



Primary hand motor representation areas in healthy children, preadolescents, adolescents, and adults[☆]

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ABSTRACT

The development of the organization of the motor representation areas in children and adolescents is not well-known. This cross-sectional study aimed to provide an understanding for the development of the functional motor areas of the upper extremity muscles by studying healthy right-handed children (6–9 years, $n = 10$), preadolescents (10–12 years, $n = 13$), adolescents (15–17 years, $n = 12$), and adults (22–34 years, $n = 12$). The optimal representation site and resting motor threshold (rMT) for the abductor pollicis brevis (APB) were assessed in both hemispheres using navigated transcranial magnetic stimulation (nTMS). Motor mapping was performed at 110% of the rMT while recording the EMG of six upper limb muscles in the hand and forearm. The association between the motor map and manual dexterity (box and block test, BBT) was examined. The mapping was well-tolerated and feasible in all but the youngest participant whose rMT exceeded the maximum stimulator output. The centers-of-gravity (CoG) for individual muscles were scattered to the greatest extent in the group of preadolescents and centered and became more focused with age. In preadolescents, the CoGs in the left hemisphere were located more laterally, and they shifted medially with age. The proportion of hand compared to arm representation increased with age ($p = 0.001$); in the right hemisphere, this was associated with greater fine motor ability. Similarly, there was less overlap between hand and forearm muscles representations in children compared to adults ($p < 0.001$). There was a posterior-anterior shift in the APB hotspot coordinate with age, and the APB coordinate in the left hemisphere exhibited a lateral to medial shift with age from adolescence to adulthood ($p = 0.006$). Our results contribute to the elucidation of the developmental course in the organization of the motor cortex and its associations with fine motor skills. It was shown that nTMS motor mapping in relaxed muscles is feasible in developmental studies in children older than seven years of age.

Abbreviations

APB abductor pollicis brevis
ADM adductor digiti minimi
BB biceps brachii
BBT Box and block test
CoG Center of gravity
CST corticospinal tract
ECR extensor carpi radialis
FCR flexor carpi radialis
FDI first dorsal interosseus
fMRI functional magnetic resonance imaging

MEG magnetoencephalography
MEP motor evoked potential
MRI magnetic resonance imaging
M1 primary motor area
MSO maximum stimulator output
nTMS navigated TMS
rMT resting motor threshold
SCD scalp to cortex distance
SMA supplementary motor area
S1 primary sensory area

[☆] Some of these results have been presented in abstract form previously.

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1. Introduction

The cortical motor areas include the primary motor cortex (M1), the region responsible for the execution of movement and its concrete aspects, as well as premotor areas that provide cognitive, sensory, or motivational inputs for motor behavior (comprehensively reviewed by Picard and Strick, 2001). The sensory cortex is tightly connected to the motor cortex (Eidelberg 1969; Nudo et al., 1995; Terao et al., 1995), and together with premotor cortex, they are considered as the main executive loci for simple voluntary movements (Gerloff et al., 1998; Witt et al., 2008). There is both electrophysiological and functional connectivity between the M1 and premotor areas as well as supplementary motor areas (SMA), parietal cortex and cerebellum (Wessel et al., 1997; Akkal et al., 2007; Narayana et al., 2012; Genon et al., 2017). Connectivity studies have revealed that dorsal premotor areas constitute a mosaic with several functions such as motor learning, imagery and planning of motor tasks, and action formulation, which is related to hand preference (Lega et al., 2020). This interconnectivity is thought to provide the flexibility necessary to modify the existing network to accommodate a behavioral change (Sanes and Donoghue 2000).

The primary somatosensory and motor cortices are somatotopically organized, such that specific body parts are represented separately and adjacent to other body parts, resulting in a body map (Penfield and Boldrey 1937; Schott 1993; Plow et al., 2010; Card and Gharbawie 2020). Large-scale somatomotor organization with topographic scaffolding is a fundamental principle of early development. This phenomenon is established prenatally and provides the protoarchitecture of the entire brain. It both directs and constrains experience-driven modifications with changes in connectivity (Dall'Orso et al., 2018; Arcaro et al., 2019). The relative position, distance, and overlap between body-part representations constitute a somatotopic layout that differs in the different terminals of the sensorimotor network (Wassermann et al., 1992). Somatotopy is also found in the cerebellar cortices and putamen (Hahamy and Makin 2019). Functional somatotopy is a balance between discrete peaks of individual muscles, their distribution as well as a within-limb overlap of representations (Schabrun et al., 2015). However, the exact somatotopy is also questioned and challenged with action maps (Graziano 2016). There is a considerable overlap between the hand and forearm territories in healthy individuals (Marconi et al., 2007; Plow et al., 2010), potentially providing opportunities for coordinated movements and efficient synergies, while the somatotopic distinctiveness of centers of within-limb representations may be involved in ensuring fine control. The presence of multiple areas of high excitability (peaks in maps) or somatotopically discrete centers is thought to reflect the potential for synergistic, intermuscular coordination and complex movement strategies, and conversely, it is important for finely individuated movement (Te et al., 2017).

Handedness is one behavioral trait that affects somatotopy (Nudo et al., 1992; Nicolini et al., 2019) providing clues to the asymmetrical organization of the human brain (Toga and Thompson 2003; Kong et al., 2018). The neural basis and timing of hemispheric lateralization during development are far from being fully understood (Wilson et al., 2010; Dennis and Thompson 2013; Cochet 2016; Kong et al., 2018). In addition to the primary and secondary motor and sensory areas (Cochet 2016), a study using resting-state functional magnetic resonance imaging and diffusion tractography combined with connectivity-based parcellation detected asymmetry in the organization, functions and connectivity between hemispheres in the dorsal premotor cortices (Genon et al., 2017; Genon et al., 2018). A neurodevelopmental TMS study has revealed a decrease in asymmetry with age, favoring earlier maturation of the dominant hemisphere (Garvey et al., 2003). In adults, trends towards a larger representation in the dominant hemisphere have been described in most published studies (Triggs et al., 1999; Coppì et al., 2014; Chieffo et al., 2016), possibly related to better dexterity, whereas in both preadolescents and adolescents, a trend

toward a reduced motor map was observed for the left, dominant hemisphere (Grab et al., 2018).

Neuromotor function plays an essential role in normal cognitive development and is frequently abnormal in children with developmental disabilities. Noticeable gains in motor functions are made throughout the early school years; the fine-tuning and continued improvement of motor skills, reflected as better quality and speed of motor movements, occur up until the age of 30 years (Fietzek et al., 2000). The most dynamic period of motor performance development ends at 10 to 12 years of age (Fietzek et al., 2000). In structural terms, cortical thickness in M1 attains its peak at the age of about nine years, followed by the SMA (~10 years) and most of the frontal pole (Shaw et al., 2008). Thereafter, the cortical thickness starts to taper-off, and this process asymptotes around 14 years of age (Vandekar et al., 2015). However, the corticospinal tract (CST) reaches its fully mature state earlier than other white matter tracts (Dennis and Thompson 2013). The CST exhibits a leftward asymmetry; its maturation reveals differences between the sexes, but there does not seem to be any relationship to age-related changes in manual skills (Herve et al., 2009).

The motor maps can be assumed to be fundamentally the same in children and adults, but little is known regarding how local network properties within M1 evolve during maturation and how these are associated with concomitant changes in the motor repertoire. There are some key findings emerging from animal models on the use-dependent map plasticity related to the learning of motor skills (Nudo et al., 1996; Kleim et al., 1998). It has been suggested that the emergence of fine motor control is associated with a relative broadening of connectivity between functionally diverse cortical motor neurons and changes in synaptic properties that could enable the emergence of smaller independent networks (Biane et al., 2015; Arcaro et al., 2019). However, this proposal is at odds with other evidence from animal experiments which indicate that the very broad motor connectivity and distributions in the fetus and newborn appear to be tightly refined during early development (probably during the first 2–3 years (Martin 2005)). Neuroplastic changes due to physical practice or mental rehearsal may lead to either a reduction or expansion of cortical representations of actively used muscles (Schieber 2001; Kleim et al., 2004; Adkins et al. 2006; Vaalto et al., 2013) or to some kind of altered overlap (Tyc et al., 2005).

In humans, the functional motor map and its developmental plasticity can be assessed by several non-invasive brain mapping methods (Narayana et al., 2015a). Neuronavigated transcranial magnetic stimulation (nTMS) is one such form of a direct method. TMS has an evident potential with both diagnostic and therapeutic applications in pediatric neuropathologies (Frye et al., 2008; Hameed et al., 2017), such as for assessing plasticity in prenatal, perinatal, or pediatric stroke (Walther et al., 2009; Staudt 2010; Kirton et al., 2016). Though nTMS mapping has proved advantageous in the clinical examination of the presurgical evaluation of the eloquent areas in both children and adults (Säisänen et al., 2010; Vitikainen et al., 2013; Kaye and Rotenberg 2017), the motor maps have not been determined at different stages of development in the healthy brain. Developmental studies using other methods have shown higher activation of the bilateral sensorimotor cortex, parietal areas, the SMA and the cerebellum in adults in comparison to children using functional magnetic resonance imaging (fMRI) (Mall et al., 2005). On the other hand, a magnetoencephalography (MEG) study that examined the generators and areas for motor control in typically developing children and adolescents, found evidence for the involvement of SMA and cerebellar cortices in addition to M1 (Wilson et al., 2010).

The aim of this study was to assess the functional cortical representation of upper extremity muscles at rest in healthy right-handed individuals at different stages of development from school age to adulthood by using nTMS, especially during the critical period for the development of fine motor skills i.e. preadolescence. The motor map results were correlated with manual dexterity. Based on the results from previ-

Table 1

Gender and age, handedness, scalp-to-cortex distance and manual dexterity scores (number of blocks moved in one minute), resting motor thresholds as maximum stimulator output (%). Mean \pm SD (range). Significance indicates differences between age groups (ANOVA, post-hoc SIDAK). Significant asymmetrical differences within age group are indicated with **bold font** (paired *t*-test).

	Children(n = 10)	Preadolescents(n = 13)	Adolescents(n = 12)	Adults(n = 12)	Significance	
Gender (male/female)	5 / 5	7 / 6	6 / 6	6 / 6	–	
Age (years)	7.7 \pm 0.4 (6.8–8.3)	10.9 \pm 0.4 (10.2–11.8)	15.8 \pm 0.8 (14.3–17.0)	28.0 \pm 3.8 (22.3–33.7)	$F = 232.07$ $p < 0.001$ a,b,c,d,e	
Edinburgh handedness	31.1 \pm 8.5 (11–40)	32.7 \pm 5.7 (26–36) [n = 3]	28 \pm 5.2 (23–40)	29.0 \pm 7.1 (17–40)	$F = 0.401$ $p = 0.753$	
Scalp-to-cortex distance (mm)	Left hemisphere	7.89 \pm 1.37 (5.3–9.7)	9.41 \pm 0.82 (7.3–10.3)	12.46 \pm 1.85 (9.1–14.6)	15.23 \pm 2.50 (11–19.5)	$F = 34.47$ $p < 0.001$ b,c,d,e
	Right hemisphere	7.44 \pm 1.26 (5.1–8.8)	8.28 \pm 1.08 (6.4–10.0)	10.93 \pm 1.66 (7.3–13.4)	12.78 \pm 2.57 (9.0–16.7)	$F = 20.95$ $p < 0.001$ b,c,d
Box and block test (score)	Left hand	51 \pm 8 (43–66)	65 \pm 6 (56–75)	73 \pm 11 (53–87)	83 \pm 8 (71–105)	$F = 27.67$ $p < 0.001$ a,c,d,e
	Right hand	55 \pm 6 (46–65)	65 \pm 6 (55–75)	77 \pm 14 (56–94)	84 \pm 7 (75–98)	$F = 23.37$ $p < 0.001$ a,b,c,d
Resting motor threshold (%)	Left hemisphere	67 \pm 17 (42–96)	52 \pm 12 (31–75)	42 \pm 8 (30–54)	41 \pm 7 (32–54)	$F = 12.42$ $p < 0.001$ a,b,c
	Right hemisphere	68 \pm 11 (55–94)	53 \pm 11 (30–73)	40 \pm 8 (25–56)	40 \pm 6 (32–49)	$F = 21.15$ $p < 0.001$ a,b,c,d,e

^a children and preadolescents,.

^b children and adolescents,.

^c children and adults,.

^d preadolescents and adolescents,.

^e preadolescents and adults, ^f adolescents and adults.

ous developmental and animal studies on motor learning and plasticity, we hypothesized that topographic maps would be essentially the same between children and adults though the relative size of motor representation areas might change, and in addition that the overlap between hand and arm representation might increase with development. Furthermore, the map measures were correlated with the improvements in fine motor abilities.

2. Materials and methods

2.1. Participants

This study was carried out in the premises of Kuopio University Hospital, in the Departments of Clinical Radiology and Clinical Neurophysiology, and is part of an earlier study (Määttä et al., 2017; Säisänen et al., 2018). Thirty-five healthy right-handed participants (range 7–17 years), with the genders uniformly distributed, were studied (demographics in Table 1) and compared with 12 young adults (22–33 years old). The participants in the youngest age groups were recruited from a general population sample of predominantly normal-weight children from the city of Kuopio. Adolescents were recruited from pupils in the 8th grade from the nearest comprehensive school. One ambidextrous boy was considered in this study as right-handed. The exclusion criteria were neurological or psychiatric disorders, previous central nervous system (CNS) infection or trauma, medications with known CNS effects, or any contraindication of TMS (Rossi et al., 2009). All participants were informed about the nature of the study. After having received a detailed description of the procedure, the participants provided written informed consent. Consent was also provided from the guardian in the case of a participant being under 15 years of age. The study was approved by the Research Ethics Committee of the Hospital District of Northern Savo (48/2010). All procedures performed were in accordance with the ethical standards of the institutional and/or national research committee (ethical permission 48/2010).

2.2. Motor task

The Box and Block Test (BBT) was used to assess motor speed and skill (Mathiowetz et al., 1985). This task requires the participant to move

as many blocks as possible within 60 s from one side of a box to the other. Each hand was assessed separately, beginning with the dominant hand.

2.3. MR imaging

Subjects were scanned with a 3.0 T MRI -scanner (Philips Achieva TX; Philips Healthcare, Eindhoven, The Netherlands). Structural three-dimensional T1-weighted MR-images were acquired (TR 8.07 ms, TE 3.7 ms, flip angle 8°, 1 \times 1 \times 1 mm³ resolution) for TMS navigation. An experienced neuroradiologist screened all the structural MRIs for focal changes before nTMS examination. Scalp-to-cortex distance (SCD) was assessed in mm to the depth of gray matter surface using the navigation software (Määttä et al., 2017).

2.4. Navigated TMS

nTMS was performed with an eXimia stimulator and a biphasic figure-of-eight coil combined with a navigation system (3.2 research version, Nexstim Plc., Helsinki, Finland) in both hemispheres in a randomized order. A more thorough description of the stimulator setup is provided in our previous paper (Säisänen et al., 2018). TMS-induced motor evoked potentials (MEPs) were recorded using disposable Ag-AgCl surface electrodes placed on the abductor pollicis brevis (APB), abductor digiti minimi (ADM), first dorsal interosseus (FDI), extensor carpi radialis (ECR), flexor carpi radialis (FCR), and biceps brachii (BB) using a belly-tendon montage. Throughout the measurement, muscle activity was monitored on-line and recorded by stimulus-locked EMG (Nexstim Plc., Helsinki, Finland). First, the optimal cortical representation site (“hotspot”) of the APB was determined (Säisänen et al., 2008). The hotspot was the stimulation site where the MEPs of greatest amplitude were elicited repeatedly. At that site, by using the optimal coil orientation, the individual resting MT (rMT) was determined using a threshold hunting paradigm TMS Motor Threshold Assessment Tool 2.0 (Awiszus 2003; Awiszus and Borckardt 2012) as a percentage of the maximum stimulator output (%-MSO). Mapping of motor representation areas was performed at the stimulation intensity of 110% of rMT of the APB with the aid of a grid (size 5 \times 5 mm per square) that was

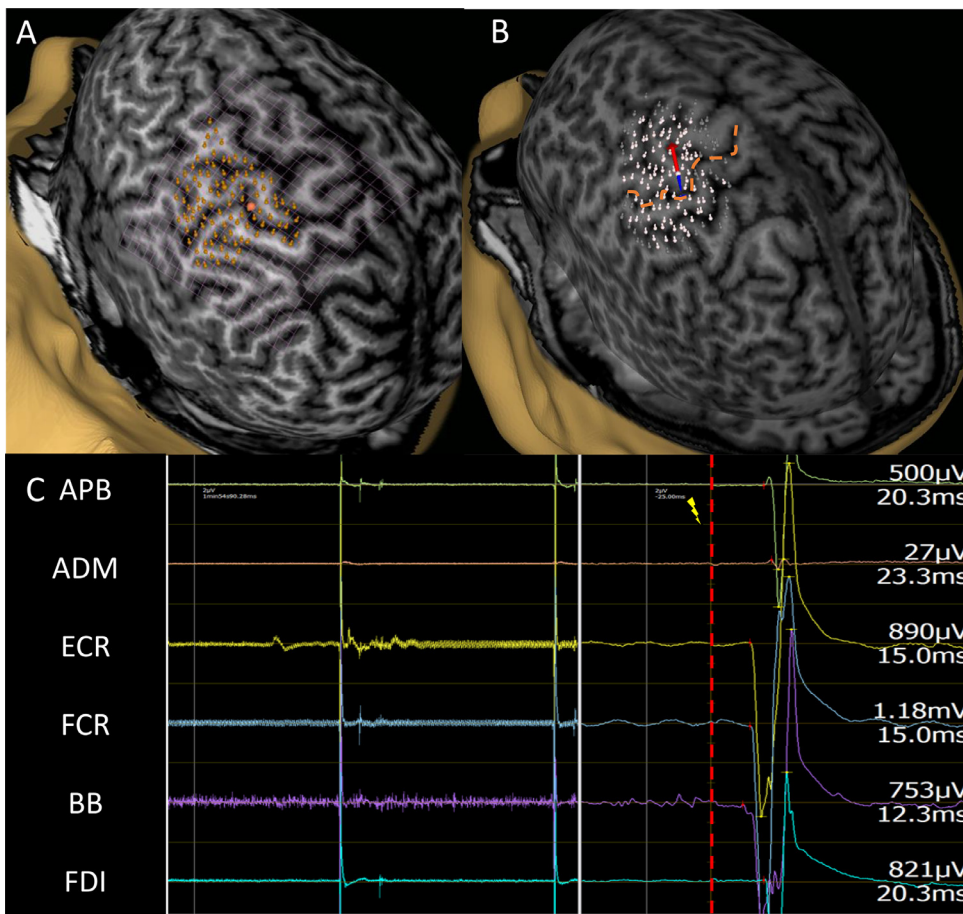


Fig. 1. A Example of the mapping at 110% of rMT using the grid targeted around the hotspot. B Example of the resulting map: white dots are locations eliciting MEP response in any upper limb muscle; gray dots are negative sites. The arrow shows the hotspot with electric field orientation. Orange dashed lines indicate the central sulcus. C EMG was recorded; running online (left panel) and TMS-triggered responses individually for the APB (uppermost), FDI, ADM, ECR, FCR, BB, and FDI (the lowest). Stimulus is given at the moment of the red dashed line. APB = abductor pollicis brevis; ADM = adductor digiti minimi; ECR = extensor carpi radialis; FCR = flexor carpi radialis; BB = biceps brachii; FDI = first dorsal interosseus.

individually centered to the hotspot. One stimulus was applied per spot, extended until there was a rim of stimulation sites eliciting no more MEPs (Fig. 1) (Säisänen et al., 2015). The coil orientation was approximately 45° to midline, perpendicular to the nearest sulcus, the software ensuring optimal tilting. The interstimulus interval was 3 to 5 s. The duration of the whole measurement session (including the explanation of the procedure, attaching the electrodes, performing the co-registration etc.) was approximately two hours. The coarse mapping at the beginning of the session and the following MT determination lasted about twenty minutes per hemisphere. Thereafter, the durations of bilateral motor mapping sessions ranged from 9 to 33 min, the mean was 18 min depending on the size of the map and the subjects' ability to remain relaxed. A tolerability questionnaire was administered immediately following the session (Supplementary Table 1).

2.5. Data analysis

The MEPs with amplitudes of $>50 \mu$ V in relaxed muscles were accepted as responses (Rossini et al., 2015; Groppa et al., 2012). The representation areas for hand (APB, FDI, ADM) and arm (ECR, FCR, BB) were calculated for each subject using the spline interpolation method (Julkunen 2014), and their ratio was studied. The relative overlap (%) was determined as the area where stimulation elicited a response in both hand and arm muscles divided by the union of the hand and arm muscle representation area (i.e. the total area, at least either hand or arm produced a MEP), the so-called Jaccard index.

The Centers-of-Gravity (CoGs) (Wassermann et al., 1992) for motor representation of each hand muscle were determined using the MRI coordinate space of the eXimia software that utilizes the LAS (i.e. left, anterior, superior) coordinate system, which has an origin in the right-posterior-inferior corner of the image. The individual CoG coordinates

and MRIs were spatially normalized to standard space (origin in anterior commissure, x coordinate positive values in right, y coordinate forward positive, z coordinate up positive) using the SPM8-software running on Matlab 7.4 (The Mathworks, Natick, USA). Two templates were used depending on age. The template used for children and preadolescents was created with the TOM toolbox on SPM using the NIH reference data (Wilke et al., 2008). For adolescents and adults, the standard MNI template provided by SPM was used. To determine the variation between the CoGs within the groups, ellipsoids of the 90% confidence interval were fitted to the clusters of the individual sites by estimating the lengths and directions of the ellipsoid main axis based on chi-square distribution using Matlab (Niskanen et al., 2010).

2.6. Statistical analysis

The differences in rMT, SCD, and BBT scores across the age groups were analyzed with ANOVA and the post hoc Sidak test. Interhemispheric differences in rMT, SCD, and BBT scores within the age groups were evaluated with the paired *t*-test since these variables were normally distributed. The representation extents, ratios of areas and overlap (not normally distributed, Kolmogorov-Smirnov) were tested with univariate general linear model for effects of age, hemisphere and their interaction. The CoGs of the muscle-specific coordinates were evaluated using the nonparametric Kruskal-Wallis test and post hoc pairwise comparisons with Bonferroni correction, and interhemispheric differences with Mann-Whitney U test. The Levene test was used to evaluate the variation within age groups in CoG of the APB anterior-posterior y coordinate and the relative overlap within the age groups. Spearman correlation and partial correlations adjusted for age or BBT were used to test the associations between the hand/arm ratios, overlap, motor dexterity, and age. Associations and differences with *P*-values <0.05 were considered

Table 2
Hotspot locations of the APB in each age group shown as percentage (%).

	Children	Preadolescents	Adolescents	Adults
Precentral gyrus	60	50	71	92
Central sulcus	40	38	29	0
Postcentral gyrus	0	12	0	0
Premotor area	0	0	0	8

statistically significant. The statistical analyses were performed with the SPSS software, Version 22 (IBM Corporation, Somers, NY, USA).

3. Results

3.1. Safety and tolerability

The TMS was well tolerated by all participants with no significant side effects. Minor complaints were reported by seven participants i.e. tiredness ($n = 4$), irritation of the stimulated site ($n = 2$), and excess compression from the tracker band ($n = 2$).

3.2. Hotspot locations

The APB hotspots were usually found in the precentral gyrus (68%) or in the central sulcus (27%) (Table 2). There was a posterior-anterior shift with age; the locations in the central sulcus or posterior to that location were more common in the younger age groups, whereas adults' APB hotspots were found in the precentral gyrus (Table 2). The most common location was the hand knob, more specifically, its lateral corner (representative examples from each age group are shown in Fig. 1). SCD increased with age ($F = 20.9$ on the right hemisphere, 20.8 on the left, $p < 0.001$) (Table 1). SCD exhibited a statistically significant asymmetry (longer distance on the left, dominant hemisphere) in all age groups other than in the children (Table 1).

3.3. Motor thresholds and feasibility

The rMTs of the APB are shown in Table 1, and the individual rMTs and feasibility to perform motor mapping in the youngest age group are displayed in Table 3. The rMT was too high to be assessed in the youngest participant aged 6.8 years. Despite using the maximum stimulator output in one participant (G4), 106% and 108% of rMT was reached and used for mapping instead of the intended stimulus intensity, and for this subject, only the results for CoGs are included in the motor map group analysis. This participant also had MEPs of high amplitude in the ECR during mapping. Based on this finding, we performed an additional rMT estimation using a method of 50 μ V-level rMT for the ECR muscle (Julkunen et al., 2011) in children to ensure that the rMTs in the ECR were not lower than in the APB ($p = 0.582$ for right, $p = 0.969$ for left hemisphere, paired t -test) (Table 3). The input-output curves have been assessed earlier (Säisänen et al., 2018). The mean rMT for the APB did not differ between the hemispheres, but some subjects (equally distributed in all age groups) exhibited a substantial interhemispheric difference in rMT (left hemisphere – right hemisphere) ranging from –18 to 19%-MSO.

3.4. Hand and arm representation extents, their ratio, and overlap

The motor representations were located mainly on the precentral gyrus spreading to regions both anterior and posterior to that location, but not extending to the SMA (Figs. 2 and 3). Motor map results at individual level are shown in Fig. 3. A preadolescent subject is taken as an example, and her muscle-specific maps shown in Fig. 3B. Fig. 3A shows the mapped areas bilaterally, and in the left hemisphere the locations where MEPs were elicited in all six recorded muscles. The same kind of map on multi-joint responses is shown for another (adult) subject in

Fig. 3C. The mapping results at group level are shown in Table 4. In children, the hand/arm ratio was below one and increased with age, i.e. children differed significantly from adolescents and adults ($F = 6.157$, $p = 0.001$) (Table 4). The relative overlap of the hand and arm representations was less in children than in the other age groups ($F = 7.864$, $p < 0.001$) (Fig. 4).

3.5. CoG

The muscle-specific normalized CoG-coordinate volumes are shown in Fig. 5. When visually evaluated, the 90% confidential ellipsoid was anatomically narrow and oriented in the antero-posterior direction in children. In preadolescents, the CoGs used for fitting were more scattered resulting in a larger ellipsoid. Thereafter the ellipsoid decreased in size and concurrently, its shape changed from a circle in adolescents to being oval-shaped in adults as a result of the uniformly clustered CoG locations. In one adult, all of the CoGs were in premotor areas.

The CoG coordinate for the APB, the main muscle of interest, was separately examined (Table 5). The medio-lateral x-coordinate exhibited an age-related effect: this CoG moved in a medial direction in the left hemisphere ($p = 0.006$, post hoc pairwise test with Bonferroni correction between preadolescents and adults). In the right hemisphere, the x-coordinate moved slightly in the lateral direction, but this change was not statistically significant (Table 5). All other muscles showed a similar effect of age in the latero-medial direction in the left hemisphere, with the post hoc evaluation highlighting revealing the difference between preadolescents and adults (Supplementary Table 2). No shift was found in the right hemisphere. By using the absolute values of the coordinates, we detected interhemispheric differences in adolescents and adults (Table 5). The CoG for the APB was more medial in the left hemisphere than in the right hemisphere in adolescents and adults, and more posterior in the left hemisphere compared to the right in adolescents.

The antero-posterior y-coordinate showed a trend towards an anterior shift with age in the APB in the left ($p = 0.076$, Table 5), and a statistically significant difference for the ECR and BB (Supplementary Table 2). In the right hemisphere, only the BB revealed this kind of anterior shift associated with age ($p = 0.004$). It is noteworthy that there is extensive variation in the anterior-posterior orientation, specifically in preadolescents and children ($F(3,42) = 5.803$, $p = 0.002$ on the right, non-significant on the left hemisphere). Adolescents exhibited asymmetry, with the y-coordinate on the left being located in a more posterior location ($p = 0.045$).

3.6. Correlations between manual dexterity, age and the motor map parameters

Manual dexterity improved with age ($p < 0.001$, Table 1). The dexterity of the right hand was statistically significantly better than that of the left in children and adolescents ($t = 2.483$, $p = 0.017$). The hand/arm ratio in the right hemisphere correlated positively with the BBT score of the contralateral hand ($\rho = 0.522$, $p < 0.001$); a trend was found for the left hemisphere (Fig. 6). This correlation in the right hemisphere remained when adjusted for age ($r = 0.346$, $p = 0.020$), but age no longer correlated with the hand/arm ratio when adjusted for the BBT score. The relative overlap of the right hemisphere correlated with the BBT score for the contralateral hand ($\rho = 0.500$, $p < 0.001$), but in the left hemisphere this was evident only as a trend (Fig. 6). The correlation disappeared when adjusted for age but remained for overlap and age when adjusted for the BBT score ($r = 0.306$, $p = 0.041$).

4. Discussion

This cross-sectional study describes the cortical maturation and development of hand motor representation areas and their intrinsic organizational principles using nTMS mapping in an age range from childhood

Table 3

Individual resting motor thresholds (rMT) of abductor pollicis brevis (APB) in the children. The rMTs too high for mapping at 110% of rMT are indicated with **bolded** text. Mapping was performed at the maximum stimulator output in G4, corresponding to 106% and 108% of rMT. Mapping was not performed for B5 and only on the right hemisphere for G2.

Subject	Age(years)	Mapping	rMTfor APB(%)		Estimated rMT for ECR (%)	
			L hemisphere	R hemisphere	L hemisphere	R hemisphere
G1	7.7	Both hemispheres	42	55	42	NA
G2	7.9	R hemisphere	96	77	NA	NA
G3	8.3	Both hemispheres	65	66	60	61
G4*	7.8	Both hemispheres at 100% MSO	92	94	NA	NA
G5	7.4	Both hemispheres	64	60	60	54
B1	8.1	Both hemispheres	62	66	60	63
B2	7.3	Both hemispheres	59	62	60	59
B3	7.8	Both hemispheres	64	66	65	66
B4	7.7	Both hemispheres	61	70	56	NA
B5	6.8	no	>100	>100	NA	NA

B=boy, G=girl, ECR = extensor carpi radialis, L = left, R = right, rMT = resting motor threshold. NA = not assessed.

* During mapping, large MEPs were elicited in ECR muscles indicating that the rMT of arm muscles was probably lower than that of APB.

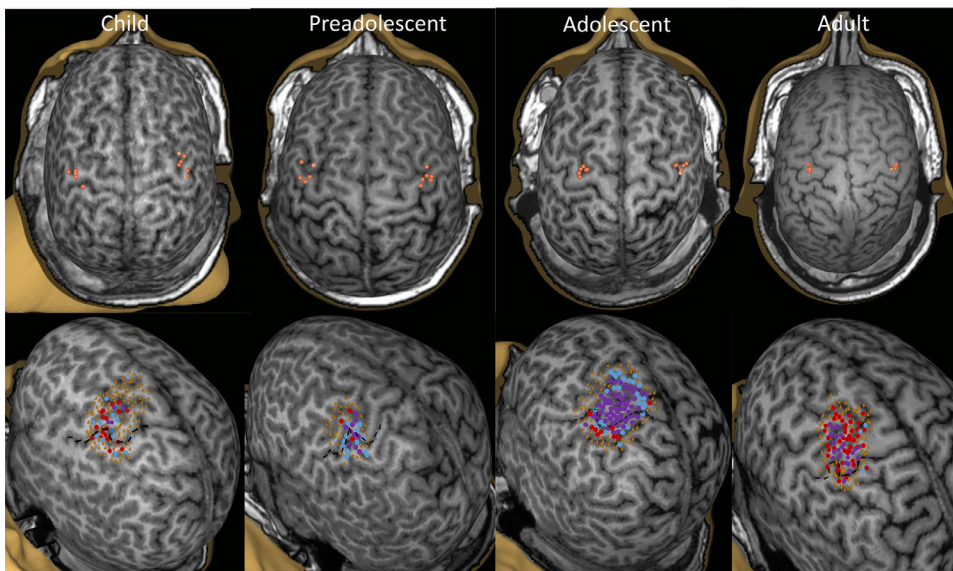


Fig. 2. Upper panel shows representative, qualitative examples of CoG for each muscle. In children, the CoGs are scattered. In an adult brain, the CoGs are close to each other in the precentral gyrus, near the lateral corner of the omega-shaped hand knob. Lower panel shows raw data on individual maps in each age group. Red indicates MEPs in hand, blue in arm and purple in both. The subjects in upper and lower panel are not the same, but randomly chosen.

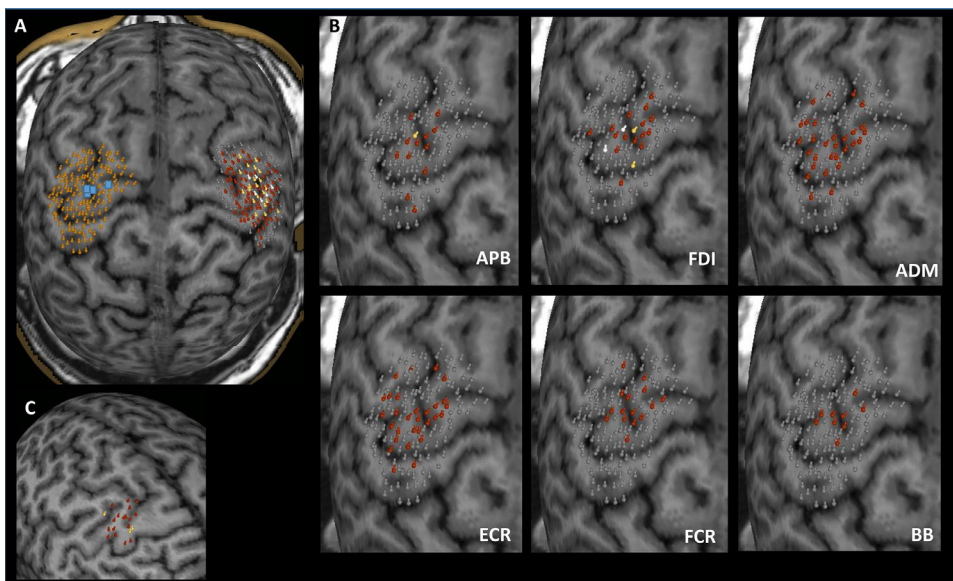


Fig. 3. The mapping data on one preadolescent. **A** All stimulated locations bilaterally. In the left hemisphere the sites where MEPs were elicited in all six muscles are indicated with blue squares. The sites where MEPs were elicited in any of the muscles are shown with heat map, white >1 mV, yellow 500–1000 μ V and red 50–500 μ V. **B** Muscle-specific maps. **C** A similar map showing the multi-joint responses (MEPs elicited in all muscles) for an adult subject.

Table 4

Motor mapping results reported as mean (standard deviation) for hand and arm extents and their ratio and overlap as absolute area and as percentage. Significance indicates difference between age groups (general linear model). No interaction was found between age and hemisphere.

	Children	Preadolescents	Adolescents	Adults	Significance
Extent hand (cm²)	6.83 (3.03)	6.90 (3.04)	8.26 (3.38)	8.62 (3.34)	$p = 0.158$
Extent arm (cm²)	10.18 (3.48)	7.68 (4.17)	7.75 (4.72)	7.90 (3.56)	$p = 0.195$
Ratio hand/arm (-)	0.684 (0.24)	1.035 (0.43)	1.283 (0.59)	1.224 (0.48)	$F = 6.157$ $p = 0.001$ ^{b,c}
Overlap area (cm²)	5.86 (2.70)	6.32 (3.26)	6.61 (3.85)	6.74 (2.87)	$p = 0.857$
Overlap (%)	0.47 (0.13)	0.63 (0.13)	0.61 (0.16)	0.69 (0.12)	$F = 7.864$ $p < 0.001$ ^{a,b,c}

^a children and preadolescents,

^b children and adolescents,

^c children and adults.

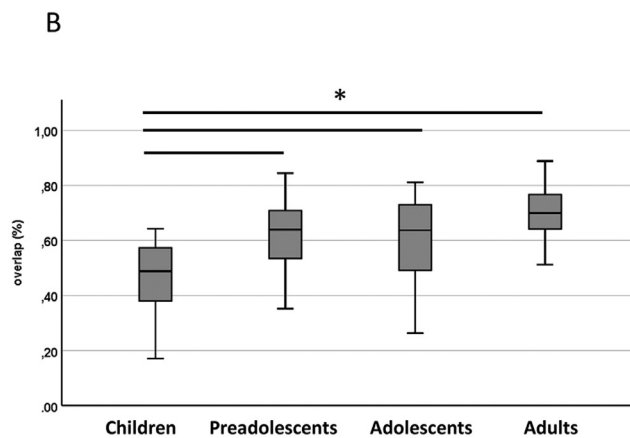
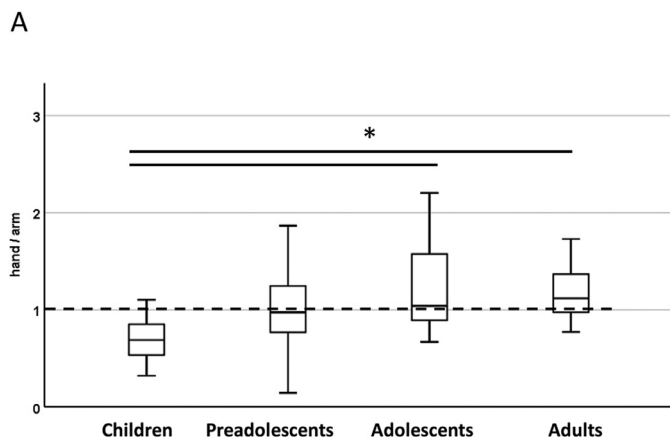


Fig. 4. A Hand/arm ratio and **B** overlap in different age groups.

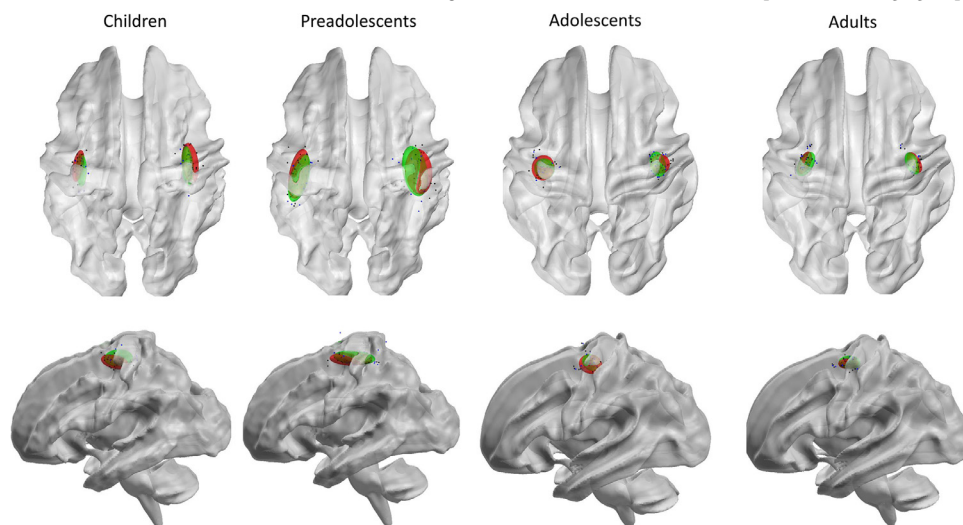


Fig. 5. Ellipsoids showing the locations of centers-of-gravity (CoGs) of hand and arm muscles with 90% confidence interval in normalized standard brain from two different orientations. The children's template was used for children and preadolescents; the adult template was used for adolescents and adults. In the two youngest age groups, the volume was spread also posteriorly to the central sulcus. The dots outside the adult ellipsoid (in the premotor area) were those of one male subject. The surface is that in the border between the gray and white matter. Black dots and red ellipsoid indicates the hand, blue dots and green ellipsoid indicates arm muscles.

Table 5

Normalized coordinates (in mm) for CoG of the APB presented as mean (standard deviation). Origin is in anterior commissure, x is the medial-lateral orientation (left being negative) and y is anterior-posterior (negative increasing posteriorly). Significance indicates difference between age groups (Kruskal-Wallis, post-hoc paired test with Bonferroni correction). Interhemispheric differences were examined with Mann-Whitney U, significant differences were found for x in adolescents ($p = 0.020$), y in adolescents ($p = 0.045$) and x in adults ($p < 0.001$), shown in bold font.

		Children	Preadolescents	Adolescents	Adults	Significance
x coordinate	Left hemisphere	-36.3 (2.4)	-38.5 (5.4)	-36.1^a (4.6)	-32.7^c (2.8)	$p = 0.006$ *
	Right hemisphere	38.5 (2.4)	39.8 (4.9)	40.6 (4.2)	40.6 (4.2)	$p = 0.414$
y coordinate	Left hemisphere	-12.6 (7.0)	-17.1 (10.3)	-14.9^b (3.9)	-10.6 (4.2)	$p = 0.076$
	Right hemisphere	-10.2 (8.4)	-16.5 (8.3)	-11.7 (3.2)	-10.5 (3.4)	$p = 0.371$

The significance (*) is between preadolescents and adults.

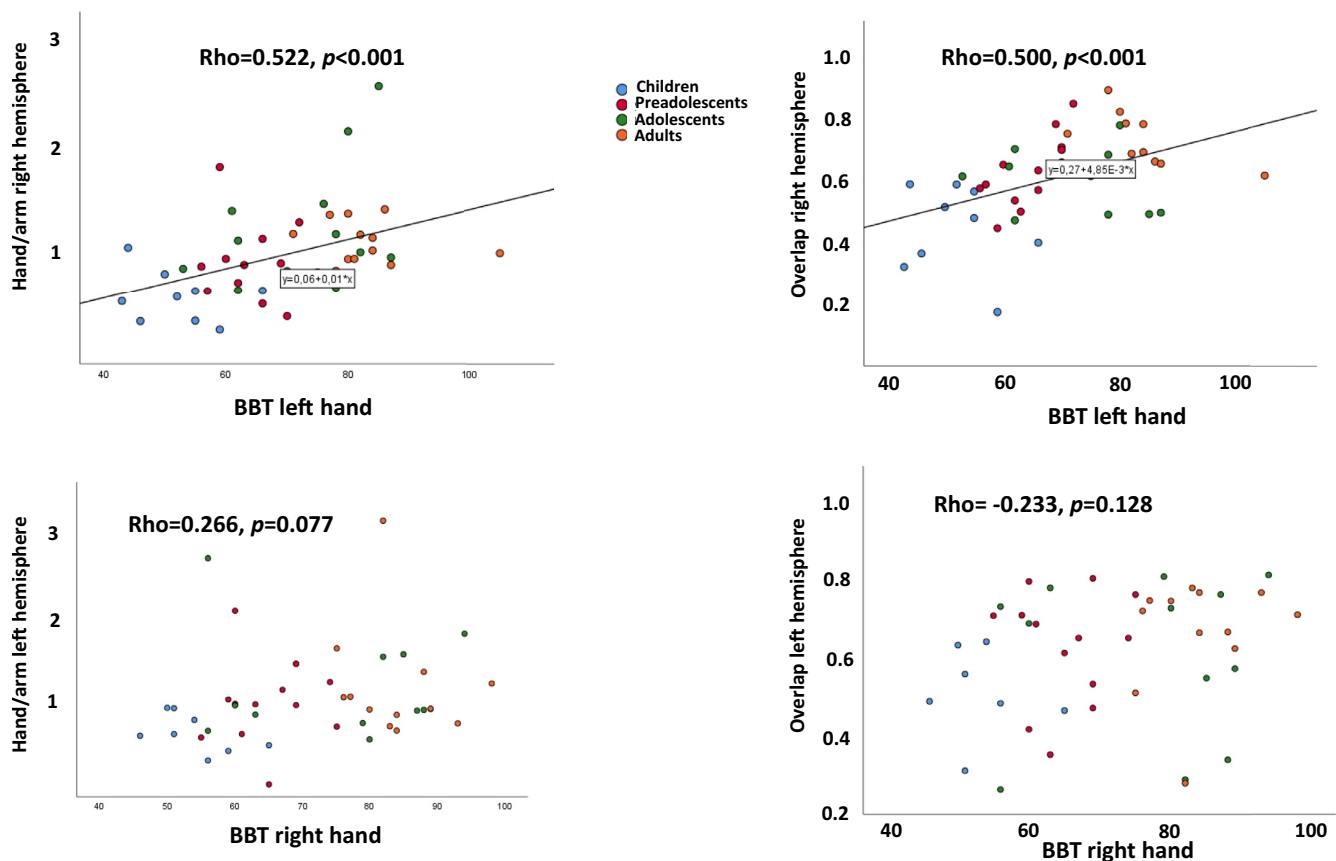


Fig. 6. Significant correlations between hand/arm representation areas and overlap with manual dexterity of the contralateral hand were found in the right hemisphere. No such correlations were found in the left, dominant hemisphere. Colors indicate different age groups.

to young adulthood. Both an expansion and the extensive variation in the CoGs of the upper limb muscle representations were found in children aged 10–12 years, which is the time window when fine motor abilities improve. Thereafter, the CoGs of the motor representations became more concentrated with age. In the left hemisphere, the CoGs were located more laterally in preadolescents and then shifted medially with age. The hand and forearm muscle representation ratio increased with development; in the right hemisphere, this is associated with greater fine motor ability. The hand and forearm muscle representations overlapped less in children compared to other age groups.

4.1. Hotspot location

In adults, the APB hotspot was found in the expected location in the precentral gyrus or anterior to that location, usually in the lateral corner of the hand knob (Ahdab et al., 2016; Reijonen et al., 2020a). Our observation of the premotor hotspot in one adult is not unique (Ahdab et al., 2016). In children, the hotspot was most often located in the central sulcus, and there was a posterior to anterior shift with age. We also observed an unexpected posterior to anterior trend with age, similar to that occurring in the location of the hotspot. Previous developmental work has indicated that both the CoG calculated based on the motor maps as well as the hotspot are valuable parameters, providing a robust tool for estimating the targeted site (Grab et al., 2018). In their seminal modeling work, Fox and colleagues applied a column-based model and found the hotspot location in adults to be deep in the sulcus (Fox et al., 2004). Later on, this location was specified to the anterior wall of the central sulcus toward the gyral lip, though subsequently it was claimed that several issues such as axon bending needed to be considered (Gomez-Tames et al., 2020). There was an expected effect of

age on the SCD, and the used simplified spherical EF model, instead of a realistic head model, may have introduced uncertainties into the mapping result (Beauchamp et al., 2011; Danner et al., 2012; Julkunen et al., 2012).

4.2. Anatomical map location

Anatomically, the hand motor representations were found in M1 around the hand knob, extending slightly frontally to the premotor areas. However, this is mainly due to the spread of the electric field and not a separate connection from the remote premotor area (Teitti et al., 2008). In our study, a coil orientation of approximately 45° was used in all subjects. The EF modeling study revealed that the preferential coil orientations are either perpendicular to the gyrus or toward the CoG or the hotspot on the top of the gyri and furthermore, coil orientation can be crucial for the accuracy of motor mapping (Reijonen et al., 2020b). The functional motor maps were not extended to non-primary motor cortices such as the SMA, which has an important role in complex movement (Shibasaki 2012). In practice, short bursts of high-frequency TMS stimulation are needed to transiently disturb the motor function when assessing the effects on the SMA (Schramm et al., 2019). Activity in the SMA (and cerebellum) in addition to the contralateral precentral gyrus has been observed in brains undergoing maturation among children aged 8–15 years using a simple motor task in an MEG (Wilson et al., 2010). An fMRI study using wrist stimulation detected large activation areas including the SMA in preterm infants (Dall’Orso et al., 2018). Instead, another fMRI study revealed larger activated areas including the SMA and cerebellum in adults in comparison to children (Mall et al., 2005). Not only the type (precision vs. power grip) of task (Ehrsson et al., 2000), but the modalities (fMRI vs. TMS vs.

MEG) and functional connectivity networks differ and the results are not directly comparable to other published reports (Wang et al., 2020; Weiss Lucas et al. 2020).

4.3. Topographic specificity, action maps and motor primitives

When examining the individual maps in each age group, children's maps were more sporadic or diverse, those of preadolescents were located postcentrally, in adolescents, the maps were larger, whereas in adults, they were much more focused and clustered near to the hand knob. However, the individual variation in motor maps was extensive, and caution is necessary when interpreting our results. Our results of diverse maps may be in line with a highly interesting branch of research that questions the topographic organization of the motor cortex, and instead suggests functionally distinct areas (action maps), that are based on coordinated effects on animal's behavior, for review see (Graziano 2016). Complex motor primitives exhibit interspecies differences, but are connected to ethologically relevant behaviors (Desmurget et al., 2014). The researchers were surprised to find a nonuniform representation of synergies by using intracortical microstimulation, where movements tended to converge toward particular postures (Overduin et al., 2012). These action maps, one in the parietal cortex, one in the motor cortex also show connectivity between them. They are partly shaped by experience, and exhibit reorganization and capability of developing new zones. However, it is still unclear whether they are largely fixed after development or does it continuously change with learning.

4.4. Hand and arm representation ratios

Regarding the hand and forearm muscle representation ratios, the children showed larger arm than hand areas, and the proportion of the hand increased with age according to our hypothesis. In a seminal mapping study of distal and proximal muscles in adults, it was observed that the APB map was larger than the FCR (Wassermann et al., 1992) whereas in squirrel monkeys, the arm area was larger than the hand (Card and Gharbawie 2020). In our study, adolescents exhibited extensive variation in this measure, which may suggest that adolescence is a period of dynamic plasticity in the motor areas. It has been claimed that cortical thinning, which is guided by a genetic timetable and by experience, predominates in adolescence and increases well beyond adolescence into middle age and is associated with white matter development and myelination (Casey et al., 2005; Giedd et al., 2009; Paus 2010; Vandekar et al., 2015). Though age dependency in the hand/arm ratio was observed, this dependency disappeared when controlled for the BBT, suggesting that the hand/arm ratio correlates with dexterity rather than with age. Earlier, we detected similar pronounced arm representation in the left, dominant hemisphere in subjects with Asperger syndrome as compared to typically developing preadolescent boys, and this was related to their poorer dexterity (Säisänen et al., 2019). The finer-scale differentiation of the individual fingers increases (i.e., topographic representations are refined) over the first two years (during early development), coinciding with the maturation of fine motor skills (Arcaro et al., 2019). In a study conducted with young and old mice, a short duration of reach training increased the area of proximal forelimb movement representations at the expense of distal representations in the young animals, this reorganization may have been partially mediated by a long-term potentiation (LTP)-like mechanism and synaptic pruning (Tennant et al., 2012).

4.5. Overlap between hand and arm muscles

The overlap is a map metrics related to the hand/arm ratio. In our study, the overlap between distal and proximal muscles varied between 40 and 80%, in line with previous publications (Melgari et al., 2008; Chieffo et al., 2016). Children displayed less overlap than those in the older age groups. The maturation of the overlap appears to occur earlier

(in preadolescence) than the hand/arm ratio (at adolescence). We observed a positive correlation between the overlap and motor skills, but this correlation disappeared when controlled for age. On the other hand, the overlap correlated with age when controlled for dexterity, suggesting that the overlap in the right hemisphere was directly related with age rather than indirectly due to dexterity. A structural plasticity study examining motor learning in rats has revealed that distal and proximal forelimb projecting neurons are intermingled in the motor cortex with an overlapping distribution, which does not change as a function of skill acquisition (Wang et al., 2011). The overlap seems to be highly important for movement coordination, while the somatotopic distinctiveness of centers of within-limb representations could ensure finely individualized control. It has been suggested that in piano players, the less extensive overlap in the dominant hemisphere and the reduced map area are reflections of the long-term plasticity related to motor learning of a skill (Chieffo et al., 2016). Although several aspects of the overlap — the task-specificity (Masse-Alarie et al., 2017), training-related plasticity (Tyc et al., 2005; Vaalto et al., 2013), as well as its presence in pathological conditions such as chronic pain (Schabrun et al., 2009; Tsao et al., 2011; Schabrun et al., 2015) — have been studied, their interpretation still needs further clarification.

4.6. Change in the distribution of muscle-specific CoGs with age

The scatter of muscle-specific CoGs was enlarged (mainly posteriorly) in preadolescent individuals as compared to children or those in the older age groups. This reorganization coincides with a dynamic period in fine motor behavior. Our previous investigations into excitability and maturation revealed that the excitability curve did not reach its plateau in preadolescents (Säisänen et al., 2018), which may point to the potential for plasticity. Preadolescence, i.e. the so-called critical window for motor functions (Shaw et al., 2008), occurs in parallel with the rapid development of executive skills of planning, working memory, and cognitive flexibility, whereas mature “cognition” is characterized by the ability to filter and suppress irrelevant information and actions in favor of relevant cues. All aspects of the movements such as movement initiation, grip force adaptation, and maximum speed of movement are not reached at exactly the same time (Forssberg 1999; Fietzek et al., 2000). Our finding of scattered CoGs in preadolescence corresponds with an imaging study that observed additional nodes which have been postulated to reflect the neural dynamics unique to the maturing motor system, probably to serve the somewhat elementary movements present during the preadolescent years (Dennis and Thompson 2013). A structural developmental study on the corticospinal tract has shown that at the age of 11, the CST origin was located in both pre- and postcentral gyrus, whereas an exclusive pre- or postcentral origin was more common in younger children (Kumar et al., 2009). On the other hand, another study revealed that the maturational direction in the CST origin was from anterior to posterior and increased complexity with age, contrasting our results (Kwon et al., 2016).

4.7. Maturation of map metrics and motor learning

We observed the CoGs to become more concentric with age, and the CoG of our main muscle of interest shifted medially with age on the left hemisphere. A reorganization of the motor map was necessary for the acquisition of a skill (motor learning), but the maintenance of the reorganized state was not necessary for the maintenance of that skill (motor performance) (Tennant et al., 2012). Lower movement thresholds may be associated with increased synaptic efficacy, and synaptic potentiation may be maintained after the map has returned to control levels (Tennant et al., 2012). During early adulthood (20 to 30 years of age), a time of optimal brain health and behavior, spontaneous beta activity of the motor cortex as studied by an MEG was at its minimum, which was suggested as to be indicative of greater neuronal efficiency

(Heinrichs-Graham et al., 2018). Adults have shown both greater integration within networks and greater segregation between networks than is the case in children (Dennis and Thompson 2013). Regions whose brain activity correlates with task performance become more focal or fine-tuned, whereas other regions not involved in the aspects of the behavior that undergo specific refinement remain unchanged by the experience or even decrease in activity with age (Casey et al., 2005). There is a dynamic interplay of simultaneously occurring progressive and regressive events; the gradual elimination of excessive connections, with the establishment and stabilizing of nascent synapses and strengthening of relevant connections with development and experience (Casey et al., 2005). The maturation of superficial white matter continues until the age of 18 years (Wu et al., 2014). The reduced interconnectivity may also generate a greater number of independent networks, supporting enhanced fractionation and resolution of movement (Biane et al., 2015). When a task has been extensively practiced, fewer neurons within the motor cortex need to be active to produce the same movement while the number of synapses is increased only in the reorganized areas of the motor cortex. Our previous large-scale study in young and old adults exploiting the same method, revealed the locations of the APB muscle hotspots around both M1 and S1 (Niskanen et al., 2010). Both pre- and postcentral gyri contain corticospinal cells, and the maps do not strictly adhere to architectonic borders (Groppa et al., 2012; Arcaro et al., 2019). It has been speculated that bilateral S1-M1 might play an important role in the preparation and execution of complex movement (Shibasaki 2012).

4.8. Map correlations with motor tasks

As a behavioral correlate of fine motor skill performance, we used a rather simple task of the BBT that involves the activation of hand and forearm muscles, and observed an expected effect of age. The dexterity of the left hand continued to improve from adolescence to adulthood, whereas the dominant right one did not improve further. Preadolescents and adults were equally good with both hands; children and adolescents were better with their dominant hands. There are several alternative fine motor tests such as the nine hole peg test (NHPT), the finger tapping test, or pinch grip tests (Bashir et al., 2014; Chieffo et al., 2016; Masse-Alarie et al., 2017; Grab et al., 2018; Sirkka et al., 2020). These tests may evaluate different aspects of the motor function and also reveal differences between the sexes during maturation (Herve et al., 2009). Overall, inter-individual variation is a major feature with different motor proficiency tasks typically encountered in developing young children aged from three to 18 years of age (Kakebeeke et al., 2018). No correlations were found related to the self-reported degree of hand preference, but it is noteworthy that all of our tested subjects were clearly right-handed.

4.9. Interhemispheric differences and asymmetry

We did not observe asymmetry in rMT. The increasing symmetry in active MT (aMT) with age suggests that the maturation of the non-dominant cortex is complete by early adulthood (Garvey et al., 2003). Our hypothesis was that the relative proportion of the left, dominant hemisphere motor map would increase with age, but map asymmetry was not observed in the total study population or in any specific age group. A robotic mapping study in participants aged eight to 18 years, but not specifically examining the effect of age, showed a trend toward reduced motor map area and volume for the dominant left hemisphere compared to the right (Grab et al., 2018). A smaller area in the dominant hemisphere has been interpreted as reflecting the more efficient organization of refined muscle representations as compared to the non-dominant side. Our previous study in preadolescents with Asperger syndrome revealed asymmetry reflected as a larger representation in the left hemisphere than the right one, as compared to control subjects whose motor maps were symmetric (Säisänen et al., 2019). The preadolescents

with Asperger syndrome also had slightly larger overlap in the left hemisphere compared to the right hemisphere (Säisänen et al., 2019). This topic is evidently somewhat controversial. Theoretically, map asymmetry could be expected since there is anatomical asymmetry in the left dorsal premotor area which has a hardly distinguishable isolated dorsal subregion (Genon et al., 2018). In adults, the increased overlap and increased area in the left hemisphere as compared to the right was considered to reflect dominance, whereas in pianists, the less and the more symmetric overlaps were postulated reflecting the long-term plasticity for motor learning (Chieffo et al., 2016). A higher overlap in the left hemisphere over the right was considered to reflect the higher training over the lifetime, including grasping, lifting, and joint stabilization (Melgari et al., 2008).

Significant correlations between the map metrics and motor performance were found only in the right, non-dominant hemisphere, although the left hemisphere showed a similar trend. A cortical plasticity study in which 1 Hz rTMS was applied to the right hemisphere detected significant differences only in the right, non-dominant hemisphere (Bashir et al., 2014). The authors suggested that this finding was in agreement with the right hemi-aging model i.e., the right hemisphere is more sensitive to aging effects than its left counterpart. Our previous results on the same data detected stronger inhibition measured with a silent period duration in the right hemisphere in children, but not in other age groups (Säisänen et al., 2018). Structurally, the cortex was located deeper in the left hemisphere compared than in the right in all age groups other than in children. This agrees with one published study in which the left precentral gyrus was found to be located deeper than the right one (Davis 2020), and an earlier study that found the left central sulcus to be deeper, and the intrasulcal surface of the precentral gyrus increased compared to the right-sided structures (Amunts et al., 1996). Subsequently, this finding was also associated with hand preference but was limited to males (Amunts et al., 2000). It needs to be mentioned that opposite findings have also been reported (Toga and Thompson 2003).

4.10. Clinical implications

The motor mapping was safe and well-tolerated in all subjects, though occasional pain or discomfort related to TMS has been reported (in children, adolescents and adults) in accordance with previous studies in younger subjects (Garvey et al., 2001; Gilbert et al., 2004; Coarkin et al., 2011; Narayana et al., 2015a; Grab et al., 2018). The risk for adverse effects in children and toddlers is similar to that in adults (Krishnan et al., 2015; Narayana et al., 2015b). Considering applicability, the performance level was suggested to be more important in predicting a successful mapping outcome than the chronological age (Narayana et al., 2015a). In pediatric populations, high stimulation intensities are required (on average 93% of the maximal stimulator output (Coburger et al., 2012) instead of relating stimulation intensity to rMT, but even 100% may not always sufficient to elicit MEPs with a focal coil despite the support of navigation (Ciechanski et al., 2017). A previous study proved the feasibility of robotic mapping and its suitability for investigating developmental plasticity, though in two children (eight years of age), the rMT was excessively high (Grab et al., 2018). Muscle activation can be used to lower the MT, but we only studied relaxed muscles, which is critical if one wishes to acquire accurate and reliable results in presurgical mappings (Lucente et al., 2018). The stimulus intensity of 110% of rMT produced quite large maps. However, we consider it appropriate since it is clearly suprathreshold, but does not produced too strong E-field and thus result in a map too large (Kallioniemi and Julkunen 2016). A lower stimulus intensity, such as 105% of rMT, could result in smaller maps in presurgical mapping; in fact this is already often done in clinical practice (Jung et al., 2019). The outcome of motor mapping in children can be more robust — whether or not an involuntary MEP is elicited. The APB seemed to be an optimal muscle of choice for motor mapping, as is the FDI, which is often used

in TMS studies (Groppa et al., 2012). The excitability threshold can be the lowest in any hand or forearm muscle, as seen for example in the left-handed boy aged 6.8 years, who had the rMTs for the APB of 91 and 100%, but those for the ADM were 62 and 78%. There is evidence in adults that at the group level, the MTs of the APB and FCR are similar (Wassermann et al., 1992), which we verified in the group of children, though the APB hotspot may not be optimal for the ECR. The motor mapping was based on the MT of the APB, and we did not determine it for each muscle separately which would be recommended if feasibly possible. An elegant study on plasticity in pianists also used an approach assessing the rMT in a hotspot in which the MEPs were elicited in either the APB or ADM (Chieffo et al., 2016).

4.11. Strengths and limitations

Compared to earlier non-navigated studies, electric field online navigation allowed us to gain accurate high-resolution anatomical information in relation to the hand knob, resulting in more stable MEPs with significantly higher amplitudes and shorter latencies (Julkunen et al., 2009). Strength in our study was that both hemispheres were examined, in a randomized order, and several distal and proximal muscles were recorded simultaneously. However, it needs to be mentioned, that secondary muscles may have different representations and overlaps as the used mapping intensity could be different from the rMT of APB.

When gathering data for computing the motor map size, we applied only a single stimulus per grid point. By using a dense grid and the software with an arrow brightness indicator that aided in holding the tilting optimal, we could reliably detect even small changes. Previously, it has been shown that the repeating of stimulus at each grid point increases the accuracy of the map measures (Cavalieri et al., 2017). However, the grid we used (0.5 cm x 0.5 cm) was denser compared to that (1 cm x 1 cm) used in Cavalieri et al. (2017), and if we were to place our stimuli within that grid, it would correspond four of our stimuli within one grid element (1 cm²). In our study, the stimulus locations were considered in conjunction with the response amplitude, and 2-dimensional spline interpolation was used, which reduces the variability of individual responses when 50 μ V streamline was taken to represent the edge of the representation area. The potential limitations of using only one stimulus per grid point arise at the edges of the motor map, where probability of induction of a response above the threshold amplitude are close to 50% and therefore, this limitation may have added unbiased noise to our map measures, but the use of dense grid defuses the above-mentioned phenomenon. The restricted number of stimuli was considered a justified compromise by keeping the duration of measurement session tolerable for the subjects. The interstimulus interval was long enough to prevent any habituation effect.

The number of participants was not large enough to permit reliable examination of differences in motor map metrics between the sexes that are known to influence brain development (Giedd et al., 2012; Dennis and Thompson 2013; Akilan et al., 2020). In a small group such as ours, the difference needs to be greater in order to result in a small p-value. However, despite of the small group sizes, we found statistically significant differences in motor map metrics. The study would have benefited if we had conducted several motor tasks. Most of the measurements were performed at the same time of the day, and to maintain the attention level, the participants watched a DVD during the TMS, but there may be variability in terms of both attention and fatigue. The leisure-time activities related to motor skills such as sports or playing an instrument were not controlled. Cross-talk from adjacent muscles when using surface electrodes may have compromised the interpretation of the results (e.g. map overlap) (Masse-Alarie et al., 2017). It should also be noted that there is extensive variability in brain structure among individuals, especially during development (Casey et al., 2005).

5. Conclusions

We evaluated the eloquent upper limb motor maps in relation to the anatomy in different age groups accurately using electric field navigated TMS and muscles at rest. Topographic maps were found to be rather analogous in all age groups without a major contribution from higher order motor areas around M1. The muscle-specific CoGs were scattered in large areas including postcentral areas in preadolescence, which is a dynamic phase in motor function improvement. Associations between the map metrics and hand dexterity were found only in the right hemisphere. Clinically, these results may provide a reference for outlining functional areas as a part of a multimodal presurgical evaluation in the pediatric population. We encourage recording of several upper limb muscles during motor mapping, as the excitability can be lower in some other hand or forearm muscle, especially in children who have a proportionally larger arm representation area as compared to the hand. nTMS was found to be well-suited for studying a developmental course in the organization of the motor cortex, and motor mapping may be useful in future studies as a biomarker of treatment-related improvement in developmental disabilities such as perinatal stroke. The development of the motor map before school age merits further investigation, and in the future, it would be extremely interesting to study the longitudinal change in the motor maps as well as other brain areas (thalamus, cerebellum, spinal cord and cognitive areas) in addition to the cortex, all of which are candidate locations for motor skill learning-induced plasticity (Tennant et al., 2012).

Declaration of Competing Interest

LS and PJ have received travel bursaries unrelated to this study from Nexstim Plc. PJ has a shared patent with Nexstim Plc. The rest of the authors declare no conflict of interest.

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Credit author statement

Laura Säisänen; involved in conceptualization of the research, analysis of the data; preparation of the manuscript

Mervi Könönen; development and design of methodology, analysis of the data; preparation of the manuscript; data visualization

Eini Niskanen; analysis of the data; preparation of the manuscript
Timo Lakka; involved in conceptualization of the research; acquisition of the financial support for the project leading to this publication.

Niina Lintu; involved in conceptualization of the research; recruiting the participants

Ritva Vanninen; involved in conceptualization of the research; acquisition of the financial support for the project leading to this publication.

Petro Julkunen; development and design of methodology, in programming, implementation of code and supporting algorithms, preparation of the manuscript

Sara Määttä; involved in conceptualization of the research; preparation of the manuscript; project administration; management and coordination responsibility; acquisition of the financial support for the project leading to this publication.

Data availability statement

The data that can be shared, i.e. anonymized, is available upon reasonable request.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.neuroimage.2020.117702.

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