CHAPTER III
MATERIALS AND METHODS

3.1 Study design

The two experiments were conducted serially. Study designs in this study were matched-pairs designs in the first experiment and randomized controlled trial in second experiment. The first experiment investigated deteriorated reach-to-grasp (RTG) performance in Parkinson's disease (PD) patients by comparing the planning time, kinematic parameters and coordination between PD and non-disabled groups. Then, the PD participants from first experiment and further recruitments were invited to train RTG in second experiment. .

After enrollment into the second experiment, participants were equally allocated into 3 groups; action observation (AO), Placebo (P), and control (C) groups. The second experiment were divided into 2 distinct phases: acquisition phase and retention test (45 minutes after the end of training phase).

3.2 Research participants and recruitments

Participants in this study were recruited from Physical Therapy Clinic Mahidol University, Siriraj hospital, and Golden Jubilee Medical Center Mahidol University. Researcher met and invited patient one per one at rehabilitation center and ask therapists who work at hospital or rehabilitation center for distributing the brochures. In case of patients agree to participate in this study when researcher met them, they were physically examined by researcher. The patients who received the brochures and agreed to participate in this study, they were screened by Phone Screen Test or by medical records prior an appointment for physical examination. Healthy non-disabled adults were recruited from elderly society at Golden Jubilee Medical Center Mahidol University and Amornchai village, or volunteers who were caregivers of recruited PD patients.

3.2.1 Inclusion criteria for non-disabled participants

1) No history of neurological or psychological dysfunction

2) No history of skeletomotor dysfunction of right arm and hand

3) Able to follow command (score more than 23 by Mini Mental State Examination or MMSE Thai version 2002)

4) Have normal or corrected to normal vision and normal hearing

3.2.2 Inclusion criteria for PD participants

1) Diagnosed with idiopathic PD or primary parkinsonism

2) Age between 35 to 80 years

3) Hoehn & Yahr stage range from II-III

4) Could understand and follow command (score of MMSE Thai version 2002>23) (151)

5) Could independently sit at least 1 hour

6) Could perform reaching for grasping the object that place 30 centimeters in front of sternum at least 1 time.

7) Have normal or corrected to normal vision and normal hearing

3.2.3 Exclusion criteria for PD participants

1) Diagnosed with secondary parkinsonism which caused by certain medicines, a different nervous system disorder, or another illness such as progressive supranuclear palsy, multiple system atrophy and drug-induced parkinsonism (e.g. Antipsychotics, Metoclopramide, Phenothiazine medications).

2) Other neurological and orthopedic conditions affecting the arm, hand or trunk that may have interfered with task accomplishment. For instances, arthritis in the dominant upper extremity or upper extremity weakness that would prevent task performance.

3) Severe action and resting tremor as evidenced by score ≥ 3 in domain of action or postural, resting tremor of hands.

4) Severe rigidity as evidenced by score ≥ 3 in domain of rigidity of hands.

5) ON/OFF medication fluctuations that would prevent performance of the experimental task or confound interpretation of the acquired kinematic data. Clinical

fluctuations part in Unified Parkinson's disease rating scale (UPDRS) was required to assess ON/OFF medication fluctuations in Parkinsonian participants.

6) PD patients who have history to treat by deep brain stimulation surgery
7) Receive upper extremity training during the time in this study
8) Psychiatric illness, alcohol or substance abuse during the time of study
9) Sustained and poor controlled depression or anxiety (measured by Thai Hospital Anxiety and Depression Scale)

3.2.4 Withdrawal criteria

1) Cannot participate to test or train completely
2) Unwilling to be participate in this study

3.3 Sample size

Before started this study, researcher conducted the pilot study for using the data to calculate the sample size by using this equation.

$$***** \quad N = \frac{2\sigma^2(Z_{\alpha/2}+Z_{\beta})^2}{(\mu_1-\mu_2)^2} \quad *****$$

N = number of participants per group

$Z_{\alpha/2}$ = Z-value when set the confidence level equal to 95% or significant level as 0.05 (=1.96)

Z_{β} = Z-value when set the power testing equal to 80 % or β equal to 0.2 (=0.842)

σ^2 = pooled variance

μ_1 = the means of parameters in PD group in the *first experiment*

= the different means of parameters between posttest and retention test of AO group in the *second experiment*

μ_2 = the means of parameters in non-disabled control group in the *first experiment*

= the different means of parameters between posttest and retention test of C group in the *second experiment*

3.4 Group allocation

3.4.1 First experiment

In this experiment, participants were PD individuals and non-disabled controls. Thus, the division depended on characteristic of participants. The data collection was started from PD group first, then recruited healthy participants who were matched in age range (± 5 years) as PD participants.

3.4.2 Second experiment

All PD participants were randomly assigned into 1 of 3 groups by stratified randomization with age range, Hoehn and Yahr stage and more affected side. Participants who have same characteristics were grouped into same stria, then participants in each block were randomly selected into each group.

3.5 Instruments

1) Electromagnetic tracking motion system (MotionMonitor, Innsport, Inc.) (Figure 3.1) with

1.1) Three six degree-of-freedom Mini-Bird sensors

1.2) Object (Square shape; width 2.5 cm, height 10 cm)

1.3) Barrier (Cylinder shape; diameter 2 cm, height 30 cm)

1.4) LED visual signal that synchronized to the system

2) A table and a height-adjustable chair

3) Video movies of hand movement which were performed by Parkinsonian models, who had stage I-I.5 of Hoehn and Yahr stage and were passed other all criteria, and natural perspectives slide show. These videos were developed by experimenter.

4) Video camera and tripod for record all performance of participants. The recorded movement were used it for discussion if occur unexpected results.

5) Evaluation form and questionnaire which will be used during the process of selection criteria and data collection.

5.1) Phone screening questionnaire

5.2) Edinburg handedness inventory

5.3) Hoehn and Yahr scale

5.4) Unified Parkinson's disease rating scale (UPDRS)

5.5) Thai Hospital Anxiety and Depression Scale (Thai HADS)

5.6) Mini mental state examination (MMSE) Thai-version 2002

5.7) Questionnaire regarding the advantages of training conditions such as the action understanding, enhancing memory, error detection, error correction and motivation.

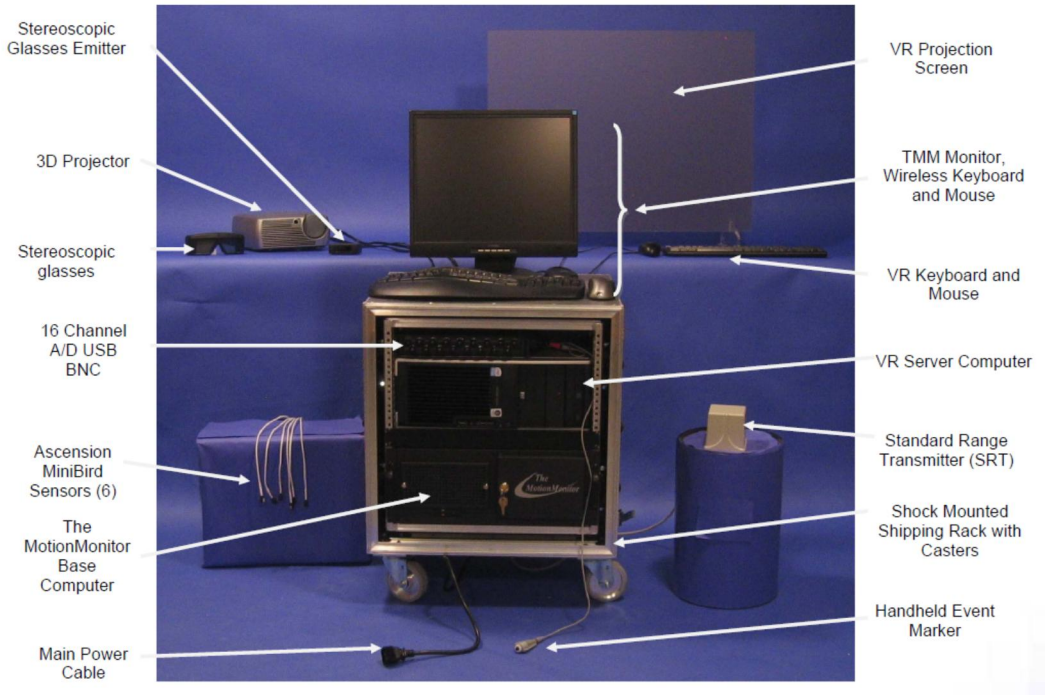


Figure 3.1 Electromagnetic tracking motion system (figure from manual guidelines of MotionMonitor, Innsport, Inc.)

3.6 Experimental procedures and tasks

3.6.1 First experiment

The procedures in this study was briefly shown in flowchart (3.11 Flowchart of procedure of the study). After all participants completed the interview screening, and

were enrolled by inclusion and exclusion criteria. They were informed about purpose and study processes. Researcher verbally instructed to the participants regarding the task and demonstrated how to reach and grasp in barrier avoidance condition for 3 times. Then, examiner attached the sensors on participants' right nail bed of thumb and index, and radial styloid process. All participants were instructed to practice before the starting of testing process for 4 trials; 2 for preferred speed and another 2 for fast speed for differentiate between both speeds. Then, they performed RTG under barrier avoidance condition (Figure 3.2) with more affected hand. They were instructed to reach immediately after seeing the visual signal and as fast as possible without a collision of the barrier, grasp the object with the thumb and index finger, lift it off the table (shoulder level) and place it to the same location. Researcher asked them to perform these movement with their best. This task was chosen because it requires transport and grasp coordination which might be impaired in PD, and was challenged and motivated task for PD patients. The data was collected for 10 successful trials. The unsuccessful trials were excluded and were recorded the number into information sheet by researcher.

3.6.2 Second experiment (see Flowchart)

All PD participants from first experiment and further recruitment were asked to participate into training session in second experiment. All participants were trained reaching for precision grasping with a randomly assigned training conditions.

At "Pretest" or prior to training sessions, dexterity part of the Wolf Motor Function test (WMFT) including lifting can, lifting pencil, lifting paper clip, stacking checkers, turning the key, and fliping cards were test with as fast as possible speed. Additional test was 10 trials the RTG with barrier avoidance condition which could be measured RTG planning, kinematics and coordination.

Then, the training sessions were done after Pretest. The duration of each training condition was approximately 45 minutes. Before training, researcher gave instructions to participants to perform the tasks the same as 6 items of WMFT as quickly as possible for 24 trials or 4 trials per item, and alternate with resting or observing depending on the groups they were allocated (see details in training condition section).

Then, "Posttest" and "Retention test" were assessed all measures again immediately and 45 minutes later after completing the training.

After complete retention test, all participants were asked to complete questionnaire regarding the advantages of training condition such as the action understanding, enhancing memory, enhancing attention, error detection, error correction and motivation for further analysis regarding the underlying mechanisms of AO.

3.7 Experimental setup

For the setting up (Figure 3.2), the patients were asked to place their index and thumb on start switch every trials before seeing the visual signal. The object to be grasped was a square shape (2.5 centimeters in width, and 10 centimeters in height) located at 30 centimeters along the midline of the trunk from the area at which the hand is placed on the start switch. The barrier was a cylinder shape (2 centimeters in diameter and 30 centimeters in height) located at 15 centimeters in to the front of a start switch and 2.5 centimeters away from the midline from the start switch to the target object.

Participants sat 2 meters in front of the television which displayed the video. However, the distance between television and participants was adjusted until they see the movie clearly.

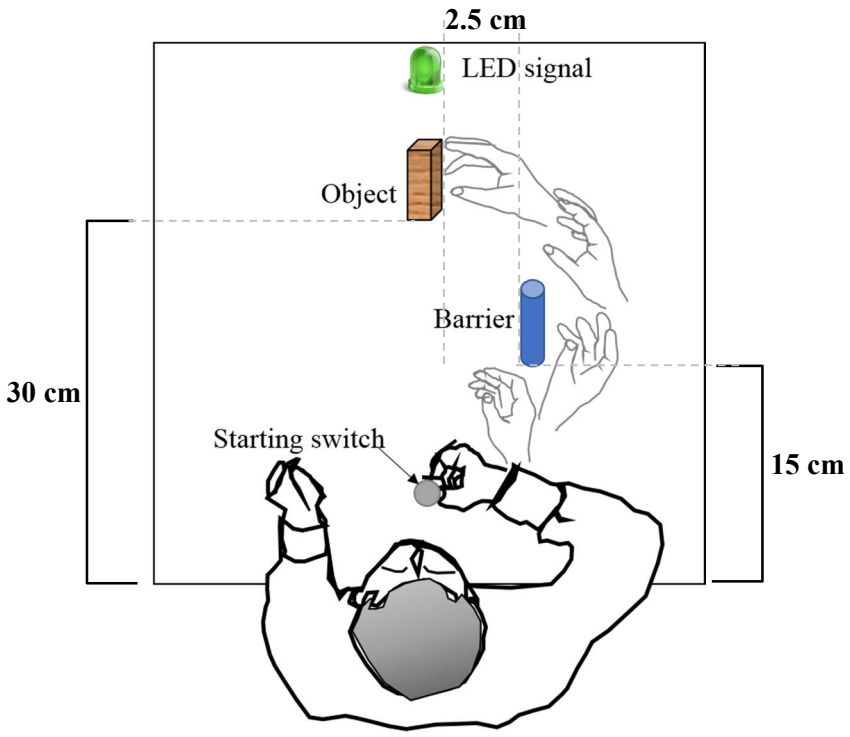


Figure 3.2 Characteristics of RTG under barrier avoidance task

3.8 Training groups

The training composes of 3 groups including

3.8.1 Action observation (AO) group

Participants were received the observation of RTG actions for 6 minute alternate with physical training for 24 trials. The training sessions consist of 4 blocks of observation and 4 blocks of physical training. Prior to training session, participants were instructed to watch video sequence carefully, imagine that they are performing without any movement of upper extremities, and reading the description in video for more clear understanding. During observing, participants saw 6 hand actions within 6 minutes (1 minute for each action). The orders of these actions were consistent in all observing blocks. Then, they were asked to perform the same action as they saw in video with as fast as possible speed during physical training period for 24 trials or 4 trials per task. The training processes were alternately between observing period and physical training as aforementioned descriptions until complete 4 physical training blocks with 4 observing periods. During observing, patients' attention were controlled by instructing the patients that they need to answer the question regarding the details in video at the end of each observing period (see appendix F).

3.8.2 Placebo (P) group

The protocol was the same as the AO group, but patients saw the video of various natural perspectives slide show instead of the video of hand movements. Participants were instructed to watch video sequence carefully.

3.8.3 Control (C) group

The training protocols was similar to the first group, but the resting was used instead of the observation. Participants were instructed to rest for 6 minutes without sleeping or any moving of UE.

3.9 Movement recording and data processing

Kinematic data during each testing time was recorded by using an electromagnetic tracking motion system (MotionMonitor, Innsport, Inc.), with six degree-of-freedom Mini-Bird sensors (Ascension Technologies).

Three sensors were attached on the upper extremity (a wrist sensor on the styloid process and other two sensors on the nail beds of the thumb and index fingers).

The sampling rate for the three sensors was 100 Hz.

During movement recording, the video camera was recorded from above and beside the testing hand of the participants.

Electromagnetic tracking motion system detected the x, y, and z coordinates of the wrist and the fingers sensors which will be then filtered to minimize distortion of the movement. All kinematic data was filtered using a zero-lag Butterworth low-pass filter with a cut-off frequency of 20 Hz.

3.10 Dependent variables and data analysis

Data analyses were performed using the SPSS for Windows release 18.0 (Chicago, IL). The significant level was set at p-value less than 0.05. Kolmogorov-Smirnov goodness-of-fit Test was used to determine the distribution of the data. Descriptive statistic was used to analyze demographic and clinical characteristic of all participants.

3.10.1 First experiment

The dependent variables in first experiment (Figure 3.3A, 3.3B) included: 1) Reaction time (RT) was time of visual signal onset until movement initiation, 2) Kinematic variables consist of 2.1) Movement time (MT) was time from movement initiation to movement termination, 2.2) Transport variables consisted of maximum transport velocity (V_{max}), time to maximum transport velocity (TV_{max}), and Deceleration time (DT) (the time from maximum velocity to movement termination), 2.3) Grasping variables consisted of maximum aperture (A_{max}), time to maximum aperture (TA_{max}), Aperture closure time (ACT) (the time from maximum aperture to the moment of object grasp) and Aperture closure distance (ACD) (the distance traveled by the thumb from maximum aperture to the time of object grasp). Figure 3.3C represents the Transport-

grasp coordination, that was assessed by using a cross correlation analysis between transport velocity and aperture size which consisted of 2 variables; highest cross correlation coefficient (r_{max}) and associated time lag for which the highest correlation coefficient (T_{max}). The high r_{max} represents the most similar pattern between transport velocity trajectory and grasp aperture. The transport velocity trajectory was shifted until reach the time at highest r_{max}, which indicate the T_{max}. In Figure 3.3C, upper left corner represents transport velocity trajectory (gray) and aperture (black) at zero time lag (T_{max} = 0 ms) with the r_{max} was 0.36. Lower left corner shows the highest correlation coefficient (r_{max} = 0.97) after move the shifted the transport velocity trajectory for 200 ms (T_{max} = 200 ms) to achieve the most similar pattern with grasp aperture trajectory. The positive T_{max} indicate that grip aperture start to open after the onset of hand's transporting and vice versa.

Average value of each variable of PD and age-matched groups were compared using one way analysis of variance (ANOVA).

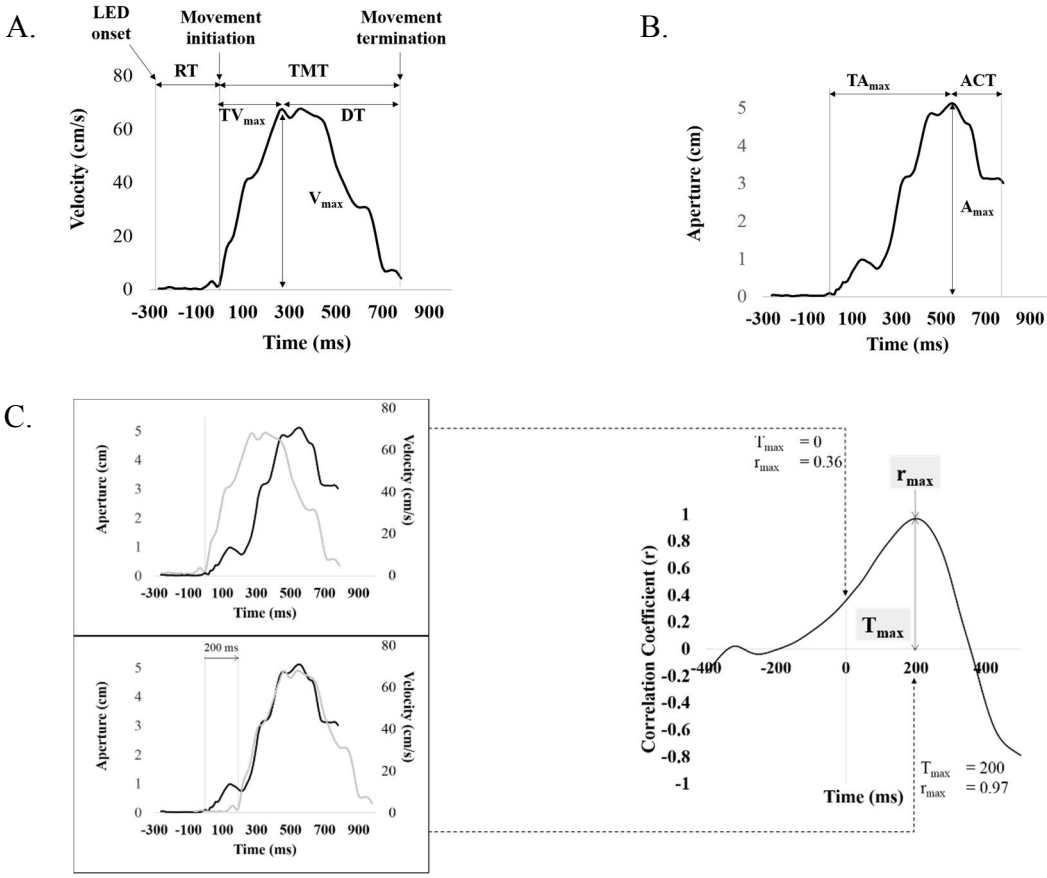


Figure 3.3 All variables of RTG planning, kinematics and coordination

3.10.2 Second experiment

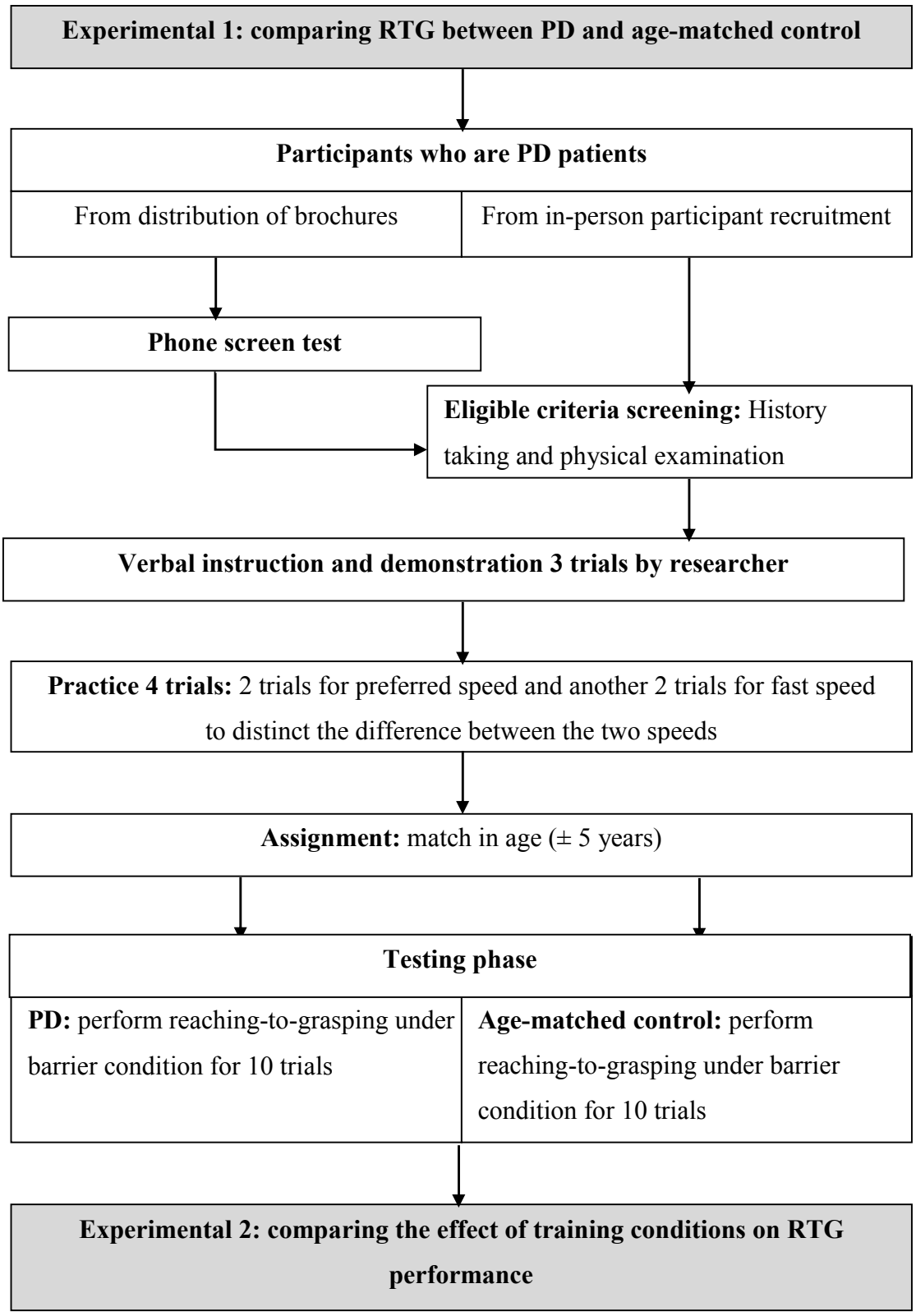
In second study, the dependent variables in trained-task was the total time of WMFT. The dependent variables in untrained-task were similar to all variables in first experiment.

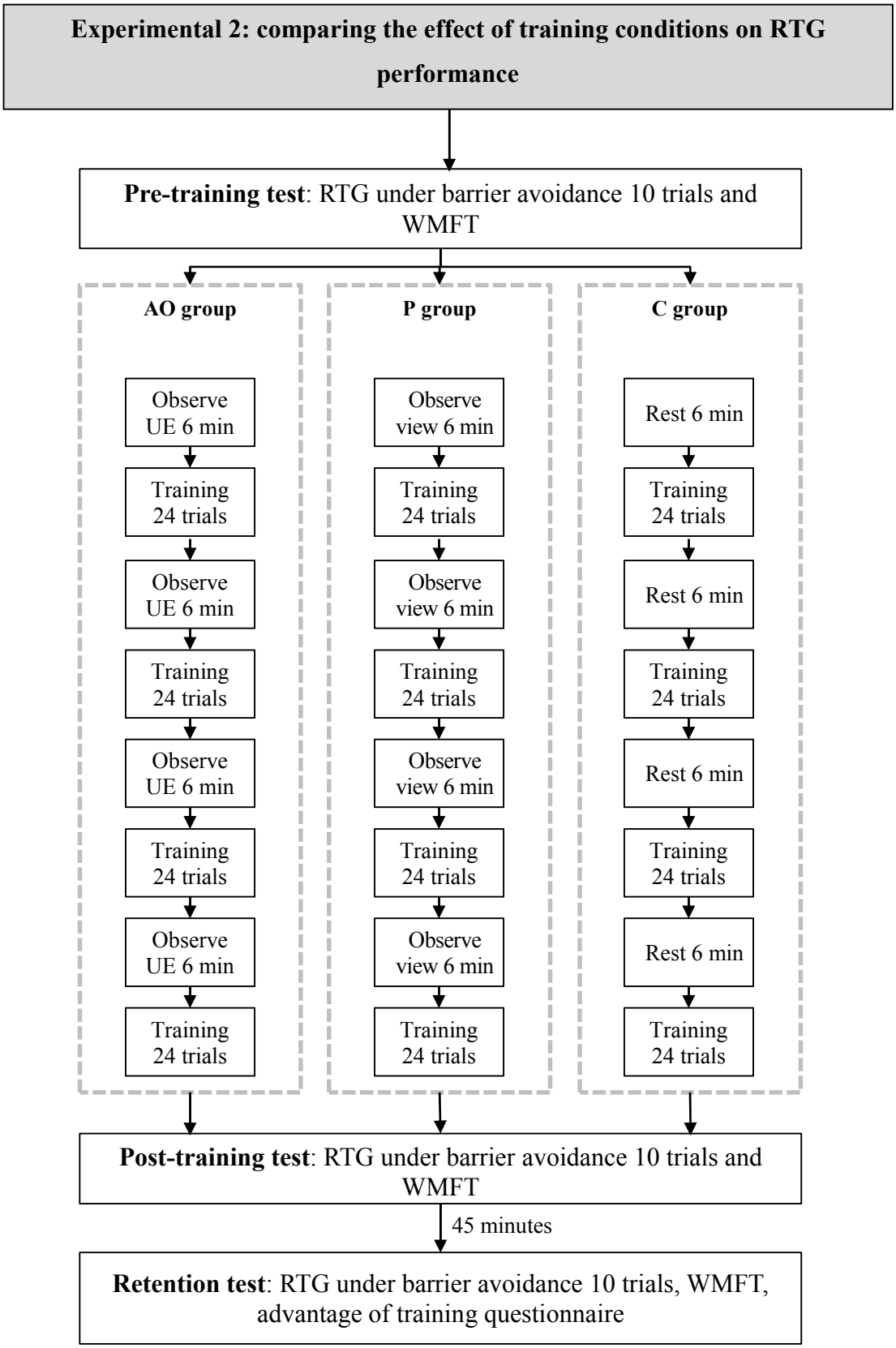
All dependent variables were analyzed separately in a mixed repeated-measure analysis of covariance (RM ANCOVA) with the factor testing time (Pretest, Posttest, Retention test) as a within-subject factor and Group (AO, P, C) as between-subjects factor and with the baseline data at Pretest was covariate. Each dependent variable at each testing time was adjusted by the mean covariate or mean Pretest data of all participants. Therefore, the baseline data at Pretest was the same across groups and the mean of dependent variables at Posttest and Retention test were also adjusted by the mean of covariate.

Multiple comparisons for repeated measures using Bonferroni were used to compare the difference of each variable between 3 testing times in each group and between 3 groups in each testing time.

If the data are not normally distributed, the non-parametric statistics was used instead.

3.11 Flowchart of procedure of the study




Experimental 2: comparing the effect of training conditions on RTG performance
Pre-training test: RTG under barrier avoidance 10 trials and WMFT
AO group
P group
C group
Observe UE 6 min
Observe view 6 min
Rest 6 min
Training 24 trials
Observe UE 6 min
Observe view 6 min
Rest 6 min
Training 24 trials
Observe UE 6 min
Observe view 6 min
Rest 6 min
Training 24 trials
Observe UE 6 min
Observe view 6 min
Rest 6 min
Training 24 trials
Post-training test: RTG under barrier avoidance 10 trials and WMFT
45 minutes
Retention test: RTG under barrier avoidance 10 trials, WMFT, advantage of training questionnaire