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Role of corpus callosum in unconscious vision

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A R T I C L E I N F O

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A B S T R A C T

The existence of unconscious visually triggered behavior in patients with cortical blindness (e.g., homonymous hemianopia) has been amply demonstrated and the neural bases of this phenomenon have been thoroughly studied. However, a crosstalk between the two hemispheres as a possible mechanism of unconscious or partially conscious vision has not been so far considered. Thus, the aim of this study was to assess the relationship between structural and functional properties of the corpus callosum (CC), as shown by probabilistic tractography (PT), behavioral detection/discrimination performance and level of perceptual awareness in the blind field of patients with hemianopia. Twelve patients were tested in two tasks with black-and-white visual square-wave gratings, one task of movement and the other of orientation. The stimuli were lateralized to one hemifield either intact or blind. A PT analysis was carried out on MRI data to extract fiber properties along the CC (genu, body, and splenium). Compared with a control group of participants without brain damage, patients showed lower FA values in all three CC sections studied. For the intact hemifield we found a significant correlation between PT values and visual detection/discrimination accuracy. For the blind hemifield the level of perceptual awareness correlated with PT values for all three CC sections in the movement task. Importantly, significant differences in all three CC sections were found also between patients with above-vs. chance detection/discrimination performance while differences in the genu were found between patients with and without perceptual awareness. Overall, our study provides evidence that the properties of CC fibers are related to the presence of unconscious stimulus detection/discrimination and to hints of perceptual awareness for stimulus presentation to the blind hemifield. These results underline the importance of information exchange between the damaged and the healthy hemisphere for possible partial or full recovery from hemianopia.

1. Introduction

A loss of vision in one hemifield of both eyes as a result of a unilateral lesion along the post-chiasmatic visual pathways and the visual cortex is known as homonymous hemianopia (Bouwmeester et al., 2007; Goodwin, 2014). The pioneering studies of Pöppel et al. (1973), Weiskrantz et al. (1974, 1995) and Weiskrantz (2004, 2009a) have shown that some patients with hemianopia can report different types of "visual" reports and behavioral responses in the blind hemifield, a phenomenon named by Weiskrantz "blindsight". So far, the study of the neural basis of this form of unconscious vision has been mainly focused on cortical, subcortical brain areas (e.g., Tamietto et al., 2012) and post-chiasmatic pathways while less is known about the contributions of the corpus callosum (CC).

Therefore, the aim of this study is to evaluate the association between properties of the CC fibers, behavioral detection/discrimination performance and level of perceptual awareness in the blind field of patients with hemianopia. This was motivated by the possibility of an important role of the CC in contributing to recover the missing visual information thanks to the intact hemisphere. Evidence of the crucial role

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of the contralesional hemisphere in the rehabilitation of unilateral cortical lesions has been provided by many studies of various cerebral functions (e.g., Voytek et al., 2010 for prefrontal cortex lesions; Johansen-Berg et al., 2002 for motor lesions). However, the role of the CC in the rehabilitation of visual cortical lesions and, in particular, of hemianopia through the intact hemisphere is still lacking. An interesting related finding has been reported by Kavcic et al. (2015) showing that the lesioned hemisphere responds in some areas interhemispherically to visual stimulation of the intact counterpart. Broadly similar results have been reported by Bollini et al. (2017); Sanchez-Lopez et al. (2019) with the addition of the finding of some reliable, although abnormal, responses by directly presenting visual stimuli to the lesioned hemisphere.

A behavioral study addressing the role of the CC after unilateral hemispheric lesions was carried out a few years ago by (Celeghin et al., 2015) in a group of patients with hemianopia by using Poffenberger (1912) visuo-motor manual reaction time task to behaviorally tap the speed of interhemispheric transfer subserved by the CC. The rationale of this task is based on the fact that in healthy humans the direct uncrossed motor response to a visual stimulus (e.g., right hemifield-right hand) is typically faster than the indirect crossed response (e.g., right hemifield-left hand) that requires an interhemispheric route. Celeghin et al. (2015) found that in the blind hemifield patients could respond quickly and correctly to the unseen stimuli but that the crossed responses were faster than the uncrossed. They hypothesized that this paradoxical result could be explained by an additional transfer of the missing visual input from the damaged to the intact hemisphere and then back again for motor response (see Celeghin et al., 2015 for a thorough discussion). This means a longer route in the uncrossed than crossed response, hence a slower reaction time.

Later, Celeghin et al. (2017), tested extensively in the Poffenberger paradigm an adult patient (GY-56 years) who presented hemianopia as a result of traumatic brain damage occurred at 8 years of age. The patient consistently showed the "blindsight" phenomenon by responding quickly and accurately to brief visual stimuli presented to the blind hemifield without reporting any hint of perceptual awareness. By means of functional magnetic resonance imaging (fMRI) and tractography they found structural and functional evidence of plastic compensatory modifications in posterior portions of the CC. This finding corroborates the hypothesis of an important role of the CC in enabling detection, albeit unconscious, of stimuli presented to a blind hemifield. We believe that an intriguing, fascinating, further question is whether the CC might in some conditions subserve conscious vision in the hemianopia hemifield.

In the present study we have attempted to verify this possibility by testing a sample of patients with chronic hemianopia who a few years beforehand underwent a study in which they were tested with a behavioral visual paradigm similar to the Poffenberger task (Sanchez-Lopez et al., 2020). We found that there were significant correlations between damage of specific cortical areas and presence of blindsight and/or rudimental perceptual awareness. On the bases of those results and of previous evidence on the importance of the CC for blindsight in hemianopics, we carried out the present study to find out what parts of the CC are crucially involved in either unconscious or conscious vision following stimulus presentation to the blind hemifield of hemianopic patients.

2. Material and methods

2.1. Participants

Twelve patients with a diagnosis of hemianopia as a result of long-standing lesion in the post-chiasmatic pathway including the visual cortex were tested in this study (4 females; mean age = 58.5±9.1 yrs). All patients had previously taken part in a study in which their behavioral performance and perceptual awareness reports were analyzed with respect to individual site and extent of brain damage, see Sanchez-Lopez et al. (2020) for details. These 12 patients were selected from the 17 patients of the above study on the basis of the quality of the MRI images that had to be sufficiently good to enable a probabilistic tractography (PT) analysis of the CC. The selection was done by the neuroradiologist without considering the results of the previous study. It is important to specify that none of the patients had direct macroscopic lesions of the CC (see Table 1 for a more detailed neuroradiological description).

Briefly, according to the inclusion criteria, patients should have had a diagnosis of homonymous hemianopia at least three months before testing and be in possession of a recent structural MRI and clinical visual campimetry. Additionally, a further MRI acquisition was carried out as part of our study. We did not recruit patients with pre-existing psychiatric or neurological disorders, substance (alcohol or drugs) use disorder, attentional impairment, such as neglect, evaluated with Bell Cancellation (Gauthier et al., 1989), Line Bisection (Schenkenberg et al., 1980), and Diller letter H cancellation (Diller et al., 1974) tests, and suspicion of cognitive decline (score equal or less than 24 at the Mini Mental State Examination; Folstein et al., 1975). All patients had normal (20/20) or corrected to normal visual acuity and were right-handed. Additionally, a group of eight control participants without brain damage (6 females; mean age = 57.9 ± 10.6 yrs) were included in the study to perform a comparison in the PT values with the group of patients with hemianopia (Table 3). Controls were tested following the same procedure as in the group of patients, but they did not perform the behavioral tasks. No differences in age ($U = 46$; $p = 0.91$) or gender ($\chi^2 = 3.33$; $p = 0.06$) were found.

The original study (Sanchez-Lopez et al., 2020) was approved by the Ethics Committee of the European Research Council and of the Verona Azienda Ospedaliera Universitaria Integrata (AOUI). Clinical details of the patients are described in Table 1 and a graphical representation of the brain lesions and visual defects are shown in Fig. 2.

2.2. Behavioral testing

Behavioral testing aimed at evaluating patients' detection/discrimination accuracy and level of perceptual awareness in a movement detection or orientation discrimination task with stimuli presented to either the blind or the sighted field. The experimental paradigm was described in detail in the paper mentioned above (Sanchez-Lopez et al., 2020).

2.2.1. Stimuli

The stimuli consisted of black and white square-wave gratings with a size of 4 × 4 degrees of visual angle, a full contrast (Michelson = 1), a spatial frequency of 0.875 cycles/degree and were presented with a duration of 250 ms on a background with a luminance equal to the mean of that of the grating = 17.7 cd/m2. The visual stimuli were stationary or moving. For the latter a temporal frequency of 8.33°/second was used. The stimuli were projected on a computer screen placed in front of the participants who were seated with their eyes at 57 cm from the central fixation point in a dimly lit room. All participants were asked to keep fixation on a central point of the screen and eye movements were monitored by means of a closed-circuit TV to control for unwanted saccadic movements. The position of stimulus presentation on the screen was determined individually for each patient considering the blind area previously detected by means of clinical campimetry and of binocular visual field mapping carried out in the laboratory (see for further details Sanchez-Lopez et al., 2020 and Table 1).

2.2.2. Movement detection and orientation discrimination tasks

The movement detection task consisted of horizontal stationary or downward moving stimuli that participants were asked to discriminate by pressing one of two different keyboard keys in a forced-choice task paradigm (one key for stationary and another for moving stimuli). The orientation discrimination task used stationary horizontal or vertical stimuli and patients were asked to discriminate their orientation by pressing one of two different keys (one key for vertical and another for

Table 1
Patients' age, gender, months from injury and neuroradiological description of the lesion. Note: M = Male; F= Female.

Patient Code (Gender)	Age	Months from Injury	Lesion Volume voxels (mm^3)	Stimulus Position (degrees) from the center of the screen	Neuroradiological Description of the Lesion (Type of lesion)
					Left lateral hemianopia
HE (M)	60	4	8763	x = 10, y = 8	Right lesion over the medial portion of occipital lobe extended to the peri-calcarine part, predominately involving the upper calcarine fissure until the cuneus. (Ischemic lesion).
FB (F)	49	17	255,095	x = 14.5, y = 8.5	Right lesion involving superior and middle occipital gyri with interruption of the optic radiation in the same side. Also, right parietal and temporal lobe lesion extended to the poro-encephalic cavity of the latter and ex-vacuo dilatation of right lateral ventricle. (Traumatic lesion).
GB (M)	65	4	26,573	x = 9, y = 7	Lesion over the right fusiform gyri, lingual and calcarine fissure. (Ischemic lesion).
GS (M)	75	6	10,872	x = 16, y = 7	Lesion of antero-superior part of the right calcarine fissure partially extended to the cuneus. (Ischemic lesion).
					Right lateral hemianopia
RF (M)	52	3	22,046	x = 6, y = 6	Left lesion over the middle and anterior part of the calcarine fissure, lingual gyrus and posterior part of fusiform gyrus. (Ischemic lesion).
SL (F)	48	79	27,770	x = 21.5, y = 6	Left lesion over the median para-sagittal part of the occipital lobe extended to the lingual gyrus and peri-calcarine fissure. (Ischemic/ hemorrhagic lesion)
					Upper left quadrantanopia
AP (M)	47	6	44,211	x = 6, y = 11	Right lesion over the anterolateral occipital lobe extended to the posterior temporal lobe, inferior part of the optic radiation and the upper part cerebellar hemisphere. (Surgical lesion).
LF (F)	49	29	3108	x = 12.5, y = 8.5	Lesion involving the right anterior portion of the calcarine fissure until the origin of parietooccipital fissure. (Ischemic lesion).
					Lower left quadrantanopia
BC (M)	69	6	39,925	x = 9, y = 3	Lesion over the right medial portion of the occipital lobe and parieto-occipital fissure extended to the fusiform gyri, lingual, occipital pole and calcarine fissure. (Ischemic lesion)
					Upper right quadrantanopia
DD (M)	56	14	38,229	x = 7, y = 7	Left lesion over the infero-lateral part of the occipital lobe extended to the lingual and fusiform gyri, involving the lateral occipital sulcus as well. (Traumatic lesion)
					Lower right quadrantanopia
LB (F)	62	5	39,344	x = 8, y = 8	Left lesion over the occipital lobe involving the calcarine fissure. (Ischemic lesion).
					Bilateral altitudinal hemianopia
AM (M)	65	34	23,959	x = 12.5, y = 9	Left and right lesion over median para-sagittal occipital lobe that involves the lingual gyrus (more evident on the right portion), a thinning of the anterior portion of the right calcarine cortex is observed as well. (Ischemic/ hemorrhagic).

horizontal stimuli), as in the movement task. All patients used for response their dominant hand (right). The stimuli were presented either in the sighted or blind visual hemifield in different blocks. The maximum number of presented stimuli for each task and participant was 480 (240 per visual field), organized in blocks of 80 stimuli (40 per condition, i.e., stationary/moving for the movement task or vertical/ horizontal for the orientation discrimination task, respectively). Intermingled with the trial stimuli there were 16 catch trials with no stimulus presented. When patients anticipated (by pressing a key before stimulus presentation) or missed the response, the trial was repeated until an on-time response was obtained.

At the end of each trial in the blind field patients were asked to verbally indicate their level of visual perceptual awareness by means of a three-level scale in which they were previously trained: *I saw nothing* = 1; *I realized that there was a stimulus, but I could not discriminate it* = 2; *I clearly saw the stimulus* = 3. In the few occasions of response 3 the trial

Table 2
Detection/discrimination accuracy and level (1-2-3) of perceptual awareness across patients in the movement detection and orientation discrimination tasks for the sighted and blind hemifield.

	Sighted Field		Blind Field	Blind Field	Blind Field					
Patient Code	Accuracy Movement Detection (%)	Accuracy Orientation Discrimination (%)	Accuracy Movement Detection (%)	Accuracy Orientation Discrimination (%)	Perceptual Awareness Movement (%)			Perceptual Awareness Orientation (%)		
					1	2	3	1	2	3
HE&	90.62	66.25	50.00	51.25	86.88	13.12	0	88.75	11.25	0
FB	89.37	82.50	48.12	47.47	100	0	0	98.75	0	1.25
GB	96.25	97.50	56.87	61.25*	99.35	0.65	0	100	0	0
GS	95.00	90.00	49.37	50.00	100	0	0	100	0	0
RF	96.87	93.75	44.82	55.83***	87.5	12.5	0	95.00	5.00	0
SL&	98.75	96.88	43.12	50.00	25	75	0	37.50	62.50	0
AP&	98.75	98.75	54.77**	56.96	27.5	70.62	1.88	65.00	33.75	1.25
LF&	98.75	97.92	58.33*	50.00	61.67	38.33	0	97.50	2.50	0
BC	93.75	97.50	42.03	48.10	98.12	0	1.88	97.50	1.25	1.25
DD&	90.62	93.75	71.25*	55.13	0.63	99.37	0	0.00	97.50	2.50
LB	98.12	98.75	54.77	56.25	91.87	6.25	1.88	96.87	3.13	0
AM&	93.12	91.25	50.62	48.75	80.63	19.37	0	61.25	38.75	0
Mean	95.00	92.07	52.01	52.58	71.60	27.93	0.47	78.18	21.30	0.52

Note: *Above chance performance considering all trials of the task; ** above chance performance considering only the trials where perceptual awareness was reported, ***above chance performance considering only the trials where no perceptual awareness was reported (binomial test at 50%; $p < .05$). & Patients showing visual perceptual awareness according with the classification described above. For statistical analysis of perceptual awareness (PA) only values corresponding to PA = 2, were analyzed.

Table 3
FA and MD values for patients (upper) and controls (lower).

Patient Code	Fractional Anisotropy			Mean Difusivity (mm2/s)		
	Genu	Body	Splenium	Genu	Body	Splenium
HE	0.26	0.26	0.25	1.15	1.15	1.13
FB	0.23	0.23	0.22	1.26	1.28	1.30
GB	0.32	0.32	0.32	1.11	1.11	1.11
GS	0.26	0.25	0.25	1.12	1.14	1.12
RF	0.27	0.27	0.26	1.10	1.10	1.07
SL	0.28	0.26	0.27	1.07	1.05	1.05
AP	0.29	0.28	0.28	1.02	1.01	1.01
LF	0.27	0.26	0.26	1.00	1.01	1.00
BC	0.30	0.31	0.31	1.21	1.13	1.09
DD	0.27	0.26	0.26	1.09	1.09	1.07
LB	0.27	0.24	0.26	1.11	1.18	1.10
AM	0.26	0.26	0.25	1.10	1.12	1.12
Mean	0.27	0.27	0.27	1.11	1.11	1.10
Controls Code						
01 (M)	0.33	0.32	0.33	1.13	1.16	1.15
02 (F)	0.33	0.35	0.34	1.01	1.02	1.01
03 (M)	0.34	0.33	0.34	1.04	1.05	1.00
04 (F)	0.32	0.35	0.34	1.13	1.02	1.05
05 (F)	0.34	0.35	0.35	1.11	1.07	1.07
06 (F)	0.31	0.31	0.31	1.10	1.15	1.05
07 (F)	0.30	0.31	0.30	1.14	1.09	1.08
08 (F)	0.29	0.29	0.29	1.16	1.28	1.22
Mean	0.32	0.32	0.33	1.10	1.11	1.08

was discarded since it usually corresponded to a shift of eye fixation. Routinely, trials were discarded considering both eye movements tracking throughout a CCTV and response of the patients indicating clearly seeing the stimulus. The number of discarded trials ranged between 0 and 2.5% at group level (see Table 2 for the percentage of perceptual awareness score for each patient).

The experimental procedure began with the presentation of a black fixation point at the center of the screen followed, with a semi-random delay ranging between 300 and 500 ms, by the presentation of a stimulus. Finally, in the sighted field a blank screen was displayed for 2500 ms while for the blind field the display of the visual perceptual awareness scale was presented in full vision lasting until response. Correct detection/discrimination responses and errors were recorded for both tasks and visual hemifields while for the blind hemifield we also calculated the percentage of visual perceptual awareness score reports (see Fig. 1).

2.3. MRI data acquisition and preprocessing

A 1.5 T scanner (Philips Ingenia, Philips Healthcare, Eindhoven, The Netherlands) with a 15-channels head coil was used for MRI acquisition. To locate the brain lesion, an anatomical image of the whole brain at high resolution of 1*1*1 mm^3 with Spoiled Gradient Echo (SPGR) 3D T1-weighted was acquired for all patients. From these images, the brain lesion was manually drawn to obtain the masks of the lesion on the native T1-weighted images of each patient using the software ITK-SNAP (Yushkevich et al., 2006). The resulting masks were registered from native to standard MNI space using linear transformation of FMRIB's Linear Image Registration Tool (FLIRT) (Jenkinson et al., 2002; Jenkinson and Smith, 2001) at 1 mm resolution. Finally, for each patient the extension of the lesion was estimated by quantifying the total number of voxels of the mask of the lesion. The results are reported in Table 1 and Fig. 3.

Diffusion weighted images were acquired by using an echo planar (EPI) sequence with the following parameters: TR 2935 ms, TE 84 ms recon voxel size 2 mm isotropic, b value 800 s/mm^2, 32 diffusion direction and a total acquisition time of 6:33 min. To extract the values of CC fibers, probabilistic tractography (PT) analysis was carried out with FMRIB's Diffusion Toolbox (FDT; Behrens et al., 2003, 2007). The regions of interest (ROIs) used as a seed were three: genu, body and splenium. Seeds were placed in CC on mid sagittal scan. We decided to select these three portions based on the main anatomical subdivision. Genu and rostrum were considered together to avoid smaller ROIs. The ROIs were manually drawn on midsagittal plane on the anatomical 3D T1 of each patient already registered in MNI 1 mm space. A modified AutoPtx script (https://fsl.fmrib.ox.ac.uk/fsl/fslwiki/AutoPtx) was applied to automatize the process (de Groot et al., 2013). This script was customized to calculate and extract the Fractional Anisotropy (FA) and Mean Diffusivity (MD) of each bundle that represent tissue microstructure in presence of neuronal damage. These measures are sensitive to diverse tissue properties, such as axonal ordering and density, degree of myelination, among others (Jones et al., 2013; Werring et al., 2000). It is known that decreased FA and increased MD indicate damaged fiber tracts with respect to before a lesion in the same person or a control group (Alves, 2018).

2.4. Statistical analysis

Initially, a comparison between patients and controls by means of the

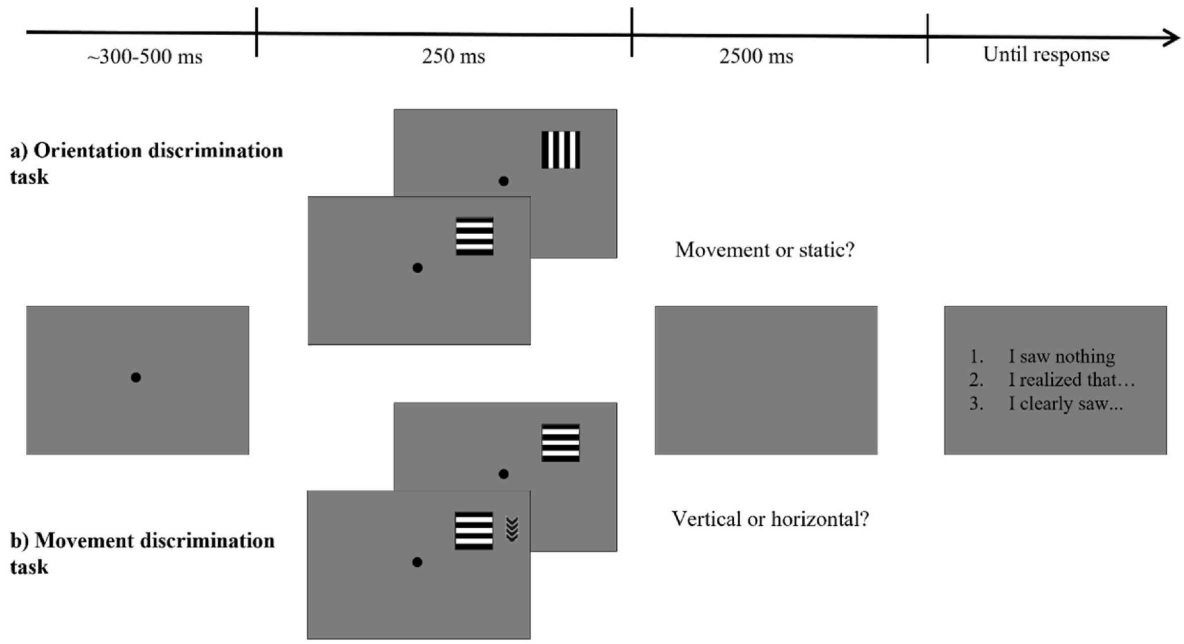

Fig. 1. Outline of orientation discrimination (a) and movement detection (b) tasks.

Mann-Whitney *U* test was performed for FA and MD values to evaluate possible differences suggesting a postlesional degree of white matter damage of the CC as compared to a group of control participants without any brain damage. Then, four different statistical analyses were performed to evaluate the association between CC PT values, detection/discrimination accuracy and visual perceptual awareness. Considering the relatively small patients' sample size ($n = 12$), a non-parametric approach was adopted. First, by using lesion volume (in voxels) and x and y axis stimulus position as covariates, we performed partial correlations of FA and MD values, with the percentage of correct responses for each task and hemifield (sighted and blind). Second, for the blind field, the above variables were correlated with the percentage of trials with scores = 2 in the visual perceptual awareness scale (*I realized that there was a stimulus, but I could not discriminate it*), again by using lesion volume and x and y axis of stimulus position as covariates. False Discovery Rate (FDR) correction was applied for each analysis and PT values category (i.e., FA. MD) to control for multiple correlations. Third, in addition to the correlation analysis, a multiple linear regression stepwise analysis was conducted using the values of detection/discrimination accuracy and visual perceptual awareness as dependent variables, the CC PT values as predictors and lesion volume (in voxels) and x and y axis of stimulus position as covariates. Fourth by using the Mann-Whitney *U* test, for FA and MD and lesion volume, we carried out, independently from the detection/discrimination tasks, comparisons between: a) left and right lesioned patients, b) patients performing above and at chance level, and c) patients with and without perceptual awareness. For b) and c) patients were classified as a function of above chance performance or presence of perceptual awareness report in the blind field in the same way as in a previous published paper (Sanchez-Lopez et al., 2020). Above chance performance was defined as a statistically significant proportion of correct responses different from chance according to a binomial test. Regarding perceptual awareness, only patients reaching a mean percentage of reports above 10 % were classified in this category (see Table 2). All data are available at DOI: 10.6084/m9.figshare.23703915.

3. Results

3.1. Detection/discrimination accuracy and perceptual awareness

Results of behavioral testing and visual perceptual awareness in movement or orientation discriminations across patients are shown in Table 2. For the *sighted visual* field, except patient HE who performed about 66 % for orientation discrimination, the mean group accuracy for movement and orientation discrimination was 95.00 % and 92.07 %, respectively. For the *blind field* the group mean accuracy for movement detection and orientation discrimination was 52.01 % and 52.58 %, respectively. By means of a binomial statistical analysis at 50%, when all trials were analyzed in the blind field, three patients showed above chance performance (i.e., they displayed a significant p value < 0.05 indicating that the proportion of correct responses was significantly higher than chance): LF and DD for movement detection and GB for orientation discrimination. Moreover, AP showed above chance performance for movement detection considering only those trials where perceptual awareness was reported. Finally, RF showed above chance performance during orientation discrimination considering only those trials where no perceptual awareness was reported.

As to presence of perceptual visual awareness in the blind field, we found a group mean of 27.93 % and 21.30 % of visual awareness reports without detection/discrimination for movement and orientation tasks, respectively. The mean for both tasks, at group level, was 24.62 %. As described above, only patients above 10 % in the mean of perceptual awareness for both tasks were classified with visual perceptual awareness, namely, HE, SL, AP, LF, DD and AM.

3.2. Probabilistic tractography (PT)

Values of FA and MD for each patient and control are shown in Table 3. For patients FA values ranged from 0.22 to 0.32, with patient FB showing the lowest values across CC segments and patients while the highest values were found in patient GB. Regarding MD values the range was 1–1.30 (mm^2/s); the lowest values were found in patient LF and the highest in patient FB.

An example of corpus callosum PT is displayed in Fig. 3 for patients FB and RF where a lower integrity of the fibers may be observed in FB with respect to RF and that is consistent with the PT values in Table 3.

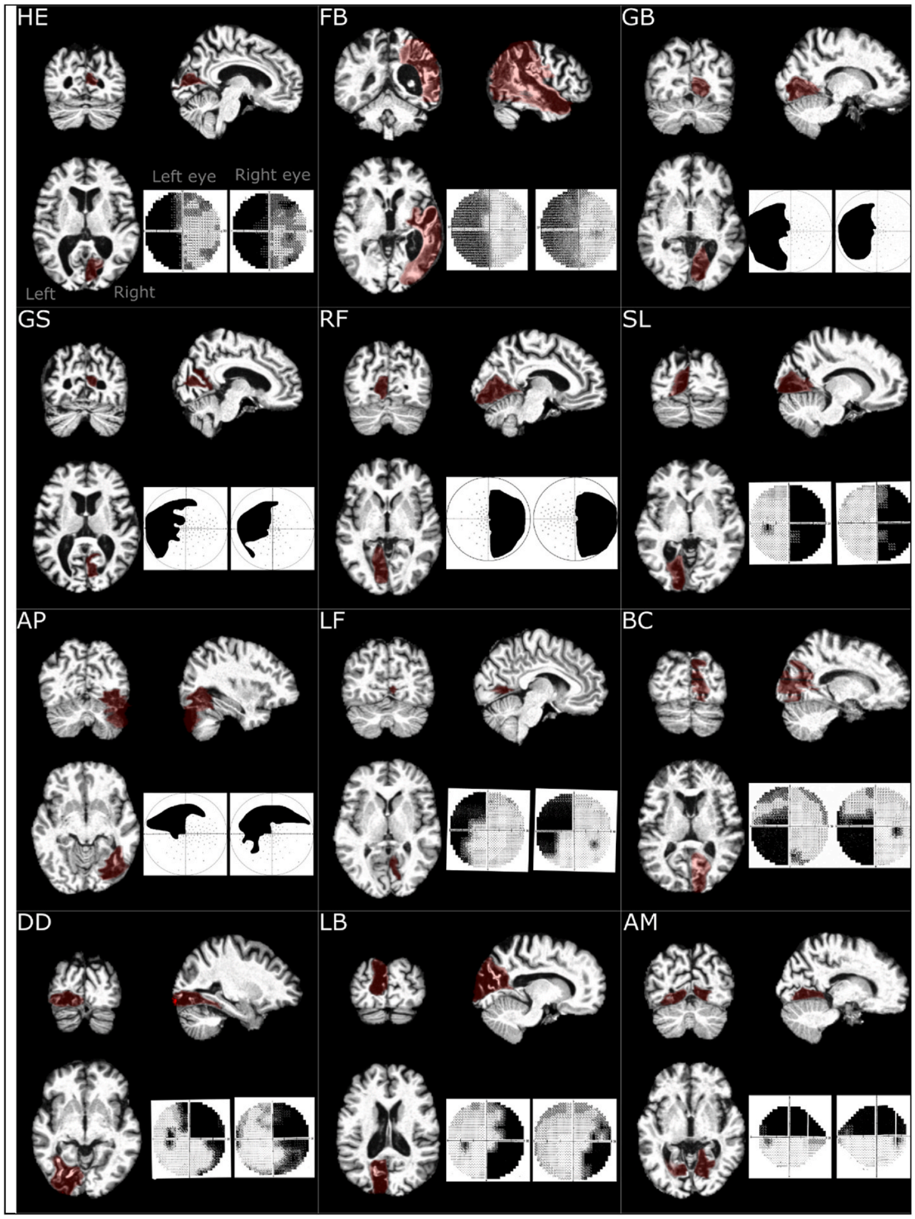

Fig. 2. Individual Structural MRI with overlapped brain lesion masks (in red) and monocular campimetry for both left and right eye (black areas represent the extension of the blind field).

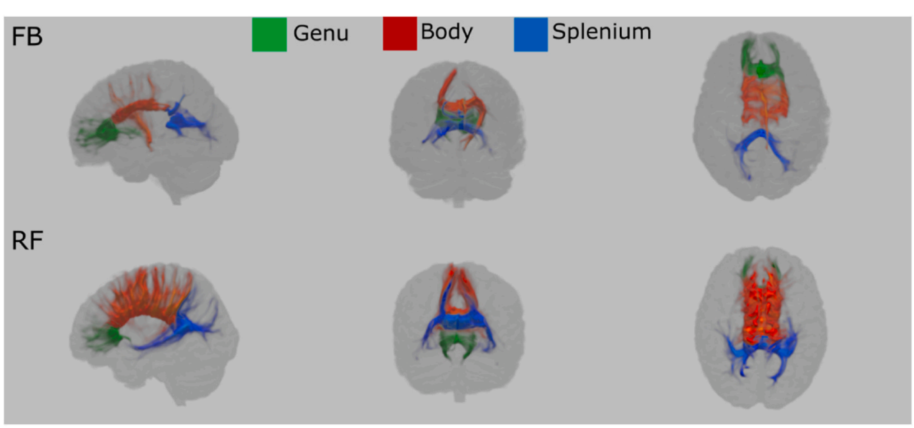

Fig. 3. Example of PT reconstruction for patients FB (upper) and RF (lower). Corpus callosum genu (green), body (red) and splenium (blue).

3.2.1. PT comparisons between patients and controls

This analysis aimed at evaluating the differences in FA and MD values between patients with hemianopia and control participants without brain damage to explore if fiber properties were affected after brain lesion in patients. Results showed significant differences in FA values where patients with hemianopia displayed lower FA in all three sections of the CC: genu (U = 6, p = <0.0001; median patients = 0.27, median controls = 0.32); body (U = 7, p = <0.0001; median patients = 0.26, median controls = 0.32); and splenium (U = 5, p = <0.0001; median patients = 0.26, median controls = 0.34). No differences in MD values were found between groups. If, as previously suggested (Jones et al., 2013), high values of FA represent increased axonal density, reduced axonal caliber and increased degree of myelination, thus suggesting a higher degree of fiber integrity, we could interpretate these differences as lower integrity of the CC fibers of patients with hemianopia with respect to participants without brain damage.

3.3. Correlation between detection/discrimination accuracy and perceptual awareness with PT

This analysis was carried out by evaluating the non-parametric correlation between PT values (see Table 3) and percentage of correct responses for sighted and blind visual fields in the movement detection and orientation discrimination tasks with the volume of the brain lesion and x and y axis stimulus position during the tasks as covariates. Only significant results are reported. Perceptual awareness was analyzed only for the blind field.

3.3.1. Sighted field

Movement: Accuracy was negatively correlated with MD, that is, the higher the performance the lower the values of MD for splenium (partial $rho_{(df = 8)} = -0.76$; FDR corrected-p value = 0.033). No reliable effects were found for FA (see Fig. 4 panel A).

Orientation: Accuracy was positively correlated with FA, that is, the higher the accuracy the higher the values of FA for genu (partial $rho_{(df = 8)} = 0.74$; FDR corrected-p value = 0.02), and splenium (partial $rho_{(df = 8)} = 0.76$; FDR corrected-p value = 0.02). Furthermore, it was negatively correlated with MD of splenium (partial $rho_{(df = 8)} = -0.77$; FDR corrected-p value = 0.02), see Fig. 4 panel B.

In sum, PT tracing for the sighted field showed significant correlations in the two tasks for genu and splenium. Clearly, this implies that the CC is involved in lateralized visual presentations to the sighted field.

3.3.2. Blind field

3.3.2.1. Detection/discrimination accuracy. No significant correlations for FA and MD in the two tasks were found.

3.3.2.2. Visual perceptual awareness. This analysis was performed by evaluating for the blind field the correlation between PT values and percentage of trials where patients reported a level of perceptual awareness = 2 (*I realized that there was a stimulus, but I could not discriminate it*).

Movement: There was a significant negative correlation with MD of genu (partial $rho_{(df = 8)} = -0.83$; FDR corrected-p value = 0.009), body (partial $rho_{(df = 8)} = -0.71$; FDR corrected-p value = 0.02) and splenium (partial $rho_{(df = 8)} = -0.65$; FDR corrected-p value = 0.02; see Fig. 5).

Orientation: No significant correlations were found between the level of perceptual awareness and the PT values for the orientation discrimination task.

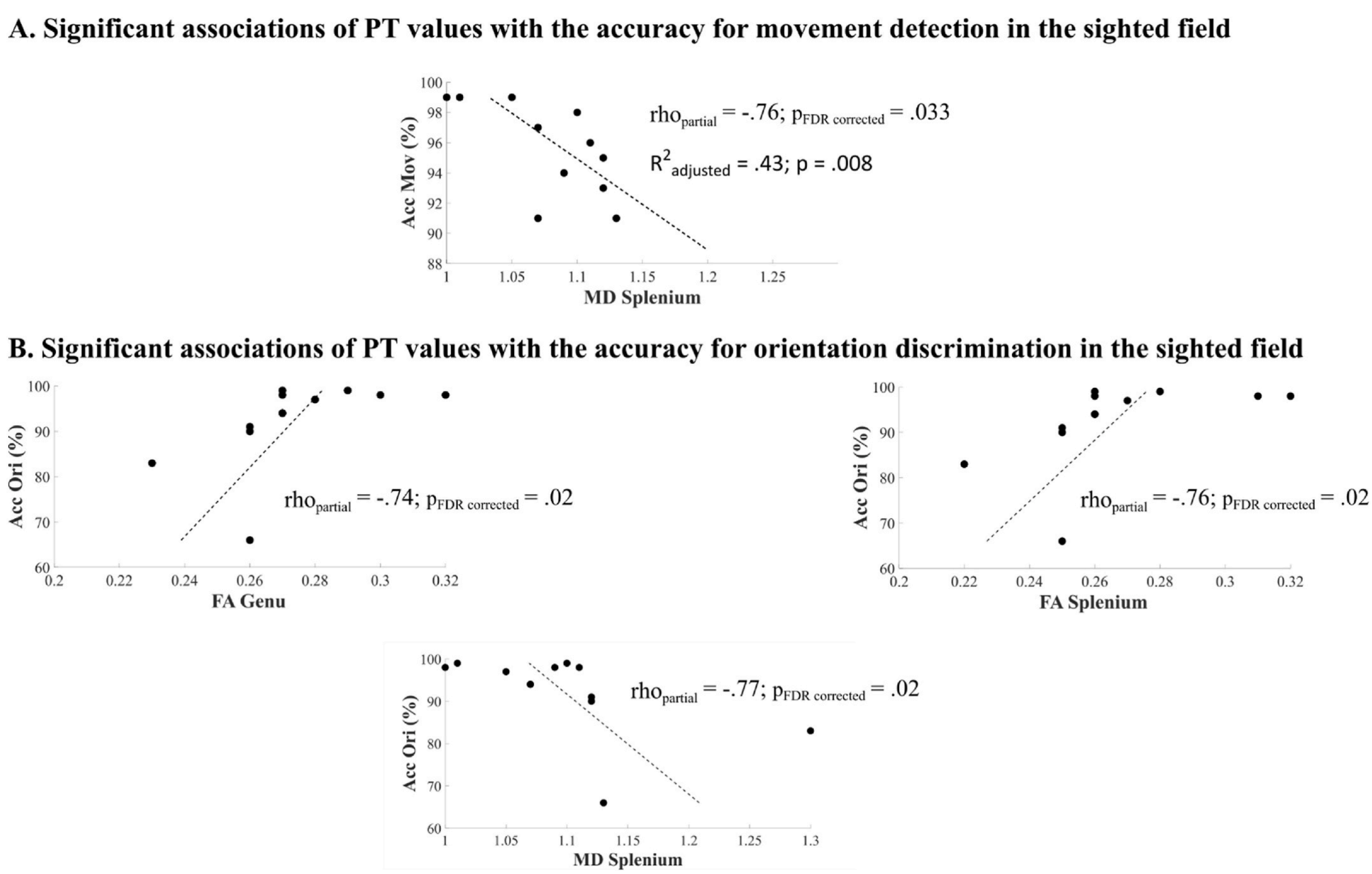

Fig. 4. Scatterplot of the associations between PT values and percentage of correct responses for the sighted visual field in the movement detection (panel A) and orientation discrimination (panel B) tasks. PT = Probabilistic Tractography; FA = Fractional Anisotropy; MD = Mean Diffusivity; Acc Mov (%) = Percentage of accuracy for movement detection; Acc Ori (%) = Percentage of accuracy for orientation discrimination.

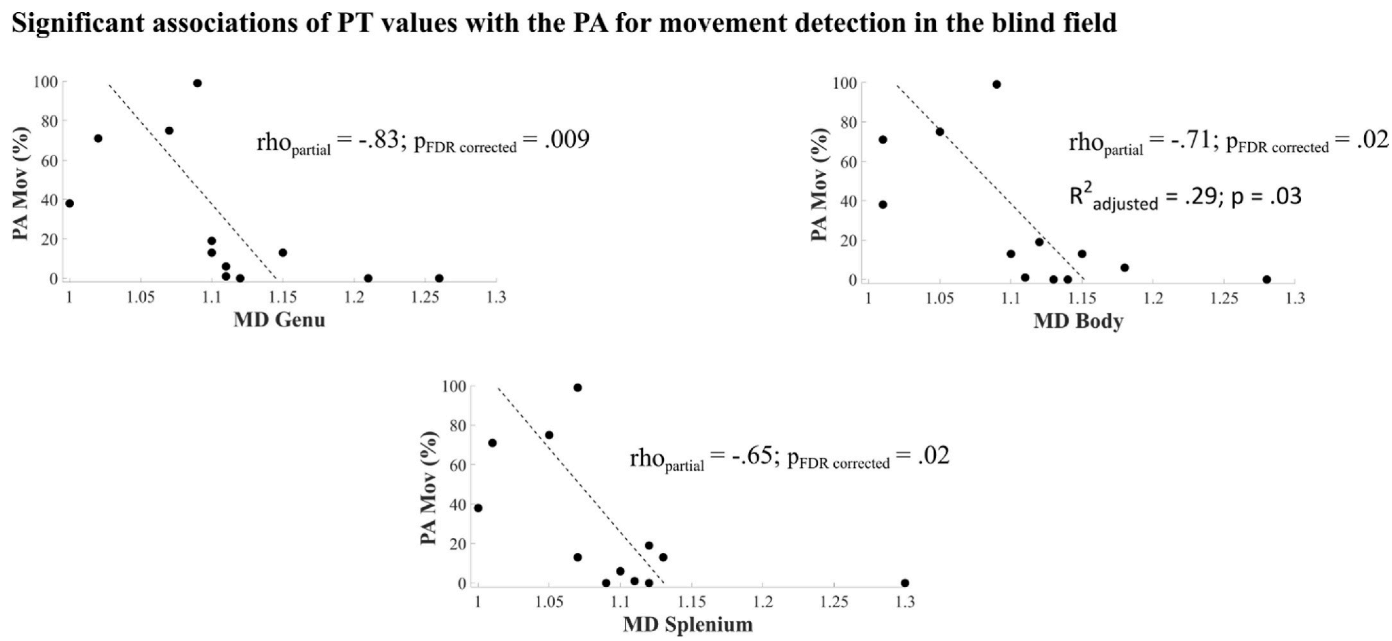

Fig. 5. Scatterplot of the significant associations of PT values with the level of perceptual awareness for movement detection task. PT = Probabilistic Tractography; PA = Perceptual Awareness; MD = Mean Diffusivity; PA Mov (%) = Percentage of trials with perceptual awareness = 2 for the movement detection task.

In sum, in the *movement task* the correlation PT-Perceptual Awareness was reliable for all three portions of the CC for movement but not for *orientation*. There were no significant effects for FA.

3.4. Multiple linear regression analysis between detection/discrimination accuracy and perceptual awareness with PT

This analysis aimed at evaluating whether detection/discrimination accuracy and/or level or perceptual awareness could be predicted as a linear function of the PT values of the CC. As described above, PT values were introduced in a multiple linear regression stepwise analysis as independent variables, using detection/discrimination accuracy and level of perceptual awareness as dependent variables, separately. Additionally, volume of the brain lesion and x and y axis stimulus position during the tasks were introduced as covariates. Only significant results are reported.

3.4.1. Sighted field

MD values of the splenium significantly predict the movement detection accuracy (R^2adjusted = 0.43; $F_{(1,11)} = 10.24$; p = 0.008; β = −32.07; SE = 10.02). No other significant results for movement detection or orientation discrimination accuracy in the sighted field were found (see Fig. 3, panel A).

3.4.2. Blind field

3.4.2.1. Detection/discrimination accuracy. As in correlational analysis, no significant results were found for FA or MD.

3.4.2.2. Visual perceptual awareness. Percentage of visual awareness during the movement detection task was significantly predicted by the MD values of the body portion of CC (R^2adjusted = 0.29; $F_{(1,11)} = 6.12$; p = 0.03; β = −288.71; SE = 116.62). No other significant results for the level of perceptual awareness for movement detection or orientation discrimination tasks in the blind field were found (Fig. 5).

3.5. Group comparisons as a function of: hemispheric side of the lesion, chance/above chance level of detection/discrimination performance, and presence of perceptual awareness

Non-parametric comparisons by means of the Mann Whitney *U* test were tested for PT values as well as total volume of the lesion. Only significant results are reported.

Hemispheric side: Patients were subdivided in two groups: 7 right-lesioned and 4 left-lesioned. Patient AM was not included in this analysis since he has bilateral lesion and bilateral visual defect. No overall significant differences were found for the hemispheric variable.

Chance/above chance: We compared patients with above (n = 5) and at chance (n = 7) performance in detection/discrimination tasks. Significant group differences were found in MD for genu (U = 5, p = 0.04; median above chance = 1.09, median at chance = 1.12), body (U = 3, p = 0.01; median above chance = 1.09, median at chance = 1.14) and splenium (U = 5, p = 0.04; median above chance = 1.07, median at chance = 1.12). No other significant differences were found.

In sum, in contrast to detection/discrimination analysis, see 3.4, here there were significant MD differences for all three callosal portions with lower values of MD for patients scoring above chance independently from type of task.

Perceptual awareness: Patients with (n = 6) and without (n = 6) perceptual awareness were compared. Results showed significant differences in MD of the genu (U = 4, p = 0.02) with patients with PA showing lower values (median = 1.08) than those without PA (median = 1.12). No other significant difference was found.

4. Discussion

Overall, an important finding of the present study is that the degree of CC fibers integrity, as inferred from PT, is positively related to the presence of unconscious stimulus detection/discrimination as well as to perceptual awareness for stimuli presented to the blind hemifield. In Table 4, we summarize the significant correlations between PT values and detection/discrimination or perceptual awareness for the three CC portions as well as significant differences when the various groups were compared.

It turns out that all three CC portions are involved in

Table 4
Summary of correlation, multiple linear regression and comparison results. Note: CC = corpus callosum; FA = fractional anisotropy; MD = mean diffusivity.

CC section	Correlation	Multiple linear regression	Comparison
	Negative		
Genu	Level of perceptual awareness for movement detection negatively correlates with MD of genu.		Patients with above chance performance show lower MD values in genu than patients with at chance performance. Patients with perceptual awareness show lower MD values in genu than patients with no perceptual awareness.
Body	Level of perceptual awareness for movement detection negatively correlates with MD of body.	MD of body predicts level of perceptual awareness for movement detection.	Patients with above chance performance show higher FA values in body than patients with at chance performance.
Splenium	Level of perceptual awareness for movement detection negatively correlates with MD of splenium.		Patients with above chance performance show higher FA values in splenium than patients with at chance performance.

interhemispheric transfer of information related to movement detection or orientation discrimination accuracy and above-chance performance or perceptual awareness. Note that the latter could not be responsible for detection/discrimination performance given that patients scoring 2 in the perceptual awareness scale reported that they did not know what they saw. Hence, awareness of stimulus detection was immaterial for a correct detection/discrimination.

Trying to pin down the cortical areas interconnected by means of the three CC portions would help understand their specific contribution to blindsight. The previous paper from our lab (Sanchez-Lopez et al., 2020) used a rationale broadly similar to that of the present study but focused on extent of damage of 12 cortical areas rather than CC anatomy. In that study we found that for above-chance detection of visual movement there were two important areas, namely, the Precuneus and the Posterior Cingulate Gyrus. For perceptual awareness, the important areas were the Striate and Extrastriate areas as well as the Posterior Cingulate Gyrus again. As to relation with the CC, Striate and Extrastriate areas, i.e., BA 17 (V1), 18 (V2), 19 (V3, V4, V5) send numerous callosal projections to homologous and non-homologous areas of the other hemisphere (see Zhang et al., 2023). Most of them run in the Splenium and this would fit well with its role in perceptual awareness found in our study. Moreover, the Splenium contains, among others, many fibers that interconnect the visual primary cortex representation of the vertical meridian and strictly adjacent areas and this fits well in with its role in the detection/discrimination tasks. However, there is evidence that the Splenium also hosts many fibers from other cortical areas, namely, temporal and parietal areas, see Berlucchi (2014) and Park et al. (2008), and this might be in keeping with a higher-level role its role in tasks performance that we have not specifically tested in the present study. By the same token, it is not surprising to find a role of the CC Body in both detection/discrimination accuracy and perceptual awareness given that it contains fibers connecting the sensory-motor cortex of the two hemispheres which are likely to play an important role in the task employed in our study, i.e., visuomotor reaction time. Finally, the contribution of the Genu is well explainable and very interesting because this portion of the CC is composed of fibers interconnecting prefrontal and frontal areas. Importantly, selective fMRI activation of the Genu has been found by several studies when using the Poffenberger paradigm (Gawryluk et al., 2011; Tettamanti et al., 2002; Weber et al., 2005), that is, a task basically similar to the one adopted in the present study.

One intriguing question that remains to be clarified is why the contribution of the CC connections is not sufficient to yield full perceptual awareness of the stimuli that some participants are able to unconsciously discriminate. The only evidence of perceptual awareness provided by interhemispheric transfer in our study is the visual sensation felt by some patients following presentation of stimuli that they cannot identify, a phenomenon similar to that defined “Blindsight type II” (Sahraie et al., 2010; Weiskrantz, 2009b). It could be hypothesized that this effect might be related to a callosal input insufficient to reach full stimulus awareness. The reason for that could rely on either quantitative and/or qualitative problems. Among the latter there might be inhibitory effects from the receiving hemisphere that is “reluctant” to deal with the weak and/or distorted signal from the damaged hemisphere.

To our knowledge this is the first study addressing the question of the contribution of the CC to behavioral performance and visual perceptual awareness in patients with hemianopia. Of course, it is important to recognize the limitations related to the sample size of our study and the variability in the type and extension of the patient’s lesion. To explore the possible effect of the presence of outliers on the results, we performed the same correlational and regression statistical analysis excluding patient FB, since this patient showed the highest lesion extension (255,095 mm^3). These exploratory analyses (not reported) showed similar results for the blind visual field: for the movement detection task a negative partial correlation between the MD value of the CC genu and the percentage of perceptual awareness was found (partial $rho_{(df = 8)} = -0.78$; FDR corrected-p value = 0.03). This correlation was confirmed by the multiple linear regression analysis where MD values for the CC genu and those of the CC body significantly predicted the percentage of perceptual awareness during the movement detection task and accuracy for the orientation discrimination task, respectively (R2adjusted = 0.29; F (1,10) = 5.65; p = 0.03; $\beta = -393.51$; SE = 165.49; R^2adjusted = 0.60; $F_{(1,10)} = 18.02$; p = 0.002; $\beta = 0.000004$; SE = 0.0000009). Considering the similarity between those results we believe that our findings are not biased by the presence of outliers and therefore we decided to include FB in the study.

In conclusion, we found a relation between the structures of the CC and unconscious pattern detection/discrimination as well as of perceptual visual feeling experienced in the blind field. This might represent an important first step for trying to stimulate the efficacy of callosal interhemispheric exchange of information with the purpose of regaining partial or full perceptual awareness in the blind field. Examples of the importance of the CC for recovery after a unilateral cortical lesion have been described in patients with spatial hemineglect following right hemisphere lesion and the intact parts of the CC are crucial for rehabilitation therapies, e.g., with prism adaptation (Lunven et al., 2019; Rossetti et al., 1998). Thus, these studies, as well as the present one, support the hypothesis (Bartolomeo and Thiebaut de Schotten, 2016; Lunven et al., 2015) that a cross-talk between hemispheres is essential for recovery of disrupted functions after unilateral lesions.

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CRediT authorship contribution statement

Javier Sanchez-Lopez: Conceptualization, Data curation, Formal analysis, Methodology, Software, Validation, Visualization, Writing – original draft. **Nicolo Cardobi:** Data curation, Formal analysis, Investigation, Software, Validation, Writing – review & editing. **Giorgia Parisi:** Conceptualization, Data curation, Investigation, Writing – review & editing. **Silvia Savazzi:** Conceptualization, Funding acquisition, Methodology, Project administration, Supervision, Writing – review & editing. **Carlo A. Marzi:** Conceptualization, Funding acquisition, Methodology, Project administration, Supervision, Writing – original draft.

Declaration of competing interest

None.

Data availability

I have shared the link to my Data at the methodology section of the Manuscript.

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