sequence2: 8 notes, 7 inter-keystroke intervals). Y-axis shows the inter-onset interval (IOI) in ms. Black lines represent the mean IOI pattern. (B) Task-related variability was measured using two parameters: the within-trial and across-trials coefficient of variation of IOI, cvIOI. (C) Scores achieved by participants during training following a median split of all 60 participants into high and low within-trial cvIOI at baseline. Trials were split into bins of 25 trials and scores were averaged within each bin. Black bars at the bottom indicate the bins of significant between-group differences (p < 0.05, FDR-corrected). (D) Same as C but for keystroke velocity, using cvKvel to do a median split of participants. E-F. Same as C-D but using the across-trials cvIOI and cvKvel, respectively. Bars around the mean display ±SEM.

6

208 To first evaluate the effect of baseline within-trial temporal variability on subsequent reward-based
209 learning, regardless of the group, we did a median split of all 60 participants based on the within-trial cvIOI,
210 averaged across trials. This analysis revealed that larger within-trial cvIOI at baseline was associated with
211 higher scores during training ($p < 0.05$, FDR-corrected; PS_{dep}= 0.91; **Figure 2C**). Corresponding with this
212 result, there was a significant non-parametric rank correlation between the values of within-trial cvIOI at
213 baseline, and also later during training – as expected, and the average scores obtained (Spearman ρ = 0.474,
214 p = 0.001, at baseline; ρ = 0.646, p = 0.00001, during training). A control analysis performed with groups of
215 low and high values of within-trial cvKvel demonstrated a non-significant difference in subsequent scores (p
216 > 0.05; **Figure 2D**).

217   We also stratified participants based on the degree of across-trials cvIOI at baseline exploration.
218 Participants whose performance exhibited a higher across-trials cvIOI at baseline achieved higher scores
219 during training ($p < 0.05$, FDR-corrected; PS_{dep} = 0.81; **Figure 2E**). Changes in across-trials cvKvel did not
220 influence subsequent reward-based learning ($p > 0.05$; **Figure 2F**).

221   Notably, the amount of within-trial variability expressed by participants in timing and keystroke velocity
222 was not correlated (ρ = 0.019, p = 0.898). Neither was the across-trials cvIOI and cvKvel (ρ = 0.021, p =
223 0.788). This supports that the temporal and velocity dimensions in our task were uncorrelated and, in
224 principle, participants could vary them separately. Participants, however, generally used a lower amount of
225 variability in Kvel relative to timing at baseline, likely due to the higher difficulty required to precisely
226 control loudness during piano performance.
227
228 **Influence of Anxiety on Baseline Variability and Subsequent Reward-based Learning**
229 Next, we assessed pair-wise differences between each experimental group (anx1, anx2), separately, and the
230 control group. Participants affected by state anxiety at baseline (anx1) achieved significantly lower scores in
231 the subsequent reward-based learning phase relative to control participants (**Figure 3A:** $p < 0.05$, FDR-
232 corrected, between-group non-parametric effect size[31], PS_{sup} = 0.78). By contrast, in the anx2 group scores
233 did not significantly differ from the scores in the control group ($p > 0.05$). Converging with the previous
234 analysis, the total average score (related to the amount of money received) achieved by anx1 participants
235 was significantly smaller than the amount received by control participants (52 [SEM 3] for anx1, 63 [3] for
236 controls, p = 0.02, PS_{sup} = 0.85). Anx2 and controls did not achieve significantly different average scores than
237 control participants (61 [3] for anx2; $p > 0.05$). A planned comparison between both experimental groups
238 demonstrated significantly higher total average scores in anx2 (p = 0.045, PS_{sup} = 0.67).
239   At baseline, anx1 used a lower degree of within-trial and across-trials cvIOI than the control group
240 (**Figure 3BC**. Within-trial cvIOI: $p < 0.05$, FDR-corrected; PS_{sup} = 0.67; Across-trials cvIOI: p = 0.032; PS_{sup} =
241 0.67). There was no between-groups (anx1-controls) difference in within-trial or across-trials variability in
242 Kvel ($p > 0.05$, **Figure 3 – figure supplement 1**). Performance at baseline in anx2 did not significantly differ
243 from performance in the control group, both for cvIOI or cvKvel, and for within and across-trials variability
244 ($p > 0.05$).
245
246 **Performance in the Training Phase: Exploration and Exploitation**
247 We evaluated whether the significant increase in scores found in each group from beginning to end of the
248 training blocks was paralleled by a significant drop in the across-trials cvIOI, reflecting exploitation of the
249 rewarded options (**Figure 3**). A 2x2 factorial analysis of the across-trials cvIOI with factors Group (anx1,
250 control) and Phase of training (block2, block3) demonstrated a significant main effect Phase and interaction
251 effect ($p < 0.05$, FDR-corrected). Further exploration of the interaction effect established that in control

7

252 participants – not in anx1 – the across-trials cvIOI dropped from training block2 to block3 (p < 0.05, FDR-
253 corrected, PS_{sup} = 0.66). A similar 2x2 analysis comparing anx2 and control groups revealed a significant
254 main effect Phase (p < 0.05, FDR-corrected), due to smaller across-trials cvIOI values in block3 in both
255 groups. Collectively, these findings support that during reward-based learning exclusively participants in the
256 anx2 and control groups went through a gradual transition from an explorative regime (characterized by
257 higher across-trials cvIOI) to an exploitative regime, in parallel to their achieving higher scores.

258 Additional similar 2x2 factorial analyses of the average score and within-trial cvIOI with the
259 abovementioned Phase and Group factors demonstrated significant main effects for Phase in all cases (p <
260 0.05, FDR-corrected: all groups had larger scores and within-trial cvIOI in the second training block), a main
261 effect Group for anx1 and controls (p < 0.05, FDR-corrected) and no significant interaction effects. This
262 finding suggested that the transition in scores and within-trial task-related variability from the first to the
263 second training blocks was similar in all groups, despite anx1 having significantly overall lower within-trial
264 cvIOI and lower scores than control participants.
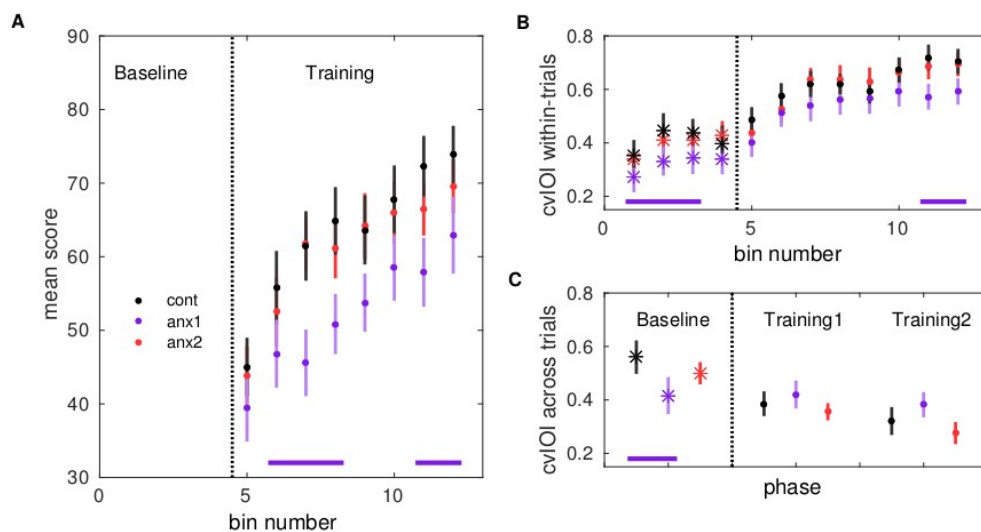
**Figure 3. Effects of anxiety on behavioral variability and reward-based learning.** The score was computed as a 0-100
normalized measure of proximity between the pattern of inter-onset intervals performed in each trial and the target
rhythm ([0.2, 1, 0.2, 1, 0.2, 1, 0.2] s). (A) Scores achieved by participants in the anx1, anx2, and control groups across
bins 5:12 (bins of 25 trials: trial range 101-300), corresponding with blocks 2 and 3 and the training phase. Participants
in anx1 achieved significantly lower scores than control participants in bins 6:8 and 11:12 (trials 125-200 and 250-300,
p < 0.05, FDR-corrected, denoted by the bottom purple line). (B) Changes in within-trial cvIOI from the exploration
phase (bins 1-4) to the training phase (bins 5-12). Participants in anx1 used smaller within-trial cvIOI than controls
during exploration (bins 1-3) and at the end of the training blocks (bins 11-12, p < 0.05, FDR-corrected). Anx2
participants did not differ from control participants. (C) Same as (B) but for the across-trials cvIOI, revealing a signifcant
drop in task-related exploration at baseline in anx1 relative to control participants (p < 0.05, FDR-corrected). Bars
around the mean show ±SEM.

Without Baseline Exploration, State Anxiety during Reward-based Learning Reduces Learning Rates.
293 Because participants in anx2 performed at a level not significantly different from that found in control
294 participants, we asked whether the initial unconstrained motor exploration at baseline might have
295 counteracted the effect of anxiety during training. To that aim, we performed a control behavioral
296 experiment with new control and anx2 groups (N =13 each). Participants in each group performed the two

8

298 training blocks 2 and 3 (**Figure1**), but without completing a preceding baseline exploration block. In anx2,
299 state anxiety was induced exclusively during the first training block, as in the original experiment. We found
300 that HRV and within-trial temporal variability were significantly reduced in anx2 relative to controls during
301 the manipulation phase ($p < 0.05$, FDR-corrected, **Figure 3 – figure supplement 2).** Moreover, anx2
302 participants achieved significantly lower scores than control participants during the first training block ($p <$
303 0.05, FDR-corrected), yet not during the second training block ($p > 0.05$). Importantly, however, overall anx2
304 participants achieved a lower average score (and monetary reward) than control participants ($p = 0.0256$;
305 $PS_{sup} = 0.64$). The degree of across-trial temporal variability did not differ between both groups, yet in the
306 control group – not in anx2 - there was a significant transition from an explorative to an exploitative regime
307 (drop in across-trials cvIOI, $p = 0.0001$, $PS_{dep} = 1$), as expected.
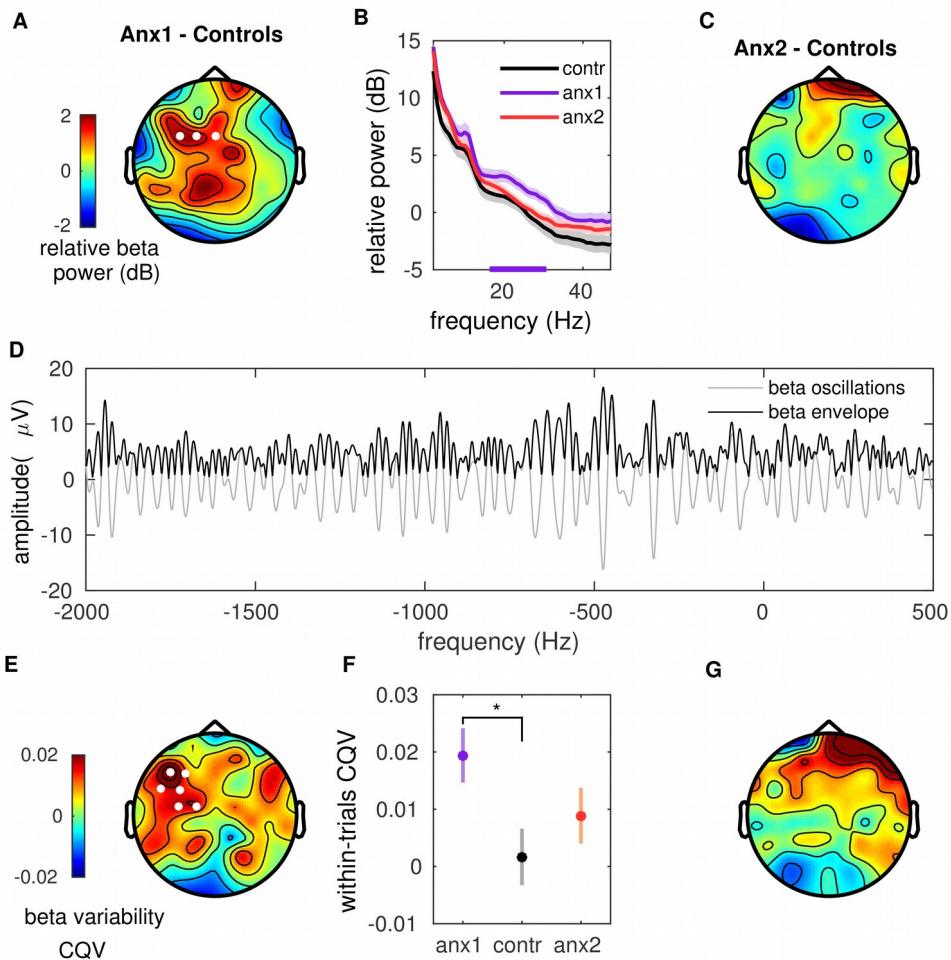
Figure 4. Sensorimotor beta activity during baseline exploration is modulated by anxiety. (A) Topographical
representation of the between-group difference (anx1-controls) in normalized beta-band power spectral density (PSD)
in dB. A larger beta-band PSD increase was found in anx1 relative to control participants in a small cluster of
contralateral sensorimotor electrodes (white dots indicate significant electrodes, two-tailed cluster-based permutation
test, $p < 0.025$, FWE-corrected). (B) Averaged PSD within 4-45Hz for each experimental and control group
corresponding to the cluster shown in (A). Beta-band power differences were additionally assessed within the broader
range 4-45Hz, revealing an effect exclusively within 17-30Hz ($p < 0.05$, FDR-corrected), denoted by the purple line at

9

344   the bottom). No significant effects outside the beta range were found. Anx2 and control participants did not differ in
345   power modulations. Shaded areas denote mean ±SEM. (C) Same as (A) but for differences in beta-band PSD between
346   anx2 and control participants. No significant clusters were found. (D) Illustration of the amplitude of beta oscillations
347   (gray line) and amplitude envelope (black line) for one representative subject and channel. (E) Scalp topography for
348   between-group differences in the coefficient of quartile variation (CQV) of the beta-band amplitude envelope, as a
349   measure of beta-band amplitude variability. We obtained one significant cluster of left sensorimotor electrodes (white
350   dots, p < 0.025, FWE-corrected), due to larger beta-band variability in anx1 than in control participants. (F) Beta-band
351   CQV index averaged within the electrodes pertaining to the significant positive cluster shown in (E). Data shown as
352   mean and ± SEM. Significant differences between anx1 and control groups are indicated by the asterisk. (G) Same as
353   (E) but for beta-band CQV differences between anx2 and control participants. No significant differences were found.

354

355   **Variability in Beta Oscillations at Baseline is Enhanced by State Anxiety**
356   We assessed whether the changes in motor variability found during baseline exploration are associated with
357   changes in sensorimotor beta-band oscillatory activity. Specifically, we tested whether within-trial variability
358   in the amplitude envelope of beta oscillations is influenced by state anxiety at baseline in anx1 relative to
359   control participants – using the coefficient of quartile variation (CQV[32]) as a measure of relative dispersion.
360   In addition, between-group differences in the averaged normalized power spectral density (PSD) of beta
361   oscillations were evaluated. Normalization of the raw PSD into decibels (dB) was carried out using as
362   reference the average PSD from the initial rest recordings (3 min). Results on the effects of anxiety on the
363   modulation of beta oscillations by feedback-locked reward processing will be reported elsewhere.
364   We found a significantly higher beta-band power in a reduced set of three channels in the contralateral
365   sensorimotor region in anx1 relative to control participants at baseline (p < 0.025, two-sided cluster-based
366   permutation test[33]; PS_{sup} = 0.73. **Figure 4A-B**). By contrast, in anx2 participants, the beta power was not
367   significantly different than in controls (**Figure 4C**, p > 0.05). No significant between-group changes in PSD
368   were found in lower (<13Hz) or higher (>30Hz) frequency ranges (p > 0.05). Crucially, in anx1, the CQV of
369   beta oscillations was significantly higher than the values in the control group across an extended set of
370   channels in the left sensorimotor region (p < 0.025, PS_{sup}= 0.80 **Figure 4D-E**). No difference in the CQV of
371   beta oscillations was found between anx2 and control participants (**Figure 4F**). Thus, the anxiety
372   manipulation during baseline exploration led to a pronounced enhancement of within-trial beta variability
373   in contralateral sensorimotor electrodes. This indicates a more irregular range of dynamic changes of beta
374   amplitude. To a lesser degree, the anxiety manipulation at this phase also increased contralateral
375   sensorimotor beta power, although in a more locally confined set of electrodes.
376   A similar analysis in the training period revealed no significant between-group beta power differences
377   (**Figure 4 – figure supplement 1**). There was, however, significantly larger within-trial beta-band variability
378   in contralateral sensorimotor electrodes in anx1 relative to control participants (p < 0.025). Accordingly,
379   despite the targeted effect of the anxiety manipulation in the anx1 group, which led to changes in HRV
380   exclusively in the baseline phase, the larger variability of beta oscillations found during baseline extended to
381   the training period as well. Anx2 participants also exhibited larger beta-band CQV values relative to control
382   participants, albeit in a region of frontal electrodes (p < 0.025).

383

384   **State Anxiety during Exploration Prolongs Beta Bursts**
385   To explore further the result of anxiety-related increases in within-trial variability in beta oscillations, we
386   assessed the distribution and duration of beta bursts. To identify bursts of beta oscillations and assess the
387   distribution of their duration, we applied an above-threshold detection method, which was adapted from
388   previously described procedures[25,27](see **Figure5A** and *Materials and Methods*). Bursts extending for at least
389   one cycle were selected. Using a double-logarithmic representation of the probability distribution of burst

10

390 durations, we obtained a power law and extracted the slope, τ, also termed "life-time" exponent[25].
391 Modelling work has revealed that a power law in the burst-duration distribution (slope τ = 1.5), reflecting
392 that the oscillation bursts or neuronal avalanches have no characteristic scale, indicates that the underlying
393 neural dynamics operate in a state close to criticality, and thus benefitial for information processing[25,34].
394   During baseline exploration, beta bursts lasted significantly longer in anx1 as compared to control
395 participants (**Figure 5B**, p < 0.025, PS_{sup} = 0.75). This effect was most pronounced in a cluster of electrodes in
396 the contralateral sensorimotor area, resembling the topography of the CQV effects (**Figure 4**). The mean
397 burst duration in these electrodes was 147 (2) ms in control participants and 168 (10) ms in the anx1 group,
398 with a difference of 20 ms corresponding with at least 2 cycles of 13Hz oscillations (5 cycles of 30Hz
399 oscillations). A further between-group comparison focusing on the distribution of burst duration
400 demonstrated that shorter bursts were significantly more frequent in control relative to anx1 participants
401 (130-194ms, p < 0.05, FDR-corrected; PS_{sup} = 0.70; **Figure5CD**). By contrast, long bursts of 630-1130ms were
402 more frequent in anx1 than control participants (p < 0.05, FDR-corrected, PS_{sup} = 0.92). The life-time
403 exponents were smaller in anx1 than in the control group at left sensorimotor electrodes (1.43 [0.30]; 1.70
404 [0.15]; p < 0.05, FDR-corrected; PS_{sup} = 0.81). No differences in mean burst duration, life-time distribution, or
405 exponents were found between anx2 and control participants. Regarding the distribution of beta bursts
406 throughout the trial, the probability in all groups increased significantly at the completion of the trial-wise
407 performance, as reported previously[28,30] (p < 0.05 in all groups, FDR-corrected; **Figure 6**). Interestingly,
408 between-group comparisons demonstrated that, during sequence performance, the probability of
409 oscillation bursts dropped in anx1 relative to control participants (p < 0.05, FDR-corrected), due to the
410 smaller rate of brief bursts in this experimental group (**Figure 6 – figure supplement 1**).

411

412 During training, the mean duration of bursts in anx2 or anx1 was not significantly different from values
413 obtained in the control group (**Figure 5E**, p > 0.05). However, long bursts were more frequent in anx2 than
414 in control participants (**Figure 5FG**, duration 630-930 and 1380-1680ms; p < 0.05, FDR-corrected, PS_{sup} =
415 0.71), supporting that each experimental group exhibited longer beta bursts relative to control participants
416 during the blocks affected by the anxiety manipulation. Within-group comparisons further confirmed this
417 outcome, demonstrating that the average burst duration was longer during baseline exploration than during
418 training in anx1 across left sensorimotor electrodes (p < 0.05, FDR-corrected, PS_{sup} = 0.73) – despite anx1
419 also exhibiting significantly more frequent long bursts during training than controls (630-770ms, p < 0.05,
420 FDR-corrected, PS_{sup} = 0.68). Also, the burst duration was significantly longer during the first block of the
421 training phase than at baseline in anx2 (p < 0.05, FDR-corrected, PS_{sup} = 0.71). In control participants, the
422 duration of beta bursts did not change across the experimental blocks (p > 0.05). Throughout the trial, the
423 probability of beta bursts did not differ between groups; yet there was a significant within-group increase in
424 burst probability from beginning to end of the trial in all channels and all groups (p < 0.05, FDR-corrected,
425 **Figure 6**). Following the feedback presentation, the burst probability dropped significantly relative to the
426 end of the trial in each group (p < 0.05, FDR-corrected) and similarly in all groups. The life-time exponent, τ,
427 did not differ between groups (p > 0.05, around 1.6 on average in all groups).
428   Lastly, smaller slope values τ – corresponding with long-tailed distributions of burst duration due to
429 the more frequent long bursts, as in anx1 – were associated with higher beta-band CQV across participants,
430 and both during exploration and during training (Spearman ρ = 0.496, p = 6 x 10^{-4} for exploration, ρ = 0.413,
431 p = 0.0011 for training; N = 60; **Figure 6 – figure supplement 2**).
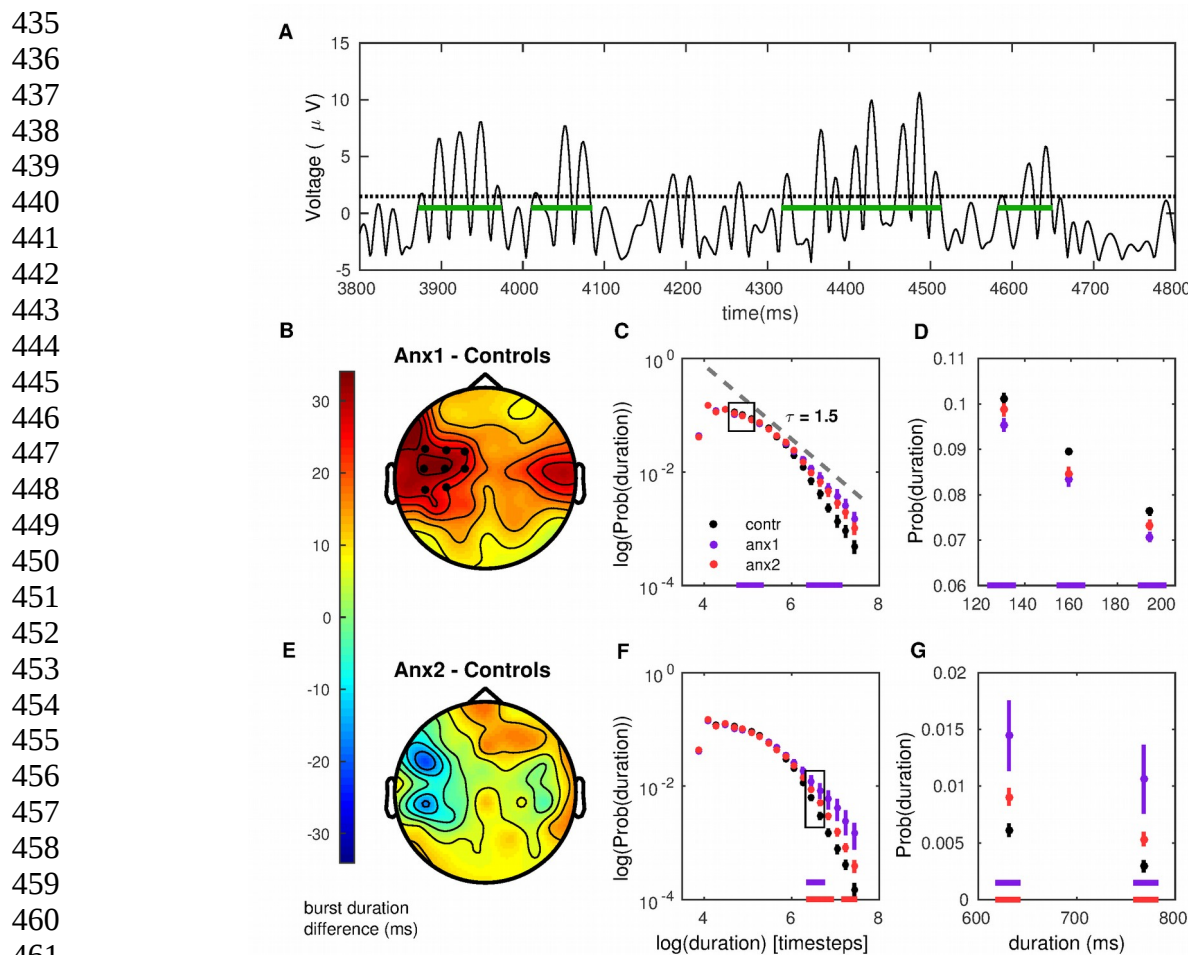
432

433

434

11

Figure 5. Anxiety during baseline exploration modulates the duration of sensorimotor beta-band oscillation bursts. (A) Illustration of the threshold-crossing procedure to detect beta oscillation bursts[25,27]. A threshold of 75% of the beta-band amplitude envelope was selected and beta bursts extending for at least one cycle were accepted. Windows of above-threshold oscillation bursts detected in the beta-band amplitude envelope (black line) are denoted by the green lines. (B) Scalp topography for between-group changes in the mean burst duration during baseline exploration. A significant positive cluster was found in an extended cluster of left sensorimotor electrodes, due to a longer average burst duration in anx1 than in control participants (20-30ms longer; Black dots indicate significant electrodes, two-tailed cluster-based permutation test, p < 0.025, FWE-corrected). (C) Probability distribution of beta-band oscillation-burst life-times within range 50-2000ms for each group during baseline exploration. The double-logarithmic representation reveals a power law within the fitted range (timesteps in logarithmic x-axis 4.09-7.62, corresponding to time windows 59.64 – 2053ms; first timestep excluded from the fit[25]). For each power law we extracted the slope, τ, also termed life-time exponent. The dashed line illustrates a power law with τ = 1.5. Significant differences between anx1 and control participants in oscillation-burst durations are denoted by the purple line at the bottom (p < 0.05, FDR-corrected). The rectangle highlights the area enlarged and displayed in the right panel (D). Data shown as mean and $\pm$ SEM. (E) Same as (B) but for differences in mean burst duration between anx2 and control groups during training. No significant differences were found. (F) Same as (C) but during training. Significant between-group differences were found for long-lived oscillation bursts within 630-930 and 1380-1680ms (anx2-controls, red bar at the bottom; p < 0.05, FDR-corrected) and 630-770ms (anx1-controls, purple bar at the bottom). (G) Enlarged display of the region of between-group significant differences highlighted by the rectangle in (F).
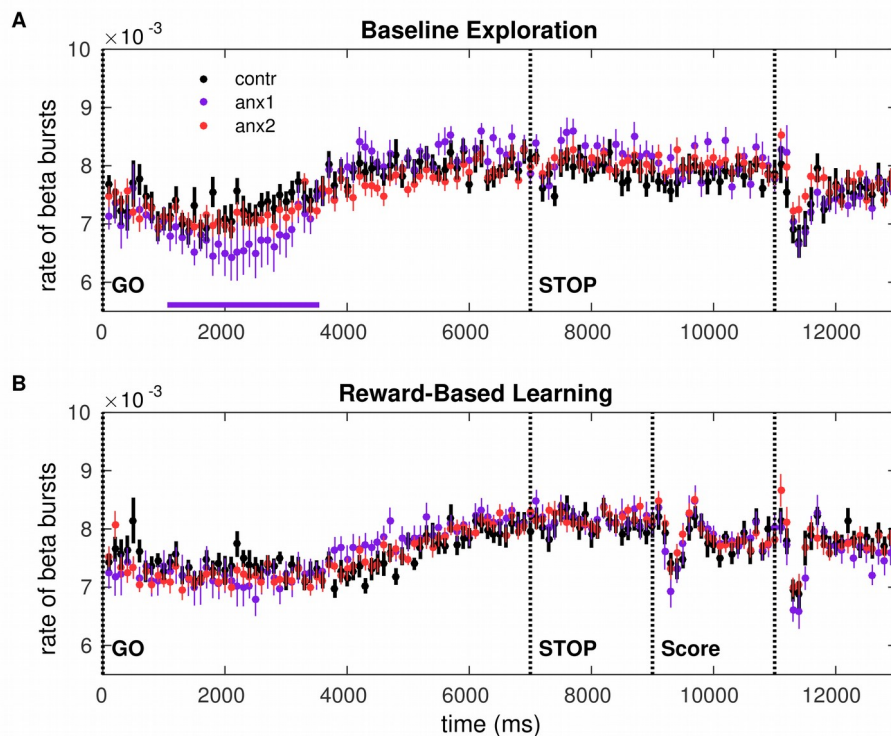
12

Figure 6. Time course of the rate of beta-band oscillation bursts throughout trial performance. (A) Rate of beta bursts during sequence performance in the baseline exploration phase. Participants completed sequence1 on average between 600 (SEM 100) and 3600 (100) ms (non-significant differences between groups, p> 0.05). The STOP signal (red ellipse on the monitor) was displayed 7000 ms after the GO signal. At 11000 ms the trial ended (the red ellipse vanished). In all groups there was a significant increase in the rate of oscillation-bursts duration following completion of the sequence performance (0-3500ms versus 3500 – 7000s trial segments, p < 0.05, FDR-corrected). In addition, between-group comparisons demonstrated a significant drop in the burst rate in anx1 participants relative to control participants during sequence performance (1100-3500 ms, denoned by the purple bar at the bottom; p < 0.05, FDR-corrected). Data display the mean and ± SEM. (B) Same as (A) but for the training period, when participants played sequence2. At 9000 ms, 2000 ms after the STOP signal, the feedback score was displayed for 2000 ms. There was a within-group significant increase in burst rate following completion of the sequence performance (0-3500ms versus 3500 – 7000s trial segments, p < 0.05, FDR-corrected) and a subsequent significant drop following feedback presentation (p < 0.05, FDR-corrected). No significant between-group effects were found.

Discussion

Our findings expand previous computational modelling and experimental work that linked anxiety levels (trait) and poorer performance, albeit in aversive environments[2-3,35]. The results demonstrate that state anxiety impaired motor variability and exploration at baseline, decreasing performance in a subsequent reward-based learning phase. Participants with larger task-related variability and exploration at baseline scored higher during the following training phase, extending recent findings on the faciliatory effect of exploration on motor learning[10,36]. Crucially, combining evidence from both experiments, we were able to show that reward-based learning is not affected by concurrent state anxiety if participants were given the opportunity of unlimited exploration during a preceding baseline phase. On the neural level, state anxiety during baseline exploration increased variability of beta oscillations in the contralateral sensorimotor

13

528 cortex; and to a lesser degree, also enhanced average beta power. Finally, bursts of sensorimotor beta
529 oscillations, a marker of physiological beta, lasted longer under the effect of anxiety, resembling recent
530 findings of abnormal burst duration in movement disorders.
531 These results thus provide the first evidence for changes in variability and burst duration of sensorimotor
532 beta oscillations mediating the effects of anxiety on motor exploration, with negative consequences for
533 reward-based motor learning.
534
535 **Anxiety constrains motor variability and exploration**
536 Previous studies manipulating psychological stress and anxiety to assess motor learning showed both a
537 deleterious and facilitatory effect[37-38]. Differences in experimental tasks, which often assess motor learning
538 during or after high-stress situations but not during anxiety induction in anticipation of a stressor, could
539 account for the previous mixed results. Here, we adhere to the neurobiological definition of anxiety as a
540 psychological and physiological response to an upcoming diffuse and unpredictable threat[1,2]. Accordingly,
541 anxiety was induced using the threat of an upcoming public speaking task reliably shown to lead to
542 anticipatory changes in heart-rate and perceived anxiety[14,29]. The analysis of HRV confirmed that the
543 experimental manipulation succeeded in modulating activity in the autonomic nervous system in
544 association with the anxiety induction during the targeted blocks. Behaviorally, state anxiety at baseline
545 reduced task-related variability within the trial but also exploration across trials. This converges with recent
546 evidence demonstrating that anxiety leads to ritualistic behavior (repetition, redundancy, rigidity of
547 movements) to regain a sense of control[14]. Crucially, however, anx1 participants continued to exhibit a
548 limited use of temporal variability and exploration during subsequent non-anxiety-related training – despite
549 this phase requiring an unrelated piano sequence performance and the HRV returning to normal levels.
550 Moreover, they achieved lower scores and an overall smaller monetary reward. By contrast, participants in
551 the control and anx2 groups who freely explored the temporal dimension during baseline achieved higher
552 scores during training. Our results thus extend previous work[10,36] on the beneficial effect of motor variability
553 on motor learning to the context of anxiety. In particular, the data support that mechanistically the anxiety-
554 induced reduction in behavioral exploration impairs performance in successive tasks that depend on
555 exploration for learning.
556   Significantly, the control experiment demonstrated that removal of baseline motor exploration leads to
557 anxiety diminishing reward-based learning, establishing the relevance of unconstrained exploration for
558 successful motor learning. Our results thus have implications for research on anxiety disorders and
559 performance anxiety, by supporting that intervention programs exploring movements during a non-anxious
560 phase could preserve subsequent motor learning when anxiety re-emerges.
561   We accounted for two sources of temporal variability. Within-trial variability was directly linked to
562 the computation of feedback scores during training. Across-trials variability was higher in participants
563 exploring different performance options in successive trials. Operationally, however, higher levels of across-
564 trials variability could reflect both an intentional pursuit of an explorative regime; or, an unintentional
565 higher level of motor noise. Similarly, motor variability in previous studies reflected contributions from
566 motor noise and intentional exploration, and it is possible that both sources of variability could be beneficial
567 for reward-based learning[10-11]. A recent study, however, established that motor learning (and decision-
568 making) is improved by the use of intended exploration, not motor noise[13]. Although our paradigm cannot
569 dissociate between intended and unintended exploration, the successful transition from an explorative to
570 an exploitative regime in anx2 and control participants from baseline to training blocks, and further during
571 the training blocks, shows they were capable of context-dependent modulation of task-related variability.
572 This outcome aligns well with animal studies where evidence shows a reduction in motor exploration when

14

573 stakes are high (high-reward situations, social context)[36,39-40]. Furthermore, the transition was paralleled by
574 an increase in within-trial task-related variability to achieve higher scores, demonstrating that separately
575 controlling within-trial and across-trials variability was possible and necessary for success. The results are
576 consistent with computational approaches to motor control emphasizing that during task performance,
577 some variables are controlled by the central nervous system, whereas others are left unconstrained[9, 41].
578

579 **Variability and burst duration of beta oscillations mediate the effects of anxiety on behavior.**
580 An important finding was that anxiety at baseline increased variability in the amplitude envelope of beta
581 oscillations during performance. This increase was observed in a region of contralateral sensorimotor
582 channels, supporting that in humans changes in sensorimotor beta variability by anxiety track the changes
583 in motor variability and exploration. Although EEG does not allow for a detailed anatomical localization of
584 the effect, the finding is consistent with the involvement of premotor and motor cortex in driving motor
585 variability and learning, as previously reported in animal studies[16-18], as well as with the changes in motor
586 cortical excitability found in anxious individuals in clinical settings[42]. Moreover, the data suggest that an
587 excessive degree of variation in the amplitude of sensorimotor beta oscillations might be detrimental for
588 performance.
589 The observed anxiety-related changes in beta variability at baseline and during training were correlated
590 with the life-time exponents of the distribution of oscillation bursts across contralateral sensorimotor
591 channels. These correlation results indicate that a tendency towards more frequent long bursts was
592 associated with more variable amplitude of beta oscillations during trial performance. A similar association
593 has been recently observed in work comparing beta oscillation properties in real and shuffled data[28]. Our
594 data demonstrate for the first time a context-dependent anxiety-related modulation of the burst
595 distribution of cortical sensorimotor beta oscillations. Although bursts of 50-100ms were the most frequent
596 in all experimental groups, the most pronounced presence of long bursts was found in anx1 during
597 exploration, and partially also during training. The outcomes thus tentatively link the more frequent
598 presence of long-lived oscillation bursts in sensorimotor regions to reduced motor exploration and learning.
599 Brief bursts of alpha and beta oscillations extending from one to several cycles have been linked to the
600 normal physiological state during rest and motor performance, respectively[25-26]. In the case of alpha
601 oscillations at rest, it has been suggested that bursts represent neuronal avalanches propagating in neural
602 networks operating near a critical state[25]. The life-time exponents reported for sensorimotor alpha
603 oscillations lies within 1.5-1.99[25], in line with the values of the beta-band oscillation-burst distribution we
604 obtained, 1.4-1.9. This range of exponents is consistent with neural dynamics operating in a state close to
605 criticality[25], which would be beneficial for information processing as it supports a balance between flexibility
606 and stability[34]. A link between beta-band oscillation bursts and information processing has also been
607 proposed in recent studies, which showed that the timing and distribution of beta bursts influence motor
608 processing on a trial-by-trial basis[26,28]. These brief bursts of beta oscillations emerge most prominently in the
609 pre- and post-movement period[26,28], which converges with the time course of burst probability in our study.
610 Alternative hypotheses posit that beta bursts contribute to inhibitory processes[43], in line with the suggested
611 anti-kinetic role of beta oscillations[44]. This interpretation would apply to the power effects in our study, as
612 anxiety at baseline increased the average beta power, which could have limited the expression of motor
613 variability in anx1 participants.
614 Interestingly, during baseline the exponents in contralateral sensorimotor electrodes dropped in anx1
615 relative to control participants, corresponding with the long-tailed distribution of burst duration in this
616 experimental group. This finding at the cortical level converges with recent data from the basal ganglia in
617 patients with Parkinson's disease, showing that beta bursts last longer in association with more severe

15

618 motor symptoms[27]. The link is also interesting considering the evidence for a role of basal ganglia variability
619 driving movement variability[19-20]. Previous songbird studies demonstrated that contextual cues such as the
620 presence of a partner alter the expression of neuronal variability during singing via modulation of dopamine
621 release[39]. In Parkinson's disease, a condition characterized by a loss of dopaminergic cells in the substantia
622 nigra, reward-based modulation of movement variability is limited[11]. Our data thus imply that
623 corresponding changes in the duration of beta-band oscillation bursts in basal ganglia structures could be
624 driving the cortical effects, thereby shaping the use of movement variability. Future work, combining
625 recordings in the human basal ganglia and cortex, should test this prediction.
626
627 In conclusion, this study provides the first evidence that contextual modulation of beta bursts and variability
628 by anxiety biases motor behavior, leading to changes in motor variability and exploration, with
629 consequences for motor learning.
630
631

Materials and Methods

633
Participants and sample size estimation
635 In the main experiment, 60 right-handed healthy volunteers (37 females) aged 18 to 44 (mean 27 years,
636 standard error of the mean, SEM, 1) participated in this study. In the second, control experiment, 26 right-
637 handed healthy participants (16 females, mean age: 25.8, SEM 1, range 19-40) took part in the study.
638 Sample size estimation can be found in S.I. Materials and Methods. Participants gave written informed
639 consent prior to the start of the experiment, which had been approved by the local Ethics Committee at
640 Goldsmiths University. Participants received a base rate of either course credits or money (£15) (equally
641 distributed across groups) and were able to earn an additional sum up to £20 during the task depending on
642 their performance.
643 We used pilot data from a behavioral study using the same experimental paradigm (data not shown) to
644 estimate the minimum sample sizes for a statistical power of 0.95, with an α of 0.05, using the MATLAB (The
645 MathWorks, Inc., MA, USA) function sampsizepwr. In the pilot study we had one control and one
646 experimental group of 20 participants each. In the experimental group we manipulated the reward
647 structure during the first training block (in this block feedback scores did not count towards the final
648 average monetary reward). For each behavioral measure (within-trial cvIOI and mean score), we extracted
649 the standard deviation (sd) of the joint distribution from both groups and the mean value of each separate
650 distribution (e.g. m1: control, m2: experimental), which provided the following minimum sample sizes:
651 Between-group comparison of within-trial cvIOI and mean score parameters (using 2-tailed t-test):
652 MinSamplSizeA = sampsizepwr('t',[m1 sd],m2, 0.95) = 18-20 participants.
653 Accordingly, we recruited 20 participants for each group in the main experiment. Next, using the behavioral
654 data from the anx1 and control groups in this main experiment (as we found large non-parametric effect
655 sizes in the anx1-control comparison, PS_{sup} in range 0.7-0.8), we estimated the minimum sample size for the
656 second, control experiment:
657 Between-group (anx1-controls) comparison of within-trial cvIOI and mean score parameters (using 2-tailed
658 t-test): MinSamplSizeA = sampsizepwr('t',[m1 sd],m2, 0.95) = 13 participants.
659 Therefore for the second control experiment we recruited 13 participants in each group.
660
Apparatus and Materials
662 Participants were seated at a digital piano (Yamaha Digital Piano P-255, London, United Kingdom) and in

16

663   front of a PC monitor in a light-dimmed room. They sat comfortably in an arm-chair with their forearms
664   resting on the armrests of the chair. The screen displayed the instructions, feedback and visual cues for start
665   and end of a trial. Participants were asked to place four fingers of their right hand (excluding the thumb)
666   comfortably on 4 pre-defined keys on the keyboard. Performance information was transmitted and saved as
667   Musical Instrument Digital Interface (MIDI) data, which provided time onsets of keystrokes relative to the
668   previous one (inter-onset-interval – IOI in ms), MIDI velocities (related to the loudness, in arbitrary units,
669   a.u.), and MIDI note numbers that corresponded to the pitch. The experiment was run using Visual Basic
670   and additional parallel port and MIDI libraries.
671   The sequence patterns for the baseline exploration and training blocks were designed so that the key
672   presses would span a range of four neighbouring keys on the piano (**Figure 1A**).
673

674   **Experimental design**
675   In all blocks, participants initiated the trial by pressing a pre-defined key with their left index finger. After a
676   jittered interval of 1-2 s, a green ellipse appeared in the centre of the screen representing the "go" signal for
677   task execution. Participants had 7 s to perform the sequence which was ample time to complete it before
678   the green circle turned red indicating the end of the execution time. If participants failed to perform the
679   sequence in the correct order or initiated the sequence before the "go" signal, the screen turned yellow
680   (**Figure 1B**). In blocks 2 and 3 during training, performance-based feedback in form of a score between 0
681   and 100 was displayed on the screen 2 s after the red ellipse, that is, 9 s from the beginning of the trial. The
682   performance-based feedback (scores) provided participants with information regarding the target
683   performance. The pattern of inter-onset-intervals (IOIs) was used to assess the timing or rhythm of the
684   performance, whereas the MIDI keystroke velocity was used to quantify the dynamics (changes in
685   loudness).
686   The target rhythm consisted of a pattern of alternating short and long IOIs: $t \equiv$ [0.2, 1, 0.2, 1, 0.2, 1, 0.2] s
687   (7 IOIs for an 8 notes-long sequence). The score was computed using a measure of proximity between the
688   pattern of IOIs performed in each trial (p) and the rewarded rhythm. Specifically, we computed the norm of
689   the differences between adjacent IOI values (MATLAB function *diff*) for the performed pattern
690   $normDp = \lVert diff(p) \rVert$ and, separately, for the target pattern $normDt = \lVert diff(t) \rVert$. Next, the score
691   was calculated using this expression:

692
$$score = 100\, e^{-\lvert normDp - normDt \rvert}$$

693   In practice, different rhythm patterns could achieve the same reward, as any pattern of IOIs leading to the
694   same $normDp$ value obtained identical scores. The scores correlated with the difference between the
695   cvIOI of the performed and target patterns (ρ = 0.53, p < 0.0001), and accordingly, same values of cvIOI led
696   to identical scores. Participants were unaware of the existence of various solutions and their performance
697   demonstrated that they approached a single-solution. The rationale for accepting different timing patterns
698   as maximally rewarded solutions was our aim to enable a steady learning rate in all participants, by
699   diminishing the difficulty that would be associated with requiring one single solution.
700

701   **Anxiety Manipulation**
702   Anxiety was induced during block1 performance in group anx1, and during block2 performance in the anx2
703   group by informing participants about the need to give a 2-minute speech to a panel of experts about an
704   unknown art object at the end of that block[14]. We specified that they would first see the object at the end
705   of the block (it was a copy of Wassily Kandinsky' Reciprocal Accords [1942]) and would have 2 min to
706   prepare for the presentation. Participants were told that the panel of experts would take notes during their
707   speech and would be standing in front of the testing room (due to the EEG setup participants had to remain

17

708   seated in front of the piano). Following the 2-min preparation period, participants were informed that due
709   to the momentary absence of panel members they instead had to present in front of the lab members.
710   Participants in the control group had the task to describe the artistic object to themselves, not in front of a
711   panel of experts. They were informed about this secondary task at the beginning of the exploration phase.
712

713   **Assessment of State Anxiety**
714   To assess state anxiety we acquired two types of data: (1) the short version of the Spielberger State-Trait
715   Anxiety Inventory (STAI, state scale X1, 20 items)[45] and (2) a continuous electrocardiogram (ECG, see EEG
716   and ECG recording session). The STAI X1 subscale was presented four times throughout the experiment. A
717   baseline assessment before the start of the experiment before the resting state recording was then followed
718   by an assessment immediately before each experimental block to determine changes in anxiety levels. In
719   addition, we a continuous ECG recording was obtained during the resting state and experimental blocks to
720   assess changes in autonomic nervous system responses. The indexes of heart rate variability (HRV,
721   coefficient of variation of the inter-beat-interval) and mean heart rate (HR) were evaluated, as their
722   reduction has been linked to changes in anxiety state due to a stressor[29].
723

724   **EEG, ECG and MIDI recording**
725   EEG and ECG signals were recorded using a 64-channel (extended international 10–20 system) EEG system
726   (ActiveTwo, BioSemi Inc.) placed in an electromagnetically shielded room. During the recording, the data
727   were high-pass filtered at 0.16 Hz. The vertical and horizontal eye-movements (EOG) were monitored by
728   electrodes above and below the right eye and from the outer canthi of both eyes, respectively. Additional
729   external electrodes were placed on both left and right earlobes as reference. The ECG was recorded using
730   two external channels with a bipolar ECG lead II configuration. The sampling frequency was 512 Hz. Onsets
731   of visual stimuli, key presses and metronome beats were automatically documented with markers in the
732   EEG file. The performance was additionally recorded as MIDI files using the software Visual Basic and a
733   standard MIDI sequencer program on a Windows Computer.
734

735   **EEG and ECG pre-processing**
736   We used MATLAB and the FieldTrip toolbox[46] for visualization, filtering and independent component analysis
737   (ICA; runica). The EEG data were highpass-filtered at 0.5 Hz (Hamming windowed sinc finite impulse
738   response [FIR] filter, 3380 points) and notch-filtered at 50 Hz (847 points). Artifact components in the EEG
739   data related to related to eye blinks, eye movements and the cardiac-field artifact were identified using ICA.
740   Following IC inspection, we used the EEGLAB toolbox[47] to interpolate missing or noisy channels using
741   spherical interpolation. Finally, we transformed the data into common average reference.
742   Analysis of the ECG data focused on detection of the QRS-complex to extract the R-peak latencies of each
743   heartbeat and use them to evaluate the HRV and HR measures in each experimental block.
744

745   **Analysis of power spectral density and variability of oscillations**
746   We first assessed the standard power spectral density (PSD, in mV^2/Hz) of the continuous raw data in each
747   performance block and separately for each group. The PSD was computed with the standard fast Fourier
748   Transform (Welch method, Hanning window of 1s with 50% overlap). The raw PSD estimation was
749   normalised into decibels (dB) with the average PSD from the initial rest recordings (3 min). Specifically, the
750   log normalized PSD during the performance blocks was calculated as the natural logarithm of the quotient
751   between the performance-block PSD and the resting state power.
752   In addition, variability of cortical beta-band (13-30Hz) activity in each performance block was assessed using

18

753   the coefficient of quartile variation (CQV[32,48]). The CQV is a descriptive statistic based on the first (lower) and
754   third (higher) quartile of the data:

755

$$\text{CQV} = \frac{Q_3 - Q_1}{Q_3 + Q_1}$$

756

757   The difference Q_3-Q_1, termed interquartile range, is a measure of the dispersion of the data when ranked. To
758   measure the CQV of beta oscillations, the amplitude envelope of the instantaneous analytic signal was
759   computed after applying the Hilbert transform to the bandpass-filtered raw data (12–35 Hz; Hamming
760   windowed two-way least-squares FIR filter applied with the eegfilt.m routine from the EEGLAB toolbox. See
761   **Figure 4D**) spanning the full continuous recording in each performance block. Next, from the total beta-
762   band amplitude envelope we selected data epochs of 10s corresponding to each performance trial (trial of
763   7 s, post-performance period of 3 s). This step provided 100 epochs during baseline exploration and 200
764   epochs during training. The beta-band CQV index was computed for each of these single-trials, and was
765   then averaged across trials within each block.

766

767   **Extraction of beta-band oscillation bursts**
768   The time series of beta-band amplitude envelope obtained in the CQV analyses were used to detect
769   oscillation bursts. We followed a procedure adapted from previous work on oscillation bursts[25,27]. In brief,
770   we used as threshold the 75% percentile of the amplitude envelope of beta oscillations (after
771   rectification)[27]. Amplitude values above this threshold were considered to be part of an oscillation burst if
772   they extended for at least one cycle (50ms: as a compromise between the duration of one 13 Hz-cycle [76
773   ms] and 30 Hz-cycle [33 ms]). Threshold-crossings that were separated by less than 25 ms were considered
774   to be part of the same oscillation burst. As an additional threshold the median amplitude was used in a
775   control analysis, which revealed similar results (significantly more frequent short bursts in control relative to
776   anx1 participants but less frequent long bursts, p < 0.05, FDR-corrected), as expected from previous work[25].
777   Importantly, because threshold crossings are affected by the signal-to-noise ratio in the recording, which
778   could vary between the baseline and training blocks, we selected a common threshold across all
779   experimental blocks separately for each participant[27].
780   Distributions of the rate of oscillation bursts per duration were estimated using equidistant binning on a
781   logarithmic axis with 20 bins between 50-2000 ms. In all participants the double-logarithmic representation
782   of the distributions of burst duration followed a decaying power-law with slope values, τ, in the range 1.4-
783   1.9, in agreement with previously reported values for sensorimotor alpha bursts[25].

784

785   **Statistical Analysis**
786   Between-group comparison focused on each experimental group, separately, and the control group
787   (contrasts: anx1 – control, anx2 – control). Differences between experimental groups anx1-anx2 were
788   evaluated exclusively concerning the overall achieved monetary reward. When appropriate, we tested main
789   effects and interactions for factors Group (anx, control) and Phase (baseline, training) using a 2x2
790   synchronized permutations test[49]. This analysis was complemented with non-parametric permutation tests
791   to assess differences between conditions or between groups in the statistical analysis of behavioral or
792   neural measures. To evaluate differences between sets of multivariate EEG signals corresponding to two
793   conditions or groups, we used two-sided cluster-based permutation tests[33] and an alpha level of 0.025.
794   Control of the family-wise error rate was implemented in these tests to account for the problem of multiple
795   comparisons[33]. When multiple testing was performed with permutation tests and synchronized
796   permutations, the FDR was controlled at level q = 0.05[30].

19

References.

1. Grupe DW, Nitschke JB (2013) Uncertainty and anticipation in anxiety: an integrated neurobiological and psychological perspective. Nat Rev Neurosci 14:488–501.

2. Bishop SJ (2007) Neurocognitive mechanisms of anxiety: an integrative account. Trends Cogn Sci 11(7):307–316.

3. Bishop SJ (2009) Trait anxiety and impoverished prefrontal control of attention. Nat Neurosci 12(1):92–98.

4. Baumeister RF (1984) Choking under pressure: self-consciousness and paradoxical effects of incentives on skillful performance. J Pers Soc Psychol 46(3):610-620.

5. Beilock SL, Carr TH (2001) On the fragility of skilled performance: what governs choking under pressure? J Exp Psychol Gen 130: 701–725, 2001.

6. Eysenck MW, Calvo MG (1992) Anxiety and Performance: The Processing Efficiency Theory. Cogn Emot 6: 409-434.

7. van Beers R, Haggard P, Wolpert D (2004) The role of execution noise in movement variability. J Neurophysiol 91: 1050-1063.

8. He K et al (2016) The Statistical Determinants of the Speed of Motor Learning. PLoS Comput Biol 12(9):e1005023.

9. Todorov E, Jordan MI (2002) Optimal feedback control as a theory of motor coordination. Nat Neurosci 5: 1226–1235.

10. Wu HG et al (2014) Temporal structure of motor vriability is dynamically regulated and predicts motor learning ability. Nat Neurosci 17(2):312–321.

11. Pekny SE, Izawa J and Shadmehr R (2015) Reward-dependent modulation of movement variability. Journal of Neuroscience, 35(9): 4015-4024.

12. Sutton RS, Barto AG (1998) Reinforcement Learning: An Introduction: MIT press.

13. Chen X, Mohr K, Galea JM (2017) Predicting explorative motor learning using decision-making and motor noise. PLoS Comput Biol 13(4):e1005503.

14. Lang M, Krátký J, Shaver JH, Jerotijević D, Xygalatas D (2015) Effects of Anxiety on Spontaneous Ritualized Behavior. Curr Biol 25(14):1892–1897.

15. Morgan KN and Tromborg CT (2007). Sources of stress in captivity. Applied animal behavior science, 102(3-4): 262-302.

16. Churchland MM, Afshar A, Shenoy KV (2006) A Central Source of Movement Variability. Neuron 52(6):1085–1096.

17. Mandelblat-Cerf Y, Paz R, Vaadia E (2009) Trial-to-trial variability of single cells in motor cortices is dynamically modified during visuomotor adaptation. J Neurosci 29:15053–15062.

18. Santos FJ, Oliveira RF, Jin X, Costa RM (2015) Corticostriatal dynamics encode the refinement of specific behavioral variability during skill learning. eLife 4. doi:10.7554/eLife.09423.

19. Kao MH, Doupe AJ, Brainard MS (2005) Contributions of an avian basal ganglia–forebrain circuit to real-time modulation of song. Nature 433(7026):638–643.

20. Ölveczky BP, Andalman AS, Fee MS (2005) Vocal Experimentation in the Juvenile Songbird Requires a Basal Ganglia Circuit. PLoS Biol 3(5):e153.

20

842   21. Herrojo Ruiz M et al (2014) Beta-band amplitude oscillations in the human internal globus pallidus
843   support the encoding of sequence boundaries during initial sensorimotor sequence learning.
844   Neuroimage, 85: 779-793.
845   22. Bartolo R, Merchant H (2015) β oscillations are linked to the initiation of sensory-cued movement
846   sequences and the internal guidance of regular tapping in the monkey. J Neurosc 35(11):4635-40.
847   23. Tan H, Jenkinson N, Brown P (2014) Dynamic neural correlates of motor error monitoring and
848   adaptation during trial-to-trial learning. J Neurosci 34(16):5678–5688.
849   24. HajiHosseini A, Rodríguez-Fornells A, Marco-Pallarés J (2012) The role of beta-gamma oscillations in
850   unexpected rewards processing. Neuroimage 60(3):1678–1685.
851   25. Poil SS, van Ooyen A and Linkenkaer-Hansen K (2008). Avalanche dynamics of human brain
852   oscillations: relation to critical branching processes and temporal correlations. Human brain
853   mapping, 29(7): 770-777.
854   26. Feingold J, Gibson DJ, DePasquale B, Graybiel AM (2015) Bursts of beta oscillation differentiate
855   postperformance activity in the striatum and motor cortex of monkeys performing movement tasks.
856   Proc Natl Acad Sci 112(44):13687–13692.
857   27. Tinkhauser G, et al. (2017) Beta burst dynamics in Parkinson's disease OFF and ON dopaminergic
858   medication. Brain 140(11):2968–2981.
859   28. Little S , Bonaiuto J, Barnes G and Bestmann S (2018) Motor cortical beta transients delay
860   movement initiation and track errors. *bioRxiv*: 384370. Preprint, posted August 9, 2018.
861   29. Feldman PJ, Cohen S, Hamrick N, Lepore SJ (2004) Psychological stress, appraisal, emotion and
862   cardiovascular response in a public speaking task. Psychology & Health 19(3):353-68.
863   30. Benjamini Y, Krieger AM and Yekutieli D (2006). Adaptive linear step-up procedures that control the
864   false discovery rate. *Biometrika*, *93*(3), pp.491-507.
865   31. Grissom RJ, Kim JJ (2012): Effect Sizes for Research: Univariate and Multivariate Applications, 2nd
866   ed. New York: Taylor & Francis.
867   32. Iscan Z, Nazarova M, Fedele T, Blagovechtchenski E, Nikulin VV (2016) Pre-stimulus alpha oscillations
868   and inter-subject variability of motor evoked potentials in single-and paired-pulse TMS paradigms.
869   Frontiers in human neuroscience 10:504.
870   33. Maris E, Oostenveld R (2007): Nonparametric statistical testing of EEG-and MEG-data. J Neurosci
871   Methods 164:177–190.
872   34. Chialvo DR (2010) Emergent complex neural dynamics. Nature physics 6(10):744.
873   35. Browning M, Behrens TE, Jocham G, O'reilly JX, Bishop SJ (2015) Anxious individuals have difficulty
874   learning the causal statistics of aversive environments. Nat Neurosc 18(4):590.
875   36. Dhawale AK, Smith MA, Ölveczky BP (2017) The role of variability in motor learning. Annu Annu Rev
876   Neurosci. 40:479-98.
877   37. Hordacre B, Immink MA, Ridding MC, Hillier S (2016) Perceptual-motor learning benefits from
878   increased stress and anxiety. Hum Mov Sci 49:36–46.
879   38. Vine SJ et al (2013) Evaluating stress as a challenge is associated with superior attentional control
880   and motor skill performance. J Exp Psychol Appl 19(3): 185-194.
881   39. Woolley SC, Rajan R, Joshua M, Doupe AJ (2014) Emergence of context-dependent variability across
882   a basal ganglia network. Neuron 82(1):208-23.
883   40. Kao MH, Wright BD, Doupe AJ (2008) Neurons in a forebrain nucleus required for vocal plasticity
884   rapidly switch between precise firing and variable bursting depending on social context. J. Neurosci
885   28:13232–13247.
886   41. Scholz JP, Schöner G (1999) The uncontrolled manifold concept: identifying control variables for a

21

functional task. Exp Brain Res 126:289–306.

42. Wassermann EM, Greenberg BD, Nguyen MB, Murphy DL (2001) Motor cortex excitability correlates with an anxiety-related personality trait. Biol Psychiatr 50(5):377-82.

43. Shin H, Law R, Tsutsui S, Moore CI, and Jones SR (2017) The rate of transient beta frequency events predicts behavior across tasks and species. Elife 6: e29086.

44. Engel AK, Fries P (2010) Beta-band oscillations—signalling the status quo?Curr Opin Neurobio 20(2):156-65.

45. Spielberger CD (1970) STAI manual for the state-trait anxiety inventory. Self-Evaluation Questionnaire 1-24.

46. Oostenveld R, Fries P, Maris E, Schoffelen JM. FieldTrip: open source software for advanced analysis of MEG, EEG, and invasive electrophysiological data (2011) Comput Intell Neurosci 2011:156869.

47. Delorme A, Makeig S. EEGLAB: an open source toolbox for analysis of single-trial EEG dynamics including independent component analysis (2004) J Neurosci Methods 34(1):9-21.

48. Bonett DG (2006) Confidence interval for a coefficient of quartile variation. Comput Stat. Data Anal 50: 2953–2957.

49. Basso D, Chiarandini M and Salmaso L (2007) Synchronized permutation tests in replicated I× J designs. Journal of Statistical Planning and Inference 137(8): 2564-2578.

22
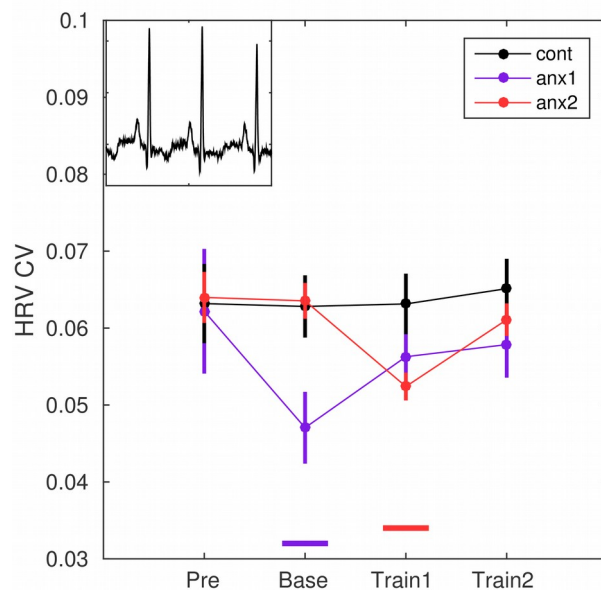
Figure 1 – figure supplement 1. Heart-rate varibility (HRV) modulation by the anxiety manipulation. Average HRV measured as the coefficient of variation of the inter-beat-interval is displayed across the experimental blocks: initial resting state recording (Pre), baseline exploration (Base), first block of training (Train1), last block of training (Train2). Relative to Pre, there was a significant drop in HRV in anx1 participants during baseline exploration ($p < 0.05$, FDR-corrected, $PS_{dep} = 0.81$). In anx2 participants the drop in HRV was found during the first training phase, which was affected by the anxiety manipulation ($p < 0.05$, FDR-corrected, $PS_{dep} = 0.78$). In addition, relative to the control group, anx1 demonstrated a significantly lower HRV at baseline ($p < 0.05$, FDR-corrected, $PS_{dep} = 0.75$). The second experimental group, anx2, exhibited a significant drop in HRV relative to controls during the first training block ($p < 0.05$, FDR-corrected, $PS_{dep} = 0.71$). These results demonstrate a group-specific modulation of anxiety relative to controls during the targeted blocks. No changes in mean heart-rate were found ($P > 0.05$). Neither was the STAI state anxiety subscale able to dissociate between the different phases in each group or between-groups ($p > 0.05$; mean values within 29-37 in all groups and experimental blocks). This is likely due to the habituation of the participants to the questionnaire.

23

953
954
955
956
957
958
959
960
961
962
963
964
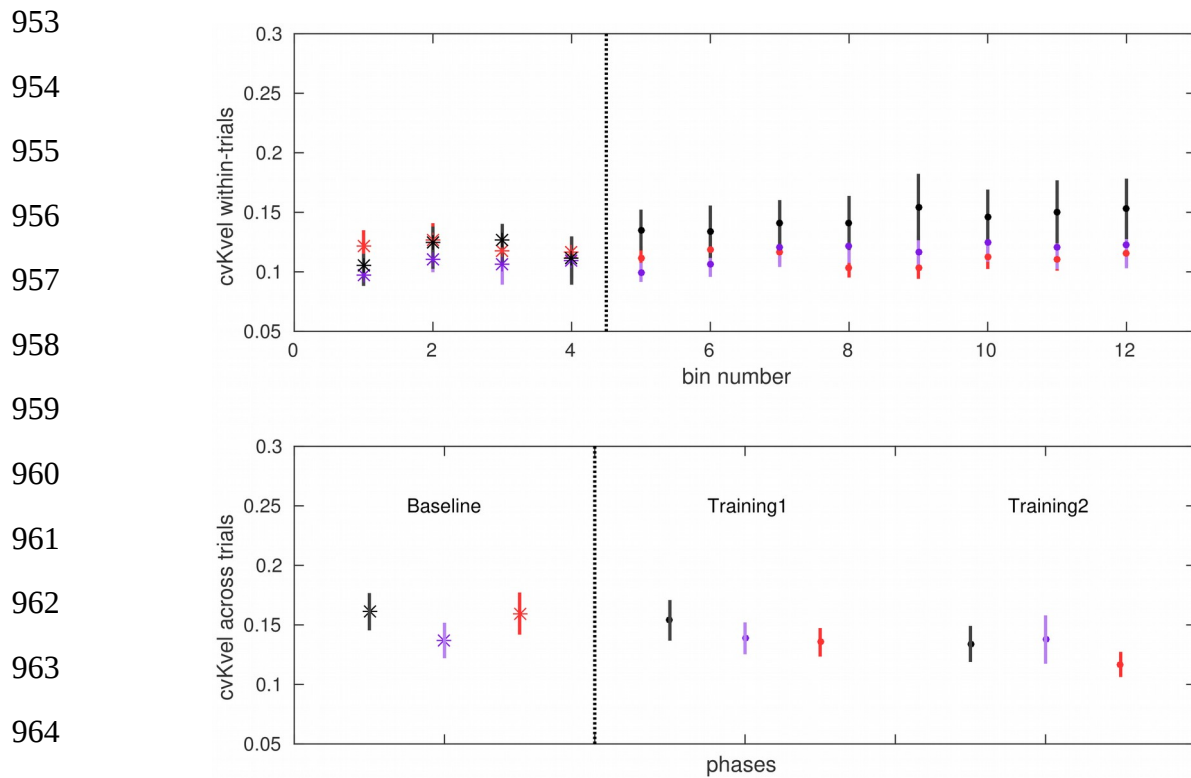
965 **Figure 3 – figure supplement 1. Non-Task related variability in keystroke velocity.** Top: Within-trial variability in Kvel
966 across the experimental blocks (trials were split into bins of 25 trials and values were averaged within each bin). No
967 significant between-group differences were found (p > 0.05). Bars around the mean display ±SEM. Data for control
968 participants are shown in black, wherease data in purple / red indicate values in the anx1 and anx2 groups,
969 respectively. Bottom: Same as in the upper panel but for the across-trials cvKvel. No significant between-group
970 differences were found either (p > 0.05).

971

24
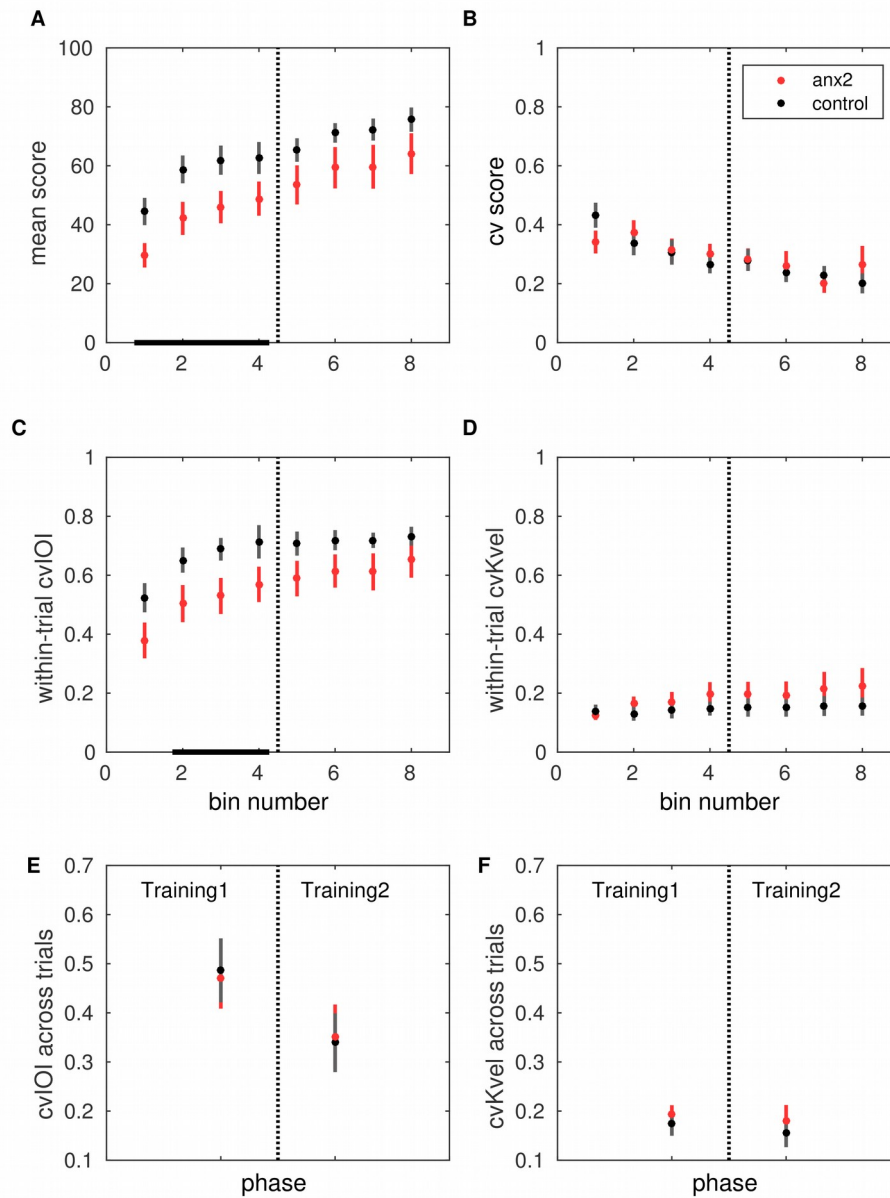
Figure 3 – figure supplement 2. Control experiment: Effect of anxiety on variability and learning after removal of the baseline exploration phase. Results of the control experiment in which new anx2 and control participants completed the reward-based learning phases without a prior exploration phase. Panels A/C/E are displayed as Figure 3A-C. Significant between-group differences are denoted by the black bar at the bottom ($p < 0.05$, FDR-corrected). (B) Coefficient of variability of the average score, showing similar dispersion in both groups. (D) Same as Figure 3 – figure supplement 1. (E) There was a significant drop in across-trials cvIOI for controls, not for anx2 participants ($p < 0.05$, FDR-corrected). (F) Same as Figure 3 – figure supplement 1.
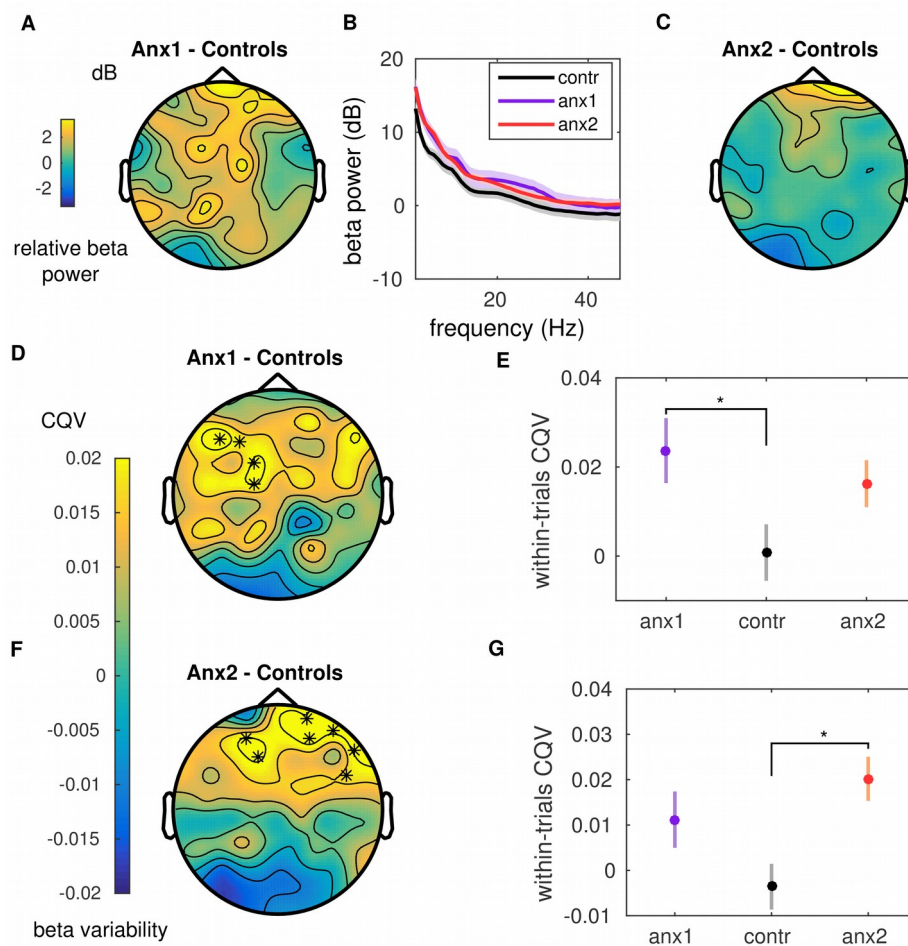
Figure 4 – figure supplement 1. Beta power and variability during reward-based training. Displayed as Figure 4: Power changes are shown in panels A-C. No between-group differences were found (p > 0.05). Changes in variability of beta amplitude envelope (Beta CQV) are shown in panels D-G. Anx1 participants had larger beta CQV values than control participants across sensorimotor electrodes (p < 0.025, two-tailed cluster-based permutation test, denoted by the asterisks). Anx2 participants also had larger beta CQV than control participants, albeit in a region of frontal electrodes (p < 0.025, two-tailed cluster-based permutation test, denoted by the asterisks).

Figure 6 – figure supplement 1. Rate of beta bursts as a function of burst duration (range 50 – 2000ms) and time during trial-wise performance (trial lengh 0 – 12000ms). (A) Rate of oscillation bursts in one representative subject during exploration. (B) Same as (A) but during training.
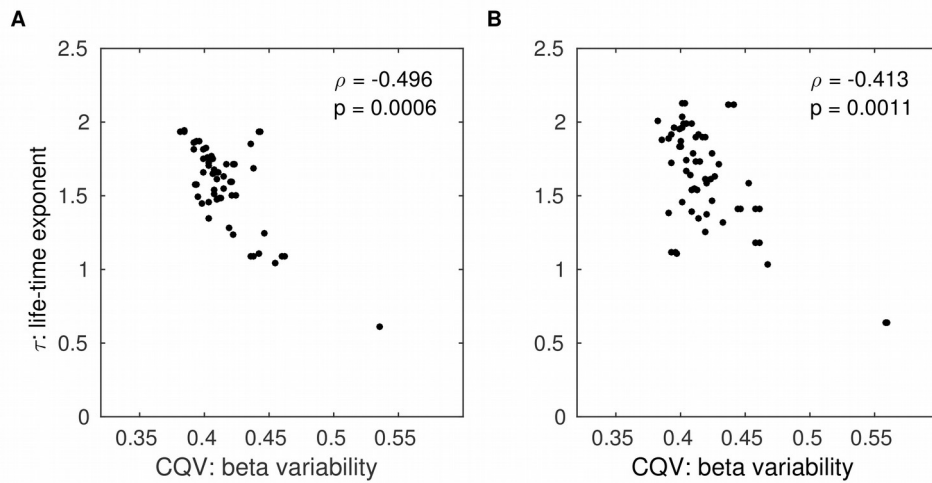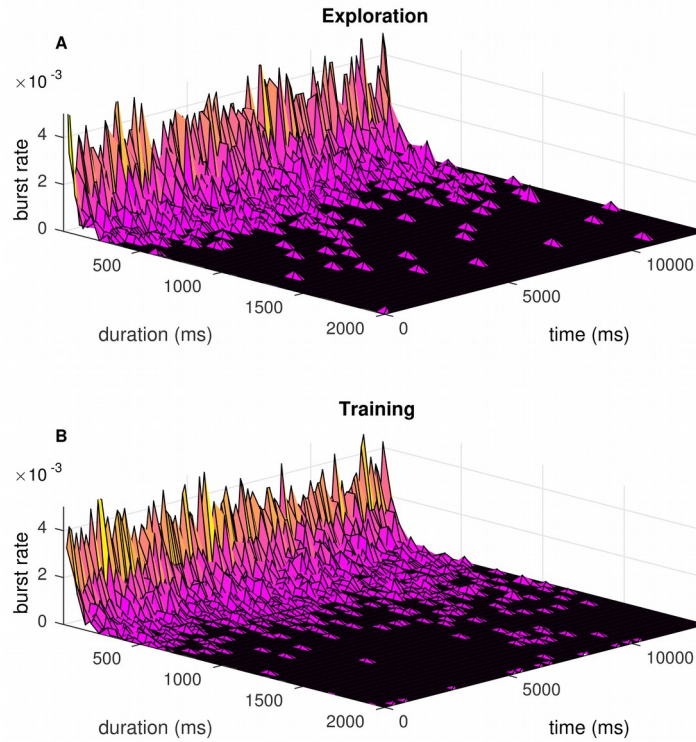

Figure 6 – figure supplement 2. Association between the life-time exponent of the beta-bursts distribution and the measure of variability of the beta amplitude envelope. Non-parametric rank correlation (Spearman ρ) across all 60 participants between the life-time exponent, τ, of the oscillation-bursts distribution and the beta CQV index during exploration (A) and training (B).

27