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Enhancement of the focal depth in anatomical photography

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Limited depth of field is one of the crucial disadvantages of macro photography because some details of the imagined object are blurred. This paper presents the benefits of using an algorithm which enhances focal depth in the close-up views of anatomical structures. The applied technique was based on combining a set of images of the same object (temporal bone) taken on different focal planes. In effect, a single image was generated which presented all details sharply across the photographed object. The extended depth of field of the composite image was reconstructed by CombineZP Image Stacking Software. (Folia Morphol 2011; 70, 4: 260–262)

Key words: depth of field, depth of focus, image fusion, digital photos

INTRODUCTION

Depth of field is defined as the distance between the nearest and farthest objects in a scene that appear as sharp within the image. The following factors have a direct relationship with depth of field: the diaphragm opening of the lens, the focal length of the lens, and the distance between the lens and the subject.

Recent advances in digital photography have enabled close-up images that have both high resolution and large depth of field. Nevertheless, depth of field still remains a big problem when photographing small objects. In macro photography the depth of field is very shallow. In particular, microscopy imaging suffers from limited depth of focus. An increase in magnification significantly decreases the depth of field [4]. Therefore, close-up views of anatomical structures become problematic because not all details can be seen sharply.

Fortunately, specific algorithms have been developed to fuse images having various planes of focus, thus obtaining a completely focused image with virtually extended depth of field. The algorithm of extended depth of field automatically determines which of the overlapping images is focused the best. Then it selects only the best focused image areas, which are combined into a single picture in which all details are visibly sharp.

Traditional extended depth of field algorithms rely on a high-pass criterion that is applied to each image in the stack. However, there are a variety of methods dealing with the process of image fusion to obtain images with a greater depth of field [2, 3, 9].

The aim of this study was to evaluate the usefulness of an algorithm which enhances focal depth to improve the image quality of photographed anatomical objects. This technique was tested on digital images of the temporal bone because this bone shows multidirectional organisation, and usually macro photography of its anatomical details produces blurred areas in the picture.

MATERIAL AND METHODS

Application of the extended depth of field was tested on the images of the temporal bone isolated from a dry human skull. The intracranial aspect of

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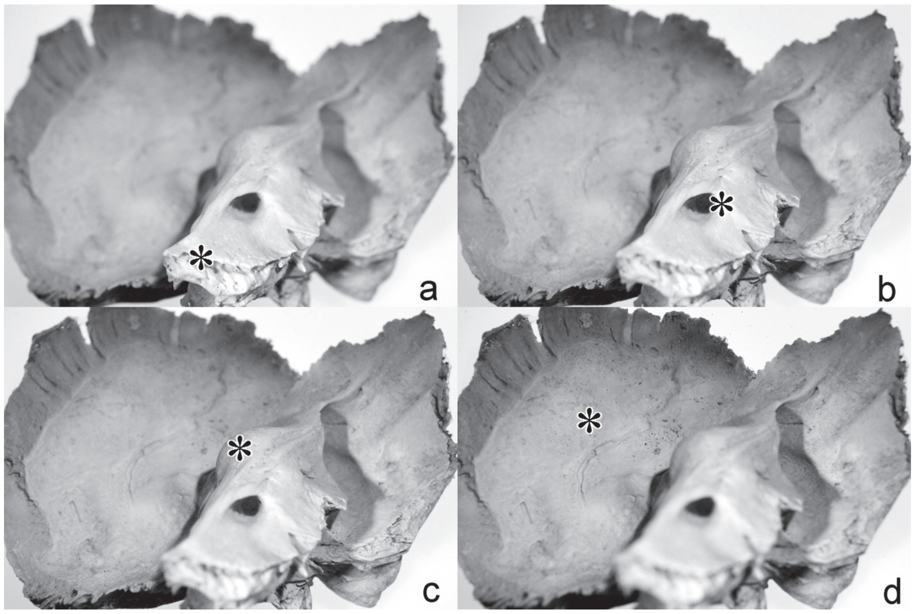

Figure 1A–D. Serial images of the temporal bone with different focal plane; asterisks mark the area in focus. Note that parts of the images are blurred (out of focus).

the temporal bone was subjected to digital photography to capture most of the anatomical details. The camera (Canon EOS 5D) was subsequently focused on four areas of the temporal bone, and the following were photographed: the petrous apex, the internal acoustic meatus, the arcuate eminence, and the inner surface of the squama. This procedure is called "focus stacking" because a set of images is shot by gradually incrementing the focusing distance across the object. As a result, singular details are seen sharply but in different images. The obtained photos are then aligned (their content overlaid pixel by pixel) by the computer program. A composite image is generated from the sharpest regions from each of the previously obtained separate images. The process of digital composition of the selected parts of each image in focus was performed by CombineZP Image Stacking Software by Alan Hadley (GNU Public Licence). This software was downloaded from the web page: www.hadleyweb.pwp.blueyonder.co.uk//CZP/News.htm.

RESULTS

In traditional digital photography each acquisition of the temporal bone shows certain regions of the specimen in and out of focus (Fig. 1). A set of 4 images was obtained by sweeping focus from the petrous apex to the squama of the temporal bone (Fig. 1A, D). In the first picture (Fig. 1A) the foreground (anterior part of the petrous bone) is sharp while the background (temporal squama) is blurred. Conversely, the fourth picture (Fig. 1D) presents a sharp background and blurred foreground. Subsequent images (Fig. 1B, C) can be regarded as average focus. In these two cases, the quality of the image can be acceptable. However, there are parts of the object in which anatomical details are not visible sharply. These are: the petrous apex, the internal acoustic meatus, the posterior part of the pyramid, and partially the squama with squamous suture.

The optical disadvantage was corrected by the algorithm of the image fusion, which allowed us to obtain a single picture of the temporal bone without blurred areas. The final image of the temporal bone presented in Figure 2 is the most satisfactory. All details of the temporal bone are easily distinguishable. Both the foreground (the petrous part) and the background (temporal squama) are sharp, and subsequent anatomical details are sharply visible. This is the effect of enhancement of the focal range, which was achieved by the digital assembly of images that contained in-focus parts of images which covered the whole depth range of an object (Figs. 1, 2).

DISCUSSION

In anatomical photography it is desirable to obtain the entire image in sharp focus. Thus, a large depth of field is appropriate to capture and visuali-

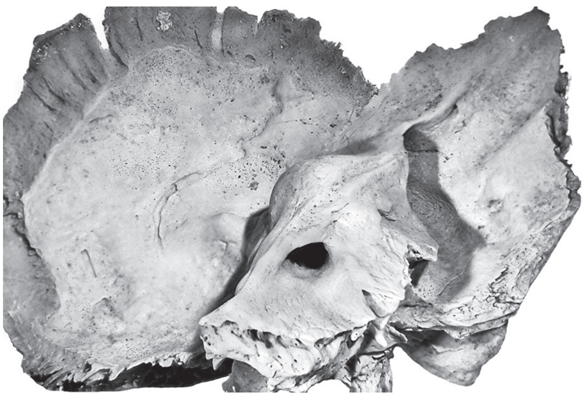

Figure 2. Composite deep focus image of the temporal bone, assembled from 4 separate images presented in Figure 1. Note that all areas of the image are sharp.

se in one image the spatial organisation of the observed anatomical structures. Unfortunately, one of the main problems in optical imaging is the limited depth of focus. This is an essential obstacle to acquire, in focus, in a single image plane, objects that are located at different distances [7]. The solution to this problem seems to be focus stacking, which enhances virtually depth of field. This technique has been used in microscope systems, and has become particularly beneficial in imaging thick objects. Using the extended depth of focus technology may increase by six to eight times the depth of focus of standard systems [1].

Focus stacking is regarded as a powerful technique, but it also has some limitations. These are: the photographed object must be motionless (a steady tripod is necessary to keep the camera still), a high precision focusing device is necessary, and specialised software is required to combine the stack of photos into a composite image.

Nevertheless, it is worth applying this procedure in anatomical photography to obtain better pictures. The argument for adopting this technique in anatomical photography is obvious because image quality is significantly enhanced. Thus, interpretation of the image becomes easier and is not biased [8]. In turn, high quality images can be subjected to automated quantification or become a reliable source of information in pathological analysis [5, 6].

Anatomical photography requires more depth of field than can be obtained in typical photography. Therefore, image stacking seems to be a helpful procedure because a composite digital image reveals all details in sharp focus across the object. A perspective view of many details and the precise capture of their spatial relationship is a clue to understanding the topography of anatomical structures. Hence, good quality photos are helpful in diagnostic and anatomical education. Application of the focus stacking algorithm opens a wide range of possibilities in careful imaging of morphological features of biological objects.

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