Three-dimensional motion analysis of arm-reaching movements in healthy and hemispinalized common marmosets

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Abstract

Spinal cord injury (SCI) is a devastating neurological injury. At present, pharmacological, regenerative, and rehabilitative approaches are widely studied as therapeutic interventions for motor recovery after SCI. Preclinical research has been performed on model animals with experimental SCI, and those studies often evaluate hand and arm motor function using various indices, such as the success rate of the single pellet reaching test and the grip force. However, compensatory movement strategies, involuntary muscle contraction, and the subject's motivation could affect the scores, resulting in failure to assess direct recovery from impairment. Identifying appropriate assessments of motor impairment is thus important for understanding the mechanisms of motor recovery.

In this study, we developed a motion capture system capable of reconstructing three-dimensional hand positions with millimeter and millisecond accuracy and evaluated hand kinematics during food retrieval movement in both healthy and hemispinalized common marmosets. As a result, the endpoint jerk, representing the accuracy of hand motor control, was asserted to be an appropriate index of upper limb motor impairment by eliminating the influence of the subject's motivation, involuntary muscle contraction, and compensatory strategies. The result also suggested that the kinematics of the limb more consistently reflects motor restoration from deficit due to spinal cord injury than the performance in the single pellet reaching test. Because of

recent attention devoted to the common marmoset as a nonhuman primate model for human diseases, the present study, which clarified arm-reaching movements in spinalized marmosets, provides fundamental knowledge for future therapeutic studies.

Keywords: Spinal cord injury, Reaching kinematics, Upper limb motor impairment, Jerk

1. Introduction

Spinal cord injury (SCI) is one of the most devastating neurological injuries. The number of patients with SCI is estimated to total 2–3 million worldwide [1]. More than half of human SCIs involve cervical segments, causing incomplete lesions in about 75% of cases [2]. Because SCI results in sensorimotor dysfunction in the areas of the body that are innervated by the spinal cord below the injury site, cervical SCI produces serious upper limb motor deficits. At present, pharmacological [3-7], regenerative [8-11], and rehabilitative [5,7,12-17] approaches are widely studied as therapeutic interventions for the recovery of upper limb motor deficits caused by SCI.

Preclinical feasibility studies on loss and recovery mechanisms for forelimb motor function have been performed with experimental cervical SCI in rodents [3,5-7,11-16] and nonhuman primates [4,8-10,17], and various methods have been established to evaluate impaired upper limb motor function. For example, single pellet reaching tasks evaluate upper limb motor function based on a number of pellets retrieved within a certain time or the retrieval success rate [4,5,7,11-18]. With respect to several models of cervical SCI in nonhuman primates, bar grip force [8,19,20] and open field scoring [9,10] have been used to evaluate upper limb motor impairment. These studies reported some extent of motor recovery. However, learned compensatory movement with limited recovery of original motor function causes overestimation of the scores in the single pellet reaching and open field tests. Grip force can be affected by the subject's

1 motivation and involuntary muscle contraction due to muscle strain. Thus, these
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3 conventional methods may fail to directly assess upper limb motor impairment, though
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5 appropriate assessment of motor impairment in animal disease models is important for
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7 understanding loss and recovery mechanisms of motor function [21].
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13 Limited numbers of studies have focused on detailed arm-reaching movements in
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15 spinalized nonhuman primates [18,22] and rodents [14,23,24]. These studies recorded
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17 the two-dimensional (2-D) position of the hand joints during food retrieval movement
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19 at less than 67 frames/s. Evaluation indices were then calculated using the data on the
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21 hand positions, such as the reaching time [18,22], average reaching speed [18,24],
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23 hand preshaping [23], reaching path distance [18], and an analog rating of movement
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25 components [14,24]. However, there are several limitations to this strategy. First,
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27 kinematic analysis of arm-reaching movement is limited to the 2-D space. Second,
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29 because our preliminary data demonstrated that marmosets typically performed food
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31 retrieval movements within 600 ms, the motion capture data of less than 67 frames/s
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33 would have an insufficient temporal resolution to properly compute the derivatives of
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35 the position (e.g., velocity, acceleration, and jerk). Third, although detailed
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37 arm-reaching movements in model animals with experimental SCI have been evaluated
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39 using many indices, the indices suitable for evaluating upper limb motor impairment
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41 itself are unclear.
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Thus, the goals of the present study were 1) to establish a camera-based motion capture system, capable of obtaining the data on the three-dimensional (3-D) positions of hand joints during arm-reaching movement with millimeter spatial resolution and millisecond temporal resolution and 2) to identify an index that can evaluate upper limb motor impairment by eliminating the influence of the subject's motivation, involuntary muscle contraction, and compensatory strategies.

In this study, we made unilateral spinal cord lesions at the cervical level in adult common marmosets (*Callithrix jacchus*), a small New World monkey. Compared with Old World monkeys, marmosets are significantly smaller in body size (300–500 g), easier to handle, and faster sexual maturation (12–18 months) [25]. Considering the recent attention devoted to the common marmoset as a nonhuman primate model for human diseases, the present study provided a high-quality motion capture system and an appropriate index for evaluating upper limb motor impairment, as well as fundamental knowledge for future therapeutic studies.

2. Methods

2.1 Animals and surgical procedure

Ten adult common marmosets (249–418 g; 5 males, 5 females) were used in the present study. The marmosets were assigned proper names as shown in Table 1 and housed individually under a 12-h/12-h light/dark cycle with food and water available *ad libitum*, except on the day before surgery and the single pellet reaching test. All interventions and animal care procedures were performed in accordance with the Laboratory Animal Welfare Act and *The Guide for the Care and Use of Laboratory Animals* (National Institutes of Health, Bethesda, Maryland, USA) and were approved by the Animal Study Committee of Keio University (IRB approval number 11006).

The surgery was performed under general anesthesia induced by intramuscular injections of ketamine (50 mg/kg; Sankyo, Tokyo, Japan) and xylazine (5 mg/kg; Bayer, Leverkusen, Germany) and maintained by isoflurane (Foren; Abbott, Tokyo, Japan). The animal's pulse and arterial oxygen saturation were monitored during the surgical procedures. In the hemisection group, after a laminectomy at the C3, C4, or C5 level, the dura mater was opened longitudinally, and the entire dorsoventral extent of the left side of the spinal cord was cut at the segment C3/C4, C4/C5, or C5/C6 using a surgical blade ($n = 4$; 2 males, 2 females). The control group in this study was a naïve control without any surgical intervention.

2.2 Single pellet reaching task

To afford a food retrieval movement using injured hands, a clear acrylic food table (Figure 1A) was newly developed. The table was 140 mm in width and 80 mm in depth, and it was raised 11 cm from the floor of the cage. A transparent wall (140 mm × 80 mm) was located at the front edge of the table relative to the marmoset. A small opening (25 mm × 25 mm) was located at the bottom left of the transparent wall, enabling the marmoset's left hand to reach a pellet. The pellet (5–7 mm in diameter) was placed in a shallow hole (8 mm in diameter, 2 mm deep), which was located 43 mm to the right (62 mm deep) of the left front corner. A blocking plate (100 mm × 25 mm) was placed at the left edge of the table to prevent pellet retrieval with the right hand.

Marmosets were trained for 30 min everyday (5 days a week) to grasp the pellets in their home cage until the baseline scores reached a plateau (>90% success rate) prior to undergoing cervical cord hemisection. After cervical cord hemisection, the marmosets were unable to reach and grasp the pellets for at least 3 weeks. Postlesion training was initiated 4 weeks after the injury, and it was performed for 10 min twice a week to avoid forgetting the task. We confirmed that by training for 10 min twice a week, the control subjects could maintain a success rate of >90% before conducting the experiment with the injured marmosets.

2.3 Motion capture system

The 3-D position of the left hand was recorded with 18 reflective markers (1.6-mm sphere; 3x3 Designs, British Columbia, Canada) and tracked using three infrared cameras (MEMRECAM GX-3; NAC Imaging Technology, Tokyo, Japan), which were synchronized through an internal trigger and operated at 1000 frames/s with millimeter accuracy. The reflective markers were attached onto the marmosets by two experimenters: one experimenter manually held the marmoset's body, while the other shaved the hair on the back of the left hand, placed drops of instant jelly adhesive, and fixed the markers on the distal interphalangeal (DIP) joints of the left index, middle, ring, and small fingers; proximal interphalangeal (PIP) and metacarpophalangeal (MP) joints of the left thumb, index, middle, ring, and small fingers; center of the back of the left hand; and left, middle, and right edges of the left hand wrist of each marmoset (Fig. 1B). Each camera was set at the following positions: 35–40 cm from the food table with a 30–60° elevation and at different positions to ensure that at least two cameras could always track all markers.

The marmosets' left hand movement during the single pellet reaching task was recorded by the infrared cameras in a healthy state or 8 weeks after the left cervical hemisection. Each marmoset participated in the series of two experiments in the following order: practice session and recording session. In the practice session, the single pellet reaching task was performed 10 times to ensure that each marmoset was

1 acclimated to the experimental environment. In the recording session, a reaching
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3 movement that took longer than 2.5 s to complete (including grasp and retrieve
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5 movements) was excluded from the analyses because of the limitations of the data
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8 buffer of the high-speed camera system. In addition, the data for when the marmosets
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10 could not grasp the pellet were not recorded for standardizing the reaching movements.
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13 Eventually, the reaching movements were recorded six times in this session. We did
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15 not repeat the recording session. The markers were attached to the marmoset's left
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17 hand after the practice session. They were not replaced during the recording session
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19 and were removed after the session ended. The recording session was completed within
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21 30 min, because marmosets began attempting to remove the markers as time passed.
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32 To convert the motion capture results into actual 3-D coordinates, a 15-point
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34 calibration chart (NAC Imaging Technology), for which the positional relationship
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36 between each point of the calibration chart in the actual 3-D coordinates was
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38 previously determined, was captured by the three high-speed cameras before and after
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40 the motion capture experiment. Because the size of the calibration chart was sufficient
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42 to cover the 3-D space through which the marmoset's hand passed during the reaching
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44 movement, the effect of optical distortion was negligible when converting the motion
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46 capture results into actual 3-D coordinates.
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2.4 Video analyses

Movias Neo (NAC Imaging Technology) was used to convert the motion capture results into actual 3-D coordinates and calculate some physical variables such as the hand position, movement speed, and finger joint angle. The left hand position in frame t $(p_x(t) p_y(t) p_z(t))$ was calculated from the marker on the middle finger MP joint, and the reference point $(p_{xr} p_{yr} p_{zr})$ was set at a center of the hole in which the food pellet was placed. The relative hand position $(P_x(t) P_y(t) P_z(t))$ and $P(t)$ were calculated as follows:

$$P_x(t) = p_x(t) - p_{xr} \quad P_y(t) = p_y(t) - p_{yr} \quad P_z(t) = p_z(t) - p_{zr}$$

$$P(t) = \sqrt{p_x(t)^2 + p_y(t)^2 + p_z(t)^2} - \sqrt{p_{xr}^2 + p_{yr}^2 + p_{zr}^2}.$$

The x , y , and z coordinates are defined in Figure 1C-1. The absolute speed of the left hand movement $V_A(t)$ was calculated as follows:

$$V_x(t) = \frac{x(t) - x(t-1)}{s(t) - s(t-1)} \quad V_y(t) = \frac{y(t) - y(t-1)}{s(t) - s(t-1)} \quad V_z(t) = \frac{z(t) - z(t-1)}{s(t) - s(t-1)}$$

$$V_A(t) = \sqrt{V_x(t)^2 + V_y(t)^2 + V_z(t)^2},$$

where s is the time for frame t . The absolute angle of the index finger PIP joint $\theta(t)$ was calculated as follows:

$$\mathbf{V}_1(t) = \mathbf{P}_D(t) - \mathbf{P}_M(t) \quad \mathbf{V}_2(t) = \mathbf{P}_D(t) - \mathbf{P}_P(t)$$

$$\theta(t) = 180 - \cos^{-1} \left(\frac{\mathbf{V}_1(t) \bullet \mathbf{V}_2(t)}{\|\mathbf{V}_1(t)\| \|\mathbf{V}_2(t)\|} \right),$$

where $\mathbf{P}_D(t)$, $\mathbf{P}_P(t)$, and $\mathbf{P}_M(t)$ indicate 3-D positions of the markers on the index

finger DIP, PIP, and MP joints, respectively. The hand positions, absolute speed, and joint angle were filtered using a low-pass, 8th-order Butterworth filter (cutoff frequency, 100 Hz) with zero-phase lag.

Evaluation indices were calculated using MATLAB 2012b (MathWorks, MA, USA), including the reaching time, average reaching speed, the maximum and minimum PIP joint angles of the left index finger, the highest hand position, the distance that the hand traveled during the reaching movement, the endpoint of the reaching movement in the x and y coordinates, endpoint error, and normalized jerk integrated over the reaching movement. The reaching time reflected the length of the reaching phase, which encompassed the time required move from $P(t) \leq 50$ mm (Fig. 1C-2) to the minimum value of $P(t)$ (Fig. 1C-5). The pullback phase was defined as the time required to move from the minimum value of $P(t)$ (Fig. 1C-5) to $P(t) \leq 50$ mm (Fig. 1C-7). The average reaching speed was the mean value of $V_A(t)$ within the reaching phase. The minimum finger joint angle was the minimum value of $\theta(t)$ during the reaching phase (Fig. 1C-4), and the maximum finger joint angle was the maximum value of $\theta(t)$ during the pullback phase (Fig. 1C-6). The highest hand position was equal to the maximum values of $P_z(t)$ during the reaching phase (Fig. 1C-3). The distance that the hand traveled during the reaching movement (D) was calculated as follows:

$$D = \int_{t_i+1}^{t_f} \sqrt{(p_x(t) - p_x(t-1))^2 + (p_y(t) - p_y(t-1))^2 + (p_z(t) - p_z(t-1))^2} dt,$$

where t_i and t_f are the initial and the end times of the reaching phase, respectively.

The endpoints in the x and y coordinates were the values of $P_x(t)$ and $P_y(t)$ at the end of the reaching phase, respectively (Fig. 1C-5). The endpoint error was the 3-D distance from the middle finger MP joint to the center of the hole in which the food pellet was placed at the end of the reaching phase. To assess normalized jerk, the square of the magnitude of jerk integrated over the reaching movement (C_J) was calculated as follows:

$$C_J = \frac{1}{2} \int_{t_i}^{t_f} \left\{ \left(\frac{d^3 p_x}{dt^3} \right)^2 + \left(\frac{d^3 p_y}{dt^3} \right)^2 + \left(\frac{d^3 p_z}{dt^3} \right)^2 \right\} dt.$$

Before the integration, time series jerk data were filtered using a low-pass, 8th-order Butterworth filter (cutoff frequency, 100 Hz). C_J varies with the movement duration and movement distance, and accordingly, is not a useful measure of reaching performance when comparing movements of different distances or durations [26]. Kitazawa et al. proposed a normalized integral of jerk (N_J) to eliminate the limitation [27].

$$N_J = \frac{C_J \times (t_f - t_i)^5}{\{P(t_f) - P(t_i)\}^2}$$

2.5 Behavioral evaluation

2.5.1 Success rate for the single pellet reaching task

To compare the difference between motor impairment and motor performance in

the same movement, we evaluated the success rate for the single pellet reaching task on the day before the motion capture experiment. Marmosets attempted to retrieve a food pellet from the hole of the food table 20 times. The success rate was expressed as a percentage of the ratio between the number of food pellets that the subject ate with the left forelimb and the number of trials [the number of food pellets that the subject ate/20×100].

2.5.2 Open field scoring

The open field test was performed in accordance with the procedure described by Kitamura et al [9]. The test was conducted by the same two examiners on the same day of the motion capture experiment. The test was separated into seven parts: Upper limb: the weight bearing in stance and walking, hand position and placement in walking, reach and grasp performance, and somatosensory function; Lower limb: the range of motion and somatosensory function; and Trunk: the stability. A lower score indicates a greater deficit, and the maximum score is 30.

2.6 Statistical analyses

2.6.1 Group analyses

All evaluation indices such as the reaching time, average reaching speed, maximum and minimum PIP joint angles, distance that the hand traveled during the reaching

1 movement, normalized jerk, endpoints of the reaching movement in x and y
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3 coordinates, endpoint error, and highest hand position were classified into two groups,
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5 those with the left cervical hemisection and the control (healthy). Fisher's exact
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7 probability test was used for all indices.
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17 2.6.2 Individual analyses

18 All motion capture-related indices were analyzed by the Kruskal-Wallis test. If this
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20 test yielded a significant difference, a post-hoc test was performed using Fisher's exact
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22 probability test with Bonferroni correction. The Type I error was set at 0.05.
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32 2.7 Section preparation and confirmation of lesion extent

33 For histological confirmation of the extent of cervical hemisection, animals were
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35 anesthetized with pentobarbital sodium (100 mg/kg) and transcardially perfused with
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37 0.1-M phosphate-buffered saline (PBS, pH 7.3) and 4% paraformaldehyde in 0.1-M
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39 PBS. Spinal cords were removed; postfixed; and exposed to 10, 20, and 30% sucrose in
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41 0.1-M phosphate buffer at 4°C. Transverse frozen sections (50- μ m thick) were
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43 collected serially from C3–C6 spinal cord segments. Centering on the lesion site (250
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45 μ m rostrocaudally), sections were picked and Nissl stained with 1% cresyl violet and
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47 were observed under a BZ-8000 fluorescence microscope (Keyence, Osaka, Japan)
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49 with objective lenses (CFI Plan Apo, $\times 10$; 0.45 numerical aperture; Nikon, Tokyo).
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3. Results

3.1 Extent of the lesions

The extent of the lesions is indicated as dark gray regions in Figure 2A. The lesions of Hemi-A and Hemi-B were located at the C3/C4 and C4/C5 junctions, respectively. The lesions encroached into the left side of the spinal cord and slightly into the right ventral funiculus. Both Hemi-A and Hemi-B displayed sparing of a small region of the lateral funiculus. The lesion of Hemi-C was located at the C5/C6 junction, where it encroached into the left side of the spinal cord, but it spared a small region of the lateral funiculus and the intermediate zone of the gray matter. The lesion of Hemi-D was located at the C4/C5 junction, where it partially encroached into the left side of the spinal cord. We confirmed that the ventromedial funiculus and intermediate zone of the gray matter were spared.

3.2 Arm reaching kinematics

3.2.1 Group analyses

The reaching time and distance that the hand traveled during the reaching movement were significantly longer for the injured marmosets than for the healthy marmosets ($P < 0.001$ in each case). The average reaching speed of the injured marmosets was significantly slower than that of the healthy marmosets ($P < 0.001$). The minimum PIP joint angle was significantly larger in the injured marmosets than in

the healthy marmosets ($P = 0.008$), whereas no significant difference in the maximum PIP joint angles was observed between the groups ($P = 0.40$). The injured marmosets displayed a larger normalized jerk than the healthy marmosets ($P < 0.001$). The endpoint value in the x coordinate was significantly larger in the injured marmosets than in the healthy marmosets ($P = 0.008$). These data suggested that the reaching movements of the injured marmosets ended further left of the food table in comparison with those of the healthy marmosets. However, there was no significant difference in the endpoint in the y coordinate ($P = 0.60$). The magnitude of the 3-D endpoint error in the injured marmosets was significantly larger than that in the healthy marmosets ($P = 0.032$). The highest hand position during the reaching movement was significantly greater in the injured marmosets than in the healthy marmosets ($P = 0.008$). The results of group analyses are summarized in Table 2.

3.2.2 Individual analyses

Individual data revealed that the healthy marmosets could execute the reaching movement in a significantly shorter time and faster speed than the injured marmosets (Figs. 2B and 2C). In particular, the reaching time and the average reaching speed of two injured marmosets (Hemi-A, the C3/C4 lesion and Hemi-B, the C4/C5 lesion) were significantly greater and lesser, respectively, than those of all healthy marmosets. The other injured marmosets (Hemi-C, the C5/C6 lesion and Hemi-D, the C4/C5

partial lesion) also showed significant differences compared with several healthy marmosets. No significant difference was observed between any healthy marmoset pairs.

With respect to the minimum PIP joint angle, two injured marmosets (Hemi-A and Hemi-C) showed significant differences compared with five of six healthy marmosets (Fig. 2D). Hemi-B and Hemi-D demonstrated significantly larger minimum PIP joint angles compared with four and one of six healthy marmosets, respectively. The Kruskal-Wallis test yielded a significant difference with respect to the maximum PIP joint angle, but no consistent difference between the healthy and injured marmosets was observed ($P = 0.001$; Fig. 2E). Both the minimum and maximum PIP joint angles significantly differed between several pairs of healthy marmosets.

Figure 3A presents the *xy*-plane hand trajectories during the reaching movements. These movements featured a smoother and more direct path in the healthy marmosets compared with those in the injured marmosets. Quantitative analysis revealed that the hand trajectories in the injured marmosets, excluding Hemi-D, were significantly longer than those of several healthy marmosets (Fig. 3B). The magnitude of normalized jerk, an index of movement smoothness, was significantly larger in Hemi-A than in all healthy marmosets (Fig. 3C). We also found that the reaching movements of Hemi-B, C, and D were jerkier than those of several healthy marmosets. Among the healthy marmosets, individual variations in the normalized jerk and the distance traveled by

the hand were not confirmed.

Figure 4A illustrates the reaching endpoint in the xy -plane. The reaching movements of the injured marmosets ended farther left in comparison with those of the healthy marmosets ($P < 0.001$). On the contrary, the left cervical cord hemisection had little effect on the control of the reaching endpoint of depth ($P = 0.15$). It was demonstrated that Hemi-C exhibited a larger endpoint error than one of the healthy marmosets (Fig. 4B). Figure 5A presents transient changes in hand height during the reaching movement. We confirmed that the highest hand position during the reaching movement tended to be higher in the injured marmosets than in the healthy marmosets. This was also significantly different between several pairs of healthy marmosets (Fig. 5B).

3.3 Behavioral evaluation in general tests

Healthy marmosets displayed a success rate exceeding 95% in the single pellet reaching test, whereas that of the hemispinalized marmosets ranged between 55% and 70% (Table 1). In the open field test, all healthy marmosets achieved the maximum score of 30, whereas the score of the hemispinalized marmosets ranged from 19–22 (Table 1). In all injured marmosets, deficits were found in the tests for reaching and grasping performance and the somatosensory function of the upper and lower limbs (Supplementary Table 1).

4. Discussion

4.1 Strength of the newly established motion capture system

We succeeded in developing a novel method for tracking the hand position of common marmosets based on attached markers on the hand. This technique allows reconstruction of the 3-D positions of the marmoset's hand joints during arm-reaching movements with millimeter and millisecond accuracy. Our novel acrylic table enabled marmosets to retrieve a food pellet only with their left hands. All marmosets achieved a success rate greater than 90% in the single pellet reaching task within 2 weeks, suggesting that marmosets can easily be trained to use the food table.

The present results suggested that hand kinematics can be evaluated from the high-resolution tracking data of marmoset arm-reaching movements. For example, the length of the reaching movement was longer and the speed of the reaching movement was slower in marmosets with cervical cord hemisection than in healthy marmosets. We assumed that the increase in the duration and the decrease in the speed of reaching movements were caused by damage to the both efferent and afferent neural pathways, which disrupted the ability to control muscle force (e.g., exerting a certain level of force to execute the desired hand movement) [28]. Furthermore, as the same results have also been observed in both animals with experimental SCI [18,22,24] and human patients with SCI [29], we considered that the newly established system was capable of detecting dysfunction in the arm-reaching movements in marmosets with neurological

injury.

4.2 Efficacy of evaluation indices

We found that the minimum PIP joint angle was significantly larger in the injured marmosets than in the healthy marmosets, whereas no significant difference in the maximum PIP joint angles was noted. These results suggested that the injured marmosets experience impaired finger extension ability but not impaired finger flexion ability. Our speculation is that fibers, which decussate at the medulla and then generate collaterals that recross the midline from the intact to the denervated side at the spinal level, increased the number of connections to flexor motor neurons, but not to extensor motor neurons, after cervical hemisection. To support this notion, Rosenzweig et al. found a remarkable increase in the number of midline-crossing fibers after a C7 spinal cord hemisection in macaque monkeys [30]. However, these monkeys displayed spontaneous recovery of both finger flexion and extension muscle activities. We speculated that these inconsistent results were because of difference in lesion levels and species type. Our present results also indicated that finger extension ability tended to be better in the marmoset with the C4/C5 partial hemisection than in the other injured marmosets. It would be because in the marmoset with the C4/C5 partial hemisection the preserved ipsilesional anterior corticospinal tract, which was well pronounced in the spinal segments C8 and Th1 that innervate the hand muscles after

unilateral pyramidal lesions in macaque monkeys [31], played a role in the maintenance of finger extension ability. We herein argue that the minimum PIP joint angle is a potential indicator of corticospinal tract integrity.

Decreases in the time-integrated squared jerk indicate improvement in the movement smoothness [32]. Normalized jerk is therefore often used as a measure of the quality of hand motor control. The magnitude of normalized jerk during arm-reaching movements increases in both animals and humans with dysfunctions of the central nervous system, such as cats with cerebellar lesions [27] and humans with Parkinson's disease [33] or chronic stroke [34]. Our results demonstrated that the magnitude of normalized jerk was significantly larger in the marmosets with SCI than in the healthy marmosets, suggesting inaccurate hand motor control in the hemispinalized marmosets.

The hand trajectory to the target in the injured marmosets was less direct than that in the healthy marmosets. We consider that the inaccuracy of hand motor control leads to a more zig-zag-type hand movement, resulting in a less direct reaching trajectory. Another possible cause of the less direct trajectory is that the injured marmosets selected a long reaching path as a compensatory strategy adopted to overcome their sensorimotor deficits. We postulated that the injured marmosets executed reaching movements by selecting a trajectory involving minimum torque change, which can minimize energy costs [35]. Although normalized jerk can directly assess inaccurate

1 motor control, it is difficult to distinguish whether the lengthening of the distance that
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4 the hand traveled is because of a compensatory strategy or inaccurate hand motor
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11 Increases in 3-D endpoint error are observed in human patients with central motor
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13 structure lesions [36]. We found that the marmosets with cervical hemisection
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15 exhibited a larger 3-D endpoint error than the healthy marmoset in the group analysis,
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17 whereas in the individual analysis, a significant difference was observed only between
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19 the C5/C6 hemispinalized marmoset and a single healthy marmoset. In the present
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21 study, reaching movements lasting longer than 2.5 s, and those in which the marmosets
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23 could not grasp the pellet were excluded from the analysis. The endpoint errors could
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25 have been underestimated, especially in the injured marmosets. We therefore presumed
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27 that the endpoint measurement is insufficient as an indicator of upper limb motor
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29 impairments, which was consistent with the idea of McKenna et al [37].
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41 Because the common marmoset has a limited capacity for independent digit
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43 movement and lacks precision grip, we considered the height till which the hand was
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45 lifted during reaching movements, i.e., to match the conformation of the hand to the
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47 target, as an indicator of preshaping, although the grip aperture between the index
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49 finger and thumb is generally used as an indicator of preshaping. In the present study,
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51 the highest hand position during reaching movements was significantly higher in the
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53 injured marmosets, suggesting that spinalized marmosets might perform excessive
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1 preshaping movements, as observed in human patients with central motor structure
2 lesions [38]. However, it is known that the angular positions of the two joints
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8 (shoulder and elbow) involved in arm-reaching movements differ widely according to
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11 the workspace in which movements are performed [39]. Thus, the maximum hand
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14 height was possibly affected by both the lesion extent and the body posture during
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17 arm-reaching movements. Because the body posture during the reaching movement
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20 was not regulated in the present study, it may have caused the individual variation in
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23 the highest hand position among the healthy marmosets.

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26 In addition, our results demonstrated that the reaching time, the average reaching
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29 speed, and the endpoint jerk appeared to be associated with the lesion extent, i.e.,
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32 marmosets with C5/C6 and C4/C5 partial hemisections tended to show better results
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35 than those with C3/C4 and C4/C5 hemisection. This is because C5/C6 hemisection and
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38 C4/C5 partial hemisection spared the ipsilesional descending axons, which are
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41 involved in reaching movements, compared with hemisections in the other marmosets.
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44 In the marmoset with C4/C5 partial hemisection, the ventromedial pathway, which may
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47 include reticulospinal and anterior corticospinal tracts, was not damaged. The
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50 marmoset with C5/C6 hemisection may be able to voluntarily control the deltoid and
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53 biceps muscles, which are innervated by the C5 level in humans [40]. However, because the
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56 speed and duration of the movement are affected by motor motivation [41,42], we asserted
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59 that the endpoint jerk was appropriate for evaluating upper limb motor impairment by
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eliminating the influence of subjects' motivation, involuntary muscle contraction, and compensatory strategies.

4.3 Comparison with general tests

The success rate in single pellet reaching testing, which is an evaluation index of hand motor performance, was lower in the hemispinalized marmosets than in the healthy marmosets. However, the success rate appeared to be unassociated with the extent of the lesion and the magnitude of normalized jerk, which was proposed as an evaluation index of upper limb motor impairments. This would be because motor performance after a neurological injury is poorly correlated with actual motor impairment of the hands [43].

Open field scoring measures the gross reach-and-grasp function by the maximum height of hand elevation and pen grasping [9]. The present result showed that the open field score tended to be associated with the extent of the lesion, although the difference in scores among four injured marmosets was extremely small. We argue that the open field scoring is insufficient for detecting small differences in upper limb motor impairments that can be evaluated by the present camera-based motion capture system.

4.4 Limitations of this motion capture system

Cervical hemisection induces damage in both ascending and descending fibers.

1 The results of the open field scoring demonstrated that the hemispinalized marmosets
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4 exhibited both motor paralysis and somatosensory loss. However, the evaluation
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8 indices, which were calculated from the motion capture data on reaching movement,
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11 are unsuitable for isolating the existence of both motor paralysis and/or somatosensory
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14 loss.
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17 Placing retro-reflective markers on the same exact positions in different animals is
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19 almost impossible. Variance in the attached positions of the markers could have
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22 contributed to the apparently inconsistent outcomes. The effect would be limited to the
23
24
25 evaluation indices calculated with a single marker, such as the reaching speed and
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28 reaching endpoint, while having an impact on the index calculated with three markers,
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31 resulting in large variation in finger joint angles between healthy marmosets. To
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34 minimize other effects such as individual differences in the hand size and functional
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37 range of motion of the PIP joint, which is known to be $60 \pm 12^\circ$ in healthy humans [44],
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40 on the variation in finger joint angles, this variable should be compared before and
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43 after lesioning in the same individual.
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5. Conclusion

We succeeded in reconstructing the 3-D positions of the marmoset's hand joints during arm-reaching movements with millimeter and millisecond accuracy. The results indicated that the magnitude of normalized jerk integrated over the entire reaching movement, a measure of the quality of hand motor control, was appropriate for evaluating upper limb motor impairment by eliminating the influence of subjects' motivation, involuntary muscle contraction, and compensatory strategies. The present study also suggested that success rates in the single pellet reaching test and open field scoring test are inadequate for assessing upper limb motor impairment. Considering the recent attention devoted to the common marmoset as a nonhuman primate model for human diseases, the newly established motion capture system has the potential to significantly contribute to elucidating the recovery mechanism of voluntary hand movement following a corticospinal injury. This provides fundamental knowledge for future studies on therapeutic strategies such as stem cell transplantation, antagonists for axonal regeneration inhibitors, and rehabilitation.

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Tables

Table 1. Marmosets used in the present study

Marmoset (Mark in figures)	Status	Days from onset	Open field scoring	Success rate of single pellet reaching task
Intact-A (○)	Healthy	-	30	100%
Intact-B (□)	Healthy	-	30	95%
Intact-C (◇)	Healthy	-	30	100%
Intact-D (◁)	Healthy	-	30	100%
Intact-E (▽)	Healthy	-	30	95%
Intact-F (△)	Healthy	-	30	100%
Hemi-A (◆)	Left C3/C4 hemisection	56	20	55%
Hemi-B (■)	Left C4/C5 hemisection	54	19	70%
Hemi-C (◄)	Left C5/C6 hemisection	56	21	60%
Hemi-D (►)	Left C4/C5 partial hemisection	54	22	60%

C3, C4, C5, and C6: cervical vertebra 3, 4, 5, and 6, respectively.

Table 2. Results of group analysis

	Healthy		Injured		Significance
	Median	25%–75%	Median	25%–75%	
Reaching time [ms]	113	95–145	459	335–527	$P < 0.001$
Average reaching speed [mm/s]	477	397–597	154	129–211	$P < 0.001$
Minimum PIP joint angle [deg]	17.0	11.0–27.8	33.6	29.5–49.8	$P = 0.008$
Maximum PIP joint angle [deg]	91.0	77.2–101.2	82.2	76.4–91.2	$P = 0.40$
Hand traveled distance [mm]	56.5	52.0–60.0	70.6	63.5–80.8	$P < 0.001$
$\log_{10}$ (normalized jerk)	4.72	4.32–5.01	7.45	6.75–8.40	$P < 0.001$
3D endpoint error [mm]	16.6	14.6–18.4	18.7	16.9–19.6	$P = 0.032$
Endpoint in x coordinate [mm]	-0.9	-2.9–1.7	3.9	2.0–6.1	$P = 0.008$
Endpoint in y coordinate [mm]	2.8	0.6–4.5	-0.2	-2.1–4.8	$P = 0.60$
Highest hand position [mm]	23.0	19.6–25.4	29.4	25.4–31.0	$P = 0.008$

25%–75%: the 25th and 75th percentiles. PIP: proximal interphalangeal.

Figure legends

Figure 1. Experimental setup and definition of the movement phase. (A)

Experimental setup. (B) Positions of the retro-reflective markers. (C) Definition of the movement phase and evaluation indices. The origin of the 3-D coordinate system in the motion capture experiment was defined as a center of the hole in which the food pellet was placed (1). The movement phase was determined by the distance between the origin of the coordinate system and the marmoset's middle finger metacarpophalangeal joint. The reaching phase was the time required to move from a distance equal to 50 mm (2) to the minimum distance, namely the endpoint (5) and vice versa for a pullback phase, (5) to (7). The reaching time was the length of the reaching phase, and the average reaching speed was the mean value of the left hand movement during the reaching phase. Motion capture data were also used to calculate the highest hand position (3), the minimum angle of the index finger proximal interphalangeal (PIP) joint during the reaching phase (4), the maximum angle of the index finger PIP joint during the pullback phase (6), the distance that the hand traveled during the reaching phase, and normalized jerk integrated over the reaching phase.

Figure 2. Lesion extent, reaching time, average reaching speed, and index finger

joint angle. (A) Lesions produced by hemisection. Reconstructions of the lesion site for all animals described in this paper; dark gray indicates the lesioned area. (B)

Duration of the reaching movement in the healthy and injured marmosets. (C) Average speed of the reaching movement in the healthy and injured marmosets. (D) Minimum angle of the index finger proximal interphalangeal (PIP) joint during the single pellet reaching task. (E) Maximum angle of the index finger PIP joint during the single pellet reaching task. Each column corresponds to one individual. A bar above the marks indicates a significant difference ($P < 0.05$) between a pair of individuals. * $P < 0.05$, different from all healthy marmosets. ** $P < 0.05$, different from all healthy marmosets, except Intact-B. *** $P < 0.05$, different from all healthy marmosets, except Intact-D.

Figure 3. Reaching trajectory in the xy-plane and evaluation indices related to the reaching trajectory. (A) Hand trajectories of the healthy (upper panel) and injured (bottom panel) marmosets during the reaching movement. Grey circles indicate the positions at which the food pellet was placed. (B) The distance that the hand traveled during the reaching movement. (C) Normalized jerk integrated over the reaching movement. Each column corresponds to one individual. A bar above the marks indicates a significant difference ($P < 0.05$) between a pair of individuals. * $P < 0.05$, different from all healthy marmosets. ** $P < 0.05$, different from all healthy marmosets, except Intact-B.

Figure 4. Endpoint and its error in the reaching movement. (A) Endpoint of

reaching movement. Grey circles indicate the position at which the food pellets were placed. (B) Endpoint error. Each column corresponds to one individual. A bar above the marks indicates a significant difference ($P < 0.05$) between a pair of individuals.

Figure 5. Transient changes in the hand height and highest hand position during reaching movement. (A) Transient changes in the hand height of the healthy (upper panel) and injured (lower panel) marmosets during reaching movement. Time zero in each graph indicates the end of reaching movement. (B) The highest hand position during reaching movement. Each column corresponds to one individual. A bar above the marks indicates a significant difference ($P < 0.05$) between a pair of individuals.

Figure1

Figure2

Figure3

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