Pain modulation by illusory body rotation: a new way to disclose the interaction between the vestibular system and pain processing

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Abstract

BACKGROUND: Clinical and experimental evidence advocates a structural and functional link between the vestibular and other sensory systems. For instance, visuo-vestibular and vestibular-somatosensory interactions have been widely reported. However, whether visual inputs carrying vestibular information can modulate pain is not yet clear. Recent evidence using natural vestibular stimulation and moving visual stimuli points at an unspecific effect of distraction.

METHODS: By using immersive virtual reality (VR), we created a new way to prompt the vestibular system through the vision of static visual cues, studying the possible interaction with pain. Twenty-four healthy participants were visually immersed in a virtual room which could appear with five different degrees of rotation in the sagittal axis, either toward the right, left or with no rotation. Participants' heat pain thresholds and subjective reports of perceived body rotation, sense of presence and attention were measured.

RESULTS: 'Being' in a tilted room induced the sensation of body rotation in our participants, even though they were always in an upright position. Importantly, we also found that rotating the visual scenario can modulate the participants' pain thresholds, determining a significant increase when a *left* tilt is displayed. Additionally, positive correlations between the perceived body midline rotation and pain threshold were found, and all VR conditions were equally distractive.

CONCLUSIONS: Vestibular information present in static visual cues can modulate experimental pain according to a side-dependent manner and bypassing supramodal attentional mechanisms. These findings may help refining pain management approaches based on multimodal stimulation.

Introduction

The vestibular system has a central role in the control of body posture, balance and eye movements (Lopez, 2015). A plethora of brain areas are involved in the processing of vestibular inputs and many of these are associative areas. For instance, the posterior insular cortex (PIC) has been recently shown to be a key visuo-vestibular integration area (Frank and Greenlee, 2018), while the putamen, insula, intraparietal sulcus (IPS), the parieto-insular vestibular cortex (PIVC), the secondary somatosensory cortex (SII), premotor cortex and supramarginal gyrus, receive inputs from both vestibular and somatosensory projections (Bottini et al., 1995; Lopez, 2015). Importantly, shared neural pathways could underlie the modulatory effects of vestibular stimulation on other sensory domains. For example, evidence shows that artificial vestibular-stimulation such as caloric vestibular-stimulation (CVS) can affect the processing of various stimuli (Ferrè et al., 2015b). This type of stimulation has been recently found to reduce pain with varying lasting effects in those with central post-stroke pain (McGeoch et al., 2008; McGeoch and Ramachandran, 2008; Ramachandran et al., 2007; Spitoni et al., 2016), persistent pain and allodynia (Ngo et al., 2015), experimental acute pain (Ferrè et al., 2015a) and headaches (Wilkinson et al., 2017). Yet, CVS is not the only way to probe the vestibular system and influence pain. Vestibular-stimulation via physical rotation has been shown to increase heat pain threshold in participants (Macrea et al., 2016), in a way similar to the effect on pain thresholds seen in healthy participants undergoing CVS (Ferrè et al., 2013). Hence, the effects of vestibular stimulation on pain seems to be quite robust. However, in the recent experiment by Macrea and colleagues, it was found that pain-reducing effects were recorded not only following vestibular stimulation via physical rotation, or by the vision of a moving visual scene, but also during the vision of randomly moving dots, which are not meant to probe the vestibular system. The authors interpreted their results as a generic analgesic effect due to distraction (Macrea et al., 2016). Thus, whether pain modulation can be obtained via visuo-vestibular stimulation remains an open question. Seminal electrophysiological studies on monkeys have shown that not only that early vestibular pathways (otolith system) respond to static body tilts, but also that tonic discharges deriving from the tilted position may last as long as the body stays in the same position (Fernandez et al., 1972; Fernandez and Goldberg, 1976). Furthermore, it has been suggested that the otolith system comes into play also during the vision of tilted static scenes (Pansell et al., 2006). Thus, we set out to examine the effect of visuo-vestibular stimulation on pain threshold during the vision of a static tilted virtual room. Importantly, given the results obtained in Macrea's study, we wanted to rule out the possible contribution of attention on pain.

Since during conflict between two signals, only one is selected and used as the perceived verticality (Sierra-Hidalgo et al., 2012), we maximized the contribution of the visual input using an 'immersive' visual scenario in virtual reality (VR). We hypothesised that 'being' in a tilted room would induce a sensation of having the body midline tilted in our participants and led to an increase of their pain threshold. Furthermore, we wanted to test whether bigger tilts (15 vs 30 degrees) would lead to bigger pain modulations. Finally, given the right-hemisphere dominance of the vestibular cortical network (Brandt and Dieterich, 1999; Dieterich et al., 2017), we explored whether the side of rotation, (right or left) could be a factor.

Material and methods

Participants

The sample size for this experiment is similar to a comparable study (Macrea et al., 2016) and was estimated using G*Power (Faul et al., 2007). Considering a 'within subjects' design, a medium effect size (f) of 0.25, an α error probability of 0.05, a power (1- β error probability) of 0.8 and 5 measurements, the calculated sample size was = 21. To have a perfect balancing of the order of conditions across subjects, twenty-four healthy participants (5 males and 18 females; age range: 18-47, mean±standard deviation: 25.67±5.86 years) were recruited via student forums and word-of-mouth and were included in the final data analysis. In this cross-sectional study all the participants were right-handed (mean±SD: 93.74±10.72, range:62.5-100), as measured by the short version of the Edinburgh Handedness Inventory (Veale, 2014). They all had normal or corrected-to-normal vision, were over the age of 18, had no history of neurological disorders, and were not on any current course of medication that could interfere with pain sensitivity. This study was conducted in accordance with the Declaration of Helsinki and approved by the University of East London's Ethics Committee prior to beginning data collection.

Virtual reality system

The stereoscopic head-mounted display (HMD) utilised was an Oculus Rift (Oculus VR, Irvine, California, USA) with a resolution of 2160 x 1200 per eye and a field of view of 100 degrees displayed at 90 Hz. The environment shown within the virtual reality simulation was programmed using the Unity platform (Unity Technologies, San Francisco California, USA). Noise isolation was ensured by the administration of pink noise via headphones, with a constant volume set at 70 dB. Such volume was used to minimize distraction by environmental noise.

Thermal stimulation

The heat was administered to the left wrist as in previous research it has been evidenced that the right vestibular cortex has a hemispheric dominance in right-handed individuals (Dieterich et al., 2003; Fasold et al., 2002). Therefore the administration of pain to the left limb, contralateral to the hemisphere activated by the vestibular stimulation was presumed to have the greatest modulatory effects (Ferrè et al., 2015a; Macrea et al., 2016). Thermal heat stimuli were delivered via the use of a TSA-II Neuro Sensory Analyzer (Medoc Ltd., Ramat Yishai Israel), with a 30x30 mm thermode tied with a Velcro strap onto the palmar side of the left wrist. In line with previous pain threshold literature (Gordon et al., 2019; Mancini et al., 2011; Martini et al., 2014; Nierula et al., 2017; Zanini et al., 2017), during each condition, the probe temperature increased from regular skin temperature (baseline temperature=32°C) at 2°C/s. Participants were instructed to click a button with their right hand immediately when perceiving the heat stimulation as being painful. Upon pressing the button, the temperature reached was noted as the pain threshold and the probe temperature instantly reduced, returning to the baseline level (32°C). For safety purposes, maximal temperature was set at 51°C.

Subjective measures

4-item questionnaire. Following each visual condition, a 4-item questionnaire was administered to the participants to measure their subjective feelings experienced when exposed to that specific condition (see table 1). This questionnaire was made in the form of a 7-point Likert scale. Items I1, I2 and I4 (i.e. "sensation of body tilt", "sensation of room tilt", "attention", respectively) ranged from 1 'totally disagree' and 7 'totally agree', while for item I3 ("presence") "1" meant having the sensation of being "in the lab" while "7" meant being "in the virtual room" (see Table 1). When administering the questionnaire, the items were read aloud and presented to the participant in a random order. Some of the items were selected and adapted from a questionnaire utilised in research on virtual arm ownership and pain (Zanini et al., 2017).

Self-rotation task. The questionnaire was accompanied with the perceived self-rotation task which was used to measure how each participant perceived the inclination of their body midline to be during each VR condition. This involved the participant drawing a line using a ruler against an existing perpendicular line, where the existing line represented the participant's body midline being perfectly straight upright. To avoid the collection of spurious data, if the participant felt her/his body midline to be perfectly straight upright, s/he was asked to simply cross out the pre-existing line. The degrees of rotation between the straight and the drawn line were measured with a goniometer.

Procedure

The study took place at the University of East London, and participants were recruited between May 2018 and August 2018. The experiment consisted of five VR conditions plus one, outside VR, which served as a baseline. To minimize the effects of habituation we perfectly balanced the order of conditions across subjects so that, in the whole sample, every condition was administered the same amount of times (i.e. 4) in each position of the sequence (1st, 2nd, 3rd, 4th, 5th, 6th). Participants sat on a chair with their arms lying on a table. During each condition the participants' heat pain threshold was measured. Before the start of the real experiment, the heat thermode was secured onto the participants left forearm close to the wrist, and they were familiarised with the heat stimuli. The heat stimuli then began to increase via the thermode, and they were asked to press a button as soon as the heat was perceived to be painful. Four pain thresholds were taken during each condition. In the baseline condition, the participants were asked to focus on a white cross placed on a table in front of them while the four heat stimuli were delivered. The participants' left arm was always kept out-of-view.

During each VR condition, the participant was asked to put on the VR headset and shown a Japanese-like room with a missing front wall. As a result, the visible 'external' environment was made clear to participants, who could see a running waterfall outside the virtual room

(Fig.1). The external scenario was provided recording, with a professional digital camera (Nikon D3300), the waterfall present in the ‘Kyoto garden’ in ‘Holland Park’ (London, UK). The external scenario appeared in the ‘normal’ upright position in all VR conditions. The choice of such scenario was driven by the idea that providing a live video of a natural environment, and especially of a waterfall, it would have been clear to the participants what the ‘real’ vertical axis would have been (since the water falls following Newton’s law of gravity). Indeed, while working at an early version of the virtual scenario, we found that it was not clear when the virtual room was rotated if the room was the only visual reference. In other words, without an external comparison, ‘being’ in a rotated virtual room may not produce a sensation of tilt. Conversely, the introduction of an external, upright, scenario provides a powerful contrast which makes the rotation of the room readily clear. We also thought that the presence of a horizontal footbridge over the pond, in the ‘Kyoto garden’ scene, would have provided a further contrast with the rotated room.

Depending on the condition, the Japanese-like room would be tilted either 15 or 30 degrees to the left or to the right relative to the outside waterfall. In regards to the room tilt, these specific degrees were selected in order to avoid symptoms of motion sickness, while maintaining the effect of a tilt. This is evidenced by Dai and coworkers (Dai et al., 2003) who found that during complete body rotation, once participants rolled their heads by 45 degrees, symptoms of increasing motion sickness presented. Similarly, Neimer and colleagues (Neimer et al., 2001) were able to induce motion sickness by exposing participants to a visual field rotated 45 degrees during whole body rotation. During the current experiment, it was found that the participants were able to perceive the tilt, and only three participants reported slight symptoms of motion sickness.

At the beginning of each VR condition participants were asked to visually explore the virtual environment. When looking downwards participants could see the body of an avatar in place of their own, which mimicked their body posture, then they were asked to look straight ahead, so they could clearly see both the room and the external scene in the centre. At this point, no body parts related to the avatar were visible and they could see that the Japanese room they were 'in' had been tilted in comparison to the 'outside'. To ensure the participants' gaze was at the centre of the external scene, during each trial a letter was quickly presented for 1 second without pre-warning. Participants were asked to verbally report which letter was displayed. If the letter reported was incorrect the trial was discarded. To avoid any mistakes in hearing the following letters were selected: G, Q, O and S. During this experiment, a total of only 2 trials were discarded. To avoid a possible confounding factor related to distraction, the appearance of the letter never coincided with the temperature of the thermode above 40 degrees.

Four of the five VR conditions were with some degree of rotation (15° left, 30° left, 15° right, 30° right) compared to the 'external' scene, while a fifth condition was used as a further control condition, with the VR room shown in a normal, upright, orientation.

Immediately following the recording of the fourth heat pain threshold, the participant would be asked to remove the HMD and complete the perceived self-rotation task and the 4-item questionnaire. This was done after each VR condition. To reduce any potential source of bias participants were told that there "were no right or wrong responses" and that "the only 'right' response was the one that actually corresponded to their feelings".

---------------------Insert Figure 1 here-------------------------------

Table 1.

The 4-item questionnaire administered to the participants following each VR condition.

Category	"During the last condition…"
Sensation of body tilt	**I1. "...**there were moments in which I felt as if my body was tilted to one side (totally disagree – 1; totally agree – 7)"
Sensation of room tilt	**I2. "...**there were moments in which I felt as if the real room was tilted to one side (totally disagree – 1; totally agree – 7)"
Presence	**I3. "...**I had a strong feeling of being in the lab (1) or in the virtual room (7)"
Attention	**I4. "...**My attention was totally focussed on other things, for example on what I was watching (1), or totally on the thermal stimulus (7)"

Data handling

Single pain threshold values (in Celsius degrees) were firstly cleaned of any value below 40°C (Dubin and Patapoutian, 2010) and then averaged per each condition and participant. Due to a high inter-subject variability, an outliers check was performed. Averaged values beyond 1.5 the standard deviation from the group's mean were identified as outliers and replaced with the group's mean scores for the same condition (Zanini et al., 2017). Nineteen out of 144 values were replaced. Normalized delta scores (x) were then calculated according to the formula: x = VR condition – Baseline. Resulting data from all conditions were normally distributed according to the Shapiro-Wilk tests (all $p_s > 0.05$). The assumption of sphericity was tested and met according to Mauchly’s test ($p_s > 0.05$). A one-way repeated measures ANOVA was initially conducted on pain thresholds from all VR conditions (a single factor: “Condition”, with 5 levels), to check for any differences among all VR scenarios. A 2x2 repeated-measures ANOVA (with two factors: “Side” and “Degrees”, both with 2 levels) was then conducted on the pain thresholds obtained from the 4 VR ‘rotated’ conditions (’15 left’, ’30 left’, ’15 right, ’30 right). Post hoc multiple comparisons were made with Tukey’s HSD. The level of significance was set at $p<0.05$.

Self-rotation scores corresponded to the angles (degrees) resulting from the deviation of the line drawn by the participants from the printed vertical 0° line. Regardless of the condition, angles created by lines drawn on the left of the midline were conventionally reported as negative values, while angles on the right were reported as positive values. The scores collected after each VR condition underwent the same outliers check described above. Seventeen out of 120 scores were replaced with the group’s mean scores for each condition. The assumptions of normality and sphericity were checked for the pain thresholds. Given that the assumption of normality was violated for the ‘*no rotation*’ condition ($p<0.05$) a non-parametric Friedman ANOVA was initially used to check for any differences among all conditions. Post-hoc analysis was carried out with Conover’s test (for non-parametric analysis).

As per the pain thresholds, a two-way ANOVA was instead computed on the scores derived from the rotated conditions (which passed the normality test), to check for any effect of the factor “Side” and “Degrees” on the perceived self-rotation.

Questionnaire scores deriving from the 4-item questionnaire were averaged across subjects per each item and condition. The resulting mean scores were subjected to Friedman ANOVAs (per each item separately), with “Condition” as the only factor with 5 levels. Post-hoc analysis was carried out with Conover’s test.

A correlation analysis between the pain thresholds and the perceived self-rotation scores for the rotated conditions was conducted using Pearson's *r*. Instead, given the ordinal nature of the questionnaire scores a correlation analysis between pain thresholds and questionnaire scores was performed with Spearman's rank correlation coefficient.

Results

Pain thresholds

The one-way repeated-measures ANOVA on the pain thresholds obtained during all VR scenarios disclosed an effect of the factor "Condition" ($F_{4,92}$=2.66, *p*=0.038, η^2_p=0.104), so that the various visual scenarios did have a distinct effect on our participants' pain thresholds. Post-hoc tests showed that the pain threshold were significantly higher during the '15 left' condition compared to a condition having the same degree of rotation but opposite side ('15 right'; *p*=0.024). All other comparisons were not significant ('no rot' VS '15 left': *p*=0.144; 'no rot' VS '30 left': *p*=0.944; 'no rot' VS '15 right': *p*=0.952; 'no rot' VS '30 right': *p*=0.999; '15 left' VS '30 left': *p*=0.512; '15 left' VS '30 right': *p*=0.168; '30 left' VS '15 right': *p*=0.589; '30 left' VS '30 right': *p*=0.961; '15 right' VS '30 right': *p*=0.933).

The two-way repeated-measures ANOVA on the four 'rotated' VR conditions revealed a main effect of the factor "Side" ($F_{1,23}$=5.87, *p*=0.024, η^2_p=0.203). Thus, the pain thresholds obtained in the VR *left* conditions were significantly higher than those obtained when the virtual rooms were tilted toward the *right*. On the other hand, the extent to which the virtual room was rotated did not seem to significantly affect the pain threshold. Indeed, no main effect of the factor "Degrees" was found ($F_{1,23}$=0.37, *p*=0.547, η^2_p=0.016). Yet, a trend toward statistical significance was found for the interaction between the two factors ("Side"x"Degrees": $F_{1,23}$=3.388, *p*=0.079, η^2_p=0.128), with a "15 left" condition reporting the highest pain threshold (see Fig.2).

Since the focus of the present study was mainly on pain thresholds, to ascertain the goodness of our findings we also calculated the Bayes factor (BF). In fact, *p*-values, effect sizes and Bayes factors usually co-vary with each other but they do not represent identical measures: for instance traditional ANOVAs' *p*-values ranging between 0.01 and 0.05 may often correspond to a Bayes factor which expresses only an "anecdotal" evidence in favour of the alternative hypothesis (Wetzels et al., 2011). Therefore, we run a Bayesian 2x2 repeated measures ANOVA with the factors "Side" and "Degrees" to assess the evidence for or against the presence of the effect found with the traditional ANOVA. Among the different models, the only one receiving support from the data in favour of the alternative hypothesis (H_1) was the one with the factor "Side" alone ($BF_{10} = 4.41$), while all the other models did not receive support ("Degrees": $BF_{10} = 0.24$; "Side+Degrees": $BF_{10} = 1.14$; "Side+Degrees+ Side*Degrees": $BF_{10} = 1.04$). In accordance with Jeffreys' nomenclature (Jeffreys, 1983) a model taking into account the factor "Side" alone would provide "*substantial*" evidence in favour of rejecting the null hypothesis.

-----------Insert Fig. 2 here------------

4-item questionnaire

Boxplots of the embodiment and attention scores are depicted in figure 3. The Friedman ANOVA computed on I1 scores, relative to the sensation that the real room was tilted, showed a significant effect of the factor "Condition" (χ^2_4=38.52, p<0.00001). As expected, post hoc tests revealed that the sensation that the real room was tilted during the 'no rotation' condition was significantly lower compared to the other visual conditions (i.e. 'no rot' VS '15 left': p<0.001; 'no rot' VS '30 left': p<0.001; 'no rot' VS '15 right': p<0.001; 'no rot' VS '30 right': p<0.001). No other comparison was significant ('15 left' VS '30 left': p=0.43; '15 left'

VS ‘15 right’: *p*=0.50; ‘15 left’ VS ‘30 right’: *p*=0.35; ‘30 left’ VS ‘15 right’: *p*=0.14; ‘30 left’ VS ‘30 right’: *p*=0.89; ‘15 right’ VS ‘30 right’: *p*=0.11).

I2 scores indicated the sensation of having one’s body midline tilted. The Friedman ANOVA reported a significant effect of the factor “Condition” (χ^2_4=37.93, *p*<0.001). Again, as expected, post hoc tests revealed that the ‘no rotation’ condition was significantly different from all other visual conditions (i.e ‘no rot’ VS ‘15 left’: *p*<0.001; ‘no rot’ VS ‘30 left’: *p*<0.001; ‘no rot’ VS ‘15 right’: *p*<0.001; ‘no rot’ VS ’30 right’: *p*<0.001). We also found a significant difference between ‘15 right’ VS ‘30 right’ (*p*=0.028). No other comparison was significant (‘15 left’ VS ’30 left’: *p*=0.32; ‘15 left’ VS ‘15 right’: *p*=0.64; ‘15 left’ VS ‘30 right’: *p*=0.08; ‘30 left’ VS ‘15 right’: *p*=0.15; ‘30 left’ VS ‘30 right’: *p*=0.43).

I3 scores indicated the so-called ‘sense of presence’ (SoP), i.e. the feeling to be in the virtual scenario rather than in the real room. Since SoP has been shown to be highly variable even within the same subject, to estimate the proportion of participants who had reported a SoP, an average score from all conditions was calculated per each participant. All average scores above 4 were considered to signal the existence of a SoP (7-point Likert scale). According to this criterion, twenty-two out of twenty-four participants (91.66%) reported a clear SoP. On the other hand, the Friedman ANOVA reported no significant effect of the factor “Condition” (χ^2_4=1.49, *p*=0.82). Thus, all virtual scenarios successfully induced a high SoP in our participants (all group mean scores, per each condition, between 5-6).

I4 scores measured where the participants’ attention was oriented. Mean scores across conditions indicated a general trend of the participants to be engaged with the visual scenario but at the same time aware of incoming heat stimulation. All scores were quite similar across conditions and this similarity is highlighted by the Friedman ANOVA which showed no significant effect of the factor “Condition” (χ^2_4=2.82, *p*=0.58).

-----------Insert Fig. 3 here------------

Perceived self-rotation

The Friedman ANOVA computed on self-rotation scores showed a strong significant effect of the factor "Condition" ($\chi^2_4=32.21$, $p<0.00001$). As expected, post hoc tests revealed that the sensation of having one's body (midline) rotated was significantly weaker in the 'no rotation' condition compared to all other 'rotated' conditions ('30 left' $p=0.002$, '15 right' $p=0.004$, '30 right' $p=0.048$) with the exception of the '15 left' condition ($p=0.32$). The multiple comparisons also revealed that the '15 left' condition induced a significantly weaker sensation to be rotated toward the left compared to the '30 left' condition ($p=0.037$), while there was no such difference between the '15 right' and the '30 right' conditions ($p=0.34$; see fig.4).

A two-way repeated-measures ANOVA conducted on the perceived self-rotation scores in the four 'rotated' VR conditions showed a main effect of the factor "Side" ($F_{1,23}=28.52$, $p<0.001$, $\eta^2_p=0.554$) with the 'right' conditions inducing a greater degree of (perceived) rotation. The ANOVA also highlighted a trend toward significance for the factor "Degrees" ($F_{1,23}=3.27$, $p<0.084$, $\eta^2_p=0.125$) but the interaction between the two factors was non-significant ($F_{1,23}=1.27$, $p=0.27$).

-----------Insert Fig. 4 here------------

Correlation analysis

A significant positive correlation was found between I2 (body tilted sensation) scores and pain thresholds collected during the 'no rotation' condition ($\rho=0.42$, $p=0.041$). However, this correlation is likely to be spurious since *all* but four subjects scored '1' at the I2. A significant negative correlation was found between I4 (attention) scores and pain thresholds collected during the ‘30 left’ condition ($\rho=-0.54$, $p=0.006$). All other correlations between the questionnaire scores and the pain thresholds were not significant (for I1 – ‘no rot’: $\rho=0.28$, $p=0.17$; ‘15 left’: $\rho=-0.10$, $p=0.63$; ‘30 left’: $\rho=-0.33$, $p=0.11$; ‘15 right’: $\rho=0.08$, $p=0.68$; ‘30 right’: $\rho=-0.23$, $p=0.28$; for I2 – ‘15 left’: $r=0.21$, $p=0.32$; ‘30 left’: $r=-0.08$, $p=0.69$; ‘15 right’: $r=0.08$, $p=0.68$; ‘30 right’: $r=-0.09$, $p=0.68$; for I3 – ‘no rot’: $\rho=-0.01$, $p=0.95$; ‘15 left’: $\rho=-0.23$, $p=0.27$; ‘30 left’: $\rho=-0.06$, $p=0.78$; ‘15 right’: $\rho=-0.05$, $p=0.80$; ‘30 right’: $\rho=0.10$, $p=0.63$; for I4 – ‘no rot’: $\rho=-0.01$, $p=0.94$; ‘15 left’: $\rho=-0.39$, $p=0.06$; ‘15 right’: $\rho=-0.17$, $p=0.41$; ‘30 right’: $\rho=-0.05$, $p=0.81$).

The analysis of the correlation between the perceived self-rotation scores and the pain threshold reported a significant negative correlation for the ‘15 left’ condition, which was, by far, the condition with the highest pain thresholds ($r=-0.42$, $p=0.04$). It has to be noted that, given the arbitrary negative signs attributed to the *left* angles, all negative correlations can be interpreted as a positive ones, so that at bigger rotations corresponded higher pain thresholds. To make this clearly available to the reader, left-related negative signs were reported as positive in Fig.5.

All other correlations between the perceived self-rotation scores and the pain thresholds were not significant (‘no rot’: $r=-0.31$, $p=0.13$; ‘30 left’: $r=0.89$, $p=0.67$; ‘15 right’: $r=0.15$, $p=0.47$; ‘30 right’: $r=0.12$, $p=0.55$).

-----------Insert Fig. 5 here------------

Discussion

This study investigated the effect of the vision of static visuo-vestibular cues on pain threshold. By visually ‘immersing’ participants in rotated virtual rooms, we aimed at prompting the vestibular system inducing a sensation of body tilt and a modulation in the perceived pain. Two major results were unveiled. Firstly, we found that the mere vision of a tilted environment in immersive VR can lead to the sensation that one's body is rotated. In a previous VR study, Blom and colleagues recorded no clear sensation of visual manipulation or body rotation after 15 degrees rotations of the virtual world along the frontal axis (Blom et al., 2014). Here we show that rotations of the virtual world along the sagittal axis, especially when this is contrasted with the vision of a normally upright 'outside' world (in our case the 'Kyoto garden'), is effective in inducing in the participants the sensation that their body midline is tilted.

Secondly, and more importantly, 'being' visually immersed in a tilted toom can induce alterations in pain perception. Interestingly, what we found is that this effect is side-dependant: a rotation towards the left yields higher pain thresholds compared to a right tilt. However, the magnitude of the rotation does not seem to play a role. Indeed, bigger tilts toward the left did not yield higher pain thresholds. In fact, on average, higher pain thresholds were reported during the '15 left', not the '30 left' condition.

Pain modulation by vestibular stimulation has already been described in previous studies. For example it has been shown how such modulation can be elicited by CVS (Ferrè et al., 2013, 2015a; McGeoch and Ramachandran, 2008), centrifugation (Aranda-Moreno et al., 2019; Macrea et al., 2016) or optokinetic stimulation (Macrea et al., 2016). Nevertheless, to our knowledge, this is the first study to show that the vision of static visual scenes, carrying vestibular information, differently modulates pain. It could be argued that the vision of a rotated static visual scene may not be enough to activate the vestibular system. Yet, from behavioural, clinical and neuroimaging data, it seems clear that recruitment of the vestibular system operated by vision may not only arise from moving visual stimuli but also for static visual scenes. For instance, in a study measuring ocular torsion in response to static tilts of visual scenes (from 0° to 45° in steps of 15°), Pansell and colleagues found out that the vision of all tilted images induced a torsional response in their participants, which increased with stimuli angle. The authors interpreted this finding as evidence in support of the recruitment of the vestibular otolith system, which would regulate the oculo-torsional response during the vision of static tilted visual scenes (Pansell et al., 2006). After all, the visual system is the predominant sensory system to maintain optimal postural balance (Grace Gaerlan et al., 2012), and clinical reports provide useful insights into this. For example, in the so-called "room-tilt illusion", a rare syndrome which often follows cerebral ischemia, a false cortical integration of vestibular and visual cues leads the patient to experience either paroxysmal 90° or 180° tilts of the visual scene, without any alteration of the physical properties of the visual objects (Sierra-Hidalgo et al., 2012). This static visual alteration of the spatial references can last from seconds to hours (Sierra-Hidalgo et al., 2012). Neuroimaging data have evidenced an altered functional activity of both the vestibular and the visual cortices during this illusion (Kirsch et al., 2017). A visuo-vestibular convergence in the brain is also well-known and it has been reported in the medial superior temporal area (Bremmer et al., 1999; Takahashi et al., 2007) and in the parieto-insular vestibular cortex in monkeys (Grüsser et al., 1990; Guldin and Grüsser, 1998).

As aforementioned, the pain-modulatory effect operated by the vision of the tilted room, found in our study, is side-dependant: a room's rotation towards the left yields higher pain thresholds compared to a right tilt. This lateralized effects found after the stimulation of the vestibular system have been previously reported in the scientific literature. For instance, it has been shown how *left* but not right vestibular stimulation, produces transient remission of visuo-spatial hemineglect symptoms (Marshall, 2009; Rode et al., 2002), improves anosognosia among individuals with schizophrenia spectrum disorders (Gerretsen et al., 2017), ameliorates tactile hemianesthesia (Bottini et al., 2005; Vallar et al., 1990), or even reduces unrealistic optimism (McKay et al., 2013). In behavioural studies, the effects of left vestibular stimulation by CVS are also seen on pain perception (Balaban, 2011; Ferrè et al., 2015a; McGeoch et al., 2008). It has been suggested that CVS reduces central pain by activating the parieto-insular vestibular cortex. As this area is anatomically adjacent to dorsal posterior insula, it may cross-activate it to suppress the neural activity in the anterior cingulate cortex (McGeoch et al., 2008)**.** Nonetheless**,** the lateralized effects of vestibular stimulation on pain are not completely clear. One explanation may lie on the dominance of the vestibular system, in the right-handers, in the right-hemisphere and the right vestibular meso-diencephalic circuitry (Dieterich et al., 2017). However, when it comes to the present results, it is less clear whether being visually immersed in a left-titled environment can stimulate the right side of the brain. Indeed, to our knowledge, there are no studies that have investigated the possible lateralized effects on the brain of the vision of left- or right-titled environments. So, we can only speculate that the vision of a left-titled room activates the right side of the brain (and vice-versa).

Our results may contribute shedding light on the analgesic effect of vestibular-related stimulation. Crucially, we not only report that the vision of static visuo-vestibular cues can modulate pain, but also that this modulation is unlikely to be ascribed to different attentional levels. Indeed, as expected, the ratings of attention in our experiment did not differ among VR conditions. Consequently, the observed pain modulation cannot be attributed to the distracting effects of a specific visual scene. What could determine the pain modulation instead, is the sensation of having one's body tilted, as highlighted in the correlations with the pain thresholds. It has to be noted that the sensation of being tilted can bring about analgesic effects only when this sensation is mild. In our case this was evident when the room was tilted 15° to the left. At bigger rotations, a sensation of anxiety due to the possibility of falling down could kick in, hampering the possible analgesic effects deriving from the vestibular stimulation. Indeed, it is well known that anxiety have pain increasing effects (Ploghaus et al., 2001). Unfortunately, we haven't collected any data relative to anxiety, so we can only suggest this mechanism as a possible explanation. On the other hand, this account fits well with our results, which show lower pain thresholds when bigger body-rotations were experienced (see fig.5).

The clinical relevance of the present findings could be significant: vestibular stimulation, mainly through CVS, has been shown to be effective in reducing a number of chronic pain conditions (Aranda-Moreno et al., 2019; McGeoch et al., 2008; McGeoch and Ramachandran, 2008; Ngo et al., 2015). Unfortunately, CVS has the downside of triggering possible side effects including nausea, headaches and vomiting (Kelly et al., 2018). To our knowledge, the VR-based protocol used in this study does not produce any of those undesirable symptoms. Indeed, although the use of immersive VR could sometimes be linked to the presence of nausea, dizziness and disorientation (Kourtesis et al., 2019), these symptoms are often triggered by very long exposures to VR and/or bad synchronization between head movements and the corresponding visual scenario. In the present study, the exposure to a specific VR condition was very brief (around 2 minutes), and the greater pain modulatory effect was found during a rotation of only 15 degrees, which, coupled with the fact that we used static images, is very unlikely to evoke any of the aforementioned symptoms. In addition, the clinical usefulness of our approach could even be greater if used in conjunction with galvanic vestibular stimulation, which is another technique that is potentially free from side effects and has been shown to effectively reduce experimental pain (Hagiwara et al., 2020).

However, our approach has its limitations too. For instance, our subjects showed quite a big interindividual variability in terms of perceived self-body rotation and pain modulation. Thus, a possible intervention based on the current model may not work for all. On the same line, it is worth mentioning that a couple of participants perceived some degree of rotation even during the “no rotation” condition. Therefore, carry-over effects may be more prominent in some subjects, especially in ‘within-subjects’ designs. We also have to keep in mind that our sample was constituted of young adults, so the generalizability of our findings to other age ranges (for ex. adolescents or older adults) should be tested.

To conclude, the current piece of research has shown how the vision of static visual scenes can modulate pain, likely through the engagement of the vestibular system. Previous VR studies showed how such technique can represent a valid tool for pain modulation, not only by capturing attentional resources (Hoffman et al., 2000; Maani et al., 2011) but also through the use of virtual embodiment (Martini, 2016; Martini et al., 2013, 2014, 2015; Matamala-Gomez et al., 2019; Nierula et al., 2017; Zanini et al., 2017). The present experiment expands the existing options of VR as a pain management tool and, with its caveats and limitations, contributes to increase the knowledge in a key area of pain treatment, which relates to non-pharmacological interventions by multisensory integration.

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References

Aranda-Moreno, C., Jáuregui-Renaud, K., Reyes-Espinosa, J., Andrade-Galicia, A., Bastida-Segura, A.E., González Carrazco, L.G. (2019). Stimulation of the Semicircular Canals or the Utricles by Clinical Tests Can Modify the Intensity of Phantom Limb Pain. *Front Neurol* 10, 117.

Balaban, C.D. (2011). Migraine, vertigo and migrainous vertigo: Links between vestibular and pain mechanisms. *J Vestib Res Equilib Orientat* 21, 315–321.

Blom, K.J., Arroyo-Palacios, J., Slater, M. (2014). The Effects of Rotating the Self Out of the Body in the Full Virtual Body Ownership Illusion. *Perception* 43, 275–294.

Bottini, G., Paulesu, E., Gandola, M., Loffredo, S., Scarpa, P., Sterzi, R., Santilli, I., Defanti, C.A., Scialfa, G., Fazio, F., Vallar, G. (2005). Left caloric vestibular stimulation ameliorates right hemianesthesia. *Neurology* 65, 1278–1283.

Bottini, G., Paulesu, E., Sterzi, R., Warburton, E., Wise, R.J.S., Vallar, G., Frackowiak, R.S.J., Frith, C.D. (1995). Modulation of conscious experience by peripheral sensory stimuli. *Nature* 376, 778–781.

BRANDT, T., DIETERICH, M. (1999). The Vestibular Cortex: Its Locations, Functions, and Disorders. *Ann N Y Acad Sci* 871, 293–312.

BREMMER, F., KUBISCHIK, M., PEKEL, M., LAPPE, M., HOFFMANN, K.-P. (1999). Linear Vestibular Self-Motion Signals in Monkey Medial Superior Temporal Area. *Ann N Y Acad Sci* 871, 272–281.

Dai, M., Kunin, M., Raphan, T., Cohen, B. (2003). The relation of motion sickness to the spatial-temporal properties of velocity storage. *Exp Brain Res* 151, 173–189.

Dieterich, M., Bense, S., Lutz, S., Drzezga, a, Stephan, T., Bartenstein, P., Brandt, T. (2003). Dominance for vestibular cortical function in the non dominant hemisphere. *Cereb Cortex* 994–1007.

Dieterich, M., Kirsch, V., Brandt, T. (2017). Right-sided dominance of the bilateral vestibular system in the upper brainstem and thalamus. *J Neurol* 264, 55–62.

Dubin, A.E., Patapoutian, A. (2010). Nociceptors : the sensors of the pain pathway Find the latest version : Review series Nociceptors : the sensors of the pain pathway. *J Clin Invest* 120, 3760–3772.

Fasold, O., von Brevern, M., Kuhberg, M., Ploner, C.J., Villringer, A., Lempert, T., Wenzel, R. (2002). Human vestibular cortex as identified with caloric stimulation in functional magnetic resonance imaging. *Neuroimage* 17, 1384–1393.

Faul, F., Erdfelder, E., Lang, A.-G., Buchner, A. (2007). G*Power 3: a flexible statistical power analysis program for the social, behavioral, and biomedical sciences. *Behav Res Methods* 39, 175–191.

Fernandez, C., Goldberg, J.M. (1976). Physiology of peripheral neurons innervating otolith organs of the squirrel monkey. I. Response to static tilts and to long-duration centrifugal force. *J Neurophysiol* 39, 970–984.

Fernandez, C., Goldberg, J.M., Abend, W.K. (1972). Response to static tilts of peripheral neurons innervating otolith organs of the squirrel monkey. *J Neurophysiol* 35, 978–987.

Ferrè, E.R., Bottini, G., Iannetti, G.D., Haggard, P. (2013). The balance of feelings: Vestibular modulation of bodily sensations. *Cortex* 49, 748–758.

Ferrè, E.R., Haggard, P., Bottini, G., Iannetti, G.D. (2015a). Caloric vestibular stimulation modulates nociceptive evoked potentials. *Exp Brain Res* 233, 3393–3401.

Ferrè, E.R., Walther, L.E., Haggard, P. (2015b). Multisensory Interactions between Vestibular, Visual and Somatosensory Signals. *PLoS One* 10, e0124573.

Frank, S.M., Greenlee, M.W. (2018). The parieto-insular vestibular cortex in humans: More than a single area? *J Neurophysiol* 120, 1438–1450.

Gerretsen, P., Pothier, D.D., Falls, C., Armstrong, M., Balakumar, T., Uchida, H., Mamo, D.C., Pollock, B.G., Graff-Guerrero, A. (2017). Vestibular stimulation improves insight into illness in schizophrenia spectrum disorders. *Psychiatry Res* 251, 333–341.

Gordon, C., Barbullushi, A., Tombolini, S., Margiotta, F., Ciacci, A., Yosef, L.S., Barker, L., Martini, M. (2019). Visuo–tactile stimulation, but not type of movement, modulates pain during the vision of a moving virtual limb. *Pain Manag* 9, 449–460.

Grace Gaerlan, M., Alpert, P.T., Cross, C., Louis, M., Kowalski, S. (2012). Postural balance in young adults: The role of visual, vestibular and somatosensory systems. *J Am Acad Nurse Pract* 24, 375–381.

Grüsser, O.J., Pause, M., Schreiter, U. (1990). Vestibular neurones in the parieto-insular cortex of monkeys (Macaca fascicularis): visual and neck receptor responses. *J Physiol* 430, 559–583.

Guldin, W.O., Grüsser, O.J. (1998). Is there a vestibular cortex? *Trends Neurosci* 21, 254–259.

Hagiwara, K., Perchet, C., Frot, M., Bastuji, H., Garcia-Larrea, L. (2020). Cortical modulation of nociception by galvanic vestibular stimulation: A potential clinical tool? *Brain Stimul* 13, 60–68.

Hoffman, H.G., Doctor, J.N., Patterson, D.R., Carrougher, G.J., Furness, T.A. (2000). Virtual reality as an adjunctive pain control during burn wound care in adolescent patients. *Pain* 85, 305–309.

Jeffreys, H. (1983). *Theory of probability* (Clarendon Press).

Kelly, E.A., Stocker, C., Kempton, C.M., Dierking, D.M., Fehlberg, H.E., Adams, M.E.

(2018). Vestibular Testing. *Otol Neurotol* 39, 1222–1228.

Kirsch, V., Keeser, D., Becker-Bense, S., Karali, T., Ertl-Wagner, B., Brandt, T., Dieterich, M. (2017). Vestibular and visual cortex activity during room tilt illusion. *J Neurol* 264, 70–73.

Kourtesis, P., Collina, S., Doumas, L.A.A., MacPherson, S.E. (2019). Validation of the Virtual Reality Neuroscience Questionnaire: Maximum Duration of Immersive Virtual Reality Sessions Without the Presence of Pertinent Adverse Symptomatology. *Front Hum Neurosci* 13, 417.

Lopez, C. (2015). Making sense of the body: The role of vestibular signals. *Multisens Res* 28, 525–557.

Maani, C. V, Hoffman, H.G., Morrow, M., Maiers, A., Gaylord, K., McGhee, L.L., DeSocio, P.A. (2011). Virtual reality pain control during burn wound debridement of combat-related burn injuries using robot-like arm mounted VR goggles. *J Trauma* 71, S125-30.

Macrea, L.M., Macauda, G., Bertolini, G., Straumann, D., Brugger, P., Maurer, K., Palla, A., Lenggenhager, B. (2016). Reducing pain by moving? A commentary on Ferrè et al. 2013. *Cortex* 78, 167–169.

Mancini, F., Longo, M.R., Kammers, M.P.M., Haggard, P. (2011). Visual distortion of body size modulates pain perception. *Psychol Sci* 22, 325–330.

Marshall, R. (2009). Rehabilitation Approaches to Hemineglect. *Neurologist* 15, 185–192.

Martini, M. (2016). Real, rubber or virtual: The vision of "one's own" body as a means for pain modulation. A narrative review. *Conscious Cogn* 43, 143–151.

Martini, M., Kilteni, K., Maselli, A., Sanchez-Vives, M. V (2015). The body fades away: investigating the effects of transparency of an embodied virtual body on pain threshold and body ownership. *Sci Rep* 5, 13948.

Martini, M., Perez-Marcos, D., Sanchez-Vives, M. V. (2013). What colour is my arm? Changes in skin colour of an embodied virtual arm modulates pain threshold. *Front Hum Neurosci* 7, 1–5.

Martini, M., Perez-Marcos, D., Sanchez-Vives, M. V (2014). Modulation of pain threshold by virtual body ownership. *Eur J Pain*.

Matamala-Gomez, M., Diaz Gonzalez, A.M., Slater, M., Sanchez-Vives, M. V. (2019). Decreasing Pain Ratings in Chronic Arm Pain Through Changing a Virtual Body: Different Strategies for Different Pain Types. *J Pain* 20, 685–697.

McGeoch, P.D., Ramachandran, V.S. (2008). Vestibular stimulation can relieve central pain of spinal origin. *Spinal Cord* 46, 756–757.

McGeoch, P.D., Williams, L.E., Lee, R.R., Ramachandran, V.S. (2008). Behavioural evidence for vestibular stimulation as a treatment for central post-stroke pain. *J Neurol Neurosurg Psychiatry* 79, 1298–1301.

McKay, R., Tamagni, C., Palla, A., Krummenacher, P., Hegemann, S.C.A., Straumann, D., Brugger, P. (2013). Vestibular stimulation attenuates unrealistic optimism. *Cortex* 49, 2272–2275.

Neimer, J., Eskiizmirliler, S., Ventre-Dominey, J., Darlot, C., Luyat, M., Gresty, M.A., Ohlmann, T. (2001). Trains with a view to sickness. *Curr Biol* 11, 549–550.

Ngo, T.T., Barsdell, W.N., Arnold, C.A., Chou, M.J., New, P.W., Hill, S.T., Nunn, A.K., Brown, D.J., Gibson, S.J., Miller, S.M. (2015). Bedside neuromodulation of persistent pain and allodynia using caloric vestibular stimulation: an effectiveness trial. *J Neurol Sci* 357, e91.

Nierula, B., Martini, M., Matamala-Gomez, M., Slater, M., Sanchez-Vives, M. V. (2017). Seeing an Embodied Virtual Hand is Analgesic Contingent on Colocation. *J Pain* 18, 645–655.

Pansell, T., Sverkersten, U., Ygge, J. (2006). Visual spatial clues enhance ocular torsion response during visual tilt. *Exp Brain Res* 175, 567–574.

Ploghaus, A., Narain, C., Beckmann, C.F., Clare, S., Bantick, S., Wise, R., Matthews, P.M., Rawlins, J.N.P., Tracey, I. (2001). Exacerbation of Pain by Anxiety Is Associated with Activity in a Hippocampal Network. *J Neurosci* 21, 9896–9903.

Ramachandran, V.S., McGeoch, P.D., Williams, L., Arcilla, G. (2007). Rapid relief of thalamic pain syndrome induced by vestibular caloric stimulation. *Neurocase* 13, 185–188.

Rode, G., Tilikete, C., Luaute, J., Rossetti, Y., Vighetto, A., Boisson, D. (2002). Bilateral vestibular stimulation does not improve visual hemineglect. *Neuropsychologia* 40, 1104–1106.

Sierra-Hidalgo, F., de Pablo-Fernández, E., Herrero-San Martín, A., Correas-Callero, E., Herreros-Rodríguez, J., Romero-Muñoz, J.P., Martín-Gil, L. (2012). Clinical and imaging features of the room tilt illusion. *J Neurol* 259, 2555–2564.

Spitoni, G.F., Pireddu, G., Galati, G., Sulpizio, V., Paolucci, S., Pizzamiglio, L. (2016). Caloric vestibular stimulation reduces pain and somatoparaphrenia in a severe chronic central post-stroke pain patient: A case study. *PLoS One* 11, 1–14.

Takahashi, K., Gu, Y., May, P.J., Newlands, S.D., DeAngelis, G.C., Angelaki, D.E. (2007).

Multimodal Coding of Three-Dimensional Rotation and Translation in Area MSTd: Comparison of Visual and Vestibular Selectivity. *J Neurosci* 27, 9742–9756.

Vallar, G., Sterzi, R., Bottini, G., Cappa, S., Rusconi, M.L. (1990). Temporary remission of left hemianesthesia after vestibular stimulation. A sensory neglect phenomenon. *Cortex* 26, 123–131.

Veale, J.F. (2014). Edingburgh Handedness Inventory—Short Form: A revised version based on confirmatory factor analysis. *Laterality Asymmetries Body, Brain Cogn* 19, 164–177.

Wetzels, R., Matzke, D., Lee, M.D., Rouder, J.N., Iverson, G.J., Wagenmakers, E.J. (2011). Statistical evidence in experimental psychology: An empirical comparison using 855 t tests. *Perspect Psychol Sci* 6, 291–298.

Wilkinson, D., Ade, K.K., Rogers, L.L., Attix, D.K., Kuchibhatla, M., Slade, M.D., Smith, L.L., Poynter, K.P., Laskowitz, D.T., Freeman, M.C., Hoffer, M.E., Saper, J.R., Scott, D.L., Sakel, M., Calhoun, A.H., Black, R.D. (2017). Preventing Episodic Migraine With Caloric Vestibular Stimulation: A Randomized Controlled Trial. *Headache J Head Face Pain* 57, 1065–1087.

Zanini, A., Montalti, M., Caola, B., Leadbetter, A., Martini, M. (2017). Pain during illusory own arm movement: a study in immersive virtual reality. *Eur Med J* 2, 90–97.

Figure Legends

Figure 1. The virtual scenario presented to the participants, during the five VR conditions. The room is a Japanese-like room with a missing front wall showing an external garden with a running waterfall. Only the room where the participants ‘were in’ rotated in the different conditions, while the external garden remained in its normal upright position.

Figure 2. The pain thresholds recorded during the five VR conditions. The columns and vertical error bars represent the group means and standard errors of the normalized pain thresholds in each experimental condition. Asterisks indicate significant comparisons (* $p < 0.05$)

Figure 3. The box and whisker plot of the scores reported in the 4-item subjective questionnaire for each experimental condition, as measured on a 7-point likert scale.

Figure 4. The figure presents the angles reported during the perceived self-rotation task, reflecting the average perceived tilt of body midlines during each VR condition. The bars displayed in correspondence of the necks represent the standard errors.

Figure 5. Correlation plots. The graph on the left shows a significant positive correlation between the item 2 (“I2”, body tilted sensations) scores and the normalized pain thresholds during the “no rotation” condition. On the right, a significant positive correlation is shown between the perceived self-rotation scores and the normalized pain thresholds during the “15-Left” condition.