# A Proposal of New Reference System for the Standard Axial, Sagittal, Coronal Planes of Brain Based on the Serially-Sectioned Images

Sectional anatomy of human brain is useful to examine the diseased brain as well as normal brain. However, intracerebral reference points for the axial, sagittal, and coronal planes of brain have not been standardized in anatomical sections or radiological images. We made 2,343 serially-sectioned images of a cadaver head with 0.1 mm intervals, 0.1 mm pixel size, and 48 bit color and obtained axial, sagittal, and coronal images based on the proposed reference system. This reference system consists of one principal reference point and two ancillary reference points. The two ancillary reference points are the anterior commissure and the posterior commissure. And the principal reference point is the midpoint of two ancillary reference points. It resides in the center of whole brain. From the principal reference point, Cartesian coordinate of x, y, z could be made to be the standard axial, sagittal, and coronal planes.

Key Words : Brain; Anatomy, Cross-Sectional; Magnetic Resonance Imaging; Imaging, Three-Dimensional; Neuronavigation

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### INTRODUCTION

The anatomist has long been interested in sectional anatomy which is the study of cut surfaces of the human organs. The sectional anatomy is based on four imaginary planes, i.e., axial (horizontal), median (midsagittal), sagittal, and coronal planes that pass through the body. The sectional anatomy is particularly important in the brain where functional topography is of utmost interest. This sectional anatomy of brain becomes increasingly important as a response to giant strides in neuroimaging technology.

Traditionally three standard anatomical planes, namely axial, sagittal, and coronal have been practiced in the brain. All these planes are based on the median plane in most anatomy books (1-3). The sagittal plane has been defined as any vertical plane parallel to the median plane. And the coronal plane is a vertical plane perpendicular to the sagittal plane, whereas axial plane is a plane perpendicular to both sagittal and coronal planes. However, these are all imaginary planes without specific landmarks or reference points. Therefore, the cutting directions of the brain have been decided by the examiner's need and intention.

After completing Visible Korean project (4, 5), we realized the axial sections of the body displayed by different authors have not been standardized (6-8). It was particularly true in head and neck. Therefore, we thought that anatomical landmarks should be explored to set a standard for the guidance of the cutting plane in human brain.

In recent year advance of neuroimaging technology with drastically increased resolution, the sectional images of the central nervous system became the main source of neurological information. Perhaps more than tens of thousands images of magnetic resonance images (MRIs) and computed tomographs (CTs) are being examined and interpreted daily in all parts of the world. In these MRIs and CTs, orbitomeatal line and canthomeatal line have been used for axial radiological images. These radiographic base lines have been selected because they allow the relatively consistent brain images without excessive bony variations. However, there is no universal standard reference point to guide the cutting direction for axial, sagittal, and coronal brain images. Therefore, the cutting direction of neuroimaging has been arbitrarily determined by the radiologists or by the request of the physicians in charge of the patients.

Considering all these uncertainties and lack of intracerebral landmarks, we thought it is necessary to set a reference system to orient the central nervous system precisely. To achieve this object, we attempted 0.1 mm serial sectioning of cadaver head including whole brain based on the proposed reference points using the anterior commissure (AC) and posterior commissure (PC), and obtained a complete set of axial, sagittal, and coronal images that could be used for the standardization of sectional anatomy of the human brain.

## MATERIALS AND METHODS

A donated cadaver was a 67-yr-old, Korean male with the height of 162 cm and the weight of 45 kg. He was a known patient of myasthenia gravis, for which he was treated for the last several years. Recently several bouts of acute upper respiratory infection were experienced. He died suddenly of cardiorespiratory arrest.

Four hours after death the cadaver was moved to Neuroscience Research Institute to take postmortem MRIs. The cadaver head was placed on the bed of 7 Tesla MR scanner (Siemens Medical Solutions, Erlangen, Germany) and the direction of the cadaver head was adjusted. Median MRI of the cadaver brain was acquired to identify centers of AC and PC. The cadaver brain was 7 Tesla MR rescanned (TR, 39 ms; TE, 17 ms; BW, 60; flip angle, 30°; matrix size,  $480 \times 576 \times 312$ ) along the axial plane passing the both AC center and PC center of median MRI. Using laser indicator of MR machine, axial line was drawn on the face of the cadaver parallel to the line that passed both AC and PC centers. Sagittal line was

also drawn on the face along the cerebral falx and longitudinal cerebral fissure (Fig. 1A).

The cadaver was then moved to department of anatomy for the serial sectioning procedure. The cadaver was put into a freezer and frozen to  $-70^{\circ}$ C. After a week, the head was separated from the main body to make a head block. The head block was put into the embedding box and the direction of the block was adjusted referring to the axial and sagittal indicator lines on the face. The embedding box was filled with the embedding agent (1,000 mL of water, 30 g of gelatin, and 0.5 g of methylene blue) which was hard frozen (Fig. 1B) (4).

On the specially-made cryomacrotome, the head block was serially sectioned from vertex to chin at 0.1 mm intervals to make sectioned surfaces (Fig. 1C). After each sectioning, the frost on the sectioned surface was cleaned using ethyl alcohol (4, 5).

The Canon<sup>TM</sup> EOS 5D digital camera (resolution 4,368 × 2,912) with Canon<sup>TM</sup> 50 mm micro lens was installed in front of sectioned surfaces. The location of the digital camera was adjusted to photograph the sectioned surface with 436.8 × 291.2 mm size (Fig. 1C). Two Elinchrom<sup>TM</sup> Digital S strobes were installed around digital camera. The position of the strobes was adjusted to keep constant brightness of the serial sectioned surfaces (4, 5).

The sectioned surface was photographed using the digital camera and was saved as tag image file format (TIFF) file (0.1 mm pixel size; 48 bit color). After cautious confirmation of each image on the computer monitor by anatomists, next step of serial sectioning was continued. The serially-sectioned images were flipped over to orient the right side of the head to the left side to coincide with the radiological convention on Adobe Photoshop<sup>TM</sup> CS3 extended version 10 (Photoshop program). Other details of the procedures were same as those in Visible Korean (4, 5). Whole procedures took us one month in winter days to acquire 2,343 serially-sectioned images from the head block.

Axial serially-sectioned images were converted from 48 bit color to 24 bit color because the in-house developed software



Fig. 1. (A) Axial and sagittal indicator lines on the face; (B) head block in the embedding box (C) which is being sectioned using cutting disc to be photographed by a digital camera.

for making sagittal and coronal images could handle only 24 bit color images. On the software, every column of all axial serially-sectioned images was stacked to make a sagittal image. Other columns were stacked to generate other sagittal images. Using the similar method, rows of all axial serially-sectioned images were cumulated to produce coronal images.

As the preliminary step of volume reconstruction on free software, both intervals and pixel size of the serially-sectioned images were increased from 0.1 mm to 1 mm; bit depth was changed from 48 bit color to 8 bit gray; outside of cerebrum was erased on Photoshop program. The cerebrum images were stacked and reconstructed by volume modeling on MRIcro version 1.4 (MRIcro program) to acquire a three-dimensional (3D) image of the cerebrum with 1 mm voxel size and 8 bit gray. Likewise 3D images of cerebral hemisphere, skull and total head were built.

#### RESULTS

#### General features of serially-sectioned images

A total of 2,343 axial serially-sectioned images was success-

fully obtained without any technical difficulty. The cadaver head including brain was basically ground off instead of being sliced, so that serial images with 0.1 mm intervals could be obtained without loss of tissue (Fig. 2A). In general, there was no distortion of brain tissue due to freezing artifact. But there was a mild subcutaneous edema and congestion in surrounding soft tissue of the skull base. The brain showed no cortical atrophy. A mild to moderate atherosclerosis of cerebral arteries was noted. Because of the atherosclerosis, larger arteries remained open although small arteries and all veins excluding dural venous sinuses were collapsed (Fig. 2B). The cerebrospinal fluid was largely emptied from the ventricle, whereas the fluid in the subarachnoid space was pooled in the cisterns. There was an anastomosis between the occipital sinus and the left internal jugular vein (Fig. 2C).

On the axial serially-sectioned images, fine brain structures like thalamic nuclei (Fig. 3A), substantia nigra, red nucleus (Fig. 3B), and also other structures like auditory ossicles (Fig. 3C) could clearly be identified. In addition, quality of sagittal and coronal images was also satisfactory, showing no difference in every detail with the original axial images even if the bit depth was converted from 48 bit color to 24 bit color.

On 3D images of cerebrum and unilateral cerebral hemi-



Fig. 2. Axial images showing (A) the head including brain, (B) the cerebral arterial wall with atherosclerosis, and (C) the anastomosis (arrow) between the occipital sinus and the left internal jugular vein.



Fig. 3. Zoomed-in axial serially-sectioned images showing (A) the nuclei of thalamus, (B) the nuclei and tracts in midbrain, and (C) the small structures in the left ear.

sphere, which were rotated on MRIcro program, individual gyrus and sulcus could easily be identified. This result helped us convince that the serially-sectioned images kept good quality and correct alignment (Fig. 4).

### Axial, sagittal, and coronal reference planes

From these 2,343 axial images, we found an image showing both AC center and PC center (Fig. 5A). We labeled this



Fig. 4. (A) Lateral and (B) medial view of the three-dimensional image of left cerebral hemisphere; (C) superior and (D) inferior view of that of the cerebrum.



Fig. 5. (A) Axial and (B) sagittal reference planes showing anterior commissure (AC) and posterior commissure (PC).

Fig. 6. (A) Sagittal and (B) coronal reference planes, both of which pass the midpoint of anterior commissure (AC) and posterior commissure (PC). Range of axial and coronal planes was indicated in the sagittal reference plane while range of sagittal planes was indicated in the coronal reference plane.

image to be the axial reference plane (0 mm), and gave all the axial images superior to the axial reference plane +sign and number representing the 0.1 mm distance from reference plane (0 mm). Those inferior images were given -sign and number. A total of +700 axial planes (+70 mm) and -640axial planes (-64 mm) was obtained, indicating the height of the brain including the brainstem to be 134 mm (Fig. 6A).

After completing the axial reference plane, we found sagittal reference plane (0 mm) coinciding with the median plane, which was already determined by the line connecting the AC center and PC center along the falx cerebri and longitudinal cerebral fissure (Fig. 5B). The right hemisphere was given +sign and numbered from sagittal reference plane. And the left hemisphere was given –sign and also numbered. A total of +700 sagittal planes (+70 mm) and -680 sagittal planes (-68 mm) was obtained, indicating 138 mm of biparietal diameter of this brain (Fig. 6B).

After completing the axial and sagittal reference plane, we tried to find the coronal reference plane (0 mm). Among coronal images perpendicular to the AC-PC line, the plane that passes the midpoint between the AC and PC was labeled to be the coronal reference plane (0 mm) (Fig. 6B). The distance from the AC to PC was 22 mm; therefore, the coronal reference plane was 11 mm back from the AC and 11 mm front of the PC. Based on this coronal reference plane, images of the anterior half of the brain were given +sign, and each image was serially numbered from the coronal reference plane. The posterior images were given –sign and also numbered. A total of +820 images (+82 mm) and -700 images (-70 mm) was obtained, indicating 152 mm of anterior-posterior diameter of this brain (Fig. 6A).

After we determined reference planes (0 mm), we examined the contents of anatomical structures in these three sections. We found interesting sets of the brain structure in each reference plane. Among axial, sagittal and coronal planes, the sagittal plane needs no further clarification. It was because this midsagittal plane is well-known and has been described in all neuroanatomy textbooks. Our midsagittal plane included most of all the superficial and deep structures in books. Perhaps the difference could be noted in the cerebellum, where vermis structures were clearly displayed with its every divisions and subdivisions (Fig. 5B, 7B).

The axial and coronal reference planes were found to contain most of cerebral lobes including the insula, basal ganglia, thalamus and midbrain that are shown in most anatomy textbooks as representative sections.

The reference axial plane showed equal proportions of the frontal, temporal and occipital lobe. Superior frontal gyrus, middle frontal gyrus and inferior frontal gyrus of triangular part (Broca area) were seen. Superior, middle and inferior temporal gyri together with medial occipitotemporal gyrus (fusiform gyrus), collateral sulcus and hippocampus with parahippocampal gyrus were also included. The full extent of insula with its central sulcus was seen. For the internal structures, other than anterior and posterior commissures, main striatum, pallidum with both internal and external segments, head of caudate nucleus, claustrum, internal capsule with its anterior limb, genu and posterior limb and external capsule were well visualized. Among thalamic nuclei, ventral lateral nucleus, ventral posterior nucleus, centromedian nucleus and pulvinar nucleus could be seen. A tip of cerebellar vermis, namely culmen could be found together with surrounding cerebellar tentorium and straight sinus (Fig. 5A, 7A).

The coronal reference plane (0 mm) showed superior frontal gyrus, middle frontal gyrus, precentral gyrus, central sulcus, postcentral gyrus, lateral sulcus and transverse temporal gyrus. It also showed superior temporal gyrus, middle temporal gyrus, inferior temporal gyrus, medial occipitotemporal gyrus, parahippocampal gyrus and collateral sulcus. Major thalamic nuclei such as ventral anterior nucleus, ventral lateral nucleus, dorsal medial nucleus and anterior nucleus were included. The subthalamic nucleus and fields of Forel were clearly shown on this image. The posterior limb of the internal capsule, striatum, and pallidum with both external and internal segments were clearly distinguishable. The mammillary body and cerebral peduncle of the midbrain were also included in this reference plane. It was found that the quadrigeminal plate became squeezed medially and the posterior corpus callosum was asymmetric, the left being larger than the right (Fig. 6B, 7C).



Fig. 7. Zoomed-in (A) axial, (B) sagittal, and (C) coronal reference planes showing important brain structures.

## DISCUSSION

In order to suggest the reference system of the brain, we tried to perform as follows.

We tried the procurement of a normal subject. The cadaver of this study was a 67 yr old man who was a known patient of myasthenia gravis. The myasthenia gravis is a neurotransmitter disease at the level of myoneural junction; it is basically a disease of peripheral nerve and muscle. There is no report that myasthenia gravis has abnormality in the central nervous system. Therefore, we thought that this subject was suitable for our study.

We took postmortem MRIs and adjusted the head block inside the embedding box for acquiring an axial serially-sectioned image showing both the AC and PC (Fig. 5A). This was very critical because without the aid of MRIs there was no way to guide the cutting direction (Fig. 1) (9).

We believe that we applied a reasonable technique involving freezing, sectioning, and photographing for acquiring serially-sectioned images with good quality (Fig. 1). The intervals of 0.1 mm and pixel size of 0.1 mm contributed for the better quality of the images (Fig. 2, 3, 5-7) than those obtained in the previous studies (4-8). It was fortunate that a total of 2,343 serially-sectioned images could be obtained from a head block without a failure.

World Congress of Anthropologists established Reid's base line (RBL) in 1884 and decreed RBL as the anatomic position of the human skull. RBL, also called Frankfurt plane, is the line passing through the inferior border of the orbit and the upper border of the external acoustic meatus, and has been used for definition of the orientation of the human skull in physical anthropology and diagnostic radiology (Fig. 8A). After introduction of CT, the radiologist developed canthomeatal line (CML) that is the line between the canthus and the midpoint of the external acoustic meatus. This line is approximately 10 degree nose up to the RBL (Fig. 8B). It is not possible to draw RBL and CML on the brain, especially when the brain is separated from the skull (10). Therefore, we need internal landmarks that can be used for the orientation of the brain.

Neurosurgeons who are interested in stereotactic surgery and also specialists of functional MRIs used some internal landmarks: Interventricular foramen-PC line and AC-PC line have been utilized to locate the specific deep nuclei of the thalamus. For AC-PC line, stereotactic atlases defined the line passing through the superior edge of the AC and the inferior edge of the PC (11, 12). However, shape of both AC and PC varies from round, oval to elliptical, and also varies in size in different individual. Therefore, the angle of the AC-PC tangential line would be different depending on the shape, size of AC and PC. Meanwhile the AC-PC central line that passes through the AC center and PC center is fairly consistent in every individual no matter how large these structures are (Fig. 5, 6A) (10).

In order to determine the relationship between the AC-PC line and the RBL and CML, we have made 3D image of total head on MRIcro program from the serially-sectioned images of this research. In the 3D image, the skull was displayed, where RBL could be drawn, and a part of skull and brain was cut out to show central AC-PC line in the median plane. Also, in another 3D image, the skin was displayed, where CML could be drawn, and a part of head was cut out to show central AC-PC line in the median plane. As a result, AC-PC line was almost parallel to the CML and showed approximately 15 degree difference from the RBL (Fig. 8). From the previous data of Visible Korean (4), we could find a similar result. To test this, we need more data including MRIs of sufficient number. The successive research dealing with enough subjects would also confirm the structural deference of reference planes between the AC-PC line, RBL and CML.

It is easy to draw central AC-PC line because both AC and PC can be easily identified in serially-sectioned images (Fig. 5). The AC and PC are particularly well visualized in MRIs. It has been known that AC and PC, identifiable in MRIs, are



Fig. 8. (A) Three-dimensional image of brain and skull made of serially-sectioned images to show anterior and posterior commissure (AC-PC) line and Reid's base line (RBL); (B) another image of brain and skin to show AC-PC line and canthomeatal line (CML).

very consistent structures that can be used for the stereotactic approach of deep internal structures of the brain. Schaltenbrand et al. and Dimitrova et al. (9, 11) used intercommissural line and midcommissural plane as the reference guides for deep nucleus structures like basal ganglia and thalamus. However, they do not mention whether intercommissural line is central or tangential. Furthermore, detailed description of the central structures is not available (9, 11). Nowinski proposed Talairach-Nowinski modification to correct the angle made by tangential intercommissural line from central intercommissural line (13).

It is logical to determine the center of a certain organ to be a reference point. After we set the midpoint of AC-PC connection, we measured the anterior-posterior, biparietal, and superior-inferior distance of the brain starting from this point. It was interesting that this was located really at the center of all three-dimensions of the brain (Fig. 6). The brain here means encephalon including myelencephalon.

Another advantage of this reference system is that the axial, sagittal, and coronal reference planes passing the midpoint of the AC and PC companion most representative brain structures. Axial and coronal reference planes in particular show almost all cerebral lobes and gyri, where the central sulcus is positioned in the center of the sections. Major motor, sensory and limbic cortex and deep nuclei could be found (Fig. 7). Therefore in the neuroanatomy class, it would be desirable to start studying the axial reference plane of brain; then to study the superior (+) and inferior (-) axial planes of brain, which are gradually changing from the axial reference plane first, and then expand anteriorly (+) and posteriorly (-).

In neuroanatomy dissection room, following brain cutting procedure is suggested according to our reference system. First, a brain is divided into two hemispheres; AC and PC are identified on the medial surfaces of the hemispheres. Second, one hemisphere, preferably the left because of cerebral dominance and identification of planum temporale, is cut along the axial reference plane passing the centers of both AC and PC. Third, the other hemisphere is cut along the coronal reference plane passing midpoint between AC and PC. Fourth, serial cuttings are made parallel to the axial and coronal reference planes, respectively. By utilizing this standardized method, brain structures could be easily identified and learned in any sections made.

The serially-sectioned images of brain can be the source of realistic brain atlas and 3D brain models. Up to now 2D images of brain atlases were neither standardized axial plane nor real body color (1, 2, 11, 12). If the new brain atlas based on the serially-sectioned images is published, these defects can be solved. In addition, the new brain atlas on new reference system could be the resource of the clinicopathologic mapping of human brain. Also, until now medical students and physicians who used virtual dissection or virtual surgery software could not be satisfied with the resolution and color of 3D models. If 3D models based on the serially-sectioned images are established, these defects would be solved as well.

In summary, authors believe that sectional anatomy has to be based on strict and reproducible axial, sagittal, and coronal reference plane. Our proposed standard consists of two ancillary reference points, which are the AC center and the PC center; and one principal reference point, which is the midpoint of the AC and the PC (Fig. 5). From this principal original reference point (0 point), which is the central point of the brain, superior-inferior planes (axial images), anterior-posterior planes (coronal images), and right-left planes (sagittal images) could be made to be standard planes (Fig. 6).

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