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ORIGINAL COMMUNICATION

Advanced Features of Whole Body Sectioned Images: Virtual Chinese Human

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Serially sectioned images of whole cadavers have become available through the work of four projects: the Visible Human Project, the Visible Korean, the Chinese Visible Human, and the Virtual Chinese Human (VCH). For the VCH, new techniques and equipment were developed and applied to two female and two male cadavers to overcome some of the limitations noted in previous studies. In this article on the VCH, the procedures described are those used on a male cadaver. The cadaver was young with little to no pathology; there were no flat back artifacts because the cadaver was frozen and embedded in the upright position. Sectioned images (intervals, 0.2 mm) were of exceptional quality and resolution (0.1 mm-sized pixels). Several specific structures were outlined (intervals, 0.6 mm) to acquire segmented images, from which surface models were constructed. The VCH data are to be distributed worldwide and are expected to encourage other investigators to produce useful three-dimensional images and develop interactive simulation programs for clinical practice. Clin. Anat. 23:523–529, 2010.

Key words: Visible Human Projects; cadaver; frozen sections; cross-sectional anatomy; computer generated three-dimensional imaging

INTRODUCTION

The Visible Human Project was the first to obtain serially sectioned images of whole human cadavers (1994, male; 1995, female). The key technique used for obtaining these sectional images at 0.33–1 mm intervals was to sequentially mill the cadaver, previously frozen in a gelatin solution (Spitzer et al., 1996; Spitzer and Scherzinger, 2006). This technique was modified by Korean investigators to prepare data of the Visible Korean project in 2002 (Park et al., 2005a, 2006). Subsequently, in 2003, the methodology was applied by separate groups of Chinese researchers to launch the Chinese Visible Human project (Zhang et al., 2004a, 2006) and the Virtual Chinese Human (VCH) (Zhong et al., 2003; Yuan et al., 2003, 2008) (Table 1). For the VCH, several unique techniques with new equipment were used to address the limitations of previous projects: the whole cadaver was frozen, embedded, and sectioned in the upright posture to acquire high-quality sectioned surfaces with the natural body contour. The techniques were applied in three young adults and a female infant. The aim of this study was to elaborate and distribute the VCH data of the four cadavers. This information can be used as an alternative resource for scientists interested in developing software for medical education and clinical

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TABLE 1. Summary of Virtual Human Data Sets Worldwide

Project	Year[a]	Laboratory	Cadavers	Intervals (mm)	Pixel size (mm)
Visible Human Project	1994	Denver, USA	1 male, 1 female	0.33–1	0.33
Visible Korean	2002	Suwon, Korea	1 male	0.2	0.2
Chinese Visible Human	2003	Chongqing, China	3 males, 3 females	0.1–1	0.2
Virtual Chinese Human	2003	Guangzhou, China	2 males, 2 females	0.1–0.2	0.1–0.2

[a]Year refers to when the sectioned images were made from the first cadaver in each project.

training. This article reports on mainly the last male cadaver among the four subjects (Table 2).

MATERIALS AND METHODS

Body Preservation

One male cadaver (VCH-M2) was selected from those donated for medical research and education with the consent of their relatives. The cadaver was a 28-year-old, with a height of 166 cm and weight of 56 kg. The patient was the subject of a court-ordered lethal injection (Table 2).

Fourteen hours after death, the first step of handling the cadaver was initiated. Without exsanguination, approximately 1.2 L of perfusion solution, a mixture of 5% phenol, 1% benzoic acid, and vermilion starch, was injected through the right common carotid artery of the cadaver. This was followed by the injection of red dye (4 g % iron oxide red) with an air pressure of 0.4 × 105 Pa.

The cadaver initially had a flat back and buttocks because of the supine position after the lethal injection. Following perfusion with the above-mentioned solution, the body was turned to face the floor and its back and buttocks were massaged for half an hour to almost full recovery of the normal body contour. The cadaver was stood upright with the back contours preserved and then sealed in a plastic bag to be frozen (−25 to −35°C) with a saturated sodium chloride solution for 72 hr.

Sectioning

After freezing, the cadaver was oriented vertically in an upright embedding mold (580 mm × 380 mm × 2,200 mm). A small amount (about 50 L) of embedding agent (gelatin: blue dye: water = 7 kg: 10 g: 450 L) was then poured into the mold to be frozen to prevent rapid volume increase of the agent from compressing the cadaver. This procedure was repeated until the embedding mold was filled with the frozen embedding agent. Prior to sectioning, the embedding mold was removed to obtain the cadaver block (Fig. 1).

The VCH laboratory for serial sectioning of the standing cadaver block was designed and constructed on the basement and ground floors of a building, where a tall cryomacrotome (JX1500A) was installed. The cadaver block was fixed to the plate of the cryomacrotome in the basement and it moved upward during the serial sectioning (Fig. 1b). To keep the cadaver block frozen, the basement itself was designed as a freezer and maintained at a temperature of −25 to −35°C. On the ground floor, the cadaver block was serially sectioned using the milling disc on the cryomacrotome. The diameter of the milling disc was 400 mm, and its rotating speed was adjusted to 850 rpm for optimal quality of the sectioned surfaces (Fig. 2a). The entire cadaver in the block was serially sectioned at 0.2 mm intervals. Frost and debris were wiped away from each sectioned surface. To facilitate manual cleaning of the sectioned surfaces, the ground floor was designed as a refrigerator with temperatures maintained between 0 and +5°C.

Photography

Serial sections were digitally photographed with a digital camera (PHASE ONE H25™) with AF 24 mm/2.8D lens (Nikon™). The resolution and color depth of the camera was 5,440 × 4,080 and 48 bits color, respectively. The digital camera captured the 544 mm × 408 mm-sized sectioned surfaces to generate 0.1 mm-sized pixel images (Table 2). The photographed surface included not only the cadaver and blue embedding agent, but also a ruler, color patch, gray scale, and image number counter (Fig. 2b).

Above the cadaver block, a strobe (Elinchrom Style1200RX™) was positioned and adjusted to provide equal illumination of all areas on the sectioned surface. The same brightness was verified by an exposure meter when the strobe flashed.

TABLE 2. Features of the Sectioned Images of the Virtual Chinese Human (VCH)

Project (year)	Sex (Age)	Intervals (number)	Resolution (pixel size)	Bit depth (file format)	One file size (total file size)
VCH-F1 (2003)	Female (19 years)	0.2 mm (8,556)	3,024 × 2,016 (0.2 mm)	24 bits color (TIFF)	18 MB (149 GB)
VCH-M1 (2003)	Male (24 years)	0.2 mm (9,232)	3,024 × 2,016 (0.2 mm)	24 bits color (TIFF)	18 MB (161 GB)
VCH-F2 (2004)	Female (10 months)	0.1 mm (4,265)	4,256 × 2,848 (0.1 mm)	24 bits color (RAW)	13 MB (53 GB)
VCH-M2 (2005)	Male (28 years)	0.2 mm (9,320)	5,440 × 4,080 (0.1 mm)	48 bits color (RAW)	127 MB (1,331 GB)

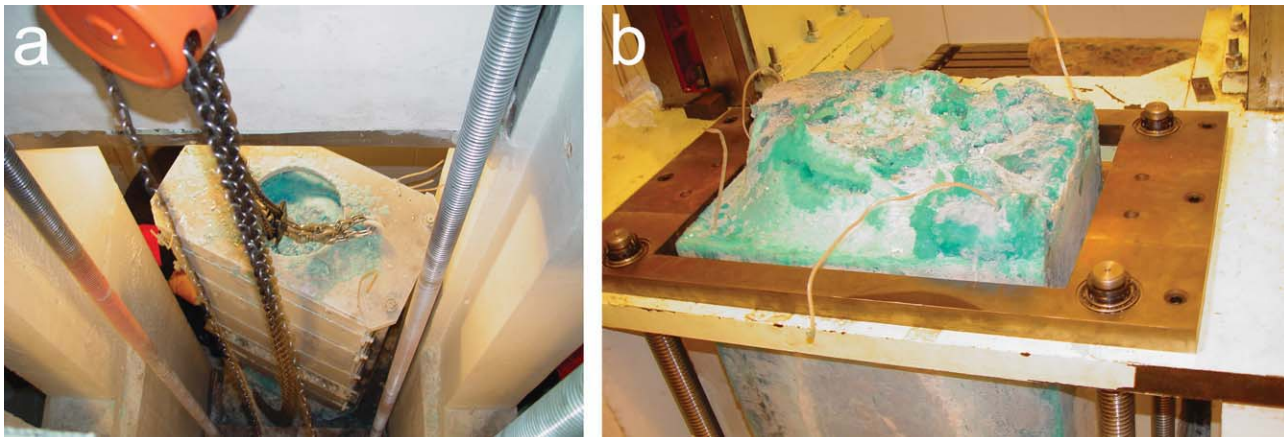

Fig. 1. Procedure for embedding the cadaver. **a:** The cadaver in the vertical position was placed inside a mold, where the embedding agent was poured and frozen. **b:** The mold was removed to produce the cadaver block, attached on the cryomacrotome. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

Photography was performed with the strobes flashing automatically under the following settings: shutter speed, 1/125 sec; aperture, f13; ISO, 100; exposure mode, programmed exposure; white balance, auto; pixel setting, 5,440 × 4,080; file format, RAW; focal distance, 1.2 m. Photographic images were transferred to the computer through IEEE-1394. On the computer screen, whether the sectioned surface was completely cleaned and whether the focus, brightness, and color of the sectioned image were correct were confirmed. After confirmation, the next serial section was obtained (Fig. 2, Table 2).

Full serial sectioning produced 9,320 sequential images with 0.2 mm intervals. The file size of each sectioned image was 127 megabytes, and the total file size of 9,320 acquired images was more than 1 terabyte (Fig. 3a, Table 2).

Segmentation

On the sectioned images, medical experts labeled important anatomic structures of the whole body in both English and Chinese.

Forty-five anatomic structures were selected for outlining. The structures were categorized as the integumentary, skeletal, muscular, digestive, respiratory, urogenital, cardiovascular, lymphoid, sensory, nervous, and endocrine systems. Using Adobe Photoshop version CS3, these structures were automatically, semi-automatically, or manually delineated in every three sectioned images to construct 3,107 segmented images (intervals, 0.6 mm) (Fig. 3b) (Park et al., 2005b).

The segmented images of every structure were stacked in sequence and surface reconstruction was

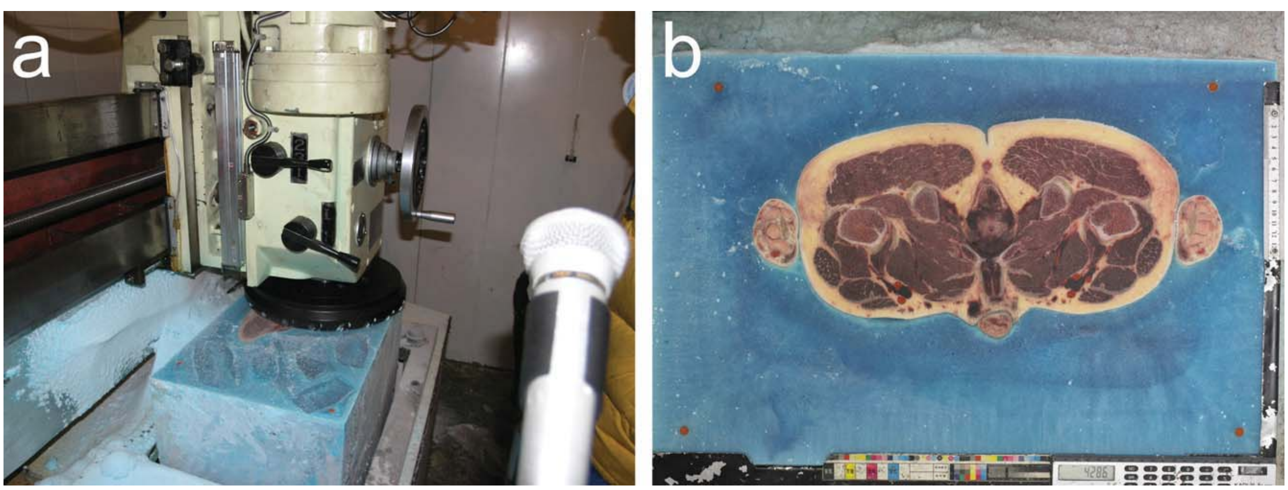

Fig. 2. Procedure for sectioning. **a:** The cadaver block on the cryomacrotome was sectioned with a milling disc. **b:** Each sectioned surface was photographed to capture the image. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

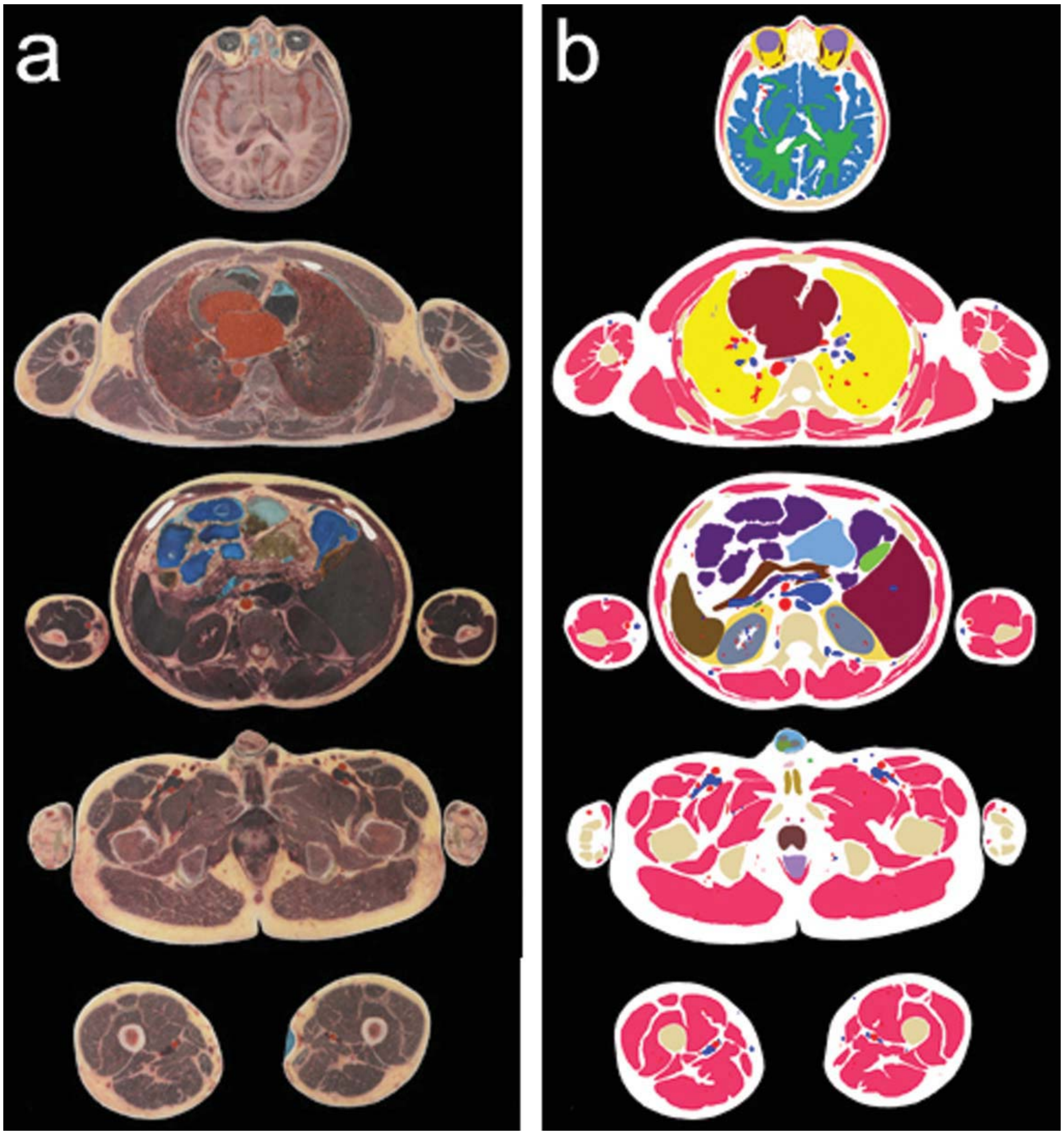

Fig. 3. **a:** Selected cross-sectional images through various regions of the body: background space surrounding the body was converted to a black color. **b:** Corresponding segmented images where specifically outlined anatomical structures were filled with different colors. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

performed using the Visualization Toolkit library™ (Kitware). This is an open-source, freely available software system for 3D computer graphics, image processing, and visualization used to create surface models of the outlined structures (Figs. 4 and 5) (Yuan et al., 2008).

RESULTS

We describe only the results from the second male cadaver (VCH-M2) who had an ordinary body size (height, 166 cm; weight, 56 kg).

There were no definitive pathological findings. This was likely due to the young age (28 years old) and the cause of death, which was execution by lethal injection. Postmortem degenerative changes were minimized thanks to the rapid perfusion following death. Further, the manner in which the specimen was prepared diminished the flat appearance of the upper back and gluteal regions in the sectioned images, segmented images (Fig. 3) as well as in the surface model of the skin (Fig. 5).

Because of the remarkable quality of the sectioned images, even small structures could be easily

Fig. 5. Surface model of the skin, which showed the natural curve of the back and buttocks similar to a living person. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

Fig. 4. Surface models of various structures including arteries.

identified and segmented. As a result, we prepared the segmented images of 45 specific body structures at 0.6 mm intervals. The red colored dye injected into the arteries greatly facilitated their segmentation (Fig. 3). From the segmented images, useful surface models were constructed (Fig. 4).

DISCUSSION

Four individual projects have successfully produced sectioned images of the whole human body: The Visible Human Project in Denver, USA; the Visible Korean in Suwon, Korea; the Chinese Visible Human in Chongqing, China; the VCH in Guangzhou, China (Table 1). Each project has its own advantages as well as limitations, and each complements the other. The increased data sets of the sectioned images are useful for the potential user's selection. Better applications for medical simulation can be accomplished by many technical and clinical groups around the world. Details of the results and applications of the three projects have been illustrated in the special issue of Clinical Anatomy in 2006 (Park et al., 2006; Spitzer and Scherzinger, 2006; Zhang et al., 2006). This article subsequently introduces the rest of the project, the VCH.

The distinguishing elements of the VCH work are the use of younger adults with little or no pathology, the incorporation of a massage step during specimen preparation to minimize artefactual flattening of the upper back and gluteal region, a trial of the new upright postures during body processing and its sectioning, and improvements in image capture of the sectioned surfaces and intervals of segmentation.

The adult cadavers in the VCH were 19, 24, and 28 years of age and generally younger than the individuals used in other projects. In the Visible Human Project, the male and female cadavers were 38 and 59 years of age, respectively (Spitzer et al., 1996). In addition, a 10-month-old female cadaver was serially sectioned (VCH-F2), and the data set from this baby could be used for the construction of pediatric simulation (Table 2).

The VCH bodies were relatively normal with limited pathology. While no cadaver is likely totally free of pathological findings and anatomical variations, attempts to find and use healthy individual is valuable. The Visible Korean cadaver died of leukemia and showed severe pneumonia (Park et al., 2005a). However, in this study, the male cadaver (VCH-M2) was executed by lethal injection, similar to the male cadaver used for the Visible Human Project (Spitzer et al., 1996). The previous cadavers (VCH-F1 and VCH-M1) died of food poisoning (Yuan et al., 2003; Zhong et al., 2003), whereas the cause of death of the female child (VCH-F2) was fetal distress. Normal anatomical models are of great use as reference images for further research.

The sectioned images of the VCH showed luster of tissue (Fig. 3a), which was achieved by injection of the fixative (a mixture of phenol, benzoic acid, and vermilion starch) into the cadaver's arteries. The positive effect of the fixation was consistent with the fact that embalmed cadavers might show more definite tissue characteristics than the frozen cadavers. The fixative also assisted in the subsequent perfusion of red dye, which was helpful in segmenting the arterial branches (Figs. 3 and 4). A similar perfusion with red dye was carried out in another Chinese laboratory (Zhang et al., 2006). During serial sectioning, the fixative was also effective for prevention of the tearing of connective tissue and reduction in the potential health risks to the technicians (Yuan et al., 2003).

Here, a novel method to revive the normal contours of the back and gluteal regions has been described. Even though the flattened surface is not a significant issue as long as the anatomical relationships are accurate, the natural contours of both the skin and underlying muscles make the simulation more realistic. The massage step and cadaver embedding procedure in the upright posture minimized the flattening artifacts (Figs. 3 and 5) (Yuan et al., 2003; Zhong et al., 2003).

The VCH project was a pioneering attempt to section the whole upright cadaver. In the Visible Human Project, upright cadavers were also sectioned. However, because of the limitation of the cryomacrotome size, the cadavers had to be divided into four parts using a saw; this caused tissue loss (Spitzer et al., 1996). In the Visible Korean and Chinese Visible Human projects, the cadavers were laid to be embedded and sectioned (Zhang et al., 2004a; Park et al., 2005a). The sectioned surfaces of the laid cadavers could be divided into superior and inferior areas. The two areas could undergo varied expansion during freezing, and differing sectioning effects on the cryomacrotome. In the VCH, a constant condition for all areas of the sectioned surfaces was intended. Therefore, the problems of prior projects were resolved by the use of new equipment including the upright embedding box and high cryomacrotome (Figs. 1 and 2).

In the latter VCH data, smaller structures could be recognized than with data from previous projects. In the former VCH research (VCH-F1, VCH-M1) and other projects, the pixel size was at least 0.2 mm (Table 1). The pixel size was reduced to 0.1 mm in the VCH-F2 thanks to the small sectioned surfaces of the infant. In the case of the VCH-M2, 0.1 mm-sized pixels were acquired by the higher resolution (5,440 $\times$ 4,080) of the digital camera used. Moreover, the new camera enabled the increase of the bit depth from a 24-bit color to a 48-bit color (Table 2). This progress of our hardware enhanced the details in the sectioned images.

Future work using 0.1 mm-sized intervals together with 0.1 mm-sized pixels might permit more sophisticated volume reconstruction because we can compose 0.1 mm-sized voxels, the unit of volume models (Park et al., 2009). We had prior experience of serial sectioning at 0.1 mm intervals with the infant subject and there was no technical problems encountered (Table 2).

The first sectional atlas with the whole body's sectioned images was published based on the information from the Visible Human Project (Spitzer and Whitlock, 1998); however, the atlas involved only male data. The second atlas included both male and female, based on the Chinese Visible Human (Zhang et al., 2004b). We succeeded the publication by two atlases featuring the female images (Tang and Dai, 2005) and the male images (Tang and Dai, 2006) from the VCH-F1 and VCH-M2 individuals. These atlases contribute not only to the understanding of sectional anatomy but also to the interpretation of CTs and MRIs. Moreover, the atlases to annotate the structures were utilized as reference for delineation of the structures (Fig. 3b) (Yuan et al., 2008).

The 0.6 mm-sized intervals of segmentation in the whole body is the first described; the segmentation process is time-consuming tedious work, and usually performed at 1 mm intervals (Park et al., 2005a; Shin et al., 2009). The segmenting of arteries with previously injected red dye was easily achieved (Fig. 3b).

The previous research of the VCH-F1 has had wide application. The application project was named the VCH clinic, which included the anatomical atlas, motion simulation, virtual endoscopy as well as a game and examination module. In addition, virtual acupuncture and image-guided neurosurgery applications were investigated based on the data from the VCH-F1 (Yuan et al., 2008). The sectioned images, segmented images, and surface models of the VCH-M2 will be used as the raw data for the VCH clinic and other applications. In particular, the virtual simulation of medical practice would likely be more valuable if equipped with a haptic device to allow users to experience the physical property of each anatomic structure (Park et al., 2008; Färber et al., 2009). In another research for computational physiology, entitled the China Physiome Project, multiparticle radiological dosimetry data were derived from the VCH data (Han et al., 2009).

The VCH is a recent addition to a diverse group including the: Visible Human Project, Visible Korean, and Chinese Visible Human. The goal of such projects is to establish a virtual image library containing data sets from the different virtual projects (Table 1). The image library will hopefully facilitate worldwide access of this information by those who in turn can develop useful three-dimensional models and software for medical education and clinical training.

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