A MICROCOMPUTER-BASED ELECTRON MICROSCOPE DIGITAL IMAGE 3D PROCESSING SYSTEM

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ABSTRACT

A Microcomputer-based Electron Microscope Digital Image 3D Processing System(EM3DPS) is described in this paper. The system mainly includes some basic image processing functions and some procedures for obtaining various electron microscope 3D data such as contrast enhancement, false color enhancement, edge enhancement, image sharping, image smoothing, image segment, relative orientation and absolute orientation based on parallel projection, the display of stereoscopic pair on the microcomputer screen, the measurement of length, angle, area, volume, digital surface model(DSM) and contour line, etc.

Since the system just needs a general microcomputer(IBM PC/AT, 80286, 80386 or its compatibles just with VGA or TVGA) instead of a conventional photogrammetric plotters or a special image processing computer, so it is more accessible to electron microscope users. In addition, the friendly user interface based on windows and on-line help system make it easier and more convinent to operate the software.

Key Words: Close-range, Electron microscope, Digital processing system, Photogrammetry, 3D, Microcomputer-based, Biostereomatric

1. INTRODUCTION

Many papers have reported the principles and methods of obtaining electron microscope 3D data by using photogrammetry, and in some scientific fields, a lot of practical problems have been solved by means of these techniques. But all of the current electron microscope 3D surveying systems are based on some special photogrammetric instruments(analogic or analytic). Although many kinds of electron microscope 3D data can be gotten on these instruments, they cann't be widely applied in the electron microscope fields widely, because it is impractical for a general electron microscope user to purchase a special photogrammetric instrument to acquire some microcosmic 3D data.

Recently, as a result of the advances in semiconductor technology and micro-electronics and consequent reduction in prices, microcomputers not only have been available in many scientific fields but also have powerful functions, thus making it possible to design a microcomputer-based electron microscope digital image 3D processing system.

This paper is concerned with some pricinples, characteristics and performance of a Microcomputer-based Electron Microscope Digital Image 3D Processing System(EM3DPS) designed by the author.

2. MATHMETICAL MODELS

2.1 PARALLEL PROJECTIVE EQUATIONS

Theoretically speaking, the EM imaging system is central projection, but as regards extremely small specimen and extremely large magnifications, an electron microscope has an extremely small field angle. So the EM imaging system should be regarded as a parallel(or near parallel)projection. According to the features of the parallel projection, the parallel projective equations can be expressed as:

$$x = a_1(X-Xo)+a_2(Y-Yo)+a_3(Z-Zo)$$
$$y = kb_1(X-Xo)+kb_2(Y-Yo)+kb_3(Z-Zo) \quad (1)$$

where x,y are the photo coordinates of photo point; X,Y,Z are the object space coordinates of the specimen; Xo,Yo,Zo are the coordinates of the original of the photo coordinate system in the object space coordinate system; ai,bi(i=1,2,3)are the parameters of rotation matrix; k is the scale factor along Y direction, taking scale factor along X direction as 1.

On current electron microscopes, the various possible

movements of the specimen stage can be controlled precisely. So in order to make the procedure of surveying more simple, one usually takes each micrograph of a stereopair just with a tilt angle ψ_1, ψ_2 respectively. In this case, the Eqs. (1) can be simplified as follows:

$$x = \cos\psi\,(X-Xo) - \sin\psi\,(Z-Zo)$$
$$y = k(Y-Yo) \tag{2}$$

According to Eqs. (2), the formula, which can be used to calculate the model coordinates from the photo point coordinates, can be expressed as:

$$X = (\sin\psi_2 x_1 - \sin\psi_1 x_r)/\sin(\psi_2-\psi_1)+Xo$$
$$Y = (y_1 + y_r)/(k_1+k_r)+Yo \tag{3}$$
$$Z = (\cos\psi_2 x_1 - \cos\psi_1 x_r)/\sin(\psi_2-\psi_1)+Zo$$

EM3DPS adopts the Eqs. (3) as the intersectional equations (similar to the real-time program in an analytical plotter).

Note: In Eqs. (3), the y coordinate axes of the left and the right micrographs coordinate system are parallel to the tilt axis of the specimen stage.

2.2 RELATIVE ORIENTATION EQUATIONS

As in conventional photogrammetry, a stereomodel of the object can be formed by means of intersection of conjugate rays established by using two micrographs of the same specimen. Because EM imaging system is regarded as parallel projection, the condition of coplanarity can not be used in micrographs relative orientation. On analytical plotters, relative orientation with EM photographs becomes quite simple if, for generating the second micrograph of a stereopair, one uses only the elements of tilt and the associated x translation. But in EM digital image surveying system, the relative orientation with the same two micrographs is not simple, because during the procedure of digitizing micrographs by using a CCD camera or a scanner, the direction of sampling can not be ensured to be perpendicular to the tilt axis of specimen stage. This means that the y axes of the screen coordinate systems of digital images (similar to the y axes of the comparator coordinate systems on analytical plotters) are not parallel to the tilt axis of specimen stage. In order to make use of the Eqs. (3) to calculate the 3D model coordinates of the specimen, the angle between the tilt axis of the specimen stage and the y axes of the screen coordinate systems of the left and the right digital images must be gotten.
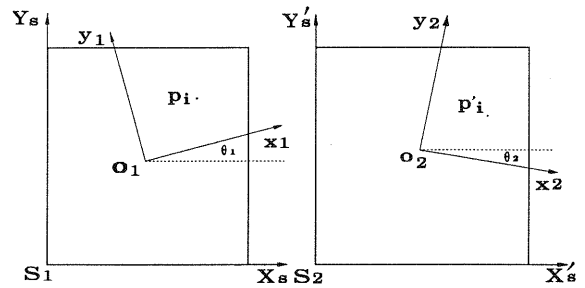

Fig. 1 Relative Orientation Geometry

This is the task of the relative orientation in EM digital image 3D surveying system. According to the parallel projective features (the planes formed by conjugate rays are all parallel to each other and perpendicular to the tilt axis of the specimen stage), the ralative orientation equation should be expressed as follows (refer to Fig.1):

$$yp_i = yp'_i \tag{4}$$

Where yp_i, yp'_i are y coordinates of p_i and p'_i in o_1-$x_1 y_1$ and o_2-$x_2 y_2$ coordinate systems. The y axes of these two coordinate systems are all parallel to the tilt axis of the specimen stage. o_1 and o_2 are a couple of conjugate points near the center of the left and the right images. S_1-$XsYs$ and S_2-$X'sY's$ represent the screen coordinate systems of the left and the right images. Eq. (4) can also be expressed by:

$$((Yp_i-Yo_1)\cos\theta_1-(Xp_i-Xo_1)\sin\theta_1)$$
$$-((Yp'_i-Yo_2)\cos\theta_2-(Xp'_i-Xo_2)\sin\theta_2)= 0 \tag{5}$$

where Xp_i, Yp_i, Xo_1, Yo_1 are the screen coordinates of the points p_i and o_1 on the left photograph; Xp'_i, Yp'_i, Xo_2, Yo_2 are the screen coordinates of the points p'_i and o_2 on the right photograph; θ_1 is the angle between o_1-$x_1 y_1$ and S_1-$XsYs$; and θ_2 is the angle between o_2-$x_2 y_2$ and S_2-$X'sY's$. After linearization, Eq. (5) becomes:

$$\frac{\partial f}{\partial\theta_1}\,d\,\theta_1 + \frac{\partial f}{\partial\theta_2}\,d\,\theta_2 + f^0 = 0 \tag{6}$$

where $\frac{\partial f}{\partial\theta_1}=-(Yp_i-Yo_1)\sin\theta^0_1-(Xp_i-Xo_1)\cos\theta^0_1$;

$\frac{\partial f}{\partial\theta_2}=(Yp'_i-Yo_2)\sin\theta^0_2-(Xp'_i-Xo_2)\cos\theta^0_2$;

$f^0 = ((Yp_i-Yo_1)\cos\theta^0_1-(Xp_i-Xo_1)\sin\theta^0_1)$
$\quad -((Yp'_i-Yo_2)\cos\theta^0_2-(Xp'_i-Xo_2)\sin\theta^0_2)$

θ^0_1, θ^0_2 are the approximate values of θ_1 and θ_2 respectively.
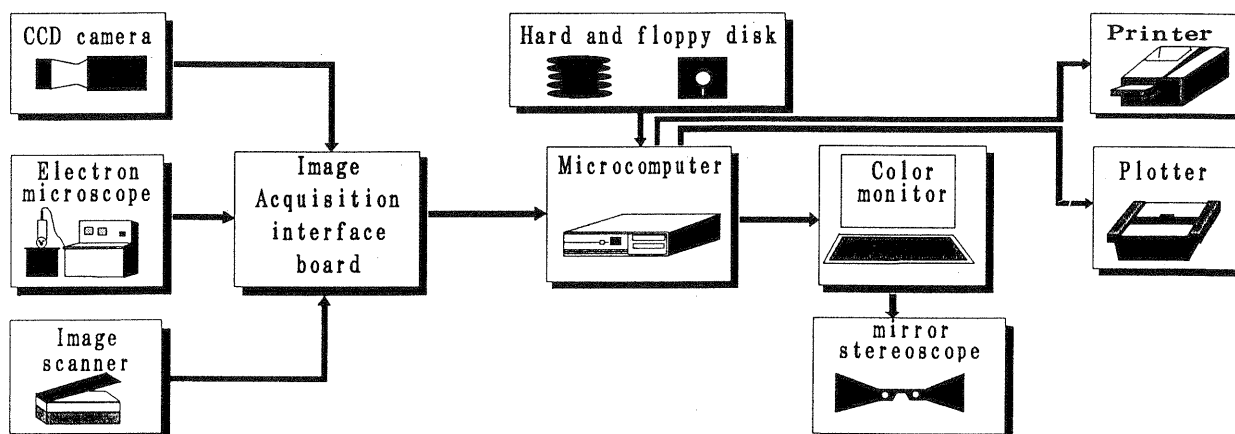
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Fig.2 Diagram of System Hardware Configuration

2.3 ABSOLUTE ORIENTATION EQUATIONS

Although EM imaging system is regarded as parallel projection, the absolute orientation equations for EM images are the same as these in conventional photogrammetry. Absolute orientation can also be done by means of microcosmic control points.

3 SYSTEM HARDWARE CONFIGURATION

The EM3DPS is functionally composed of four major components: digital image input instruments, a microcomputer, data output devices and a mirror stereoscope. Fig.2 is the diagram of system hardware configuration.

3.1 DIGITAL IMAGE INPUT INSTRUMENTS

Three types of image input instruments are used in the system. They are: an image scanner, a CCD solid camera and an electron microscope which is directly connected with a computer through an image acquisition interface board. Usually, these image input devices needn't be connected with the microcomputer installed EM3DPS. If only the format of the image data file of the input instruments is known, we can get the digital image from the floppy disk. Compared with the provious two types of image input instruments, the third has most potentials. A real-time electron microscope 3D processing system may be developed on this kind of hardware system.

3.2 MICROCOMPUTER

In order to realize various digital image processing functions and 3D data acquisition efficiently, the following microcomputer hardware components are required:

- IBM PC/AT, 80286, 80386 or its compatibles with at least 640KB memory;
- A 40MB hard disk driver and a 1.2 MB or 1.44 MB high-density floppy disk driver;
- A math coprocessor compatible with the CPU (80287 or 80387);
- A standard VGA or TVGA;
- A high resolution color monitor;
- 2MB or more expend or extend memory;
- An image acquisition board corresponding to a digital image input instrument.

3.3 DATA OUTPUT DEVICES

There are three types of data output devices in the system. They are:

- A printer. Various 2D or 3D data in table format can be output by means of the printer. Certainly, the printer can also be used to output graphics or black and white images in limited grey level. A HP LaserJet III printer has been used in the system.
- A plotter. The plotter is mainly used to output various graphic information such as contour line, perspective map and plane map, etc. A Roland DPX-3500 plotter has been used in the system.
- A high resolution color monitor. The monitor is not only the main output device in the system but also an observing device in image processing and stereomodel surveying. Various data, graphics and images can be output on the monitor.

3.4 MIRROR STEREOSCOPE

The system chooses split-screen imagery for stereomodel display, so a mirror stereoscope is required. Because in general situation, the y axis of the screen coordinate system of the left and the right digital images are not parallel to the tilt axis of the specimen stage(refer to 2.2)and the two angles
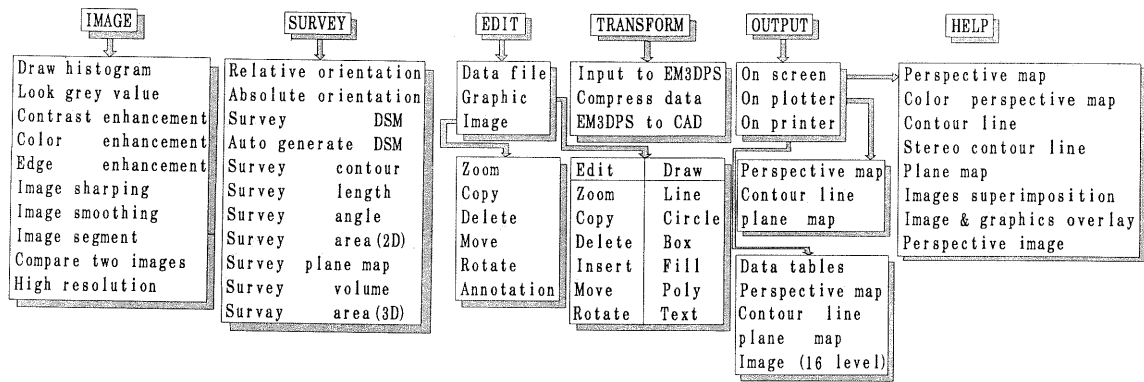
IMAGE	SURVEY	EDIT	TRANSFORM	OUTPUT	HELP
Draw histogram	Relative orientation	Data file	Input to EM3DPS	On screen	Perspective map
Look grey value	Absolute orientation	Graphic	Compress data	On plotter	Color perspective map
Contrast enhancement	Survey DSM	Image	EM3DPS to CAD	On printer	Contour line
Color enhancement	Auto generate DSM				Stereo contour line
Edge enhancement	Survey contour				Plane map
Image sharping	Survey length				Images superimposition
Image smoothing	Survey angle				Image & graphics overlay
Image segment	Survey area(2D)				Perspective image
Compare two images	Survey plane map				
High resolution	Survey volume				
	Survay area(3D)				

EDIT submenu:

Zoom	
Copy	
Delete	
Move	
Rotate	
Annotation	

Edit	Draw
Zoom	Line
Copy	Circle
Delete	Box
Insert	Fill
Move	Poly
Rotate	Text

OUTPUT submenus:

Perspective map
Contour line
plane map

Data tables
Perspective map
Contour line
plane map
Image (16 level)

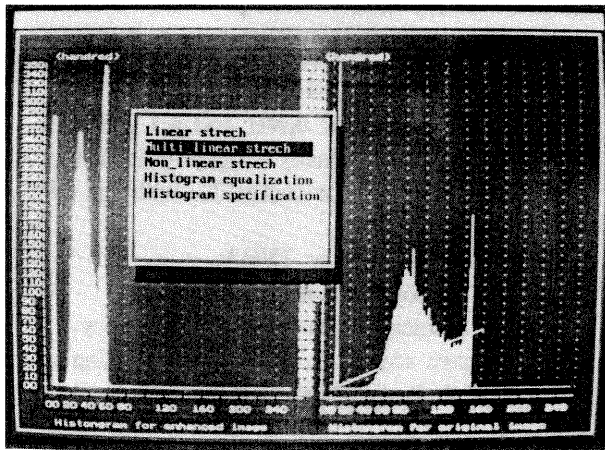Fig.3 The Diagram of The System Software

Fig.4 Screen menu display in the case of
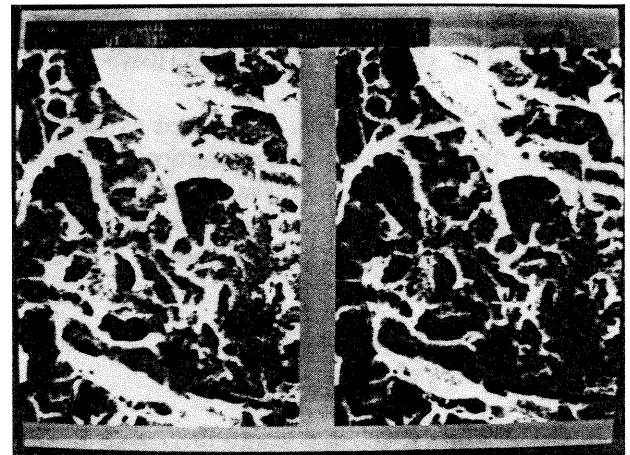contrast enhancement

Fig.5 The screen dislpay of a stereopair

θ_1 and θ_2 are not equal, it is difficult to observe stereomodel directly. There are two ways to solve this problem:

● the left and right optical system of the mirror stereoscope have the ability to rotate images.
● After the relative orientation, the left and the right digital images are rotated according to the two angles (θ_1 and θ_2) calculated by the relative orientation.

In order to make the optical system of the mirror stereoscope as simple as possible, EM3DPS uses the second method.

4. THE SOFTWARE OF THE SYSTEM

4.1 AN OVERVIEW OF THE SOFTWARE

All of the programs in the system are written in C and assembles languages, So the hardware potentials of the microcomputer have been used fully. So far, there are six main functions in the system. They are:
● Basic digital image processing functions (IMAGE);
● Orientations, 3D and 2D surveying (SURVEY);
● Data, graphic, and image editing (EDIT);
● Data file format transformation (TRANSFORM);
● Result data output (OUTPUT);
● On-line help system (HELP);

There are several sub-functions in each main function. All the functions in the system are orgnized in menu, thus making it quite easy to learn or operate the system. Fig.3 is the diagram of the system software functions and Fig.4 is the screen menu display in the case of image contrast enhancement.

4.2 CHARACTERISTICS AND OPERATIONAL ASPECTS

4.2.1 Basic Digital Image Processing. In order to improve the quality of the digital images which will be used in 3D or 2D surveying and analysing, the system provides some basic digital image processing functions. in addition to improving the image

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quality, these image processing functions can also be used to solve a number of 2D image analysis problems.

4.2.2 Orientations, 3D and 2D Surveying. Like the conventional photogrammetric surveying procedure, the operation of model surveying for EM images follow the usual steps of relative orientation, absolute orientation, and various surveying work.

At the relative orientation phase, only a part of the left and the right EM images are displayed on the screen at the same time(see Fig.5). In order to measure the screen coordinates of any photo point, the operator can not only move the two cursors on the left and the right images arbitrarily but also control the images panning and scrolling automatically. The relative orientation is solved by least squares using the Eqs.(6). Once the relative orientation is completed, the left and the right images are rotated respectively according to the θ_1 and θ_2 calculated by the relative orientation. The images used in later surveying stage are these two rotated images. Rotating the left and the right images means that the relative orientation elements(θ_1 and θ_2)have been " put" into the left and the right images respectively. So, to a given stereomodel, the relative orientation needs to be done just one time for subsequent surveying work.

Since the status of the specimen stage, during the procedure of taking the left and the right micrographs, can be controlled procisely, it is quite easy to horizontalize the stereomodel after the relative orientation. Usually, one is solely concerned with the relative information about the specimen instead of the absolute information, therefore, the offset in similarity transformation is not needed. Only in the case of the status of the specimen stage cann't be controlled procisely, the formula of similarity transformation is used in the absolute orientation with EM images.

After relative orientation and absolute orientation, various 3D data can be acquired. All 3D data processing functions have been listed in Fig.3. Surveiny DSM is the main task of EM images 3D processing(volume, 3D area and perspective image all use DSM). The system provides two ways for surveying DSM. In most situation, automatic generate DSM can be used. In this case, the stereomodel of the specimen is scannered along the lines parallel to the tilt axis of the specimen stage, and the mode coordinates are computed at regular intervals on these lines. Each new DSM point is marked on the screen by a red point superimposed on its conjugate point, this helps cheching and keeping track of the operation as it proceeds. The best correlated point is determined by the maximum value of the following equation:

$$k = \frac{(\sum a_{ij}b_{ij})^2}{\sum a^2_{ij}\sum b^2_{ij}} \qquad (7)$$

Where a_{ij} represents the grey value of pixel ij in the window of the left image and b_{ij} the grey value of pixel ij in the window of the right image.

Some times, an error correlated point may be calculated, in this case, the operator can interfere in the procedure by putting the red point on the correct conjugate point, then the automatic correlation go on.

4.2.3 Data, Graphic And Image Editing. In many cases, the users want to edit a data file, graphics and images. Certainly, the editing work can be done by using some popular text and graphics editing softwares. But this must lead to inconvenience in operating. EM3DPS provides some functions for editing data files, graphics and images. Although they are not perfect, they can meet most of the requirements.

4.2.4 Data Format Transformation. Electron microscope users have various image input instruments. To EM3DPS, the only difference of these image input instruments is the data format of their image files. In order to make EM3DPS surport as many as possible image input instruments, the system supplies a function of converting several types of image files to EM3DPS(input to EM3DPS). In adition, the cartographic plotters or printers owned by electron microscope users are also manifold. There are two methods to solve the problem of outputting the cartographic information on various plotters or printers:

● write various drawing drive programs corresponding to each kind of plotter or printer.

● take advantage of the current popular graphic softwares, since, usually, these softwares surport many types of plotters or printers.

EM3DPS uses the second method and provides a function of converting the graphic file of EM3DPS to the *.dwg files of Auto CAD(EM3DPS to Auto CAD).

4.2.5 Results outputting. The main aim of EM 3D data acquiring is to analyse the microcosmic structure of the specimen. So it is important to provides many ways for outputting various 3D or 2D data. Besides some conventional cartographic output functions, the system also provides some special 3D or 2D output functions. The perspective maps can only reflect the microcosmic structure qualitatively. In order to display the microcosmic structure qualitatively and quatitatively, the system gives two functions: color perspective map and stereo contour line(see Fig.3). The former function means that the contour line information are added into the DSM, and when the DSM is displayed perspectively, different colors are used between two contour lines. In order to reflect the microcosmic structure of the specimen more audio-visually, the system also provides a function of displaying perspective image. This function needs a DSM which includes the Z coordinate of each pixel.

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4.2.6 On-line Help System. In order to help the users, who do not understand the characteristcs and performance of EM3DPS very well, to learn how to operate the system quickly, EM3DPS provides an on_line help system. When the operator chooses HELP in the main menu, the system comes into on_line help status. At this time, the structure of the menu displayed on the screen is the same as before, but the color of the menu has been changed. This indicates that the system is in on_line help status. Now, after a system function has been chosen, some related help information will be displayed on the screen instead of performing the function. The help information has been written in two languages: English and Chinese.

5. SOME KEY TECHNIQUES

For developing an electron microscope digital image 3D processing system on a microcomputer, we must take account of some limitations of the microcomputer such as speed, memory space and the color number of the graphic card, and then use some techniques and methods to bring into full play the potentials of the microcomputer.

5.1 SPEED CONSIDERATIONS

The speed of displaying and operating images is one of the key problems in developing EM digital image 3D processing system on a microcomputer. The pulse frequancy of the microcomputer is definite, so the efficiency of programming affect the speed of displaying and operating images directly. The system chooses efficient C language as the main language and displays images by reading and writting the video memory directly. On the microcomputer without special image card, it is impossible for the two images of a stereopair to realize image roam at the same time. In order to observe all parts of the left and the right images through half of the screen, a so-called virtual roam has been used inEM3DPS. When the cursor approaches the edge of the image, new image data are automatically transmitted into the video memory from the disk. If the microcomputer is equiped with extend memory, it is suggested to build a virtual disk in the extend memory and store the EM images on it. The reason why the extend memory should be used by way of virtual disk is that this makes the EM3DPS more flexible, since the operation procedures of a physical disk and a virtual disk are the same.
To a single 1024 X 512 16 grey level EM image, VGA provides roam function in mode 12H(640 X 480 16 color). By means of the offset register in CRT control registers, a logical screen wider than the physical screen can be built. To display the image which is in the logical screen but not in the physical screen, the only thing to do is to update the startaddress register of the video memory.

5.2 MEMORY CONSIDERATIONS

Although EM images are not as large as aerial digital images, the problem of how to use the limited(640 KM in PC DOS or MS DOS) memory space in a microcomputer should also be considered in programming. The author has used different tactics to take advantage of the memory space for running the program and storing the image data.
5.2.1 Space For Running The Program. The total EM3DPS consists of about ten thousand of C original codes. In order to economize the memory space occupied by C original codes, the overlay technique has been used. As the size of data segment and code segment all exceed 64KB, the large mode is required in compiling and linking.
5.2.2 Space For Storing Image Data. VGA can only provides two types of grey level(16 and 64) to display white and black images. Since each pixel value only requires half byte to 16 grey level image, two pixel values can be compressed into one byte. This can economize half of the data space. This kind of image data file compressing can be performed in TRANSFORM of the system.

5.3 COLOR CONSIDERATIONS

Another limitation for developing EM digital image 3D processing system on a microcomputer is the color number. VGA can only display 16 colors in the highest resolution mode(640 X 480) at the same time. This mode can only be used to display 16 grey level white and black or false color images. In order to display 64 grey level images and realize images superimposition(or graphic and image superimposition), we must use mode 13H(320 X 200 256 color). In mode 13H, it is possible to perform two 16 grey level images superimposition or a few graphics and a 64 grey level image superimposition.

6. CONCLUSIONS

(1) At present, various powerful special computers for image processing and machine vision are emerging continuously. When these hardware systems are used, a better and more efficient electron microscope digital image 3D processing system can certainly be developed. Yet, as these kinds of systems are quite complex and costly, they are not readily accessible to general electron microscope users. On the contrary, the system described in this paper may be more acceptable, since the system meets the desirable requirements of flexibility, simplicity and adaptability.
(2) EM3DPS can complete various surveying tasks that can be done on conventional stereoplotters,

analogic or analytic. Moreover, it opens some interesting new aspects such as digital image processing, observing false color stereomodel, graphic and image superimposition, and digital surface model (DSM) automatic generation.

(3) A electron microscope digital image 3D processing system can be developed as a subsystem of a mature and general digital photogrammetric processing system. But it has practical significance to design a special microcomputer-based electron microscope digital image 3D processing system, since mostusers of this system are those scientists and engineers whose tasks are to research or analyse electron microscope photographs instead of other types.

(4) EM3DPS is just a fundamental system. SinceEM3DPS has been designed in the structure of modularity, the electron microscope users can develop more functions according to specific requirements in the application.

7. REFERENCES

(1) Agnard, J.P., P.A. Gagnon, and C. Nolette, 1988. Microcomputers and Photogrammetry. A new tool: The Video Plotter. Photogrammetric Engineering and Remote Sensing, Vol. 54, No.8, pp. 1165-1167.

(2) Albertz, J., and G. Koenig, 1984. A Digital Stereo Photogrammetric System. International Archives of Photogrammetry and Remote Sensing, Vol. 25, A2, Commission II, Rio de Janeiro, pp. 1-7.

(3) El-Hakim, S.F., 1985. A Photogrammetric Vision System for Robots. Photogrammetric Engineering and Remote Sensing, Vol. 51, No. 5, pp. 545-552.

(4) Gagnon, P.A., Agnard, J.P., C. Nolette and M. Boulianne, 1990. A Microcomputer-Based General Photogrammetric System. Photogrammetric Engineering and Remote Sensing, Vol. 56, No. 5, pp. 623-625.

(5) Gruen, A.W., 1989. Digital Photogrammetric Processing Systems: Current Status and Prospects. Photogrammetic Engineering and Remote Sensing, Vol.55, No. 5, pp. 581-586.

(6) Gugan, D.J., and I.J. Dowman, 1986. Design and Implementation of a Digital Photogrammetric System. International Archives of Photogrammetry and Remote Sensing, Vol. 26-2, pp. 100-109.

(7) Feng Wenhao, and Sun Lixin, 1990. Processing of Scanning Electron Microscope Image On WILD BC2 Analytical Plotter. International Archives of Potogrammetry and Remote Sensing. Vol. 28, Commission V, Zurich, Switzerland, pp.595-601.

(8) Feng Wenhao, 1985, "Non-topographic photogrammetry" (in chinese).

(9) Sanjib K. Ghosh, 1986. "Electron Microscopy— System and Applications". Photogrammetrie.

(10) Wang Zhizhuo, 1989, "Principles of Phtogrammetry (With Remote Sensing)". pp. 385-387.