Development and Evaluation of a Rat Water Maze System: Impact of Maze Size, Water Temperature and Other Parameters on Performance

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The Morris water maze (MWM) was first described over thirty years ago and is still one of the most commonly used cognition paradigms to measure spatial learning and memory in rodents. Throughout the literature, there are numerous and conflicting methodological approaches described but surprisingly very little is published concerning the impact of such procedural variables on performance. The aim of the present studies was to conduct a comprehensive assessment of variables such as pool size, rat strain, saliency of spatial and visual cues, training procedure and water opacity/temperature on a fixed platform acquisition water maze paradigm. The ultimate aim of these studies was to identify the most appropriate parameters to facilitate robust and reproducible learning curves for subsequent pharmacological studies.

For these investigations two mazes of differing diameters (1.4 and 2.0 m) were installed in separate procedure rooms that were near identical with respect to room size and internal layout. Preliminary studies focused on ensuring that both mazes were free of any potential confounding spatial bias. These studies comprised of determining the time spent in each of the four quadrants during a predetermined training time. Data from these studies demonstrated that both water mazes and their procedure rooms were free of any spatial bias that may affect performance in subsequent tests. As there are numerous publications from groups using non-oppacified water during their tests, the next study aimed to determine whether rats are able to visually locate the platform by diving. Using the small maze only, a single study was conducted using clear and oppacified water. Interestingly, there were marked differences in performance with animals trained in clear water locating the submerged platform significantly quicker than those trained in oppacified water. Unless this was previously examined, it is quite likely that false interpretations of spatial learning could have been made in these prior published studies.

Another methodological variation in published studies is the use of a pre-training day where a curtain is drawn around the tank shielding the extra-maze spatial cues and a solid visual cue is suspended over the submerged platform. The intention of this pre-training day is to reduce anxiety and associated thigmotaxic behaviors prior to spatial cue testing. In our studies, there were clear benefits of pre-training on subsequent spatial learning. Essentially, animals that were trained to locate the platform using a suspended visual cue on day 1, performed significantly better during the subsequent spatial cue training sessions. Whilst it is generally perceived that there is an optimal water temperature for water maze studies, in our studies there were no effects of water temperature (21, 23 and 25 ± 1 ºC) on spatial learning performance. During the spatial learning comparison studies between the two mazes, it become evident that performance of animals trained in the smaller of the two mazes (1.4 m) was considerably more variable between individual session and across studies. As reproducible learning curves were only evident in the larger maze, the final series of studies dedicated to examining the impact of spatial cues and rat strains were performed in the larger maze only. Surprising, whilst it is generally accepted that additional spatial cues (posters, lights etc) aid formation of enhanced spatial maps, in our studies, there was no difference in performance when these additional cues were added to the room.

The final series of studies aimed to quantify any potential performance differences between two hooded rat strains (Long Evans and Lister Hooded) and the most commonly used albino rat strains (Sprague Dawley and Wistar). Surprisingly, whilst it is generally accepted that hooded strains have enhanced visual acuity compared to albino animals, there were no significant differences between all four rat strains as measured by overall latency and path length to locate the submerged platform. There was

however a significant difference in swim speed between the Wistar strain and the other three rat strains, with Wistars consistently swimming faster across all trials/sessions. Although there were no overall significant differences in performance measures, there was however, a marked difference between the hooded strains and albino rat strains in time spent in periphery suggesting that hooded strains are less anxious in this task.

In conclusion, there were clear effects of maze size, use of visual cue, oppacified vs. clear water and subtle differences between rat strains in water maze performance. However, there were no effects of water temperature or use of additional spatial cue on performance.