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The Electron Microscope as a Structure Projector

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1. INTRODUCTION

The intuitive understanding of the process of 3D reconstruction is based on a number of assumptions, which are easily made unconsciously; the most crucial is the belief that what is detected is some kind of projection through the structure. This 'projection' need not necessarily be a (weighted) sum or integral through the structure of some physical property of the latter; in

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principle, a monotonically varying function would be acceptable, although solving the corresponding inverse problem might not be easy. In practice, however, the usual interpretation of 'projection' is overwhelmingly adopted, and it was for this definition that Radon (1917) first proposed a solution. In the case of light shone through a translucent structure of varying opacity, a 3D transparency as it were, the validity of this projection assumption seems too obvious to need discussion. We know enough about the behavior of X-rays in matter to establish the conditions in which it is valid in radiography. In this chapter, we enquire whether it is valid in electron microscopy, where intuition might well lead us to suspect that it is not. Electron–specimen interactions are very different from those encountered in X-ray tomography; the specimens are themselves very different in nature, creating phase rather than amplitude contrast, and an optical system is needed to transform the information about the specimen that the electrons have acquired into a visible image. If the electrons encounter more than one structural feature in their passage through the specimen, the overall effect is far from easy to guess, whereas in the case of light shone through a transparent structure, it is precisely the variety of such overlaps or superpositions that we use to effect the reconstruction. If intuition were our only guide, we might easily doubt whether 3D reconstruction from electron micrographs is possible: there is no useful projection approximation for the balls on a pin-ball machine! Why then has it been so successful? To understand this, we must examine in detail the nature of the interactions between the electrons and the specimen, and the characteristics of the image-forming process in the electron microscope. Does the information about the specimen imprinted on the electron beam as it emerges from the latter represent a projection through the structure? How faithfully is this information conveyed to the recorded image? These are the questions that we shall be exploring in the following sections.

2. ELECTRON–SPECIMEN INTERACTIONS

2.1. Generalities

The electron microscope specimen is an ordered array of atoms—highly ordered if the object is essentially crystalline or has been organized during the preparation process into some kind of array, and organized, or ordered more locally, in the case of isolated particles. It may be stained, in which case the light atoms of which organic matter is mainly composed will be selectively bound to much heavier atoms, but if very high resolution is desired it will probably be unstained. Its thickness will depend on its nature and on the resolution to be achieved, for reasons that will gradually became clear. The electric potential within the specimen is not uniform, since in the simplest picture, each atomic nucleus creates a Coulomb potential, screened to an extent that depends on the nature of the atom in question by its electrons. In reality, the situation is complicated by the bonding between the atoms,