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Monica Gonçalves-Garcia
Candidate

Psychology
Department

This thesis is approved, and it is acceptable in quality and form for publication:

Approved by the Thesis Committee:

Dr. Derek Hamilton, Chairperson

Dr. Marisa Silveri

Dr. Benjamin Clark

Dr. Jeremy Hogeveen

Dr. Vince Clark

Neurobiology of Human Navigation Strategies in the virtual Morris Water Task

by

Monica Gonçalves-Garcia

B.M., University of New Mexico, 2019

THESIS
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ABSTRACT

Strategies of navigation is a topic that has been investigated for decades and is still not well-understood. Organisms learn to navigate by using self-generated cues, distal cues, and proximal cues, however, how the different frames of reference are interpreted by different areas of the brain and translated into behavior is not clear. Animal studies have provided evidence for a preference for navigation by following a direction in the environment over place learning. This study investigated the performance of adolescents (mean age: 13.89) in a virtual version of the Morris Water Task with a probe trial manipulation attempting to categorize people based on a strategy of navigation preferences. Analysis of behavioral performance revealed a preference for directional responding (following a direction in the room/ using the apparatus reference frame) which is consistent with animal reports. BOLD activation showed greater activation of the left precuneus in the group with a preference for directional responding compared to those who did not show a systematic strategy for navigation. Collectively, these findings provide evidence for the development of different strategies of navigation during learning trials and the BOLD activation differences show the involvement of the network supporting navigation that includes the hippocampal formation and areas outside of the hippocampus, particularly the involvement of the precuneus in a type of navigation that involves orientation and distance estimation.

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Introduction

Navigation involves the ability to move from one point to another in the environment. Different types of navigation can be controlled by a diverse set of stimuli such as allothetic (e.g., landmark information) and idiothetic (e.g., dead reckoning which involves self-motion information) cues, by the degree of task complexity (e.g., cued-navigation vs. sequence), and by a diverse set of cognitive processes (Redish & Touretzky, 1997; Taube et al., 2013). Two types of navigation controlled by exteroceptive visual stimuli are place learning and directional responding. Tolman (1946) defined place learning as a type of spatial learning that required the formation of an internal representation of the external environment – termed a *cognitive map* – that encodes the relationships between the elements of the environment independent of the observer. In his experiments, he noted spatial (place) learning as more predominant over the type of learning that required producing motor responses, leading to the conclusion that animals have a place learning disposition. Blodgett (1949) replicated Tolman's experiments including additional maze manipulations. The results provided evidence that animals solve the tasks by directional responding – orientation toward a specific direction based on visual information- rather than true place navigation – navigating to a specific place. Both types of navigation could be acquired by the organism and could be expressed differentially according to the situation. In manipulations of spatial tasks, such as the Morris Water Task, animals tend to navigate toward a direction rather than to a specific place in the environment. The underlying neurobiological mechanisms involved in spatial behaviors were enlightened by the discovery of place cells – cells with localized firing patterns - in the hippocampus which became a centerpiece of the investigation of spatial navigation learning and memory (O'Keefe and Nadel, 1978). O'Keefe and Nadel suggested that the hippocampus was the site of storage of cognitive maps. In the subsequent years, an increased interest in studying the linkage between the hippocampus and spatial navigation processes led to the discovery of other spatial cell types associated with navigation and orientation as well as structures beyond the hippocampus that formed a network that contributes to encoding spatial information (Redish & Touretzky, 1997; Spiers, 2020) such as the entorhinal cortex and the subiculum that contain grid cells and "border" cells, retrosplenial cortex which is interconnected with the hippocampal formation and the parietal cortex (contains place cells, HD cells, and other spatial cell types) (Bermudez-Contreras et al., 2020; Burgess et al., 2002; Taube et al., 2013).

In the last century, a large body of studies investigated navigation processes as a model system approach for understanding the neurobiology of learning and memory. Even though studies of the hippocampus and surrounding structures – or homolog structures in different species of animals - have provided robust evidence to support navigation, our understanding of the neurobiological processes supporting navigation, what is learned during spatial navigation tasks, and which strategies are involved in solving spatial navigational problems remain yet to be completed.

Cognitive Mapping theory

The way organisms navigate and orient themselves in the spatial environment has been extensively studied for decades (Hamilton et al., 2007). Later studies have investigated the organisms' predisposition to navigate based on directional responding and place navigation, and the strategies involved in either type of navigation (Hamilton et al., 2007; Hamilton et al., 2008; Skinner et al., 2003).

Following Tolman's seminal paper Cognitive Maps in Rats and Men (1948), many theories have emerged attempting to explain how organisms navigate and which brain regions are required and engaged during navigation (Bennett, 1996). Tolman postulated that navigation was achieved via the establishment of a cognitive map of the environment in the rat's brain that would provide an indication of routes and paths as well as environmental relationships. Those elements combined result in the behavioral response produced by the animal (Tolman, 1948). Tolman suggested the existence of two cognitive mapping systems: a strip-like, or narrow system, and a comprehensive-like, or broad system. Although both systems could lead the animal to the goal location, the latter would provide more information for the animal to successfully navigate in a novel environment such as taking short-cuts or supporting other flexible behaviors. Expanding on the cognitive mapping theory, O'Keefe & Nadel (1978), mentioned the possible innate ability of organisms to build three-dimensional cognitive maps and the existence of two memory systems involved in navigation: locale (map-based) and taxon (route-based). Comparable to Tolman's comprehensive-like map, a locale navigational system would provide the organism with a larger amount of information allowing for more flexibility when navigating a novel environment, while in the taxon system the organism navigates by route-based strategy, which is less flexible and prone to interference.

Directional Responding and Place Navigation

Tolman et al. (1946), attempted to explicate the strategies involved in navigation and which pattern of behavior would emerge given each circumstance. The simple T-maze was used to probe the animal's predisposition to navigate to a goal location either by performing a particular motor response – defined as response learning (e.g., always turning left toward the goal location), or by navigating to a specific goal location in the room consistent with a place disposition (e.g., navigating to a particular location in the room). In the experiment, for half of the trials, rats were released from one point of the maze and had to take a right turn to the goal location. For the other half, the animals were released from the opposite extreme of the maze and had to take a left turn to the same goal location. In both scenarios, the animals learned how to navigate to the goal location regardless of the release point. The results were classified as pure place learning. The idea that animals have a predisposition to always navigate to a specific place was confronted by Blodgett et al., (1949) as he noted a confound in Tolman's experiments as an orientation toward a direction was not ruled out. Tolman's rats in the place condition were trained to navigate to the goal location from different release points. However, independent of the release point, the animals were always navigating in the same direction to the goal location. Expanding on Tolman's experiment, Blodgett et al. (1949) designed a series of manipulations of the simple T-maze with different groups to test for place, direction, and motor responses individually, as well as combinations of those. For the animals in the direction group, there were two maze placements with distinct food locations and distinct turns at the choice point. In the arrangement, only the direction from the choice point to the food location was common in both placements (Figure 1). Compared to the other groups, animals from the direction group made fewer mistakes, which was concluded to be due to a facilitation of learning when there was a common direction from choice point to food location. To disambiguate between place navigation and directional responding, Blodgett et al (1949), adapted the maze in a way that navigating to the same goal location in the environment from different release points would only be possible by following opposite directions in the environment rather than the same direction (see Figure 2 for details). Blodgett (1949) argued that the animals in Tolman's experiments did not exhibit true place navigation and demonstrated that the animals could acquire a disposition to navigate toward a direction in the environment, defined as directional responding, and learned this more readily than place navigation. Both place navigation and directional responding could result in different spatial behavior. Blodgett's theory, however, was not explored further, and 'pure' place navigation was well-accepted as the primary strategy involved in navigation for decades. In recent years, the idea of a distinction between true place navigation and directional responding has gained renovated attention. To corroborate and further expand on Blodgett's findings, Skinner et al. (2003), replicate those experiments in a square open field apparatus. Several studies (Akers et al., 2007, 2009; Hamilton et al., 2007, 2008; Knierim & Hamilton, 2011; Stackman et al., 2012; Sutherland & Hamilton, 2004) used the Morris Water Task (Morris, 1981) including task manipulation by translating the pool into a new location in the room to assess both navigation strategies. All the findings were in line with Blodgett et al. (1949) providing robust evidence for a preferred strategy of navigation based on directional responding over place navigation.

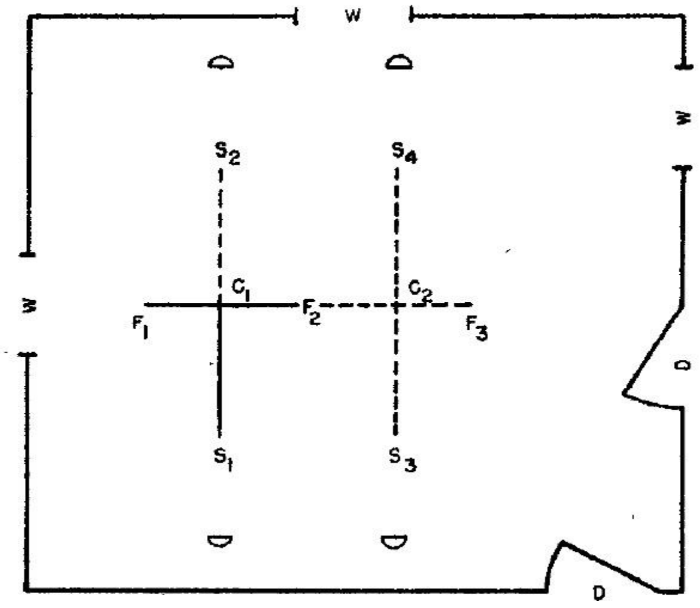


Figure 1. Figure retrieved from Blodgett et al., (1949). showing the maze placements for the animals in the direction group. S1-S4 represents start points, and F1-F3 represents the food location.

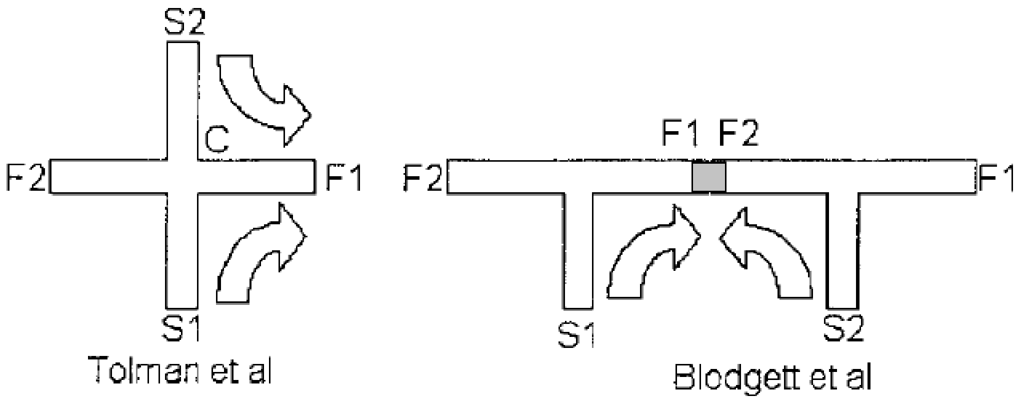


Figure 2. Adapted from Skinner et al., 2003, the grey area represents the overlap of the food location from the two maze placements. The figure on the left shows a representation of Tolman et al. (1946) where different start points (S1-S2) led to the same goal location (F1-F2) by following the same direction (not disambiguating place and direction). The figure on the right shows one of Blodgett et al. (1949) maze manipulation to disambiguate place navigation and directional responding. Different start points led to the same goal location by following opposite directions.

Morris Water Task

The Morris Water Task (Morris, 1981) has long been used as a measure of spatial navigation assessment. In the task, rodents are trained to swim to an escape platform hidden below the surface of the water. The animals are released from different starting points and learn to navigate to the platform based on exteroceptive (environmental cues) information by using the visual system. During the probe trial, the platform is absent and the animals that have mastered the task persist to swim where the platform was located. Compared to other tasks (e.g.: Radial Arm Maze - RAM), the MWT has offered evidence of greater effectiveness when assessing place navigation. The RAM, used by Olton et al. (1979), required the animals to remember which arms were already entered, and when that arm was entered, which required the involvement of a type of memory processing referred by Olton to as working memory. The MWT allows for the measurement of place learning and memory without requiring working memory processes. The MWT has become the gold standard to assess spatial learning and memory and identify the neurobiology of the behavior. Among the advantages of the MWT are: less training resulting in clear place learning, long-term retention, persistent learning regardless of conditions such as different release points or intertrial intervals, as well as no need for water or food deprivation (Hamilton et al., 2006.; Schoenfeld et al., 2017). The MWT has also been used to assess place navigation and directional responding by including a probe trial consisting of pool translation. The pool is repositioned into a new location in the room in such a way that there is a quadrant overlap between the initial position (Position 1) and the new position (Position 2), and the platform is removed. If the organism is navigating based only on the fixed spatial location of the platform relative to distal visual cues, there would be a persistent swim in the absolute region of the room where the platform was previously located. If the organism is using the distal cues to orient the trajectory of the swim, the swim would be toward a direction where the platform was previously located. Results of the pool translation probe trial done in rodents have demonstrated a preference for directional responding over place navigation suggesting that organisms have a preference to navigate by following a direction (Hamilton et al., 2007; 2008; 2009). Although the Morris Water task has been widely used in studies of spatial learning and memory, what the organisms learn during the task and how environmental cues are encoded, are not well understood.

The Morris Water Task as well as several other behavioral tasks have been adapted to a computerized version as an attempt to better understand the generality of spatial learning principles in humans and nonhuman animals (Rodriguez, 2010; Schoenfeld et al., 2017). The virtual Morris Water Task (vMWT) (Astur et al., 1998) has been of great interest and several laboratories have utilized versions of the task for different purposes such as assessing sex differences (Astur et al., 2004; Chamizo et al., 2011; Driscoll et al., 2005; Sandstrom et al., 1998; Sneider et al., 2015; Woolley et al., 2010), neuropathology (Folley et al., 2010; Hanlon et al., 2006; Kremmyda et al., 2016; Kuhn et al., 2018; Schoenfeld et al., 2017) and age differences (Driscoll et al., 2005; Li & King, 2019; Moffat et al., 2006; Reynolds et al., 2019; Schoenfeld et al., 2014) in spatial navigation task to assess learning and memory. One of the advantages of virtual tasks is greater control of task variables and environmental manipulations in a way that is not possible to do with real environments (Hamilton et al., 2009). In addition to assessing the behavioral measures, virtual tasks have also been used in combination with neuroimaging techniques such as fMRI, MEG, and PET, which allow for the investigation of the neural networks supporting navigation. An emphasis has been placed on understanding the role of the hippocampal regions and adjacent structures as there is an agreement on the central role of those structures in spatial navigation learning and memory (Iaria et al., 2003).

Studies of hippocampal lesions done in rodents have provided evidence for the role of the hippocampus in spatial navigation (Pearce et al., 1998; Stringer et al., 2005). Animals with hippocampal damage showed impairment in solving spatial navigation tasks and the results demonstrated that both place learning and directional responding were disrupted compared to sham (Packard & McGaugh, 1996; Stringer et al., 2005). The findings suggest that the neurobiological processes supporting both types of spatial learning are impaired following hippocampal damage, which confirms the involvement of the structure but does not provide evidence for the specific role of the hippocampal regions and adjacent areas in either type of learning. Findings in humans with hippocampal damage or unilateral temporal lobectomy showing navigational impairments are consistent with the idea that the hippocampus and surrounding regions are necessary for navigation (Shrager et al., 2007).

Neuroimaging and virtual navigation spatial tasks

Neuroimaging techniques combined with virtual spatial tasks provide evidence of activation of regions that are key for navigation tasks which include the hippocampus and surrounding areas, as well as parts of the basal ganglia and the posterior cingulate cortex, and the left prefrontal cortex (Maguire et al., 1999). Maguire et al. (1998) findings, in a study using PET while participants navigated in a virtual reality town, suggest that higher accuracy in complex task performance was associated with greater activation of the right hippocampus and inferior parietal cortex in non-cued trials, as well as the left lateral temporal cortex, left frontal cortex, and thalamus. Their results also showed increased activation of the right hippocampus during non-trial-following navigation. In agreement with the abovementioned findings, previous analysis of the fMRI vMWT BOLD activation of partial data of the current work – not including the pool translation probe trial - found activation of several brain regions including the hippocampus, surrounding areas in the medial temporal lobe, and frontal lobe regions during memory retrieval (Sneider et al., 2018).

Studies of spatial navigation tasks have provided evidence for similarities in behavior between humans and rodents, however, to our knowledge, no human studies of the vMWT and fMRI have assessed preferences for navigation based on the apparatus reference frame (directional) or the room reference frame (place navigation) by using the pool shift paradigm. Moreover, this study was conducted on adolescents as part of a longitudinal study. This can provide a baseline for understanding whether strategies of navigation change over time.

The current work aims to investigate the possibility of grouping people based on their preferred navigational strategy on the vMWT pool translation probe trial, which is the last block of the vMWT sequence, as well as to analyze the underlying network supporting the behaviors by fMRI BOLD activation. In the fMRI vMWT pool translation paradigm, participants are tested to establish the preferred strategies involved in navigating based on visual cues when the pool is translated from the original location (Position 1) to a new location in the room (Position 2) with one quadrant overlapping, absent platform, and the presence of the same distal cues. The participant could either swim by orienting themselves toward a direction in the room to the relative location of where the platform was previously located within the apparatus reference frame, or by place navigation – when the relationship among distal room cues is the reference frame to navigate to the absolute location of the room where the platform was previously located.

We hypothesized that all participants would learn the task similarly during offline training and that differences would emerge during the pool translation probe trial paradigm that would allow for the evidence of two distinct groups based on navigational strategies consistent with previous works done with rodents. The result of behavioral measures obtained from the probe trial was compared with the same measures from the training trials aimed to determine if there were any group differences in learning the task during the training trials. We further hypothesize that there would be differences in the degree of hippocampus activation in the group using orientation as a preferred navigational strategy versus the group using the room as a reference frame (place navigation). Even though both types of navigation are dependent on the hippocampus, it is expected a greater activation of the hippocampus in the place navigation group. Directional responding is controlled by visual information depending on circuits outside of the hippocampus. The parietal cortex is involved in orienting and directing behavior. Therefore, greater activation of that region is expected in the directional responding group.

The result of behavioral measures obtained from the probe trial was compared with the same measures from the training trials aiming to determine if there were any group differences in learning the task during the training trials. If there are fMRI differences, those could be possibly explained by behavioral differences. If the differences occur during the probe trial only, it could reflect a disruption of the pattern of navigation that participants developed during the training trials.

Methods

Participants

The sample consisted of 56 healthy adolescents (33 females); mean age = 13.89 years old. Participants were recruited from local pediatrician clinics through the Boston Children Hospital's (BCH) Research Participant Registry and local advertisements. Potential participants were screened via an online eligibility survey and completed follow-up verification to ensure they met the criteria for inclusion in the study. All aspects of the clinical research protocol were reviewed and approved by the Partners Healthcare Institutional Review Board of McLean Hospital (Belmont, MA, USA). Following a complete description of the study, all participants and their parent(s) or guardian(s) provided written assent and consent. All participants received monetary compensation after study completion.

Before scanning, participants were required to complete urine screening to rule out current psychoactive substance use (Clarity Diagnostics Drugs of Abuse Panel, Boca Raton, FL, United States) and pregnancy (QuPIS One-Step Pregnancy, Stanbio Laboratory, Inc., San Antonio, TX, United States).

Clinical assessment

The clinical interviews were conducted by trained staff using the Mini International Neuropsychiatric Interview for Children and Adolescents (MINI-KID) (Sheehan et al., 2010). The vocabulary and matrix reasoning subtests of the Wechsler Abbreviated Scale of Intelligence (WASI) (Wechsler, 1999) were administered to obtain an estimate of general intelligence. The WASI Block Design subtest was administered to assess visuospatial ability. Table 1 presents the demographics of the participants. Participants were primarily non-Hispanic whites (76.8%) with a mean age of 13.89, and 41.1% males

TABLE 1 | Participant demographics and cognitive measures.

	Total sample *(n = 56)*
Age (years)	13.89 ± 0.6
Female/Male	33 (58.9%)/23 (41.1%)
Education (years)	7.6 ± 0.8
Handedness	54R,2L
Ethnicity	96.43% non-Hispanic
Race	76.8% Caucasian
	5.36% Asian
	17.8% Other
WASI T-Scores IQ estimate (2-subtest)	115 ± 8.93
Vocabulary	60.5 ± 6.96
Matrix Reasoning	56.5 ± 6.16
Block Design	575. ± 8.75

Data represent means ± standard deviations. Ethnicity: Hispanic vs. Non-Hispanic; "Other" included the following designations: Asian/Caucasian; African American/Caucasian; American Indian or Native; Alaskan/Caucasian.

virtual Morris Water Task (vMWT)

The virtual environment was viewed from a first-person perspective and displayed on a Windows laptop. The task entailed navigating to a square platform – hidden or visible according to trial design - in a circular pool within a square room with four distal cues - one on each of the four walls of the virtual environment (Figure 3). The platform was in the southwest (SW) quadrant of the virtual pool environment for all trials. To navigate, participants used arrow keys from the laptop keyboard (right, left, and forward) during offline training and MRI compatible fiber optic response pad (fORP) box during fMRI. The offline training before performing the task in the MRI scanner was used to establish performance navigating to a hidden platform. Participants were trained to navigate to the hidden platform. With more training, the trajectories to the hidden platform become more direct. A message was displayed on the screen when the platform was found, then a new trial started from a different release point. Each block lasted 30 seconds. During each block, the participants completed as many trials as possible.

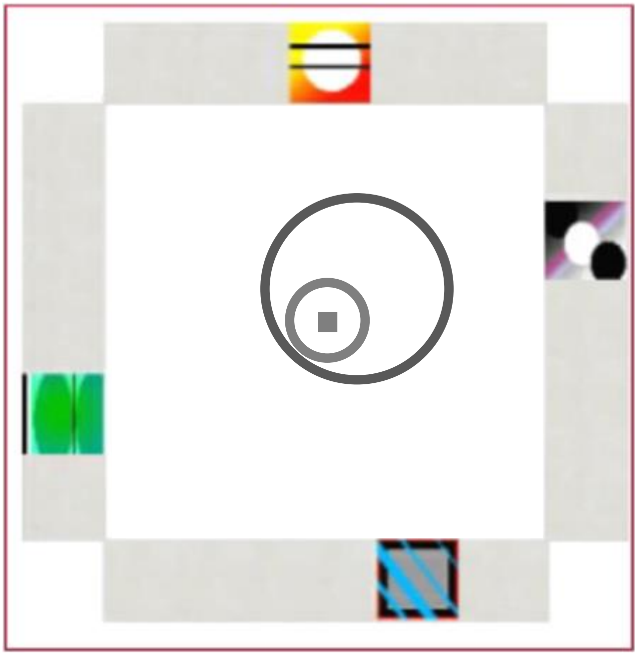


Figure 3. Representations of the offline training vMWT. Distal cues were displayed on each of the four walls. The bigger circle represents the pool location, the square represents the platform, and the smaller circle (not visible to the participants) represents the region of interest (25% of the pool diameter).

Offline training blocks

Offline training consisted of three conditions: hidden platform learning/retrieval trials, a single no-platform probe trial, and one block of visible platform motor control trials. For the learning/retrieval trials the platform was hidden below the surface of the water. The distal cues – distinctive pictures – were displayed on each of the four walls. Each of the four trials had a different release point, north, south, east, or west (pseudorandomly selected). The duration of each trial was 60 s with a 1 s inter-trial interval (ITI). If the participant found the hidden platform, a “platform found” message was presented on the screen. Failure to find the platform within 60 s (unsuccessful trial) resulted in the platform becoming visible until the participant successfully navigated to it. The single no-platform probe trial was 30 s in duration and conducted within the same environment as the learning/retrieval trials. The release point was one of the two furthest locations (N or E) from the location of the platform used during the learning/retrieval training. After completion of the probe trial, participants then completed one block of four visible platform trials, which served as a motor performance control condition. In the motor trials, the platform was visible above the surface of the water in the same virtual environment; however, the four visual cues were removed from the distal environment. The maximum duration of the motor trials was 60 s; after which the participant was prompted with a verbal message to navigate to the platform. The measures analyzed were total path length, swim latency *(this measure refers to the period between the first movement of each subject and the location of the platform),* time in region, and percent total path length in the target region (absolute or relative).

fMRI vMWT paradigm

A block design was utilized for the fMRI vMWT paradigm (Figure 6) consisting of pairs of alternating retrieval and motor trials – schematically represented in orange and green respectively – followed by a single probe trial with pool translation. Each participant had two runs of all blocks. The duration of each block was 36 s and participants try to complete as many trials as possible within that time. Each pair of retrieval and motor trials was separated by rest blocks (fixation cross, 21 s per block). For the probe trial, in which the cues in the virtual environment were the same as in the retrieval trial, only with the platform removed, participants navigated for the entire duration of the 30-s block. In addition to the behavior measures, the fMRI BOLD activation measures included six contrast of parameter estimates (COPE): Hidden > Rest, Visible > Rest, Probe > Rest, Hidden > Visible, Probe > Visible, and Probe > Hidden.

For the fMRI probe trial, the pool was translated from the original position (Position 1) to the new position (Position 2) and no platform was present (Figure 4). The probe trial duration was 36 sec. For analyses, region A represents the trained platform location in the room (distal cue) reference frame and is referred to as “absolute location”. Region B represents the location within the apparatus reference frame and is referred to as “relative location”.

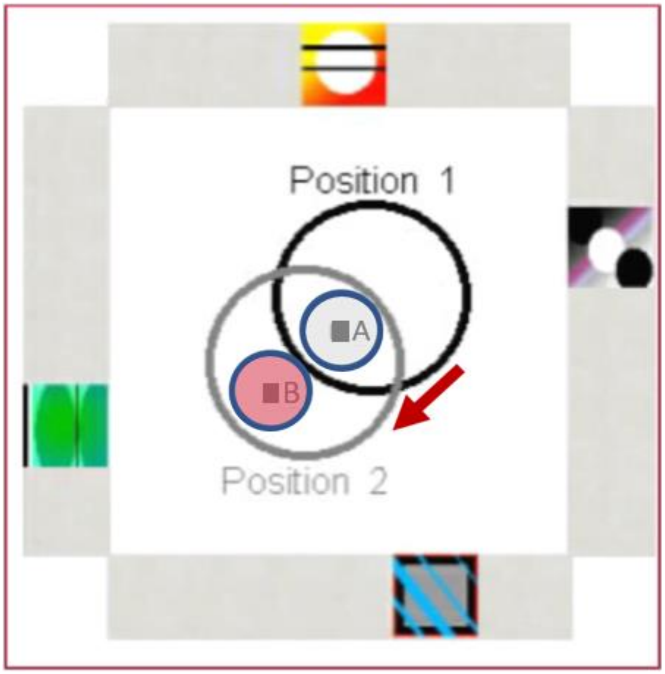


Figure 4. Pool translation during probe trial from position 1 to position 2. The circles A and B indicate the corresponding location of the platform in each position and represent the target region (25% of the pool diameter centered on the platform). The wall cues were visible, and the platform was absent.

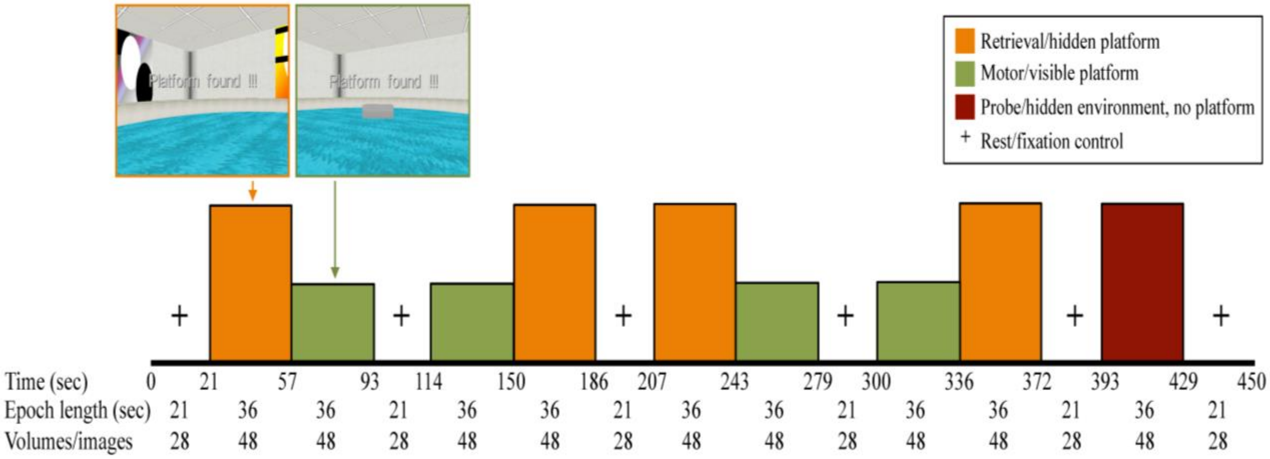


Figure 5. Experimental block design during fMRI with example of participants view. (Figure retrieved from Sneider et al., 2018)

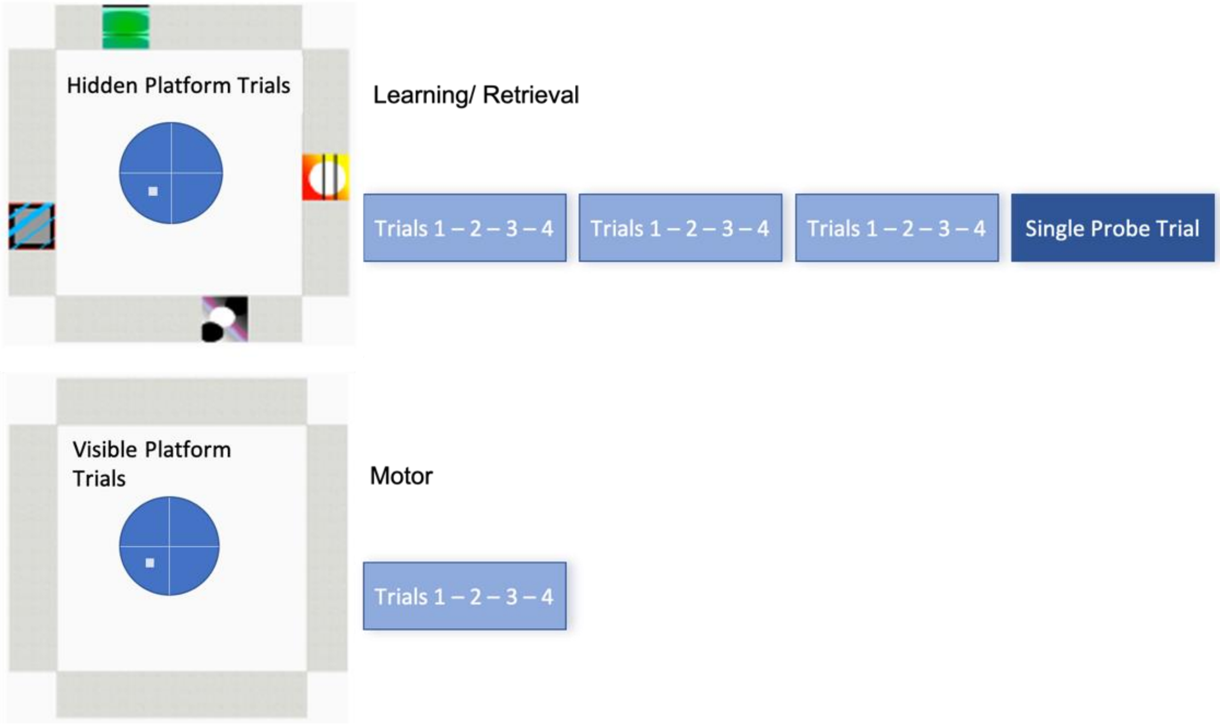


Figure 6. Representations of the offline training blocks. The top row shows the virtual environment with the 4 wall cues. The bottom row shows the motor block where the cues were removed, and the platform became visible. The quadrants division was not visible and are used for calculating navigation measures.

Preference groups characterization

To determine the preference groups (see Figure 8 for steps), the measure of time spent in each region (absolute or relative) during the fMRI pool shift probe trial was used to calculate preference scores as follows:

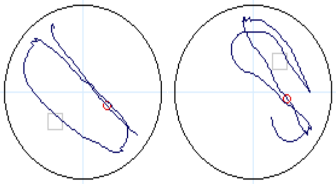
$$\boxed{\text{Preference} = \frac{(\text{time spent in apparatus region - time spent in room region})}{(\text{time spent in apparatus region + time spent in the room region})}}$$

The scores from this formula range from -1 to 1. Scores < 0 indicate a preference for navigation based on the room reference frame (place navigation) and scores > 0 indicate a preference for navigation based on the apparatus reference frame (directional responders).

The grouping criteria was that people with a score of 0.5 or higher were grouped in the “Preference group” (P) – this refers to people whose scores provided evidence for a navigation based on the apparatus reference frame. Scores of -1 – which indicates place navigation - were rare (4/56), therefore those people were grouped those who did not demonstrate a systematic preference for either location into the “non-preference group” (NP) (see supplementary figure 14 for histogram). The pool translation probe trial is the block of interest for the current work.

The first goal was to assess the evidence of two distinct preference groups based on people’s navigation strategies as assessed in the pool shift probe trial from the fMRI session. After obtaining the values for each participant from each group qualitative analysis of the swim paths was conducted. Figure 7 provides evidence for similarities in search patterns across groups supporting the grouping criteria for further analyses of this work. For each pair, the figure on the left corresponds to the learning probe trial and the figure on the right is the fMRI pool shift probe trial.

NP group



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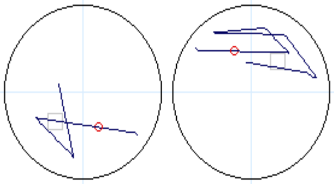

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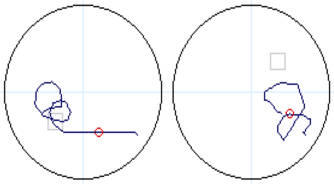

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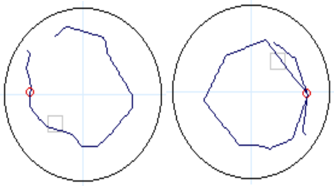

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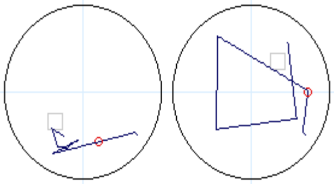

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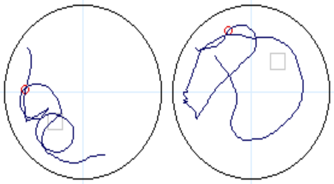

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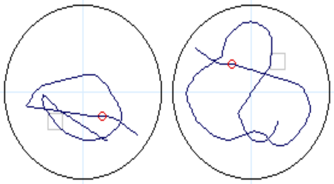

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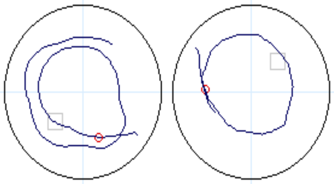

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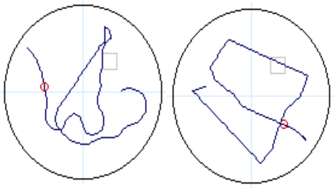

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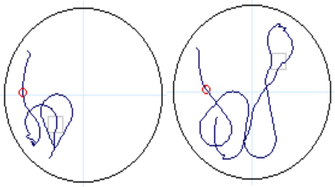

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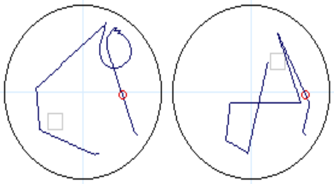

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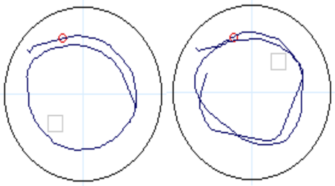

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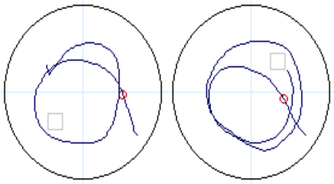

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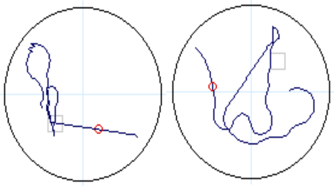

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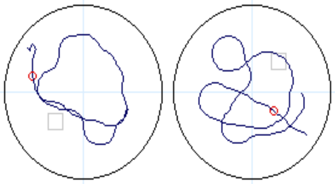

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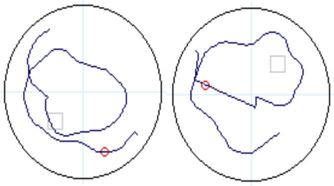

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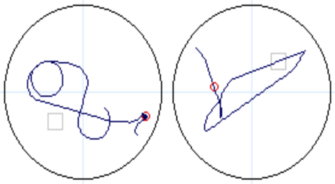

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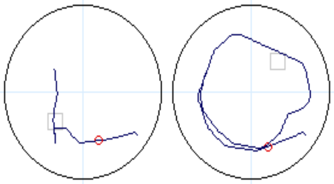

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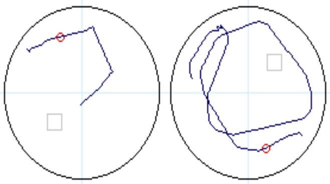

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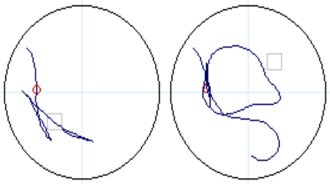

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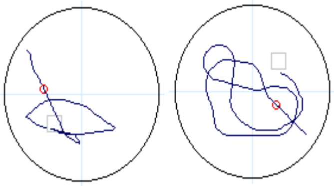

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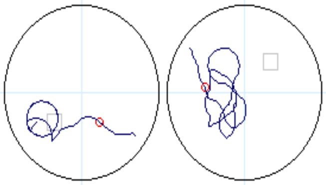

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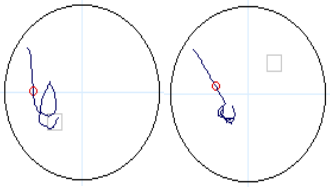

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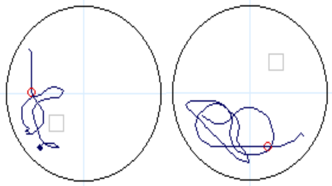

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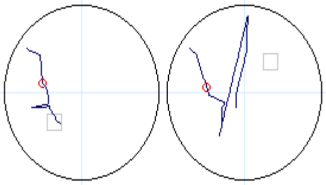

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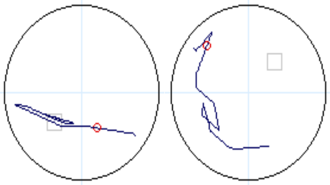

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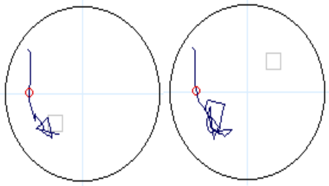

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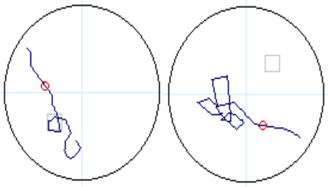

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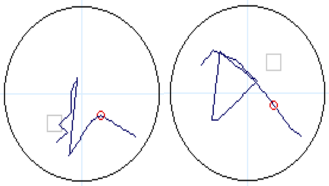

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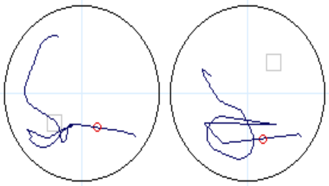

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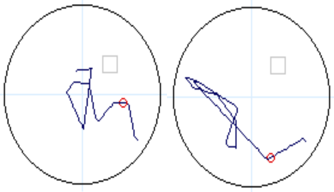

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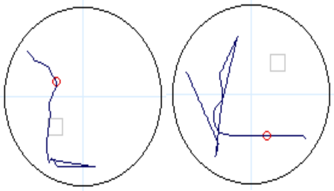

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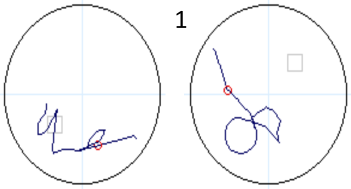

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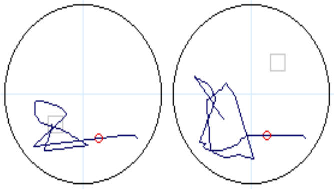

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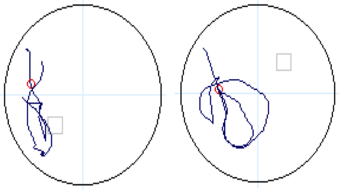

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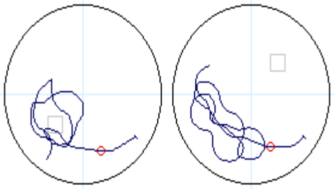

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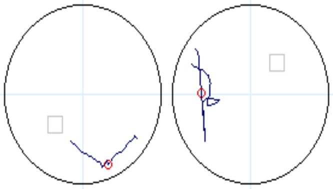

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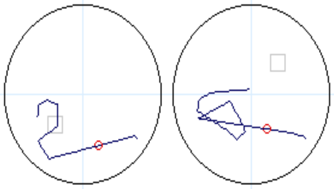

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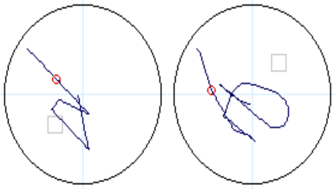

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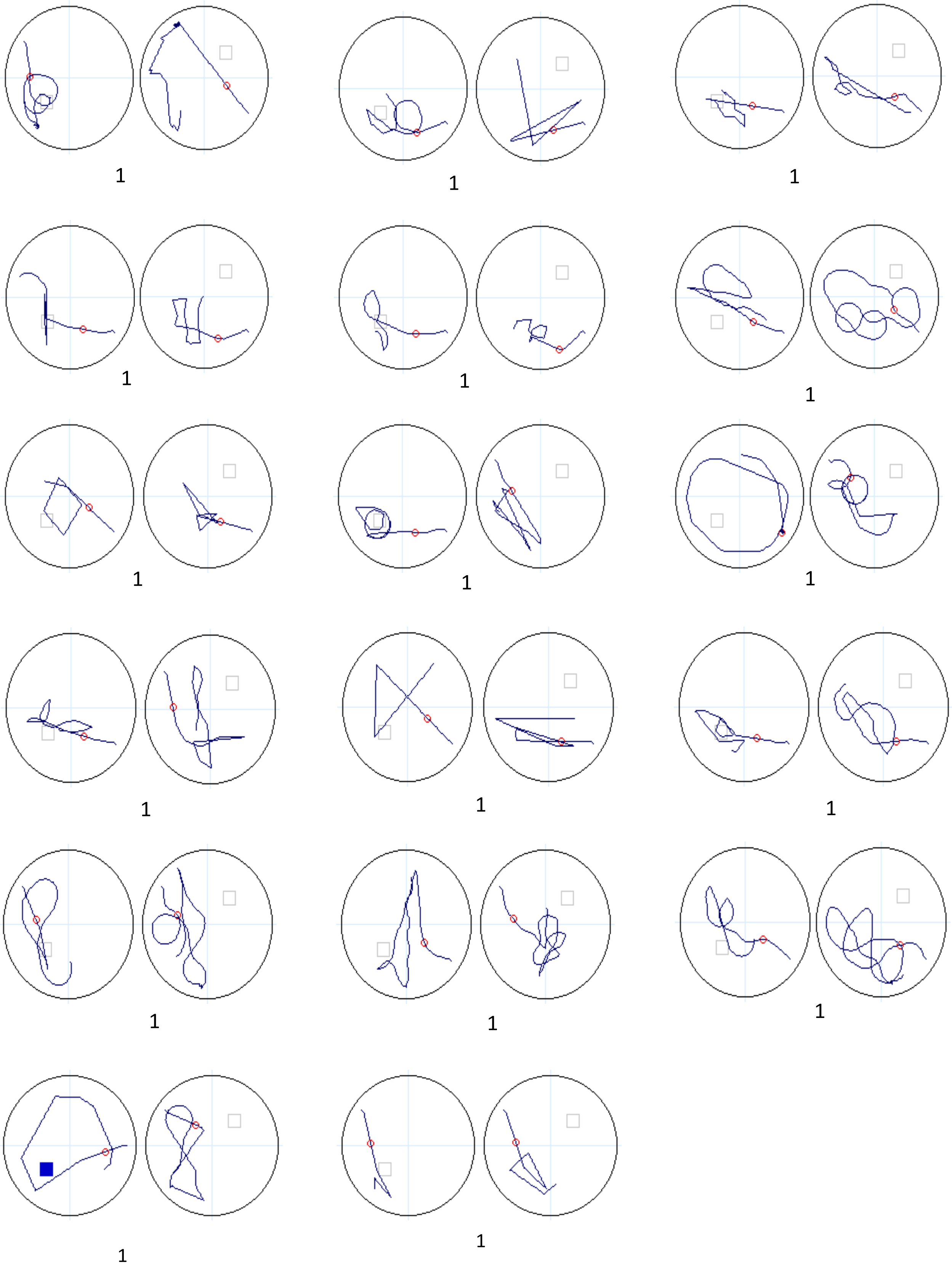

Figure 7. *Swim paths from participants from the NP and P groups with corresponding scores. There were no sex differences, therefore the groups are not split by sex. The left circle of each pair corresponds to the learning probe trial; the right circle corresponds to the fMRI pool translation probe trial.*

Functional Magnetic Resonance Imaging Acquisition and Data Processing

Brain images were acquired on a Siemens TIM Trio 3 Tesla MRI system (Erlangen, Germany) with a 32-channel head coil. High-resolution structural images were collected A T1- weighted multi-echo magnetization prepared rapid acquisition gradient echo (ME-MPRAGE) 3D sequence in four echoes (TED1.64/3.5/5.36/7.22 ms, TRD2.1 s, TID1.1 s, FAD12, 176 slices, voxel size = 1 mm x 1 mm 1.3 x mm, acquisition time D 5 min) for registration of functional images into standard space. To collect fMRI data in one 7.5 min run, whole-brain multiband gradient-echo echo-planar imaging (EPI) with BOLD contrast was used. Images were acquired in 54 interleaved oblique slices (TR/TE/FA D 750 ms/30 ms/52, FOV D 220, voxel size = 2.8 mm x 2.8 mm x 2.8 mm, multiband D 6, GRAPPA D 2). A field map was acquired at the same resolution and slice locations to allow for B0 unwarping (TR D 1000, TE D 10/12.46 ms, FA D 90, 2:44 min).

Before statistical analyses, preprocessing was performed on raw functional images using the FMRIB Software Library (FSL) software v5.0.10 (Smith et al., 2004) (FMRIB, Oxford, United Kingdom), (Groves et al., 2009) including motion correction, slice timing correction, non-brain removal, spatial smoothing (FWHM 6 mm Gaussian kernel), and grand mean intensity normalization of the 4D dataset by a single multiplicative factor. Ten volumes at the onset of the first rest block were removed to allow for signal equilibration. ICA AROMA, an independent component analysis-based denoising tool, was then used to remove motion-related components from the fMRI data (Pruim et al., 2015). Any subject with greater than 5 mm motion was excluded from further analysis. Components related to respiration and other artifacts also were identified by visual inspection of ICA components, then all motion-related and artifacts were removed from the fMRI data using fsl_regfilt. Denoised fMRI data were then temporally filtered using a Gaussian-weighted least-squares straight-line fit with a high-pass cutoff D 100 s and underwent field map-based distortion correction. Functional MRI data were registered to MNI152 standard space by first registering the fMRI images to the high-resolution structural image using boundary-based registration (BBR) and then transforming into MNI stereotaxic space using the first registration step combined with the registration information from registering the high-resolution structural image to MNI152 standard space, which was done using FNIRT.

For fMRI activation, a hierarchical voxel-wise general linear model (GLM) was conducted for each COPE with FLAME (FMRIB's Local Analysis of Mixed Effects) to assess the groups' average activation. Gaussian random field theory with cluster-based thresholding ($z = 3.1$) was done to control for family-wise error, e.g., $p < 0.05$ corrected.

Statistical Analyses

MANOVA was conducted for the offline probe trial measures (path length/ proximity/ latency/ time in quadrant/entries). Repeated measures analysis of variance (RM-ANOVA) was conducted to analyze the vMWT performance measures (total path length, swim latency, time in region, and percent total path length in target region) for trial blocks (hidden and visible), with sex and group included as between subjects' factors. The number of completed trials also was analyzed for retrieval and motor trial blocks during fMRI using repeated measures ANOVAs, with preference group and sex included as independent variables. Post hoc analyses for ANOVAs were conducted using two-sample t-tests (two-tailed) to determine the direction of the effect when the main effects or interactions were statistically significant. All statistical analyses for behavioral measures were conducted using jamovi v.1.6 (The jamovi project (2021).

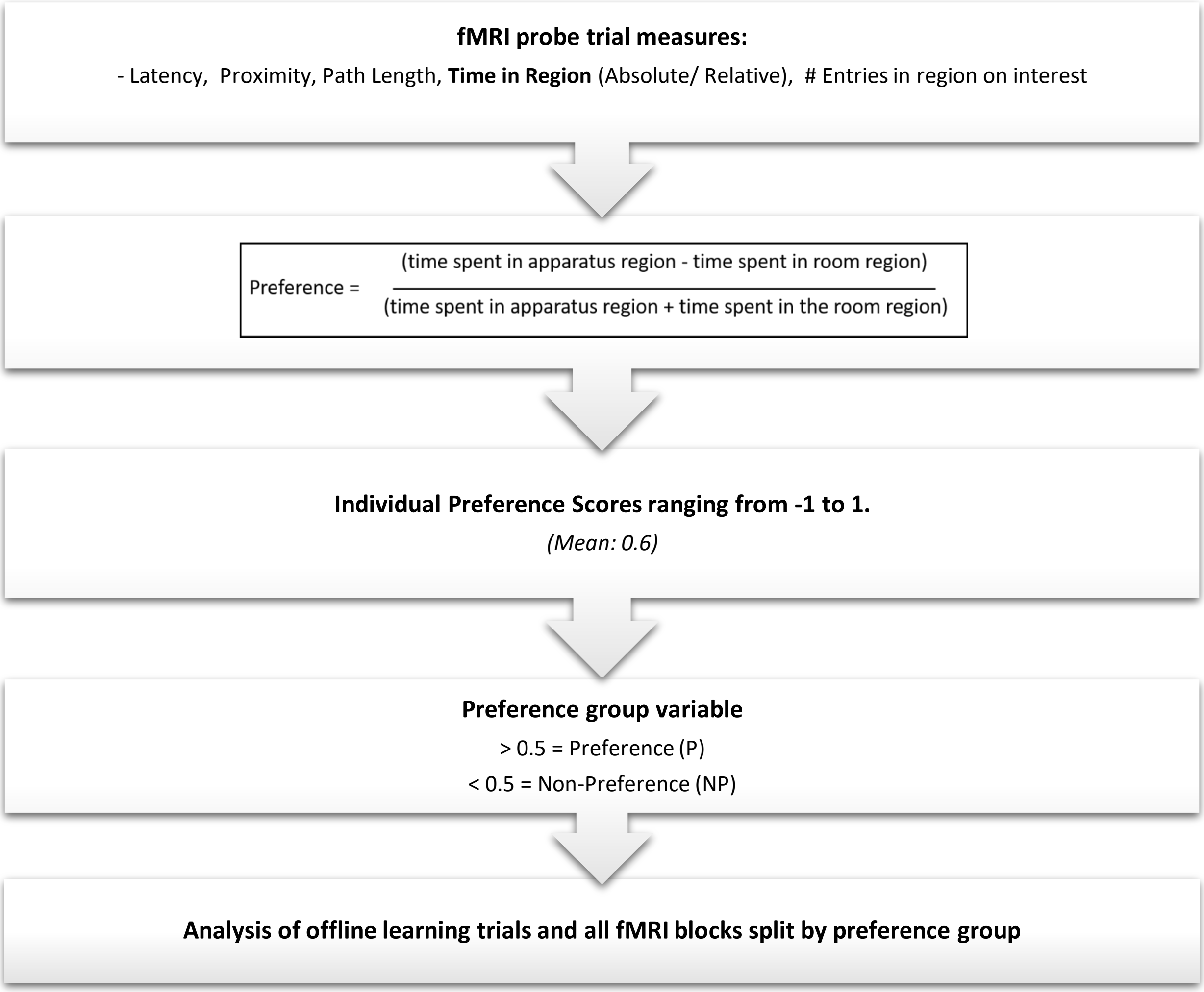

Figure 8. Flow chart showing the sequence of steps to generate the preference group variable that was used as a factor in the statistical analysis for behavioral and imaging data.

Results

Cognitive variables

MANOVA results showed no significant differences between the preference groups or sex in IQ, WASI block design, or mental rotation. All ps > 0.25.

Learning trial blocks – hidden and visible

Mean latency and path length are shown in Fig. 8. RM ANOVA revealed a main effect of trial block for latency (Greenhouse-Geisser F(1.79, 92.93) = 15.853, p <.001, η_p^2 = 0.234) and path length (Greenhouse-Geisser F(1.61, 83.83) = 8.283, p= 0.001, η_p^2 = 0.137), with both measures decreasing as a function of trial block. There was a main effect of group for path length with the non-preference group (NP) having a longer path length than the preference group (P), (F(1, 52)= 7.593, p = 0.008, $\eta_p^2 = 0.127$). There was also a main effect of sex with females having higher latencies than males (F(1,52)= 6.78, p = 0.012, η_p^2 = 0.115). There were no other significant main effects or interactions, all ps>0.11.

As described in the methods, the latency measure represents the duration between initiation of the first movement and finding the platform. In addition to the latency effects described above, RM ANOVA also revealed a main effect of trial block for first movement latency (data not shown; Greenhouse-Geisser F(1.58, 82.11) = 10.569, p <.001, $\eta_p^2 =$ 0.169) with the time to initiate the first movement decreasing as trial block increased. There was also a main effect of sex (F(1,52) = 7.999, p = 0.007, η_p^2 = 0.133) with females taking longer than males to initiate the first movement. There were no other significant main effects or interactions for time to initiate the first movement, ps> 0.34. RM-ANOVA revealed no group or sex main effects, or interactions in the visible trials for latency, first movement, or path length, ps>0.09.

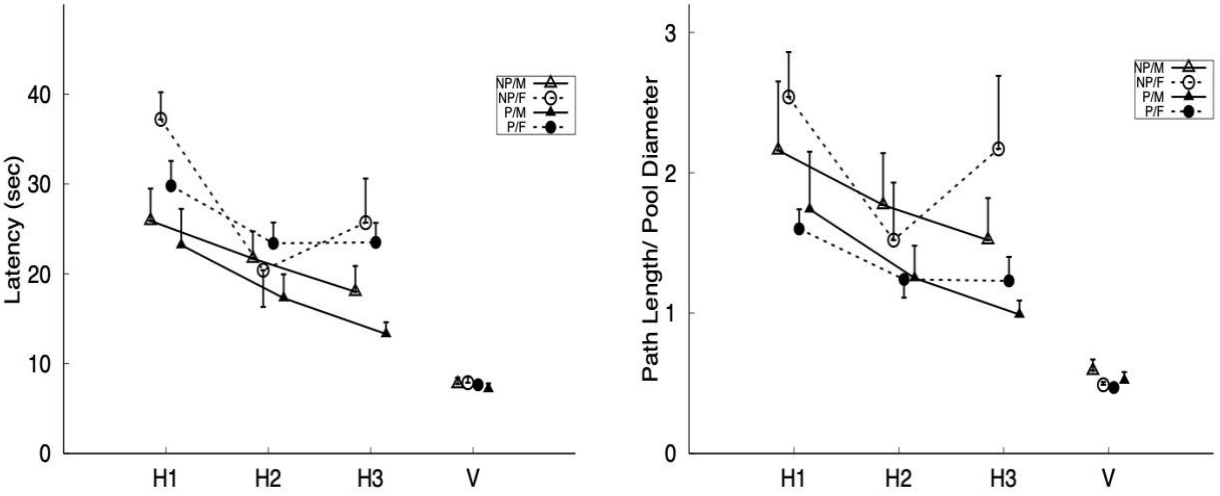

Figure 9. Mean and SEM measures during the offline probe trial with hidden and visible platform for males (solid lines and open/closed triangles) and females (dashed lines and open/closed circles) in each preference group (P = preference/ NP = non-preference). Latency (sec) across hidden (H1-2-3) and visible (V) trial blocks. Path length to enter the platform region across hidden (H1-2-3) and visible (V) trial blocks.

Offline probe trial

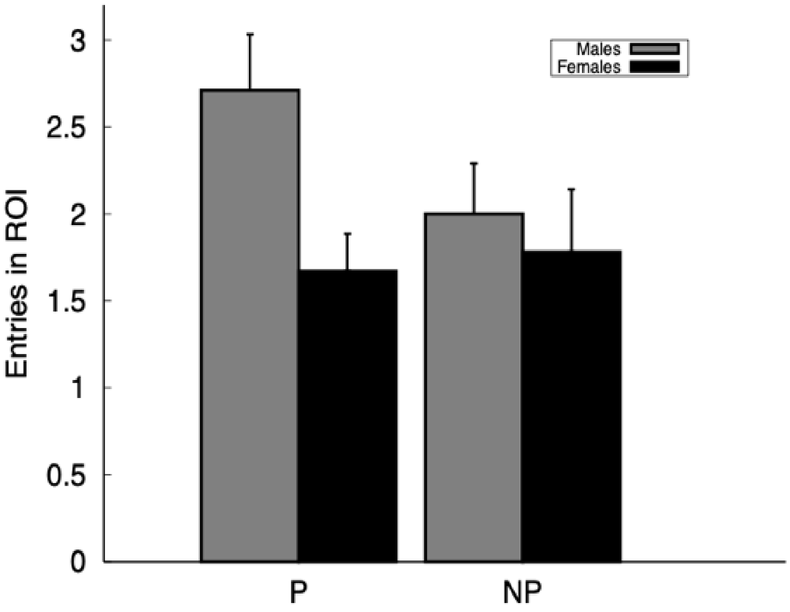

Figure 10. Mean (+ SEM) number of entries in the region of interest during the offline probe trial. Significant main effect of sex, with males having more entries than females, p=0.018.

The mean number of entries in the ROI, latency to the platform region, path length to ROI, proximity to the platform region, and time in the quadrant are shown in Fig. 10 and 11, respectively.

MANOVA results show a main effect of group (F(5,48)= 4.23, p=0.003) and sex (F(5,48)=3.05, p=0.018), but no Group by Sex interaction (p=0.35). Univariate tests show a main effect of group in the proximity to region measure (F(1,52)=7.11, p=0.010) – P group closer to the ROI -, and sex in the number of entries measure (F(1,52)=6.87, p=0.011), with males having more entries than females. All other measures were non-significant, ps>0.10. There was no significant Group by Sex interactions, ps>0.19.

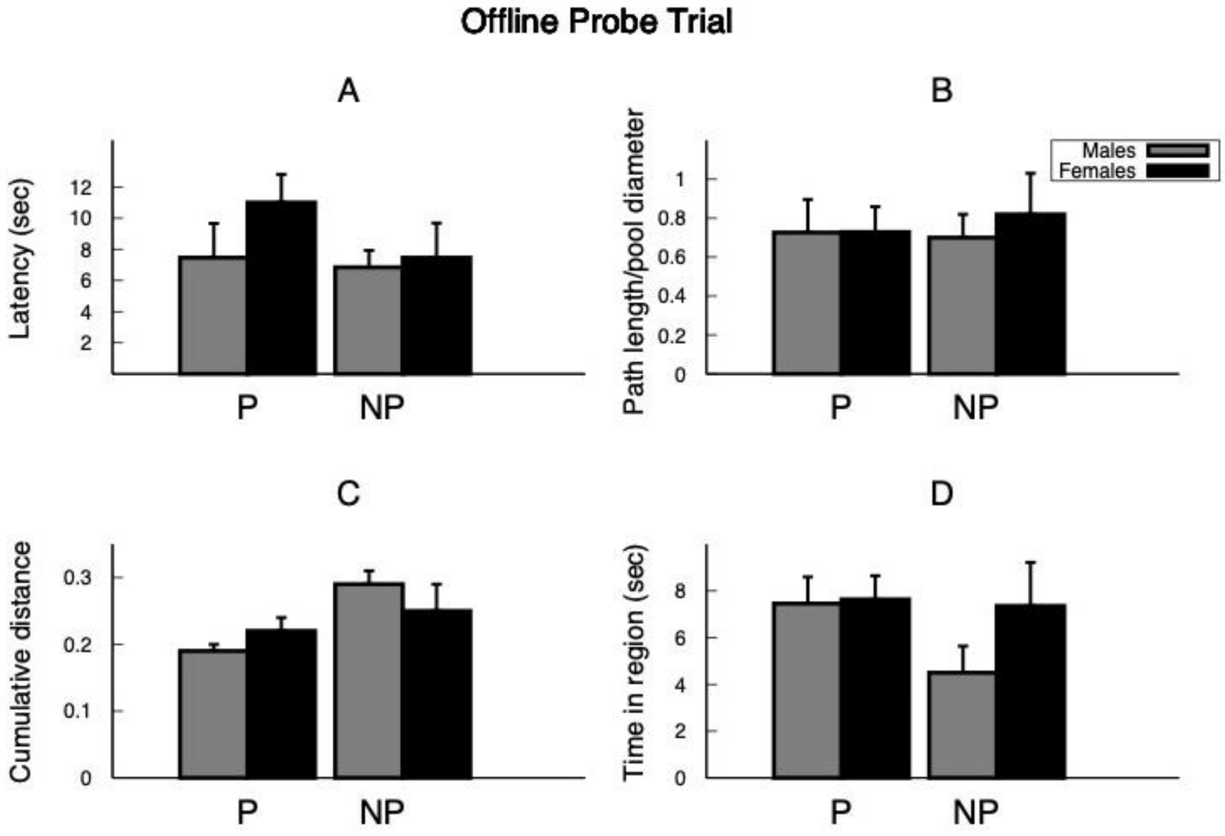

Figure 11. Mean (+ SEM) for the offline probe trial measures: A – latency(sec), B – path length to the platform region, C – cumulative distance (proximity), and D – time in region. All measures were split by sex (male in the grey bars and females in the black bars) and preference group (P – Preference/ NP – non-preference).

Trials complete (fMRI)

Figure 12 shows the mean number of trials completed during the fMRI block for each combination of group, trial type, and sex. RM-ANOVA revealed a main effect of trial type (hidden vs. visible) with more trials being completed in the hidden block 3 compared to hidden blocks 1 and 2 (F(1,52)= 3.745, p=0.012, $\eta_p^2 = 0.067$), suggesting that with repetitions, the participants effectively learned how to perform in the task. There was a main effect of group (preference > non-preference, F(1,52)=5.82, p=0.019, $\eta_p^2 = 0.101$), and sex (males > females, F(1,52)=6.84, p=0.012, $\eta_p^2 = 0.116$). There were no other significant main effects or interactions, ps>0.120.

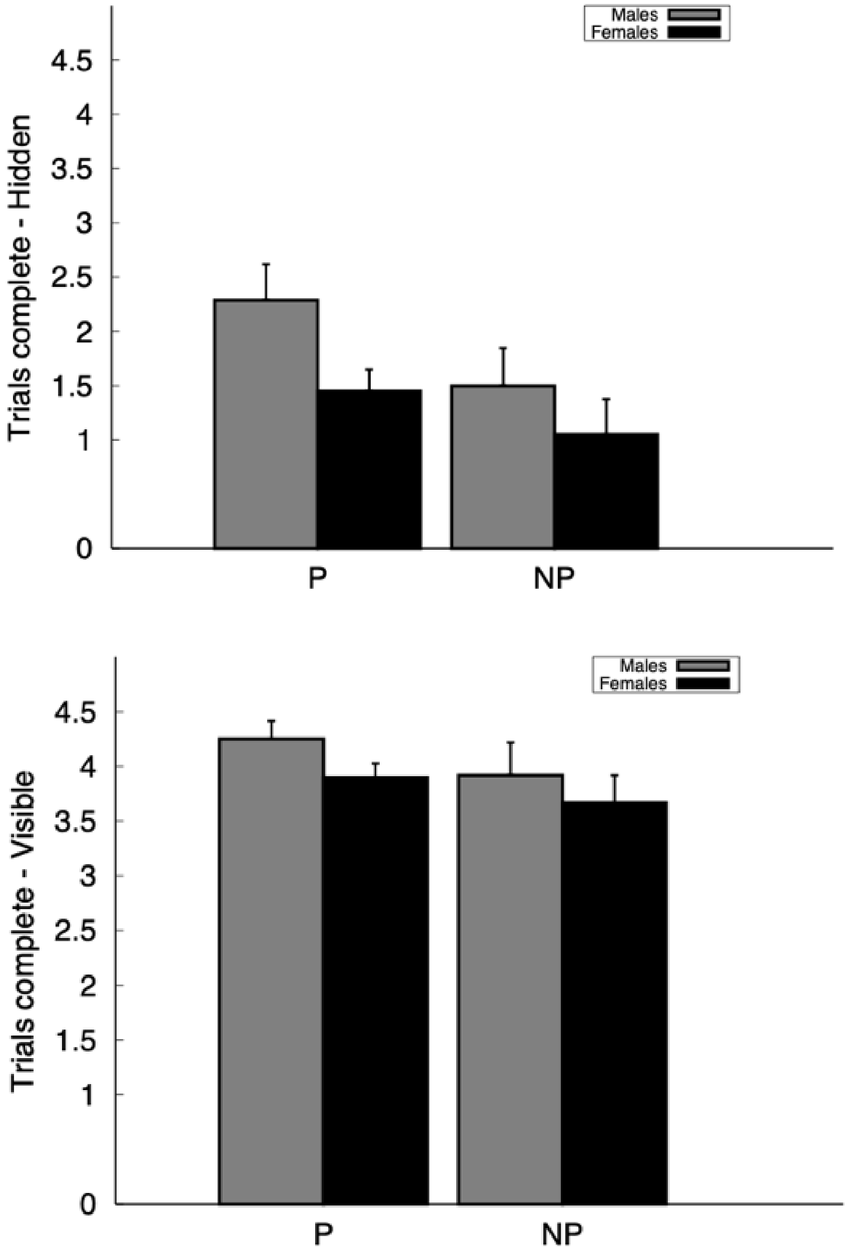

Figure 12. Mean (+ SEM) of trials complete with hidden and visible platform split by preference group and sex. There was a significant main effect of trial type (visible>hidden, p=0.012), sex (male>female, p=0.012), and preference group (P > NP, p=0.019).

fMRI vMWT probe trial

Behavioral performance

As described in the Methods, the determination of the preference group used in all analyses was based on time in region during the fMRI probe trial. Thus, the results for this variable are presented first, followed by the other measures that were not used in determining preference. For the fMRI probe trial with the pool translation, absolute and relative locations were used as the RM factors. RM-ANOVA was performed to calculate latency, path length, time in ROI, proximity to ROI, and the number of entries. Location, absolute and relative, was the RM factor.

See Fig. 13 D for mean ‘time in the region’, there was a main effect of location for ‘time in the region’ (F(1, 52)=36.761, p<0.001, η_p^2 = 0.414) with more time spent in the relative location. There was a Location by Group interaction (F(1,52)=40.809, p<0.001, η_p^2 = 0.440), with participants from the preference group for directional responding spending more time searching the relative region (Ptukey <0.001). There were no other significant interactions, ps>0.56.

See Fig. 13 A for latency, there were main effects of location (relative < absolute, F(1, 52) = 42.93 , p <.001, η_p^2 = 0.452) and group (preference < non-preference, F(1,52) = 15.169, p<0.001, η_p^2 0.226), and also a Location by Group interaction (F(1,52) = 110.025, p <.001, η_p^2 = 0.679). Post hoc tests show that the subjects with a preference for navigating with the apparatus reference frame had shorter latencies to the relative region (Ptukey <0.001). There were no significant differences between locations for the group that demonstrated no systematic preference for a strategy of navigation. There was no main effect of sex (P=0.372) and there were no other significant interactions, ps>0.35.

There was a main effect of location for proximity to the region (F(1,52) = 44.59, p<0.001, η_p^2= 0.462), as participants navigated closer to the relative location. There was a main effect of group (F(1,52) = 6.531, p=0.014, η_p^2 = 0.112), as well as Location by Group interaction (F(1,52) = 83.57, p <0.001, η_p^2 = 0.616). Post hoc tests show that the subjects with a preference for the apparatus reference frame, had navigated closer to the relative region, while the subjects from the non-preference group navigated closer to the absolute location of the platform (Ptukey<0.001).

There was a main effect of location for entries in the region (F(1,52)= 51.55, p<0.001, η_p^2 = 0.489), as there were more entries in one region compared to the other. There was a significant Location by Group interaction (F(1,52)=84.23, p <0.001, η_p^2 = 0.618). Post hoc tests indicate that participants from the non-preference group had more entries in the absolute region, while the participants from the preference group, had more entries in the relative region (Ptukey<0.001). There were no other significant main effects and/or interactions, ps>0.14.

There was a main effect of location for path length to the region (F(1,52)=21.25, p<0.001, η_p^2= 0.290) and a significant Location by Group interaction (F(1,52)= 61.98, p<0.001, η_p^2 = 0.544), with participants from the preference group having longer path lengths to the absolute region, and the participants from the non-preference group having longer path lengths to the relative location, as revealed by Post hoc tests (Ptukey <0.001). There was a main effect of group, (F(1,52)= 5.036, p=0.029, η_p^2 = 0.088), as the overall path length of the subjects in the preference group was longer than that of those in the non-preference group. There were no other significant main effects and/or interactions, ps>0.093.

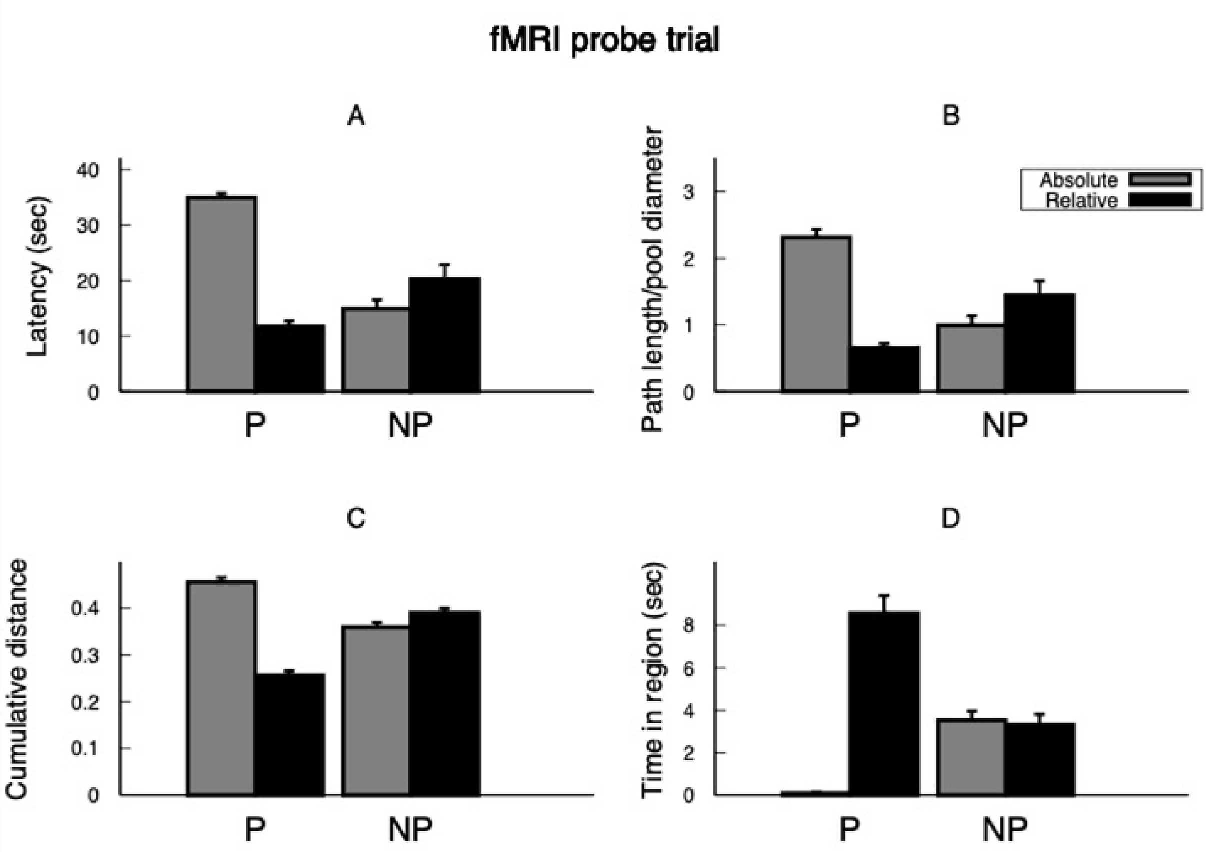

Figure 13. Mean (+ SEM) for the fMRI vMWT probe with pool shift measures: A – latency(sec), B – path length to the platform region, C – cumulative distance (proximity), and D – time in the region. All measures were split by platform region (absolute/ relative) and by preference group (P – Preference/ NP – non-preference). (For measures split by sex, see supplementary figures 15.1 and 15.2).

BOLD Activation

A contrast of the fMRI probe trial versus rest conditions with preference group (P > NP) as a factor, revealed three clusters in the left hemisphere (Fig. 14). Regions in these clusters corresponded to significant activation in the precuneus/ cuneal cortex and the superior lateral occipital cortex (OCC). The other COPEs did not reveal significantly different activation in any region. The anatomical locations of the local maxima for the clusters are summarized in Table 2.

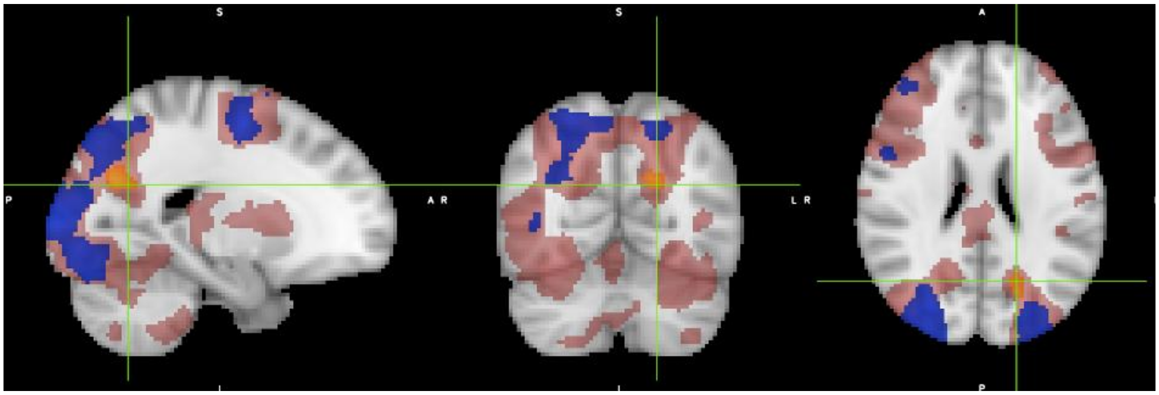

Figure 14. Regression analyses between BOLD activation (probe > rest) and preference group (P > NP) as factor. The cluster in orange shows the brain region with greater activation in the P group compared to the NP in the probe vs. rest condition ($p < 0.05$). Red is the activation for the P group and blue the areas of activation for the NP group.

TABLE 2 | Local maxima of activation: Probe vs. Rest - Preference > non-Preference as factors

Region	**Hemisphere**	**Volume (mm^3)**	**z-max** df = 55	**MNI Coordinates**		
				x	**y**	**z**
Extended region including:		226				
Precuneus/Cuneal Cortex	L		4.7	-18	-70	28
Superior Lateral Occipital Cortex	L		3.72	-30	-70	34
Superior Lateral Occipital Cortex	L		3.61	-30	-76	30

Discussion

The present findings demonstrate that a preference for directional responding is associated with distinct patterns of brain activity in the parietal cortex, specifically in the left precuneus. Many studies have shown the involvement of the parietal cortex role in the egocentric spatial information processing (Cavanna & Trimble, 2006; Clark et al., 2018; Creem et al., 2001; Grön et al., 2000; Spiers & Barry, 2015; Spiers & Maguire, 2007; Zhang & Li, 2012) as part of the network underlying navigation that includes the hippocampus (HC), parahippocampus (PHC), retrosplenial cortex(RSC), caudate nucleus (CN), and prefrontal cortex (PFC) (Clark et al., 2018; Iaria et al., 2003; Li & King, 2019; Rodriguez, 2010; Shipman & Astur, 2008; Wolbers, 2005) given the proximity of the PC to those areas. Studies of animal lesions of the PC report that the animals can learn the location of the target during spatial tasks, but are impaired in the heading direction/ orientation to the target location (impairment in proximal cue processing) (Clark et al., 2018; Save & Poucet, 2000) – which provides evidence for the involvement of the area in spatial navigation in direction and orientation processing.

Expanding from the subset data published by Sneider et al. (2018), contrasts of all the trial blocks not split by the group revealed clusters in regions that included portions of the hippocampus and parahippocampal gyrus. There was significant activation in the frontal cortex gyrus (bilateral superior frontal gyrus, bilateral MFG, ACC, paracingulate gyrus, and bilateral precentral gyrus) and the visual processing areas (bilateral fusiform gyrus, and bilateral areas of the PCC, thalamus, cerebellum, and brain stem). The activation of those areas provides evidence for the recruitment of those regions in the vMWT. After using the preference groups as factors, the results showed that both groups had similar BOLD patterns of activation not showing differences in discriminating the groups during the blocks before the probe trial. The pool translation probe trial, however, showed greater activation of the left precuneus in the P group – in addition to the other areas recruited during navigation – which provided neural evidence for group discrimination based on the strategies developed during the learning trials.

This study sought to demonstrate that people can be categorized into distinct groups based on their strategies of navigation developed in the learning phase of the task. It was hypothesized that most people would demonstrate a preference for navigating by following a direction (within the apparatus reference frame) in the virtual MWT. To discriminate strategies of navigation based on the apparatus reference frame - directional responding - or within the room reference frame - place navigation -, a probe trial manipulation in which the pool was translated into a new location in the room with one quadrant overlap was used (Hamilton, Johnson, et al., 2009; Stackman et al., 2012). The results of behavioral performance allowed for the determination of two distinct groups. Navigating by following a direction was classified as “Preference (P)”; other types of navigation – either by using the room as a reference frame or by not spending a significant amount of time in a specific location of the pool – were then classified as “non-Preference (NP)”.

Qualitative analysis of all the swim paths of both NP and P groups does not show evidence of change in the search strategy used in the learning blocks probe trial and the fMRI pool shift probe trial. The patterns of the swim paths from the learning probe trial and the fMRI pool shift probe trial for each participant are comparable. That suggests that people engaged in the same strategies in both the learning probe trials and the fMRI pool shift probe trial. In addition to the statistical analysis, qualitative analysis of the learning blocks' swim paths provide evidence for direct trajectories to the goal location with an increase of trial block for both P and NP groups supporting similar learning in both groups. However, it seems that the absence of the platform was disruptive as most people in the NP group shifted from directed trajectories to chaining or circling strategies. Engaging in those strategies means that people learned the fixed distance between the platform and the pool boundary (Astur et al., 2004; Graziano et al., 2003; Higaki et al., 2018) which is reliable and increases the probability of finding the platform (Astur et al., 2004; Graziano et al., 2003). However, circling strategy may not be a direct predictor of spatial performance (Kallai et al., 2005) as circling the perimeter of the pool is a non-spatial strategy (Astur et al., 2004; DiMattia and Kesner, 1988; Rogers et al., 2017) that does not rely on the distal cues. A few people from the NP group engaged in other types of nonspatial strategies such as scanning and random search. Most people in the P group seem to have expanded the direct trajectory to variations of focal swim and/or direct search (Rogers et al., 2017). Kallai et al., (2005) describe a temporal shift in strategies with increased experience in the task. The findings of the present study support the dynamic changes in both groups and provide evidence for the

P group persisting in using spatial strategies – distal cues for orientation and local cues for search specificity - when the pool was shifted in the room, while the NP group appears to have shifted from spatial to non-spatial strategies relying on the boundary of the pool to locate the platform.

The behavioral results demonstrated that while performing on the virtual MWT pool translation probe trial, most people navigated by following a direction while very few people navigated within the room reference frame, and some did not demonstrate a systematic strategy, spending similar time in both relative and absolute regions. The fact that only 3/56 people demonstrated a clear indication of navigation within the room reference frame - place learning -, is consistent with rodent studies that attempted to differentiate that type of learning from directional responding, concluding that place learning is difficult to acquire possibly due to starting point similarity (Akers et al., 2007; Blodgett et al., 1949; Skinner et al., 2003; Stringer et al., 2005). Studies report that animals solve the task of locating the hidden platform in the MWT by following a direction in the room (Akers et al., 2007; Hamilton, Akers, et al., 2009; Hamilton et al., 2008) rather than navigating to a specific place in the room. Hamilton et al. (2007, 2008, 2009) suggest that directional responding is preferable over place navigation because that type of navigation involves different processes controlled by both the distal room cues and the apparatus cues. Those processes can be understood as sequential in which the distal cues control the orientation of the initial search movement toward the platform followed by a refinement of the search controlled by the distance of the pool border and the platform location (Hamilton et al., 2004). An analogy made by Olton

(1979) compared navigating to a precise location in the room as a dot in the cartesian system, whereas orientation in the environment involves a vector, or following a line and a direction along an axis. It is important to note that these processes are not mutually exclusive, but rather difficult to differentiate.

Analysis of the learning blocks provided evidence that all participants included in the study learned the task before being tested in the scanner. In the hidden blocks the main effects of trial blocks for latency and path length support learning as latencies decreased and path length was shorter as trial blocks increased. Both preference groups had similar results, however, the NP group showed a longer path length compared to the P group. Sex differences were evident in the learning blocks in the latency measure (duration between initiation of the first movement and finding the platform) as females took longer than males to initiate the first movement. In the offline probe trial, males had more entries in the ROI than females, and people from the P group navigated closer to the ROI as revealed by the proximity measure. There were no other sex differences in the offline probe trial in comparison to the hidden blocks. That is possible because the females continued to improve their performance throughout the blocks as evidenced by the significant difference in latency that was significant in the hidden blocks but decreased with more blocks, which suggests that by the probe trial, the females reached a similar level of performance to males, even though numerical differences were showing a male advantage in overall performance. This is consistent with what was reported by Sneider et. al (2018), with a subset of 32 participants from the present study. Many factors could be attributed to sex differences or lack thereof. Studies of sex differences agree that males and females perform differently in spatial tasks when encoding the features of the environment by using allocentric and egocentric frames of reference. Females are more likely to rely on landmarks while males are more likely to use the geometrical information (Devan et al., 2002; Forcano et al., 2009; Nowak et al., 2015). Numerous studies provide evidence for sex differences in spatial navigation tasks (Astur et al., 1998, 2004; Forcano et al., 2009; Maguire et al., 1999; Noachtar et al., 2022; Nowak et al., 2015; Sneider et al., 2015; Woolley et al., 2010), some studies did not find sex differences (Sneider et al., 2018) or did not find it in certain conditions (Chamizo et al., 2011; Sandstrom et al., 1998; Woolley et al., 2010).

fMRI showed significantly more activation in the left precuneus/ cuneal cortex and Superior Lateral Occipital Cortex in the P group. There were no significant differences in hippocampus activation to discriminate between groups suggesting that the regardless of the strategy developed in the learning trials, the hippocampus is recruited similarly in both groups; the greater activation of the precuneus/ cuneal cortex is consistent with the hypothesis that greater activation of the parietal cortex would be evident in the P group as directional responding involves visual processing in the network that includes the hippocampus and interconnected regions. The precuneus - an area of the parietal cortex – is involved in a large array of tasks that are interconnected, such as visuospatial imagery, spatially guided behavior, and episodic memory. (Cavanna & Trimble, 2006; Zhang & Li, 2012)

It is well established that spatial navigation involves a constellation of processes. In this paradigm of vMWT, it is required that people learn the features – distal and proximal cues – of the environment to locate the hidden platform from different release points. The probe trial possibly has a higher cognitive load than the hidden trials because after learning the task and increasing the possibility to complete more trials per block, not finding the platform in the probe trial can be disruptive. The pool translation probe trial can be even more challenging as the reference frames are shifted. Not finding the platform where it was expected to be may require a change or adaptation in planned strategy that involves mental planning (Bocchi et al., 2019; Taube et al., 2013). Even though in this study the participants were not explicitly asked to engage in motor imagery, retrieving the information of the location of the platform in relation to the distal and proximal cues, implicitly requires imagination. Studies of motor imagery have shown activation of the precuneus when participants were asked to imagine walking in several settings (Malouin et al., 2003), to imagine hurdling (Ogiso et al., 2000), or imagine rotating an object or engaging in whole body self-rotation (Creem et al., 2001). The precuneus is also involved in spatial updating that contributes to the maintenance of orientation and egocentric representation of environmental cues (Galati et al., 2010). Collectively, the behavior results with the BOLD activation suggest that people from the P group were better able to use the distal cues for orientation in the environment combined with the apparatus boundaries to guide local search.

Limitations and future directions

The precuneus is part of the spatial navigation network but is also one of the hotspots of the default mode network (DMN) with a high metabolic rate at rest. The area has been of interest because of the challenges in determining whether cognitive tasks will show activation or deactivation of the area (Cavanna & Trimble, 2006; Ogiso et al., 2000; Utevsky et al., 2014). The present study was not designed to specifically evaluate the DMN. The activation of the precuneus during the spatial navigation task is consistent with reports of the parietal-retrosplenial network supporting allocentric to egocentric transformations (Bicanski & Burgess, 2018, 2020; Byrne et al., 2007; Clark et al., 2018) and the specific role of parietal-retrosplenial network in heading direction and imagined location (Baumann & Mattingley, 2010; Marchette et al., 2014). The precuneus may have distinct specialized roles within different networks beyond the DMN that yield domain-specific interpretations (Cavanna & Trimble, 2006; Salgado-Pineda et al., 2017). Therefore, future research is needed to assess patterns of activation or de-activation of the precuneus in different network contexts.

A possible explanation for the lack of sex differences in the current study could be related to the cognitive assessment scores. None of the WASI scores showed statistically significant differences, however, females performed slightly better in the block design and almost similar to males in the mental rotation task, even though the mental rotation is known to produce robust sex differences with male advantage (Parsons, 2004; Rahe & Jansen, 2022). The IQ of this cohort of adolescents was also above ~1,5 SD above average, thus the lack of sex differences may not be representative of the general population. Correlation of neuropsychological assessment scores with performance is suggested as well as replication of the study with a larger cohort that could capture more of the variability of the general population.

This study was performed in adolescents – which is novel in the field but also has no other studies available for comparison. Additional studies investigating this age group is needed, as well as replication in older population to assess differences in brain activation in this pool shift probe trial paradigm. A study done by Hamilton et al. (2008) using the same paradigm investigated a population of adults aging 18-37 years old, which provides behavioral data for comparison. Their study - similarly to animal studies - reported a preference for directional responding over place navigation, which is supported by the current study. However, their study did not include neuroimaging techniques. Studies investigating the relationship between age, strategies of navigation, and patterns of brain activation have demonstrated that with age there is a shift in spatial memory functions away from the hippocampus (Bohbot et al., 2012; Reynolds et al., 2019).

Another possible future direction could include eye-tracking to assess attention. As demonstrated by Hamilton et al. (2008) in their Experiment 4 using the vMWT, the inclusion of eye-tracking provided evidence for what people were attending to in the environment when learning the task. Their results support the sequential aspect of directional responding that involves attending to distal cues for orientation in the early phases of the trial, then switching to the boundaries of the pool for local search.

However, in their experiment with eye-tracking, they did not include the pool translation probe trial. A future study could include eye-tracking to compare attention shifts during learning trial, no-shift probe trial, and pool translation probe trials in a younger population.

Conclusion

To date, this is the first study to address strategies of navigation in healthy adolescents in the fMRI vMWT by using a pool translation probe trial. This contributes to the existing literature expanding on studies of spatial navigation - investigating networks involved in navigation including the hippocampus and beyond - commonly done in animals and older human populations. Although small, the significant difference in BOLD activation of the precuneus/ cuneal area during the probe trial - in the group that showed a preference for orientation by following a direction to the target location – helps to elucidate strategies of navigation people develop during a spatial navigation task and the neurobiology supporting the behavior. Thus, more investigation of the areas involved in spatial learning and memory – including the parietal cortex; particularly the precuneus – on how those areas contribute to each other, is needed.

Supplementary figures

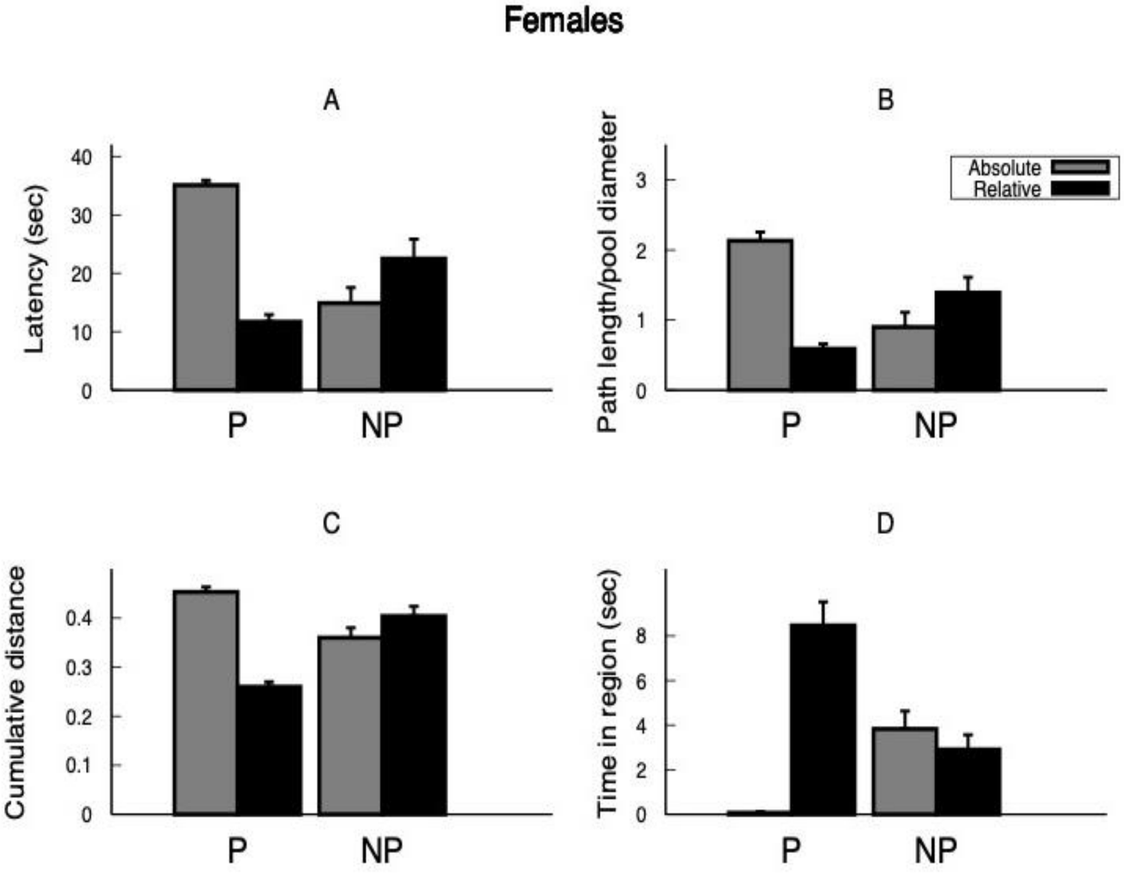

Figure 15. Mean (+ SEM) for the fMRI vMWT probe with pool shift measures: A – latency(sec), B – path length to the platform region, C – cumulative distance (proximity), and D – time in region. All measures were split by platform region (absolute/ relative) and by preference group (P – Preference/ NP – non-preference).

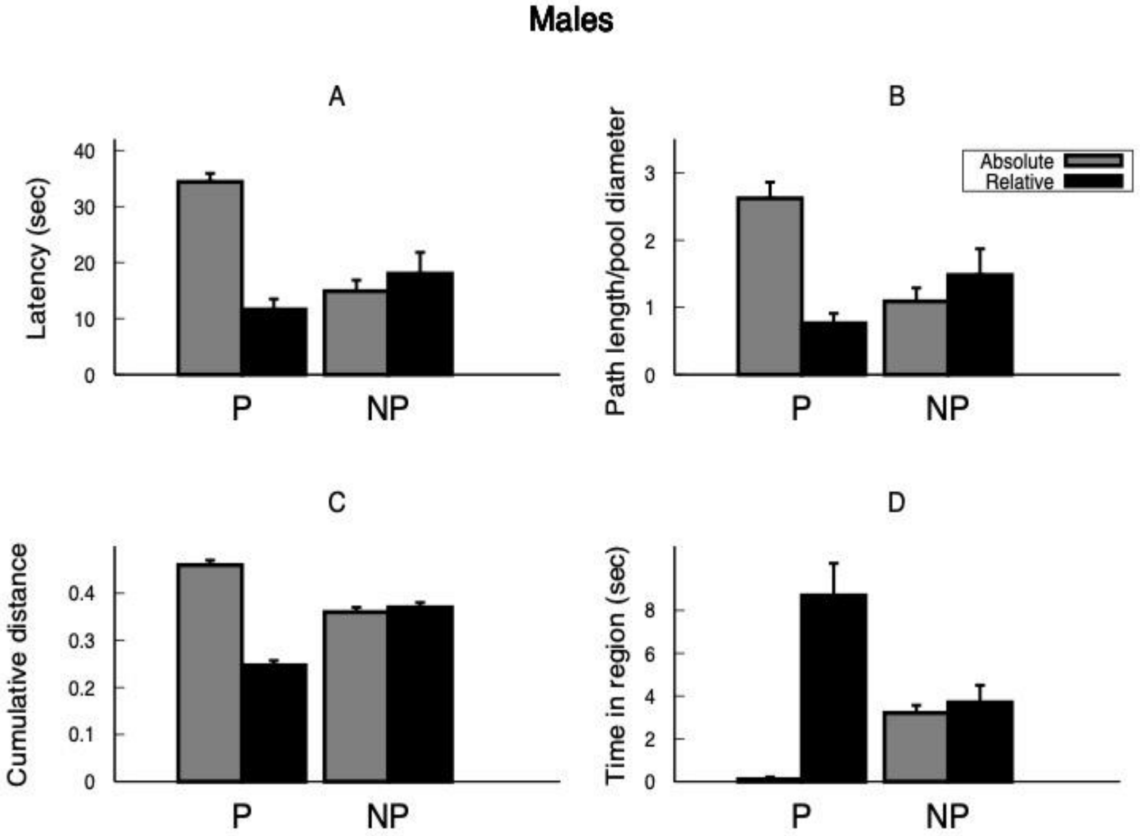

Figure 15.2. Mean (+ SEM) for the fMRI vMWT probe with pool shift measures: A – latency(sec), B – path length to the platform region, C – cumulative distance (proximity), and D – time in region. All measures were split by platform region (absolute/ relative) and by preference group (P – Preference/ NP – non-preference).

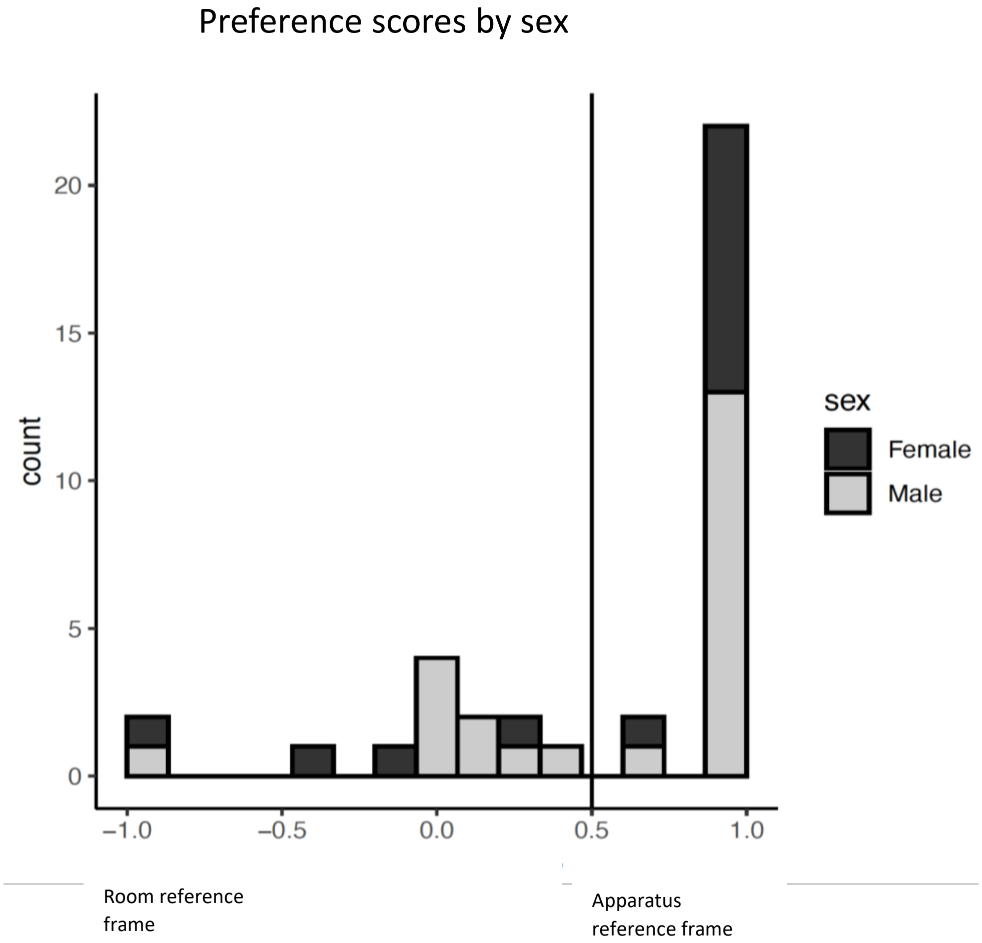

Figure 16. Histogram of preference scores ranging from - 1 to 1. The score of 0.5 was used as the threshold to split the preference groups. A score of -1 refers to the room reference frame, and a score of 1 refers to the apparatus frame of reference. Scores of -1 were rare and were grouped with those people who did not show a preference for either location. All score > 0.5 were placed in the Preference (P) group and all scores < 0.5 were placed in the non-Preference group (NP).

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