KU LEUVEN

GROEP BIOMEDISCHE WETENSCHAPPEN

FACULTEIT BEWEGINGS- EN REVALIDATIEWETENSCHAPPEN

The influence of task complexity during bimanual coordination training on behavioural performance and functional connectivity between left dorsal premotor cortex and the entire brain system

door Laura MAES
en Shanti VAN MALDEREN

masterproef aangeboden, tot het behalen van de graad van Master of Science in de revalidatiewetenschappen en kinesitherapie

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Koen Cuypers, promotor
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Opgesteld volgens de richtlijnen van Journal of Neuroscience.

WOORD VOORAF

Omdat het schrijven van een master thesis geen *one man's job* is maar een opgave waarin we werden bijgestaan door een promotor, co-promotor, vrienden, familie en kotgenoten, zouden we hen dan ook speciaal willen bedanken.

Een dikke dankjewel allemaal voor de onvoorwaardelijke steun, handige tips en ongelooflijke begeleiding. Jullie hebben ons de juiste houvast aangereikt om steeds de goede richting te kiezen, zodat wij onze thesis succesvol hebben kunnen afronden. In bijzonder willen wij onze co-promotor, Celine Maes, bedanken voor haar deskundige advies, ondersteuning en aanmoediging gedurende deze masterproef.

Ook willen we graag onze promotor, Koen Cuypers, en de volledige onderzoeksgroep Bewegingscontrole & Neuroplasticiteit bedanken voor de ervaring. De voorbije twee jaar hebben we enorm veel bijgeleerd, op wetenschappelijk gebied maar ook op persoonlijk vlak. Zo kregen we de kans om de werking van het labo te ontdekken om op deze manier meer voeling met ons onderwerp te bekomen. Daarnaast werd ook onze interesse in onderzoek aangewakkerd.

Tenslotte was het schrijven van deze thesis verbonden met onzekerheden zowel op persoonlijk als op academisch vlak. Gelukkigerwijze brachten we deze thesis niet alleen tot stand waardoor we gedurende het dusdanig schrijven van deze thesis steeds op elkaar konden terugvallen. Hierdoor hadden we het vermogen door te zetten, het beste in onszelf naar boven te halen en onszelf veelvuldig te overstijgen.

Meensel-Kiezegem, 23/05 L.M.
Schaffen, 23/05, S.V.M.

SITUERING

De masterproef is gekaderd binnen het project “Inter- and intrahemispheric interactions within the aging brain and their role in motor behaviour”, één van de lopende projecten binnen de onderzoeksgroep Bewegingscontrole & Neuroplasticiteit van de Faculteit Bewegings- en Revalidatiewetenschappen aan de KULeuven. Deze onderzoeksgroep, onder leiding van Professor Swinnen, richt zich voornamelijk op neurowetenschappelijk onderzoek met als doel het begrijpen van de controle van menselijke bewegingen onder zowel normale als verstoorde omstandigheden. Er wordt nadruk gelegd op coördinatie en cognitief uitdagende motorische taken in associatie met sensorische verwerking. Daarnaast focust men zich op acquisitie van vaardigheden en de neuro-gedragsmatige veranderingen die zich voordoen als resultaat van training. Om dit te verwezenlijken, maken de onderzoekers gebruik van brein stimulatietechnieken waaronder transcranial magnetic stimulation (TMS) alsook verscheidene medische beeldvormingstechnieken als functional magnetic resonance imaging (fMRI). Deze laatste techniek wordt ook gebruikt binnen het project waarin onze thesis kadert. Meer specifiek maakt men in dit project gebruik van task-based fMRI om hierna analyses uit te voeren die ons tot de functionele connectiviteit brengen. Hersenregio’s die een gelijktijdige verhoging/verlaging van het BOLD-signaal (d.i. de activiteit) op een bepaalde taak vertonen, worden in kaart gebracht. Hierna probeert men de connectiviteit tussen verschillende hersenregio’s met gelijkaardige functionele kenmerken na te gaan. Dit stelt ons dan in staat om de taak specifieke connectiviteit, geassocieerd met de ‘seed region’ waarin we geïnteresseerd zijn, te visualiseren. Het ontwerp van de taak bestaat steeds uit een experimentele en een controle conditie. Beiden worden gemeten binnen één scan zodat zowel het verschil in BOLD contrast uitgelokt door de experimentele als door de controle conditie gemeten kan worden (Chen & Glover, 2015). Voordien werd fMRI voornamelijk gehanteerd voor het onderzoeken van ‘resting state connectivity’. Hierbij gaat men regionale interacties na die plaatsvinden wanneer het brein in rust is. Belangrijke netwerken als het ‘default mode network’ werden hiermee in kaart gebracht (Bellec et al., 2006). ‘Task-based connectivity’ daarentegen werd veel minder intensief onderzocht. Aangezien men gedurende het grootste deel van de dag doelgerichte handelingen uitvoert, is het cruciaal om ons blikveld ook te richten naar regio’s die tijdens het uitvoeren van deze handelingen connectiviteit vertonen. Om aan deze nood tegemoet te komen, maken we gedurende ons onderzoek gebruik van ‘task-based fMRI’ om de invloed van taak complexiteit op prestatie en functionele connectiviteit tussen linker PMd en de rest van het brein gedurende bimanuele coördinatie training te onderzoeken. Dit stelt ons in staat meer inzicht te verwerven in de controle van menselijke bewegingen alsook in de training-geïnduceerde acquisitie van vaardigheden en neuro-gedragsmatige veranderingen, waardoor deze studie in de onderzoekslijn van de groep Bewegingscontrole & Neuroplasticiteit past.

Referenties

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Abstract

The role of left dorsal premotor cortex (PMd) during motor learning has been previously investigated with functional magnetic resonance imaging (fMRI) and transcranial magnetic stimulation (TMS), revealing its significance in action selection, preparation and execution of uni- and bimanual movements. Furthermore, PMd forms a key part in motor learning as well as in adaptability to task complexity. Although task complexity is known to modulate activity patterns of PMd, changes in functional connectivity patterns as well as training-induced modulations in functional connectivity are less understood. Here we examined the influence of task complexity on performance of a bimanual coordination task and functional connections incorporating left PMd during training using task-based functional magnetic resonance imaging (TB-fMRI). Sixteen healthy, young adults performed a visuomotor bimanual tracking task, containing a simple (isofrequeny) and complex (non-isofrequency) task variant, across three training sessions. TB-fMRI was acquired during the first and last day of practice. Behavioural results indicated that execution of the complex task variant resulted in a greater behavioural improvement across training as compared to the simple task variant. Second, training-related neural modulations demonstrated a decreased connectivity between PMd and visual processing regions from the first to the last day of training, in the more complex relative to the simple task variant. Conversely, increased PMd-seeded connectivity with the sensorimotor-related brain regions was observed as a function of practice. Thereby, results of the present study confirmed that left PMd is of great importance during the execution of complex tasks as well as during motor learning.

Introduction

Bimanual coordination is an essential aspect of daily life since the majority of activities performed every day require a differential contribution of each limb (e.g. driving a car, zipping your coat…). Nearly all bimanual motor skills call for practice. Motor learning is defined as a behavioural change that is expressed as an improvement in performance and acquired as a result of practice**.** Thus, learning starts during practice whereby performance immediately improves. This is called the fast within-practice phase (Kantak, Stinear, Buch, & Cohen, 2011; Solesio-Jofre et al., 2018). A second phase comprises performance gains in between subsequent practise sessions. The latter is therefore called the slow delayed phase (Kantak et al., 2011; Solesio-Jofre et al., 2018). Distributing training sessions over time is thought to strengthen the consolidation of learning (Wymbs, Bastian, & Celnik, 2016). Interestingly, the learning rate among different people and different activities is unequal. One of the factors contributing to this variability is task complexity. When task complexity is low, initial performance levels will be high, thereby reducing learning potential (Solesio-Jofre et al., 2018). In this regard, numerous studies indicated higher learning potential in complex as compared to easier task variants (Blischke & Malangré, 2017; Kuriyama, Stickgold, & Walker, 2013; Pauwels et al., 2018a). Moreover, with respect to the slow delayed phase of learning, a sufficient amount of motor complexity, combining an adequate amount of memory load and extent of movement coordination, is required to improve performance of motor tasks through sleep dependent offline learning (Blischke & Malangré, 2017; Kuriyama et al., 2013). However, as literature is inconsistent with respect to the influence of task complexity on motor learning, more research is warranted.

Research has shown that bimanual coordination skills are carried out by an entire system of brain areas, including both cerebral hemispheres (Serrien, 2008). This system consists of the supplementary motor area (SMA), premotor cortex (PMC), primary sensorimotor cortex (SM1), cingulate motor cortex (CMC) and the cerebellum (F Debaere, Wenderoth, Sunaert, Van Hecke, & Swinnen, 2004; F Debaere et al., 2001). Which brain regions of the network are activated and how they are connected, depends on various external factors (Swinnen, 2002). Examples of these external factors are task difficulty and frequency. In this regard, PMd is assumed to play a vital role in the adaptability to task complexity. As such, Debaere et al. (2004) found that activation of dorsal premotor cortex (PMd) and cerebellum augmented with increasing complexity and frequency of spatiotemporal coordination. Using a dual-site transcranial magnetic stimulation (dsTMS) paradigm, Fujiyama et al. (2016) examined the functional role of PMd to the contralateral primary motor cortex (M1) during the preparation of a complex bimanual coordination task. Results of this study demonstrated a modulation in the left PMd-right M1 interaction during the preparation of a complex task variant (non-isofrequency movement) while the interaction remained unchanged while preparing a more effortless task variant (isofrequency movement). This implies a task-dependent influence of left PMd on contralateral M1 (Fujiyama et al., 2016). These results are corroborated by functional magnetic resonance imaging (fMRI) studies showing a higher activation of PMd during complex motor activities as compared to easier activities (Meister et al., 2005). This study, investigating the influence of motor sequence complexity and long-term practice on motor representations in non-musicians and musicians, found that PMd demonstrated a higher fMRI activation during complex movements in comparison with the execution of simple motor sequence. However, this effect was only observed in non-musicians, suggesting that the involvement of PMd is associated with a higher level of visuomotor integration (Meister et al., 2005). Furthermore, also investigating motor sequence learning, Schubotz & Von Cramon et al. (2002) found that task difficulty was positively related to PMd activation, especially when the sequence length was extended. This vital role of PMd, which is part of the premotor cortex and lies between the primary motor- and prefrontal cortex (Geyer, Schormann, Mohlberg, & Zilles, 2000; Picard & Strick, 2001), can be attributed to its important influence on descending motor control (Dum & Strick, 2002). It takes part in planning of unimanual movement as well as bimanual movement including action selection, preparation and execution (Paul Cisek & Kalaska, 2005; Côté, Hamadjida, Quessy, & Dancause, 2017; Kiyama et al., 2014; Wise, 1985). This is especially the case when performing a visuomotor task (Cavina-Pratesi, 2006; Grafton, Fagg, & Arbib, 1998; Hoshi & Tanji, 2006; Terao et al., 2007).

In addition to its role in adaptability to task complexity, PMd forms a key part of motor learning (Sun, Miller, Rao, & D'Esposito, 2007). Left PMd in particular appears to perform a principal role in the selection and implementation of action plans as well as motor learning of left and right unimanual movements (P. Cisek, 2002; Hardwick, Rottschy, Miall, & Eickhoff, 2013; Kantak et al., 2011), consistent with the leading role of the left hemisphere in motor control (Schluter, Rushworth, Passingham, & Mills, 1998). It is assumed that practise demanding higher cognitive effort needs more PMd engagement (Sun et al., 2007). This significant role of PMd is demonstrated in several studies. With respect to functional connectivity, it is generally seen that a change in resting state functional connectivity (RS-FC) in young adults can be acquired through motor learning, both within and between training sessions (Solesio-Jofre et al., 2018; Woolley et al., 2015). Long-term training of bimanual skills induces a functional rearrangement of the motor system allowing planning of more complex movements without the necessity of recruiting supplementary neuronal reserves (Meister et al., 2005). This reorganization, comprising neural plasticity mechanisms, is seen in the PMC along with the SMA, M1 and various other regions (Albouy et al., 2015; Donchin, Gribova, Steinberg, Bergman, & Vaadia, 1998; Ma et al., 2010; Müller et al., 2007; Schmidt, Carroll, Hinder, Garry, & Summers, 2010; Sebastian, Rushworth, Johansen-Berg, Boorman, & O'Shea, 2007; Waddingham, Ewas, Rothwell, Ward, & Talelli, 2007).

Interestingly, investigating multimodal connectivity of PMd during motor learning using resting-state fMRI, Hardwick et al. (2013) stated that PMd might surrogate as a junction between motor control and cognition, through a functional connection with brain regions engaged in visuomotor control and executive function. Along the same lines, augmented functional connectivity between PMd, used as seed region, and the bilateral S1/M1 and the PM ipsilateral extending to contralateral S1/M1, PM and the SMA during early versus late learning was indicated by a second study. In particular, PMd established the strongest task-related modifications in interregional connectivity as compared with other prominent regions used as seed in this study. More specific, these modifications in interregional coupling are even greater during early learning in comparison with the late learning stage (Sun et al., 2007).

Thus far, the role of PMd has predominantly been investigated with brain activation (fMRI) studies as well as transcranial magnetic stimulation (TMS). Task-based functional connectivity, however, received limited attention until now. Remarkably, Kiyama et al. (2014) investigated the role of PMd in task-related functional connectivity patterns and demonstrated that performing more complex bimanual movements, i.e. anti-phase movements, required more connectivity with left PMd as compared to in-phase movements. Moreover, the authors postulated that intra- and interhemispheric connectivity is evoked with the dominant PMd as a mediator to successfully perform bimanually coordinated movements (Kiyama et al., 2014). These interregional connections, comprising both cortical and subcortical regions, are a substantial element of motor learning (Sun et al., 2007). This task complexity-dependent modulation of functional connectivity is a promising finding that needs further research.

Adjacent to the lack of studies investigating bimanual coordination by means of task-based functional connectivity, it is remarkable that the vast majority of the studies focuses on the motor network. Moreover, the insight in how the functional connectivity changes through time following training is limited. In general, training-induced modulations of resting state received more attention as compared to task-related connectivity modulations. Therefore, the aim of this study was to extend current evidence by exploring training-induced modulations of task-based connectivity patterns between PMd and the entire brain system in an easy- as compared to a more complex bimanual coordination task. To address these objectives, TB-fMRI was used during the first and last day of a 3-day training in which participants performed a bimanual visuomotor tracking task.

We speculated that the complex task variant will initially result in worse performance but will ultimately lead to a greater extend of motor learning. Furthermore, we hypothesized complexity-dependent connectivity patterns to be affected by the stage of training.

Insights in the underlying mechanisms of the functional role of PMd and its connectivity changes under the influence of training is crucial, as future research can use this knowledge to establish interventions that might enforce its function in case of deficiency (e.g. in stroke patients).

Materials and methods

Participants

Sixteen young adults (8 females, 8 men), ranging from 19 years old to 23 years old (mean age, 21.4± 1.2 years) were included in this experiment. The participants had no neurological and psychiatric disorders. Handedness was determined by means of the Oldfield Handedness scale (Oldfield, 2001) and confirmed that all participants were right-handed. A written informed consent was obtained from each participant before testing. The protocol was approved by the local ethical committee of KU Leuven, Belgium, and was in accordance with the Declaration of Helsinki (1964).

Experimental timeline

The participants were all assigned to the same training program. The training protocol consisted of 4 sessions in total: one baseline session and three training sessions (TR1, TR2 and TR3) in which participants performed a bimanual coordination task (**Figure 1**). Handedness (Oldfield, 2001) and contraindications for MRI scanning were examined during the baseline session. Subsequently, participants got acquainted with the task during the familiarization period, followed by execution of the bimanual visuomotor task in the dummy scanner to assess baseline performance. During familiarization, each coordination direction (CW, CCW, IN and OUT) was performed in the 1:1 frequency ratio. The baseline measures were followed by 3 days of training: TR1, TR2 and TR3. While the first and second training sessions were on consecutive days, a rest day between TR2 and TR3 was implemented because of practical reasons. Training was performed in an MRI scanner on TR1 and TR3, whereas TR2 took place in a dummy scanner.

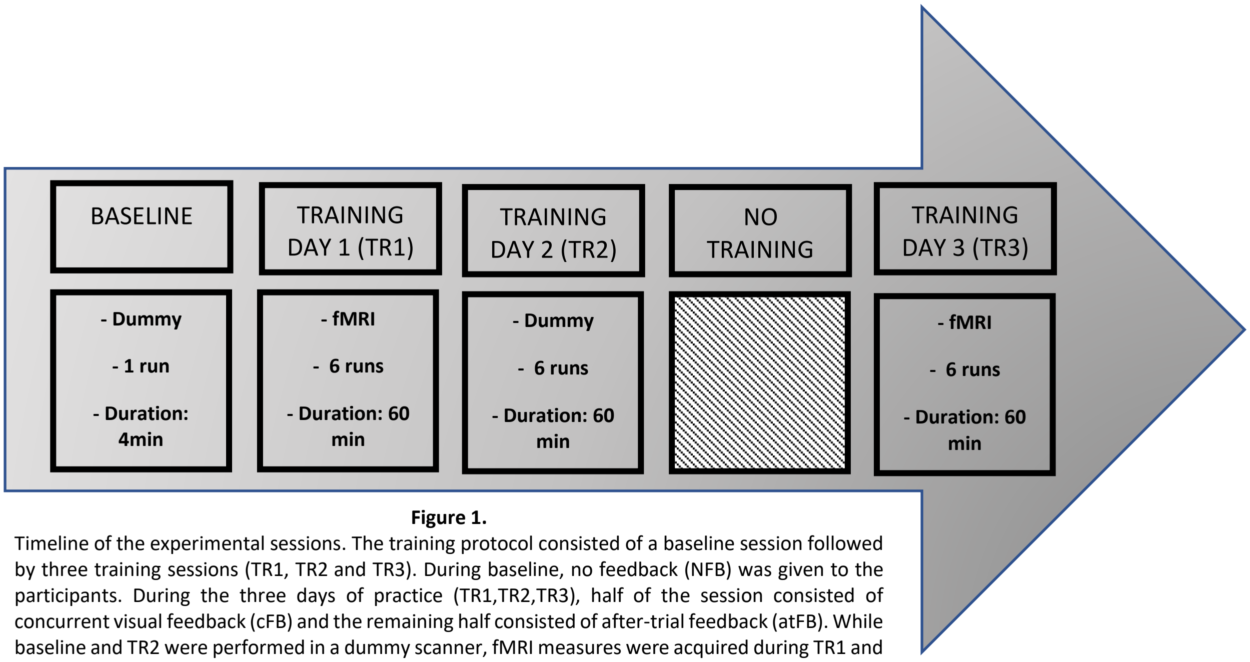

Figure 1.
Timeline of the experimental sessions. The training protocol consisted of a baseline session followed by three training sessions (TR1, TR2 and TR3). During baseline, no feedback (NFB) was given to the participants. During the three days of practice (TR1,TR2,TR3), half of the session consisted of concurrent visual feedback (cFB) and the remaining half consisted of after-trial feedback (atFB). While baseline and TR2 were performed in a dummy scanner, fMRI measures were acquired during TR1 and TR3.

Bimanual visuomotor tracking task and set-up

Task design

Subjects were positioned supine in the MRI or dummy scanner. A board with dials was placed over the legs of the participant and secured to the MRI table. To maximize comfort, the position of the elbow was adjusted to an angle of approximately 45° by moving the board forward or backwards (**Figure 2**). The subject's head was fixated by foam pillows to prevent head movements. An LCD projection on a double mirror was used to display the task. This mirror was placed in front of the subject. The task and instrumentation were adopted from the one used in previous studies done by our research group (Pauwels et al., 2018a; Pauwels, Swinnen, & Beets, 2014; Pauwels, Vancleef, Swinnen, & Beets, 2015).

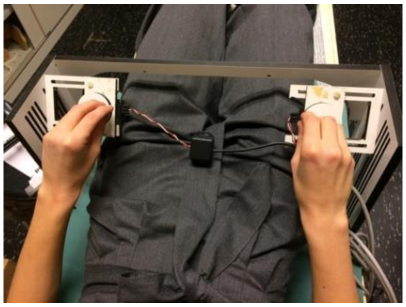

Figure 2.
The experimental set-up.

The task used was a visuomotor bimanual tracking task. The aim of this task was to follow a white target dot as it progressed over a blue target line. To this end, a board with two dials was used. Each dial consisted of a flat disc (5 cm diameter) with a vertical peg. Holding each peg between the thumb and index finger, the subject needed to rotate both dials simultaneously. The current position was indicated by a yellow cursor on the screen. The left dial corresponded with the vertical component of the yellow cursor: whereas clockwise rotation (CW) resulted in an upwards movement of the cursor, counterclockwise (CCW) rotation resulted in a downwards movement. The right dial, on the other hand, corresponded with the horizontal movement component. A CW corresponded with movement to the right and a CCW rotation corresponded with movement to the left. None of the conditions during the training protocol required inward nor outward rotation even though this was explained during familiarization. Three different task variants with a different degree of difficulty were presented. To perform the different task variants that could be distinguished based on the slope of the target line, both hands needed to move at a specific inter-hand frequency ratio. The 1:1 frequency ratio was visualized by a target line with a 45° slope. In this easy task variant, both hands needed to rotate dials at equal speed, i.e. isofrequency ratio. Adjacent to the 1:1 condition, two more complex conditions were included. That is, a 2:3 condition in which the right hand needed to rotate three times for every two rotations of the left hand and the 1:2 condition where the right hand needed to rotate the dial twice as the left hand made only a single rotation within the same time span. A schematic illustration of the different conditions is presented in **Figure 3**. Notably, participants only received information on how to move the cursor and were not informed on the presence of different frequency ratio's nor their required movement pattern.

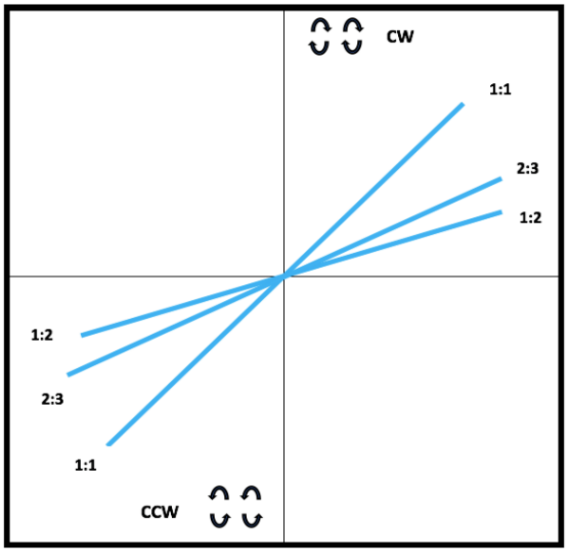

Figure 3.
Schematic image of all the possible movements during the bimanual visuomotor tracking task. The participant performed a combination of a coordination direction (CW,CCW) with a specific inter-hand frequency ratio (1:1,2:3,1:2).

Feedback conditions

To optimize learning, it was important that participants were not dependent on feedback. However, the incorporation of feedback during training was mandatory to enable learning of the task. Therefore, 3 different types of feedback were presented to the learner (Kovacs & Shea, 2011; Pauwels et al., 2018a, 2014, 2015); i.e. concurrent visual feedback (cFB), after-trial feedback (atFB) and no feedback (NFB). During the planning phase of movement (2 s), a yellow circle was presented in the middle of the screen as a cue indicating whether feedback would be provided or not. Subsequently, the white target dot, starting from the screen center, started moving along the blue target line (see 'bimanual visuomotor tracking task' for more detailed information). This 'execution phase' started when the yellow cue disappeared and lasted for 9 s. Finally, a black screen was presented in between two trials, i.e. 'intertrial interval', which lasted for 3 s. In the cFB condition, a yellow line corresponding with the participant's current position was presented during execution of the task whereupon the participant could correct his movement. In the atFB condition, feedback was given promptly after every trial during 1 s by displaying a motionless green and red line which represented the participants' performance. This line was used as an indication to show whether the relative velocity between both hands was correct (i.e. green line) or incorrect (i.e. red line). At last, in the NFB condition, no feedback was given during the execution phase nor after the task was completed. A clear overview is illustrated in **Figure 4**.

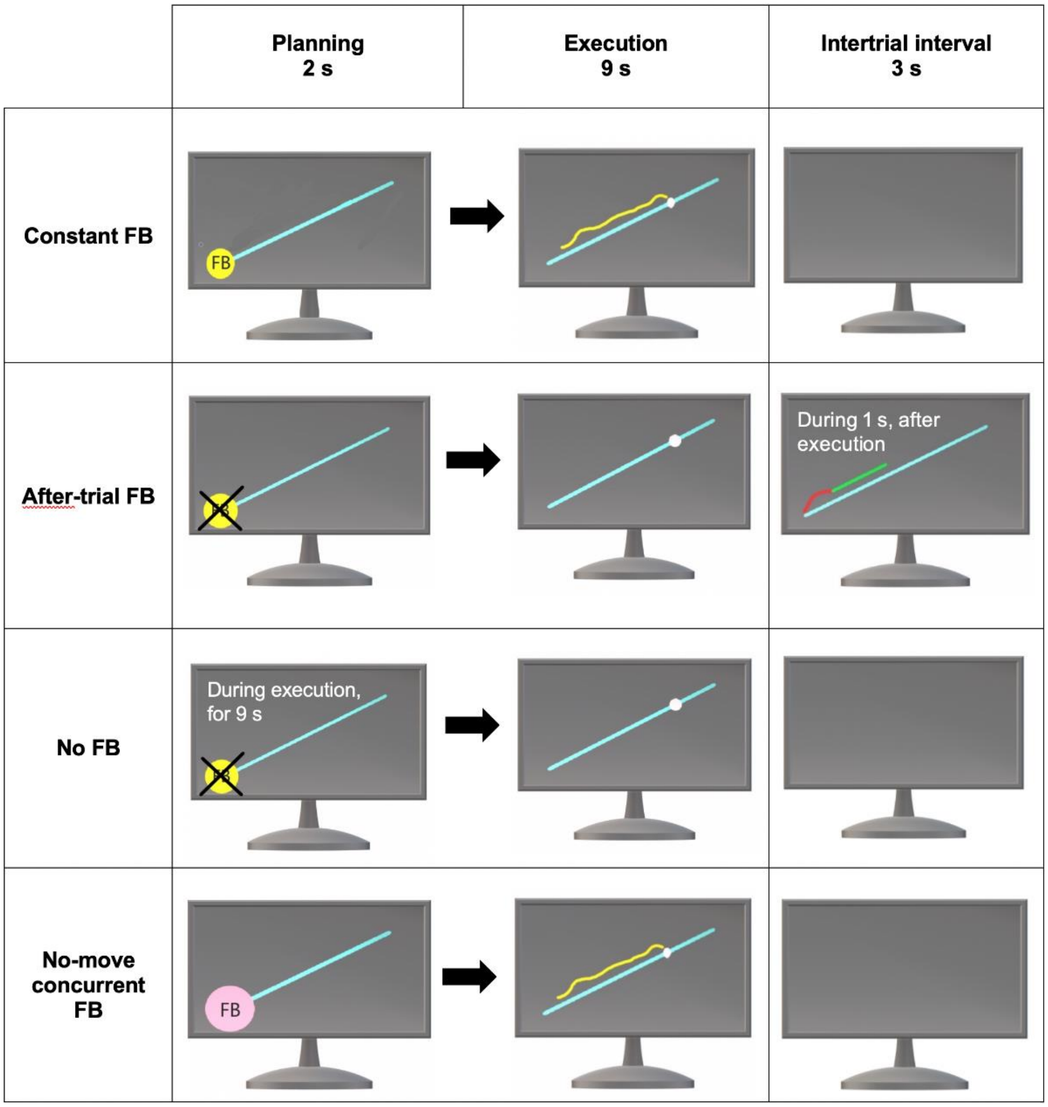

Figure 4.

Three different types of feedback were presented: concurrent visual feedback (cFB), after-trial feedback (atFB) and no feedback (NFB). During the planning phase (2 s), a yellow circle covered the target dot. A cross over the yellow circle indicated the absence of feedback during the upcoming trial. The 'execution phase' (9 s) started when the yellow cue disappeared. Subsequently, the white dot, starting from the centre of the screen, started moving along the blue target line. During the intertrial interval (3 s), a black screen was presented. In the cFB condition, a yellow line corresponding with the participants current position was presented during the execution phase. In the atFB condition, feedback was given promptly after every trial during 1 second by means of a motionless green and red line indicating whether the relative velocity between both hands was correct (green) or incorrect (red). No feedback was given during the NFB condition. Finally, the pink cue, either crossed or not, indicated the no-move trials. These trials were included to provide a baseline measure for the fMRI analysis.

Experimental outline

The behavioural outline is comparable with the behavioural design used by Pauwels et al. (Pauwels et al., 2018b).

To assess baseline performance, each subject performed four trials per frequency ratio, resulting in 12 NFB trials in total. The acquisition phase consisted of three days (TR1-TR2-TR3). Within one day, six runs with a one-minute break in between were performed. Each run contained 24 move trials and 12 no-move trials. These no-move trials, preceded by a pink cue rather than a yellow cue, were included to provide a baseline measure for fMRI analysis. The participants were exposed to a randomized practice schedule each day, i.e., the three different frequency ratios within the two coordination directions were implemented in each block. Each frequency ratio was practiced eight times per run. Half of the trials were performed with cFB and the other half with atFB (432 move trials in total). The atFB was included to maximize learning and minimize dependence on concurrent visual feedback. One practice day lasted approximately 60 minutes.

Functional magnetic resonance imaging procedure

MRI measurements were performed using a Philips Achieva 3-T magnetic resonance scanner (Philips Healthcare) with a 32-channel head coil (**Figure 5**). First, a three-dimensional T1-weighted image [magnetization prepared rapid gradient echo; time repetition/time echo (TR/TE), 9.6 ms/4.6 ms; 0.98 X 0.98 X 1.2 mm voxel size; field of view, 192 X 250 X 250; 160 coronal slices; scan time, ∼7 min] was acquired as a template for the image registration of the fMRI runs. A field map was included to address local distortions. Subsequent task-related fMRI runs comprised of 41 ascending gradient echo planar images (EPIs) for T2-weighted functional images (TR/TE, 3000 ms/30 ms; flip angle, 90°; 54 parallel axial slices with a slice thickness of 2.5 mm; interslice gap, 0.2 mm; in-plane resolution, 2.5 X 2.5 mm; 82 X 84 matrix). fMRI images were taken on TR1 and TR3 and consisted of 6 runs for each participant per day. One fMRI run lasted for 10 minutes and consisted of 24 move and 12 non-move trials.

Figure 5.
Philips Achieva 3-T magnetic resonance imaging scanner.

Data processing and statistical analysis

Behavioural data

Dependent measures

Labview (version 8.5) software was used to record behavioural data. The x and y positions of the target dot and the subjects' cursor were sampled at 100Hz and Matlab (R2014b) and Microsoft Excel 2013 were used to perform off-line analysis. Behavioural performance was assessed based on the average track deviation (ATrD) of the bimanual visuomotor task. This track deviation was estimated using the Euclidian distance between the target line (blue) and the cursor position at each moment for each trial (**Figure 6**). Ultimately, the average across all time points within each trial was calculated. The result was interpreted as followed: The lower the ATrD, the higher the performance accuracy.

Figure 6.
The average track deviation (ATrD) of the bimanual visuomotor task was estimated using the Euclidian distance between the target line (blue) and the cursor position at each moment for each trial. Ultimately, the average of all track deviations within each trial was calculated.

To acquire simplification, CW and CCW data were collapsed by virtue of the fact that different coordination directions (CW and CCW) were merely to provide an extra dimension of complexity to the task. Secondly, data were averaged across every set of three data points in time whereby 18 data points were generated (DT1, DT2, …., DT18), i.e. 6 per day of training. Moreover, to investigate the influence of task complexity, frequency ratios were divided into two task variants, i.e. an easy isofrequency (1:1) and a more difficult non-isofrequency (1:2 and 2:3) task variant. Additionally, decoupling scores were calculated by subtracting performance on the isofrequency task variant of performance on the non-isofrequency task variant. These scores represent to which extent a subject can cope with more difficult ratios in comparison with the easier ratio (1:1). To facilitate the interpretability, the absolute error measurement (ATrD) were log-transformed (base 10 logarithm).

Statistical analysis

SPSS (version 25) was used for statistical analysis. The critical probability level was set at $p < 0.05$, two sided and was applied to all analyses. Before the conduction of the analyses, the distribution of the standardized residuals was checked on normality. Initially, a paired samples t-test was carried out at baseline to examine whether the two task complexities resulted in different error scores. To investigate behavioural performance, a 18 X 2 [Time (DT1-18) X Task complexity (isofrequency (1:1), non-isofrequency

(2:3 and 1:2))] repeated-measures ANOVA was performed with Time and Task complexity as within-subject factors. Additionally, the decoupling scores were analyzed using a repeated-measures ANOVA with Time as the within-subject factor. In case of significant effects, post hoc analyses were performed using the Bonferroni correction.

Functional image processing and analysis

Neuroimaging analyses were conducted using the FMRIB Software Library (FSL 5.0).

Registration/pre-processing

The extraction of the brain from the dura and skull was performed with the Brain Extraction Tool on the T1 and field images. Preprocessing of the collected fMRI data was conducted using FSL's FMRI Expert Analysis Tool (FEAT). EPI's were realigned to the middle volume of each run by applying MCFLIRT motion correction, and a high-pass filter with a cutoff of 200 s was used. Afterwards, the B0 field map unwarping was carried out to correct geometric distortions. In addition, slice timing correction together with spatial smoothing (full-width half-maximum of 5 mm) was applied. The FNIRT registration tool was used to co-register images (EPIs) with the T1 image. This revealed which brain areas of the T1-weighted images corresponded with the activated regions of the fMRI images. This registration was carried out for each participant and each run (i.e. every day 6 registrations per participant) separately. Afterwards, this result was normalized to the standard Montreal Neurological Institute (MNI) template.

First level

The first temporal derivatives and regressors of the conditions of interest (move versus no-move) for the execution (9 s) phase of each condition were determined. Motion-induced signal variations in fMRI data were eliminated by means of ICA-based automatic removal of motion artifacts (ICA-AROMA) whereby the temporal characteristics were preserved (Sokil, Lyashuk, & Dovbush, 2016). Furthermore, additional confound explanatory variables, i.e. time series from the white matter and CSF masks, were extracted in FEAT.

Higher level

Higher level random-effects model of FSL was included to determine the coordinates of the peak activation voxel for left PMd (seed region). Firstly, we looked at differences in brain activity between younger and older adults during task performance contrasting the move to the no-move conditions. Although PMd will demonstrate activity in both older and younger adults, it is known that younger adults show a greater activity (Kiyama et al., 2014; Stewart, Tran, & Cramer, 2014). The seed region will thus correspond to the peak voxel of this 'PMd cluster'. This method of determining the seed region by comparing between young and old was initially used to explore age differences in brain activation, which is beyond the scope of this study.

Psychophysiological interaction (PPI) analysis

To explore task-related functional neural connectivity in the context of a bimanual coordination task using left PMd as a predefined seed and to investigate the changes within the connectivity during training, psychophysiological interaction (PPI) analyses were performed (Gitelman, Penny, Ashburner, & Friston, 2003). This analysis demonstrates the connectivity between one brain region, i.e. seed region, and other brain areas, called the target regions, in view of an altered psychological context.

A PPI consists of three variables, i.e. a psychological variable, a physiological variable and an interaction term of the aforementioned variables. Here, task complexity was included as the psychological variable in which distinction was made between the easy isofrequency ratio and the more complex non-isofrequency ratios (i.e. frequency ratios 2:3 and 1:2).

The physiological variable consisted of the blood oxygen level dependent (BOLD) signal time series within a sphere with a 5 mm radius around the seed region (left PMd). The PPI interaction term of interest (third variable) is the product of the two previous regressors, i.e. how the physiological variable changes when the psychological variable changes. Specifically, changes in BOLD signal dependent on changes within task complexity were investigated.

Lastly, the effect of task complexity on the functional connectivity within the first, as well as the last day of training and how this connectivity changes over time (Task complexity x Time interaction) were investigated. Therefore, a higher-level group analyses using the random-effects model of FSL was implemented. The overall training effect (TR1-TR3) was investigated by collapsing trials across the 18 runs of training. In order to integrate exclusively gray matter voxels, a grey matter mask was overlaid. During all fMRI analyses, Gaussian Random Field Theory was utilized at a cluster probability threshold of $p < 0.05$ and the cluster level using $Z > 2.3$. The ''Harvard-Oxford cortical structural atlas", the "Harvard-Oxford subcortical structural atlas", the "Juelich histological atlas" and the "Cerebellar atlas in MNI152 space after normalization with FNIRT" were used to determine which brain regions corresponded with the resulting significant clusters. Both the peak of each cluster and the possible local maxima (subclusters) were identified.

Results

Behavioural results

Baseline

The paired samples t-test revealed a difference in baseline performance between the two different levels of task complexity (i.e. isofrequency versus non-isofrequency ratio), indicating higher error scores in the non-isofrequency as compared to the isofrequency task variant (**Table 1**).

Table 1. Paired Samples T-test for the participants' baseline data in the two different task variants

All Subjects	**N**	**Mean**	**SD**	**t**	**df**	***p* value**
Isofrequency	16	0,1784	0,34492	-9,403	15	<0,001
Non-isofrequency	16	1,0981	0,09583			

Training

Prior to the statistical analysis, the assessment of normality was performed on the standardized residuals. With respect to the acquisition phase, the 2 X 18 (Task complexity X Time) repeated-measures ANOVA revealed a significant main effect of Task complexity. This result implied that the overall performance level was greater in the easy isofrequency task variant in comparison with the more complex non-isofrequency task variant. Additionally, the repeated-measures ANOVA revealed a significant main effect of Time, indicating that the error rate decreased over the course of practice. Furthermore, the analyses exposed a significant Task complexity X Time interaction. Post hoc analyses using the Bonferroni correction revealed that execution of the non-isofrequency task variant resulted in a greater behavioural improvement across training for the non-isofrequency as compared to the isofrequency task variant, especially during the first training session (p<0,05). A plot of the log-transformed average track deviation results can be found in **Figure 7** and a detailed overview can be found in **Table 2**.

Table 2. Behavioural results

	df	*F*	*p*
Acquisition phase			
ATrDlog scores			
Task complexity	1	38,076	<0,001
Time	17	20,033	<0,001
Task complexity X Time	17	2,792	<0,001
Decoupling scores			
Time	17	2,792	<0,001

In addition to these analyses, a one-way repeated-measures ANOVA containing the decoupling scores was performed with Time as the within-subject factor. A significant effect of Time was observed, indicating that the overall capability of participants to cope with increasing task difficulty improved during training.

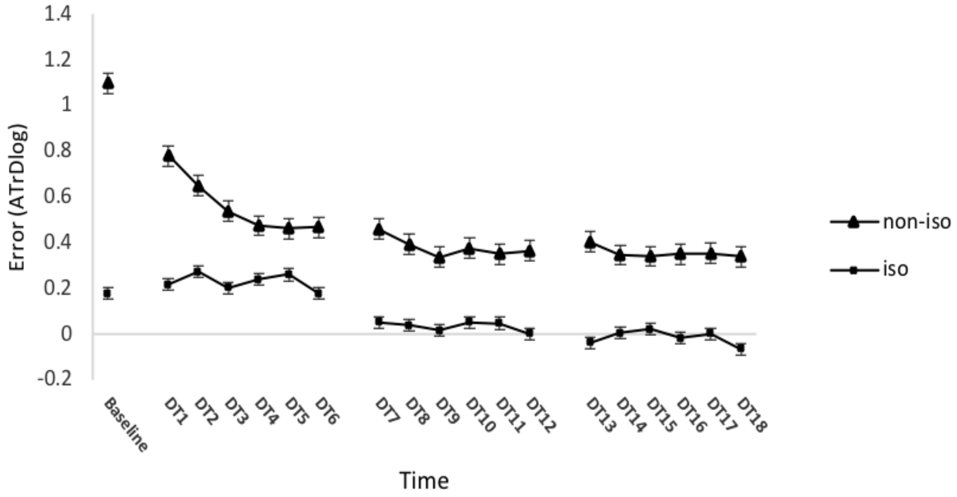

Figure 7. Behavioural results.

The error score (ATrDlog, i.e. the log transformed average track deviation) is presented over time for the isofrequency and non-isofrequency task variant [baseline and acquisition phase (DT1-DT18)]. Better execution is indicated by lower errors (ATrDlog) score. The performance level of the non-isofrequency variant was significantly lower in comparison with the isofrequency variant during baseline (p<0,001) and the acquisition phase (p<0,001). More decrease in error throughout the training of the non-isofrequency task variant was observed in comparison with the isofrequency task variant, especially during the first training session (p<0,05).

fMRI Results

Using the fMRI data obtained during task execution on the first and last day of training, psychophysiological interaction analyses were performed. These analyses provide an overview of brain areas demonstrating altered PMd-seeded connectivity when contrasting the non-isofrequency to the isofrequency task variant. First, the influence of Task complexity on PMd-seeded connectivity was investigated for TR1 and TR3 separately (Design 1). Results of these analyses are listed in **Table 3**. Second, the influence of Time on connectivity patterns was considered. Specifically, The Time X Task complexity interaction indicated which brain regions demonstrated a change in connectivity with the left PMd during the non-isofrequency in comparison with the isofrequency task variant across training, i.e. from TR1 to TR3. See **Table 4** and **Figure 8** for an overview of the results of Design 2.

Design 1: Main effect of Task complexity during the acquisition phase

Training session 1

Investigating the effect of Task complexity on PMd-seeded connectivity within the first day of training (TR1), no significant clusters were revealed. A more thorough investigation of the data revealed that, although not significant, functional connectivity between left PMd and vermis, paracingulate gyrus, left visual cortex, left cingulate gyrus, right hippocampus, right cerebral crus, right insular cortex and right primary auditory cortex were positively correlated with Task complexity, i.e. higher connectivity in the complex (non-isofrequency) as compared to the easier (isofrequency) task variant (see Supplementary Materials **Table S1**).

Training session 3

During the late phase of learning (TR3), the non-isofrequency as compared to the isofrequency task variant induced more connectivity between left PMd and left M1, extending to the left PMC, right M1 and the bilateral somatosensory cortex (S1).

Table 3. Design1: Main effect of Task complexity during the acquisition phase

Anatomical/functional region	MNI coordinates				
	X	Y	Z	Z score	*p* value
Non-iso TR1 > iso TR1	No significant clusters (see Supplementary Materials)				
Non-iso TR3 > iso TR3					
L M1, extending to L PMC, R M1 and bilateral somatosensory cortex	-30	-28	62	4.14	<0.001

MNI coordinates of the main clusters and Z scores for brain areas are presented showing a main effect of Task complexity (Z>2.3;cluster significance <0.05, corrected). Non-iso, non-isofrequency task variant; iso, isofrequency task variant; TR1, training day 1; TR3, training day 3; L, Left; R, Right; M1, primary motor cortex; PMC, premotor cortex.

Design 2: Task complexity X Time during the acquisition phase

Results of Design 2 revealed a significant Time X Task complexity interaction, indicating a decrease in connectivity between left PMd and left occipital pole and bilateral occipital cortex (for extending regions, see **Table 4)** from early learning (TR1) to late learning (TR3) in the non-isofrequency task variant as compared to the isofrequency task variant. Conversely, connectivity between left PMd and cerebellum, left M1, right PMC ad right parietal operculum cortex was higher during the late (TR3) as compared to the early (TR1) phase of learning.

Table 4. Design2: Time X Task complexity during the acquisition phase

Anatomical/functional region	MNI coordinates				
	X	Y	Z	Z score	*p* value
Non-iso (TR1>TR3) > iso (TR1>TR3)					
L occipital pole, extending to L occipital cortex inferior	-32	-96	-12	4.14	<0.001
L lateral occipital cortex superior division	-24	-72	34	3.67	0.016
R lateral occipital cortex inferior division, extending to R occipital pole	48	-80	2	4.81	<0.001
Non-iso (TR3>TR1) > iso (TR3>TR1)					
R cerebellar vermis, extending to L cerebellar vermis, bilateral anterior and left posterior cerebellar lobe	4	-70	-34	3.99	<0.001
L M1 (BA4p), extending to L S1 and L PMC	-34	-26	56	4.63	<0.001
R PMC (BA6), extending to R M1 and L PMC	0	-6	60	4.09	<0.001
R parietal operculum cortex, extending to the whole parietal operculum cortex (covering the insula)	42	-30	22	3.75	0.027

MNI coordinates of the main clusters and Z scores for brain areas are presented showing a significant Task complexity X Time interaction effect from DAY1 to DAY3 acquisition phase (Z>2.3;cluster significance: p<0.05, corrected). Non-iso, non-isofrequency task variant; iso, isofrequency task variant; TR1, training day 1; TR3, training day 3; L, Left; R, Right; M1, primary motor cortex; S1, primary somatosensory cortex; PMC, premotor cortex.

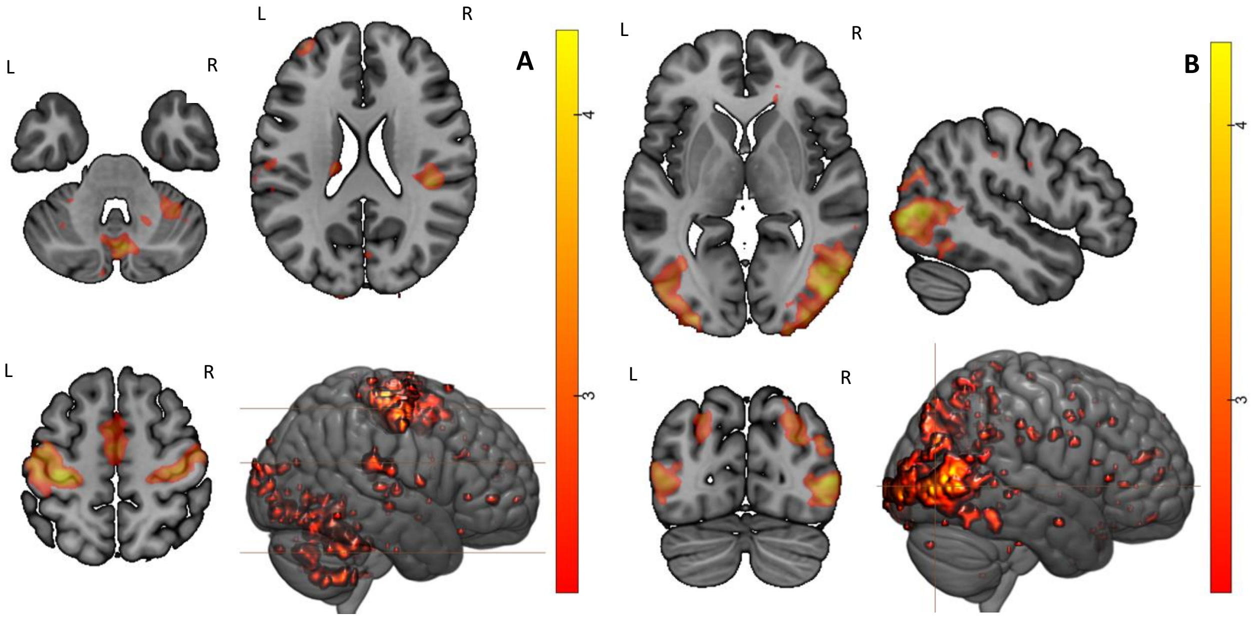

Figure 8. Time X Task complexity during the acquisition phase.

Task complexity X Time effect on functional connectivity during the third day of training is visualized using MRIcroGL by overlaying activation maps on a mni152 template. The color map illustrates Z values (Z-values between 2.3 and 4.3). **A.** The vermis, right premotor cortex BA6 and left primary motor cortex BA4p show a significant stronger connectivity with the seed region, left PMd, from TR1 to TR3 in the non-isofrequency in comparison with the isofrequency task variant. **B.** The left occipital pole and the bilateral occipital cortex show a significant lower connectivity with left PMd, from TR1 to TR3 in the non-isofrequency in comparison with the isofrequency task variant.

Discussion

This study explored the influence of task complexity on functional connectivity during bimanual coordination training using TB-fMRI. We made a comparison between an easy isofrequency task variant and a more complex non-isofrequency task variant and found that the latter one resulted in larger improvements relative to baseline after the three-day training. Additionally, the analysis of the decoupling scores demonstrated that participants were more able to cope with increasing task complexity by the end of acquisition. In line with our hypothesis, differences in task-based functional connectivity throughout practice were found, depending on the two different task variants. Observing the modifications in connectivity through time (TR1>TR3 or TR3>TR1), the non-isofrequency as compared to the isofrequency task variant induced lower connectivity between left PMd and visual processing regions during the last day as compared to the first day of training. Furthermore, stronger connections were found between left PMd and sensorimotor-related brain regions as training progressed.

Behavioural performance was differently influenced by task complexity

Baseline results are in line with previous literature indicating more accurate performance on the isofrequency as compared to the more complex non-isofrequency task variant (deGuzman & Kelso, 1991; Rueda-Delgado et al., 2017; Sisti et al., 2011b; Swinnen, Dounskaia, Walter, & Serrien, 1997). Our findings that the more complex practice variant resulted in greater improvements are in line with the findings of Sisti et al. (2011a), who found a significant Day X Frequency ratio interaction effect, meaning that task variants initially inducing greater error rates, eventually led to greater improvements. The different influence of task complexity on performance during different levels of task complexity was already investigated by Guadagnoll et al. (2004). They developed the challenge point framework, describing the effect of practice variables on motor learning. The optimal challenge point, where the potential learning benefit is maximal, is dependent on the extent of task difficulty and the skill level of the performer. This potential learning benefit is provided by an increasing task difficulty, reaching its optimal challenge point, co-occurring with an increased magnitude of information to be learned (Guadagnoll & Lee, 2004). The impact of task difficulty on reaching the optimal challenge point can thus be used to optimize motor learning processes.

Functional connectivity differed between the more complex task variant and the easier task variant

In comparison with the isofrequency task variant, training session 3 of the non-isofrequency task variant resulted in higher connectivity between left PMd and brain regions that are part of the motor association cortex (i.e. left M1 with the left PMC, right M1 and bilateral S1 as extending regions). This is in line with

Kiyama et al. (2014), who described that strong interhemispheric connections between these regions allow for complex bimanual finger movements. Because of the crucial role of PMd in adaptability to spatiotemporal task complexity (F Debaere et al., 2004) as well as its substantial influence on descending motor control (Dum & Strick, 2005), the higher connectivity between left PMd and left M1 in the more complex as compared to the easier task variant seems plausible.

Remarkably, results indicated that during the first day of training, PMd-seeded functional connectivity did not significantly differ between the easy (i.e. isofrequency) as compared to the more complex (i.e. non-isofrequency) task variant. A possible explanation could be that the participants were not yet able to make a distinction between the different task variants at training session 1, as they only received information on how to move the cursor and were not informed on the presence of different frequency ratios nor their required movement pattern. The more pronounced difference in connectivity patterns between the non-isofrequency and isofrequency task variant during training session three could be due to the fact that subjects were more capable to differentiate between task variants.

Training-induced shift in connectivity pattern

Training resulted in decreased connectivity with visual processing regions (TR1>TR3)

Our findings indicated a training-induced decrease in connectivity between left PMd and visual processing regions (i.e. bilateral lateral occipital cortex (LOC) and bilateral occipital pole) in the non-isofrequency as compared to the isofrequency task variant. The LOC plays a specific role in visual processing, namely the unification of several aspects of visual stimuli (Baars & Gage, 2010). Conscious visual processing of visual feedback, recovered by visual processing centres, forms a crucial part of motor control as well as of motor learning (Adams, 1971). This is harmonious with Thorndike's law of effect that states that reinforced actions will become reproduced and penalized actions will be discarded (Thorndike, 1933). People learn through feedback of their errors (i.e. error driven learning): by comparing the observed outcome with the desired movement, we attempt to adjust our future behaviour. This is where our vision is of crucial importance enabling us to see our target (desired outcome) and the actual outcome (Krigolson, Cheng, & Binsted, 2015). Volitional movement preparation represents one of the functions of PMd, with PMd playing a predominant role in action selection and preparation (Bestmann et al., 2008). It is therefore likely that the aforementioned regions process the visual feedback from the visuomotor task, and communicate with left PMd in order that the latter can select the desired action. Hardwick et al. (2015) already acknowledged a functional connection between PMd and brain regions engaged in visuomotor control and executive function enabling PMd to connect motor control and cognition. We noticed that the connectivity between left PMd and the visual processing centres was higher during the non-isofrequency task variant in comparison with the isofrequency variant at TR1. This stronger connectivity throughout the execution of the more complex task variant implies a reduced need for visual feedback when participants performed the easier task variant. This is consistent with the study of Pauwels et al. (2018b), that illustrates the crucial involvement of visual processing centres during random practice. To summarize, results of the present study point towards a higher involvement of visual processing areas in the early- as compared to the late stages of learning, suggesting that processing of visual information might be particularly relevant during early learning.

It is noteworthy that most studies present results of finger-sequence learning tasks (Karim Helmet T , Huppert Theodore J, Erickson Kirk I, Wollam Mariegold E, SpartoPatrick J, 2015; Kornysheva & Diedrichsen, 2014; Kuriyama et al., 2013; Ma et al., 2010; Meister et al., 2005; Schubotz & Von Cramon, 2002; Sun et al., 2007), whereas this study used a bimanual visuomotor task. The bimanual tracking task (BTT) used in our study has already been utilized in earlier motor learning studies done by our research group (Monteiro, King, Zivari Adab, Mantini, & Swinnen, 2019; Pauwels et al., 2018a, 2014; Woolley et al., 2018). As mentioned before, Pauwels et al. (2018a) found an enhanced activity in the visual processing areas during random practice as well using the same task. Examining resting state connectivity following practice of the BTT, with PMd as one of the ROI, Solesio-Jofre et al. (2018) found an augmented connectivity in young adults, in both inter- and intra-hemispheric motor network associated connections, due to short-term practice. Additionally, they investigated long-term changes after training and observed a stronger RS-FC within the right hemisphere confirming the importance of right intra-hemispheric connectivity in bimanual motor skill learning (Solesio-Jofre et al., 2018). Monteiro et al. (2019) investigated differences in network flexibility and segregations between younger and older adults. They found a training-induced general pattern of reduced cortical functional inter-network connectivity together with an increased cerebellar/subcortical task related functional connectivity in both groups although this was not significant. Remarkably, changes in connectivity strength as a result of training were not established (Monteiro et al., 2019). Our study using the bimanual visuomotor task extends the knowledge about learning-related changes in PMd-seeded connectivity as we additionally used TB-fMRI and integrated the influence of task complexity.

Training resulted in increased connectivity with sensorimotor related regions (TR3>TR1)

In the non-isofrequency as compared to the isofrequency task variants, connectivity between left PMd and typical sensorimotor-related brain areas (i.e. left M1, parietal operculum cortex, SMA, and cerebellum) increased from TR1 to TR3. Ma et al. (2010) investigated inter-regional connectivity changes between M1, SMA, PMd, basal ganglia, cerebellum and posterior ventrolateral prefrontal cortex and found a general increase in intensity with continued training. They suggest a learning-induced reorganization of the motor network indicated by an augmented inter-regional connectivity strength (Ma et al., 2010). As physiological connections between PMd and M1 were shown (Dum & Strick, 2005), this could declare the increased connectivity between PMd and M1 on TR3 as compared to TR1 found in our study.

Noteworthy, studies show that an increase in movement frequency leads to an increased activity in M1 (Blinkenberg, Bonde, Paulson, Svarer, & Law, 1995; F Debaere et al., 2004; Lutz Jäncke et al., 1998; Jenkins, Passingham, & Brooks, 1997; Kawashima et al., 1999; Norihiro Sadato et al., 1997; Wexler et al., 1997). As we aimed to investigate the influence of task complexity on functional connectivity, this movement-induced increase in activity could interfere with our results.

Practice of the non-isofrequency task variant induced more connectivity from TR1 to TR3 between left PMd and the parietal operculum. The latter one participates in macroscopic tactile sensation and manipulation (Bassetti, Bogousslavsky, & Regli, 1993; Ruben et al., 2001) and is furthermore involved in processing somatosensory information of stimuli (noxious as well as non-noxious) (Krubitzer, Clarey, Tweedale, Elston, & Calford, 1995; Mazzola, Faillenot, Barral, Mauguière, & Peyron, 2012). Moreover, it is suggested to play a role in the coordination of movements providing proprioceptive feedback (L Jäncke, Kleinschmidt, Mirzazade, Shah, & Freund, 2001; Maule, Barchiesi, Brochier, & Cattaneo, 2015). These processes may be crucial to learn a task, by converting the sensory stimuli into a motor action. As the PMd performs an important role in higher-order cognitive processes (Meister et al., 2005), we hypothesize that the PMd exchanges information with the parietal operculum to encourage learning by facilitating the execution of the motor movement. This processing of sensory information is particularly relevant in the non-isofrequency task variant as participants need to decouple the rotational speed of their left and right hand.

Various studies indicate the important role of SMA in bimanual coordination (Donchin et al., 2002; Duque et al., 2010; Immisch, Waldvogel, van Gelderen, & Hallett, 2001; Kermadi, Liu, Tempini, Calciati, & Rouiller, 1998; N Sadato, Yonekura, Waki, Yamada, & Ishii, 1997; Stephan et al., 1999; Toyokura, Muro, Komiya, & Obara, 1999; Uhl et al., 1996; Ullén, Forssberg, & Ehrsson, 2003) as well as in the communication between both hemispheres in obtaining a proper cooperation between both hands. This proper coordination is of greater importance during the more complex task variant in comparison with the easier one as both hands make the exact same movement during this task variant (F Debaere et al., 2004; Holst, 1973; Swinnen & Wenderoth, 2004). More important, besides its well-known role in motor sequence learning, SMA plays a role in the learning of sensorimotor tasks (including visuomotor tasks) (Hardwick et al., 2013). Previous research suggested that SMA possibly communicates with PMd to retrieve stored movement sequences and to initiate, alter and likely inhibit actions (Hardwick et al., 2015). As this sequence learning task study indicated a greater involvement of SMA in learned motor tasks, we speculate that this phenomenon is applicable to our visuomotor task. Altogether, we propose that the higher left PMd-SMA connectivity in the more complex task variant in comparison with the easier task variant after training (TR3>TR1) occurred because SMA is more involved in more complex, learned tasks such as the non-isofrequency task variant during the late stages of learning.

In contrast with practice during the first training session, the third training session induced more connectivity between the left PMd and cerebellum in the non-isofrequency as compared to the isofrequency task variants. The cerebellum is acknowledged as a region involved in the implicit mastering of a visuomotor task (Doyon, Owen, Petrides, Sziklas, & Evans, 1996; Doyon, Penhune, & Ungerleider, 2002). It compares the intended movement with the actual performance and can modify the movement during execution, called adaptation. Additionally, the cerebellum predicts the sensory consequences of movements. Therefore, it enforces a ‘forward model’ of the motor apparatus (Hikosaka, Nakamura, Sakai, & Nakahara, 2002; Krakauer & Mazzoni, 2011; Penhune & Steele, 2012; Shadmehr & Krakauer, 2008). In our study, mainly the vermis showed modulations under the influence of task complexity. This can be explained by the subdivision of the cerebellum where the anterior and medial parts of the cerebellum are involved in movement execution, assisting the M1 in carrying out bimanual motor tasks (Brooks & Thach, 2011; F Debaere et al., 2004). Our results are consistent with the findings of Debaere et al. (2004), who stated that PMd and the cerebellum demonstrate an apparent interaction during the manipulation of spatiotemporal complexity and cycling frequency. Furthermore, Ma et al. (2010) investigated changes in inter-regional connectivity during 4 weeks of motor learning. Consistent with results of the present study, they found that the path between PMd and the cerebellum strengthened during training (Ma et al., 2010). The authors suggest that the effective connectivity between PMd to the cerebellum might be indirect, via projections through the thalamus, as nor human or primate studies provide anatomical support for a direct connection (Ma et al., 2010; Matelli & Luppino, 1996; Matelli, Luppino, Fogassi, & Rizzolatti, 1989).

Overall, the results of the of Time X Task complexity interaction reveal an increase in connectivity with the sensorimotor-related regions during late learning versus early learning in the more complex task variant as compared to the easier task variant. We suggest that when one is more skilled, sensorimotor-related brain regions are more important. To summarize, we speculated that a more learned task needs less visual processing and more sensorimotor connectivity with left PMd.

Lastly, one could speculate that the behavioural improvements on the bimanual task are positively associated with increases in the inter- and intrahemispheric connectivity between left PMd and other significant regions of the learning network. Although a direct link between functional connectivity and behavioural performance is often suggested (Ma et al., 2010), future research should aim to invest the impact of training-induced modulations of functional connectivity on performance.

Conclusion

To the best of our knowledge, we are the first to investigate the influence of task complexity in motor learning on functional connectivity by means of TB-fMRI, rendering this study innovative in the field of motor learning. The present study demonstrated greater behavioural improvements in the more complex non-isofrequency task variant as compared to the easier isofrequency task variant. Additionally, from the early to the late phase of learning, connectivity between left PMd and visual processing regions decreased, whereas connectivity between left PMd and sensorimotor-related brain regions increased in a complex as compared to an easier bimanual task variant. These results corroborate the involvement of left PMd during the execution of complex tasks as well as during motor learning

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