Mapping from Transmission Electron Micrographs Using the Photogrammetric Plotting System

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ABSTRACT: There seems to be a growing demand within the scientific community, R&D departments, and biomedical disciplines for a better understanding of shapes and geometrical characteristics of microscopic objects. The application of photogrammetric techniques enables mapping the three-dimensional configuration of an object, thus revealing more of its geometrical characteristics. In this paper the object being mapped is a carbon black aggregate, a precisely manufactured form of polycrystalline graphite widely used in the rubber industry. Stereo micrographs of the object were taken using a transmission electron microscope (TEM) of 18,000 times magnification. These micrographs were then set up in a Zeiss stereocord G-2 interfaced through an electronic DIREC 1 to a Hewlett-Packard 9825A computer and a Hewlett-Packard 9872A plotter. It was possible, by means of this analytical system, to produce a digital terrain model (DTM) as well as longitudinal and lateral sections of the carbon black aggregate. Based on the DTM, a contour map of ten-nanometres contour interval was produced.

INTRODUCTION

Several successful attempts have been made to produce three-dimensional information from stereo electron micrographs (Boyde, 1970; Oshima et al., 1970; Ghosh, 1971; Ghosh et al., 1978). In this paper a simple analytical system (PPS) is used to extract external and internal information about microscopic objects using stereo transmission electron micrographs. This was possible due to the very nature of the imaging formation of the transmission electron microscope (TEM). The electron beam of the TEM system penetrates the object before it forms the image, thus revealing a great deal about the surface and the internal structure. Although surface details are more revealed using scanning electron microscope (SEM) micrographs (Nagaraja, 1974), the improved image definition resulting from TEM micrographs allowed surface sampling of a digital elevation model (DEM) used to produce a contour map. Meanwhile, the revealed internal structure of the aggregate enabled the plotting of longitudinal and lateral sections. This present application allows mapping of objects with dimensions below the micrometre level and can thus be classified as a micro-range photogrammetric application.

SYSTEM HARDWARE

The instrumental hardware used in performing this work may be grouped into two main classes according to their functional use. The image taking group (the Transmission Electron Microscope) and the three-dimensional reconstruction group (Photogrammetric Plotting System, Figure 1).

Transmission Electron Microscope (TEM)

The transmission electron microscope used is the Philips EM 200 model (Figure 2). The magnification...
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Taking the effect of magnification \( M \) into account, we get the expression for \( Z_L \) as follows:

\[
Z_L = \frac{I_1 P}{2 M} \sin \theta \cdot \tan \theta \tag{6}
\]

Also,

\[
X_L = X' - Z_L \cdot \tan \theta \tag{5}
\]

In case of carbon black aggregates, the maximum height difference between any two points does not exceed 0.5 \( \mu \text{m} \). Then, with the value of \( \theta \) being 7.5\(^\circ\) (value used for producing the micrographs), Equation 5 can be safely reduced to Equation 6 after considering the magnification because the second term

range of this instrument varies from approximately 500 \( \times \) to 220,000 \( \times \). The specimen stage allows for a total tilt of 60\(^\circ\), a rotation of 360\(^\circ\), as well as \( x \) and \( y \) shifts of 2.4 mm each. This movement of the stage allows the possibility of producing two stereo micrographs of the same object simply by taking the first micrograph in the normal position and the second after tilting the stage through an angle \( \theta \).

PHOTOGRAMMETRIC PLOTTING SYSTEM (PPS)

The term photogrammetric plotting system is used in this paper to describe the system shown in Figure 3, consisting of a Zeiss G-2 Stereocord, a Zeiss DIREC 1 electronic unit, a Hewlett-Packard 9825A computer, and a Hewlett-Packard 9872A plotter.

The Stereocord G-2 is an instrument used for quick and easy measurements from stereo photographs. It allows the capability of handling conventional and non-conventional photographs (Finke and Wölfang, 1977). The Stereocord is a much improved version of the Stereotope, where the function of the mechanical devices is replaced by the computer. This property makes it particularly convenient for mapping and processing of TEM micrographs taking into consideration the different geometry of the micrograph based on parallel projection (Elghazali, 1984).

The Electronic system DIREC 1 consists of a data section and a program section. It connects the stereocord to the computer through the Hewlett Packard TTL I/O. When called down by the computer, the model coordinates \((X_L, Y_L, Z_L)\) are successively transmitted from the computer to the plotter through the data interface.

The 9825A computer is well suited for real time systems and can store up to 250 K bytes of data and programs. It is characterized by a live keyboard status while a program is running and an interrupt capabilities, thus acting as a controller for the automatic plotter. The 9872A Hewlett Packard plotter ROM provides a convenient set of control measurements and produces a graphical record of the object being mapped by the Stereocord/computer combination.

MATHEMATICAL MODELING

Using the PPS analytical system, it is possible to form the model based on parallel projection principles. Parallax shifts between corresponding points in the left and right micrographs can be used to determine the spatial coordinates. The assumption is made that the field angle subtended by the electron beam hitting the specimen over the format area is so small that parallel projection geometry is applicable (Nagaraj a, 1974; Maune, 1976). In the development of the parallel projective transformations used with the PPS, a rather simplified approach was adopted. It was assumed that the object planimetric coordinates can be computed from the coordinates of the left micrograph after accounting for the tilt and magnification. This should be accepted to most users in view of the large magnification being used.

Elevations were computed based on parallax differences between corresponding points on both micrographs. Accordingly, the formula for parallel projective transformation can be derived according to Figure 4 as follows:

\[
\tan \theta = \frac{1}{2} \left( \frac{\Delta P}{\cos \theta} \right) Z_L \tag{1}
\]

where

\[
Z_L = \frac{\Delta P}{2} (\tan \theta) (\tan \theta) = \Delta P/2, \sin \theta \cdot \sin \theta \tag{2}
\]

Taking the effect of magnification \( M \) into account, we get the expression for \( Z_L \) as follows:

\[
Z_L = \frac{\Delta P}{2 M} \sin \theta \tag{3}
\]

Also,

\[
X_L = a \cdot b \tag{4}
\]

\[
X_L = \frac{x'}{\cos \theta} - Z_L \cdot \tan \theta \tag{5}
\]
...are the three-dimensional model coordinates; 
\( x', y' \) are the two-dimensional left micrographic coordinates; 
\( M \) is the magnification of the micrographs; 
\( \theta \) is the tilt angle; and 
\( \Delta P \) is the parallax difference.

Equation 8 is the basic equation used to transform the measured coordinates of the left micrograph and the parallax difference between left and right micrographs into three-dimensional model coordinates. In order to plot sections of the spatial model, it is necessary to rotate the model in space once around the \( X_L \) axis through an angle \( \omega = 90° \) to obtain longitudinal sections and another time around the \( Y_L \) axis through an angle \( \phi = 90° \) to obtain lateral sections. Figure 5 illustrates the effect of these rotations on the model, from which the following expressions for \( \omega \) and \( \phi \) rotations can be expressed:

\[
X_{\omega} = X_L \\
Y_{\omega} = Y_L \cdot \cos \omega + Z_L \cdot \sin \omega \\
Z_{\omega} = -Y_L \cdot \sin \omega + Z_L \cdot \cos \omega
\]

In matrix form Equations 9, 10, and 11 can be expressed more concisely as follows:

\[
\begin{bmatrix}
X_{\omega} \\
Y_{\omega} \\
Z_{\omega}
\end{bmatrix} =
\begin{bmatrix}
1 & 0 & 0 \\
0 & \cos \omega & \sin \omega \\
0 & -\sin \omega & \cos \omega
\end{bmatrix}
\begin{bmatrix}
X_L \\
Y_L \\
Z_L
\end{bmatrix}
\]

In the same manner, we have

\[
X_{\phi} = X_L \cdot \cos \phi - Z_L \cdot \sin \phi
\]
The purpose of stereo-photogrammetry is to establish the dimensions of an object represented in two different micrographs by means of stereoscopic viewing. The two micrographs are taken with the microscope vertical axis essentially vertical and by tilting the specimen holder. One micrograph is taken with a $0^\circ$ rotation and a predetermined angle of tilt ($\theta$) and the other is taken with $180^\circ$ rotation which, accordingly, develops an angle of tilt ($\theta$) in the opposite direction as illustrated in Figure 6. Besides the tilt angle, one has to establish the scale of the micrograph if absolute height differences between points and/or continuous mapping is needed. Because a micrograph is at a scale necessarily much larger than the object depicted, its use requires that the ratio between comparable measurements be known. To control the scale of the micrographs, a carbon replica grid mounted on a 200-mesh copper grid and made from a master diffraction grating with 2160 lines per mm in the crossed directions, was used. According to manufacturers specifications, the grid is inscribed with an accuracy of 3 percent. Thus, the interval in either direction of the grid between successive lines is $0.462963 \text{ \mu m}$, with a standard deviation of $\pm 0.013889 \text{ \mu m}$. The magnification of the used micrographs was 18000 times (18k). Knowing the values of the tilt angle ($\theta$) and the magnification ($M$), software was developed based on Equation 8 that transforms the left micrographic coordinates $x'$ and $y'$ as well as the difference in parallax $\Delta p$ between left and right micrographs into model coordinates $X$, $Y$, and $Z$. The coordinates are then plotted and heights are sampled in a regular grid of predetermined interval in both the $X$ and $Y$ directions, resulting in a digital terrain model of the carbon black aggregate (Figure 7). The program also computes the area of the outer perimeter of the aggregate as well as the perimeter length. Based on this information, accurate estimates of the aggregate volumetric and geometric configuration can be determined with a high degree of reliability. Using the orthogonality of the axes and, accordingly, no affinity or distortions are introduced.

### THREE-DIMENSIONAL MAPPING

In matrix form Equations 13, 14, and 15 can be expressed more concisely as follows:

\[
\begin{bmatrix}
X_{\phi} \\
Y_{\phi} \\
Z_{\phi}
\end{bmatrix} =
\begin{bmatrix}
\cos \phi & 0 & -\sin \phi \\
0 & 1 & 0 \\
\sin \phi & 0 & \cos \phi
\end{bmatrix}
\begin{bmatrix}
X_L \\
Y_L \\
Z_L
\end{bmatrix}
\]

(Equation 16)

Equations 12 and 16 are used to rotate the model in space through angles $\omega$ and $\phi$, respectively. If these angles are each made equal to $90^\circ$, the rotation matrices, $M_\omega$ and $M_\phi$, are reduced to the following simple forms:

\[
M_\omega = \begin{bmatrix} 1 & 0 & 0 \\ 0 & 1 & 0 \\ 0 & 0 & 1 \end{bmatrix}
\]

(Equation 17)

\[
M_\phi = \begin{bmatrix} 0 & 0 & -1 \\ 0 & 1 & 0 \\ 1 & 0 & 0 \end{bmatrix}
\]

(Equation 18)

The rotation matrices are both orthogonal; thus, the rotation of the model in space maintains the

![Diagram of model coordinate system and rotations](image)
CONCLUSIONS

This paper demonstrates the potentials of photogrammetry as a convenient tool for extracting useful information from microscopic objects. Conventionally, qualitative and quantitative information from micrographs have been based on two-dimensional analysis. It has been established from this study that additional information from the third dimension is certainly a valuable contribution. The photogrammetric plotting system proved to be a rather economic means to be used with unconventional imaging systems. The idea of combining the Stereocord with a microcomputer allows the transformation of two-dimensional image coordinates into the three-dimensional model coordinates to be shifted from hardware to software where special problems can be easily handled. The system permits detailed mapping capabilities, which has been very ably

Fig. 7. Digital terrain model sampled in a regular grid.

Fig. 8. Manual contouring based on the sampled DTM (C.I. = 10 nm).
Fig. 9. Longitudinal sections of aggregate at intervals of 3.34 nm.

Fig. 10. Lateral sections of aggregate at intervals of 3.34 nm.
demonstrated. Furthermore, the digitizing capability permits computer interfacing and subsequent rapid computations of basic object parameters. Such digital terrain modeling techniques have potentials which are considered worthy of serious in-depth investigations whenever accurate geometric information regarding microscopic objects is required.

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REFERENCES


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