CHAPTER TWO

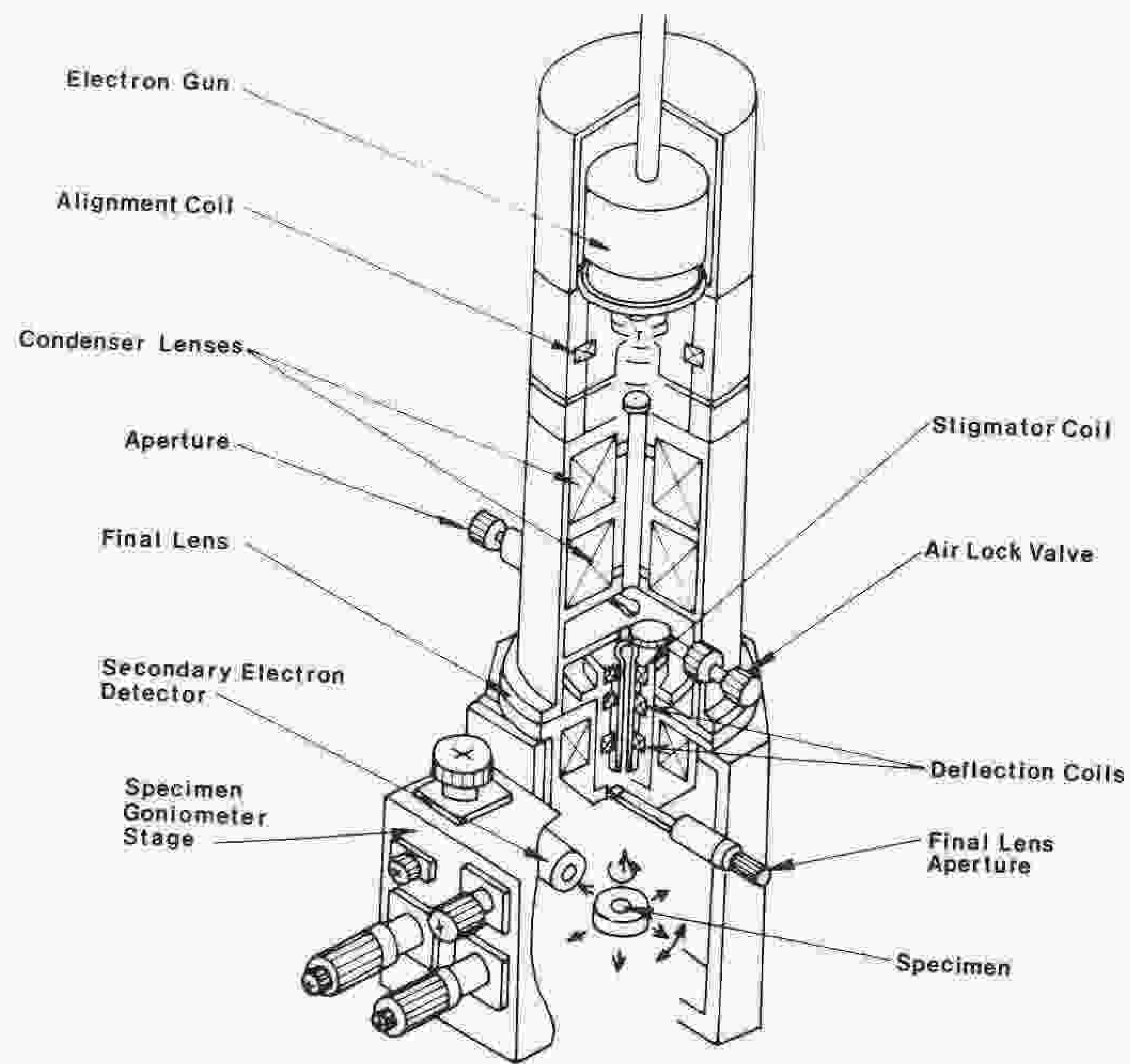
THEORY OF SCANNING ELECTRON MICROSCOPY

The previous chapter introduced the basic theory of microscopy. This chapter presents a detailed discussion of the many component parts in a typical scanning electron microscope. These components include the gun assembly, which produces a primary electron beam; the electromagnetic lenses and apertures, which focus the primary beam on the specimen; the vacuum system, which allows passage of the electron beam through the column without interference of air molecules; the specimen stage; and the signal detection and display components, which permit the observation and photography of an enlarged image of the specimen (Fig. 2-1).

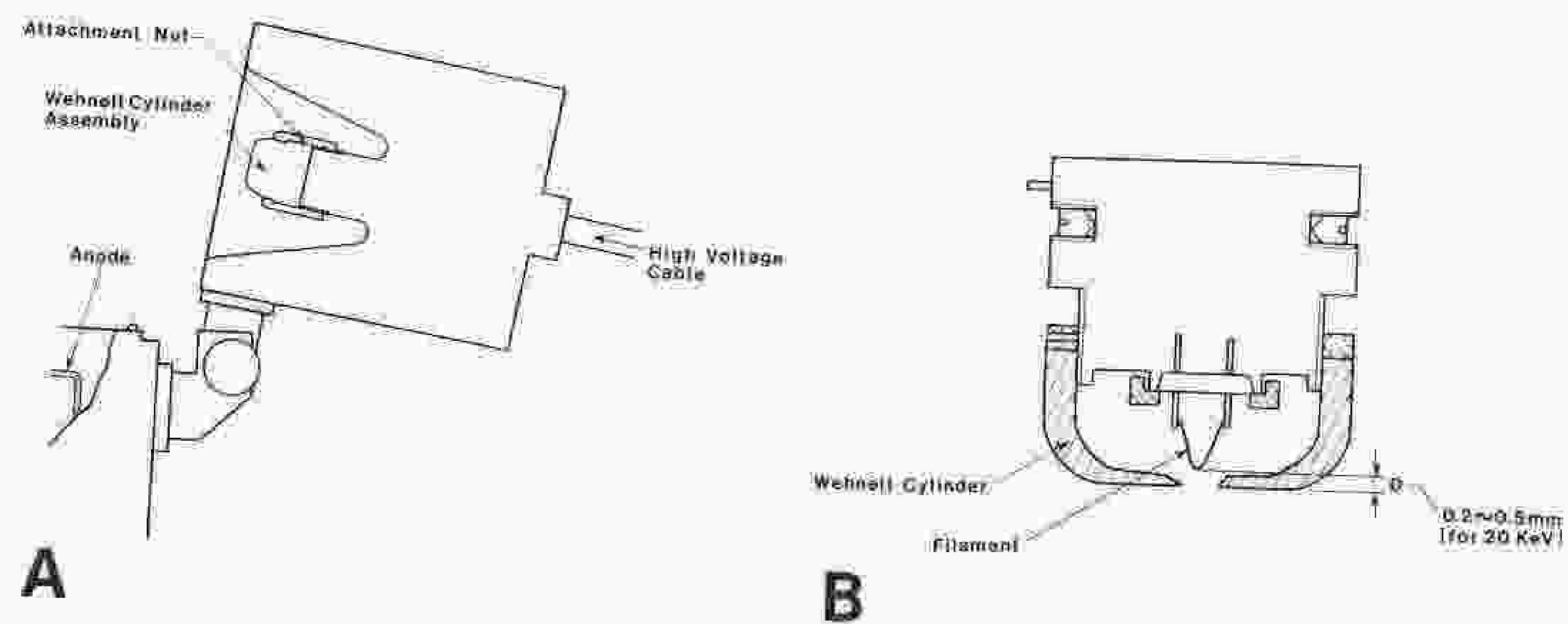
THE SCANNING ELECTRON MICROSCOPE COLUMN

THE ELECTRON GUN. The production of a high resolution image requires that a focused beam of electrons strikes the specimen. The simplest, and presently the most widely used electron gun, accounting for 95% of all electron microscopy (21), is composed of a hairpin-shaped tungsten filament (the cathode), Wehnelt cylinder, and an anode plate (Fig. 2-2). The gun and Wehnelt cylinder are connected to the negative pole of a high voltage supply and are characteristically located at the top of the microscope column. The filament is negatively charged. Thus, an electrical potential (a voltage difference) is established between the filament and the grounded anode plate. The negatively charged electrons are accelerated toward the anode. This voltage difference between the cathode and the anode plate is referred to as the "accelerating voltage." A flow of electrons from the filament (cathode), however, requires heating of the filament to overcome the work function of the metal. The emission of electrons from a filament due to heating is termed "thermionic emission."

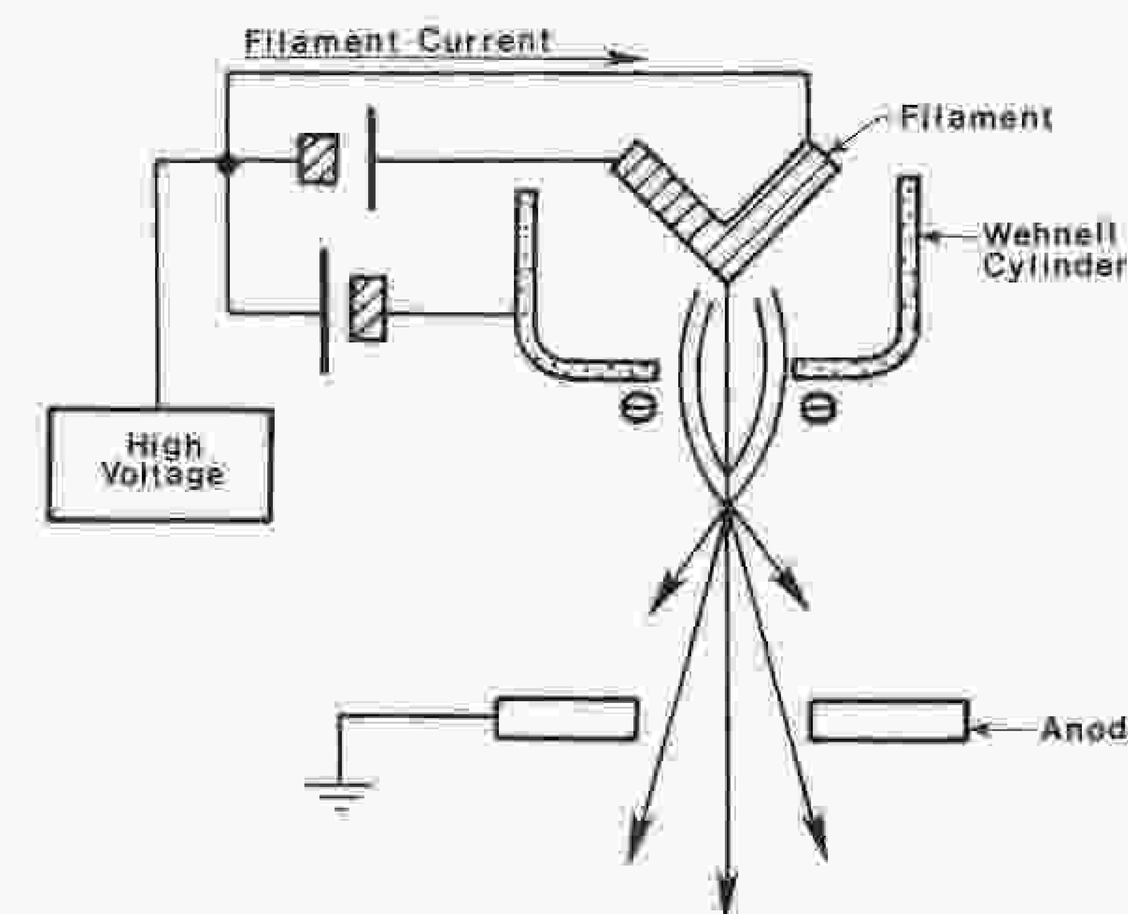
Thermionic emission results in a large spread of electrons outward from the filament tip. These electrons are contained and condensed into an electron cloud in a space between the filament tip and Wehnelt cylinder (also termed a gridcap) by a slightly greater negative charge on the cylinder than on the filament. This charge differential is referred to as "bias." The combined effect of the bias and the accelerating voltage toward the anode results in the creation and crude electrostatic focusing (focusing by electrical charge) of the primary electron beam within the gun (Fig. 2-3).



2-1. Schematic representation of a scanning electron microscope column (Courtesy of Hitachi, Ltd.).



2-2. Electron gun assembly of a scanning electron microscope. (A) Gun chamber opened revealing the electron gun assembly at the upper portion of the microscope column. (B) Wehnelt assembly and tungsten filament (Courtesy of Hitachi, Ltd.).



2-3. Thermionic emission of the electron gun depicting the effect of the negative bias of the Wehnelt cylinder and the grounded anode on the emitted electrons (Drawing courtesy of Hitachi, Ltd.).

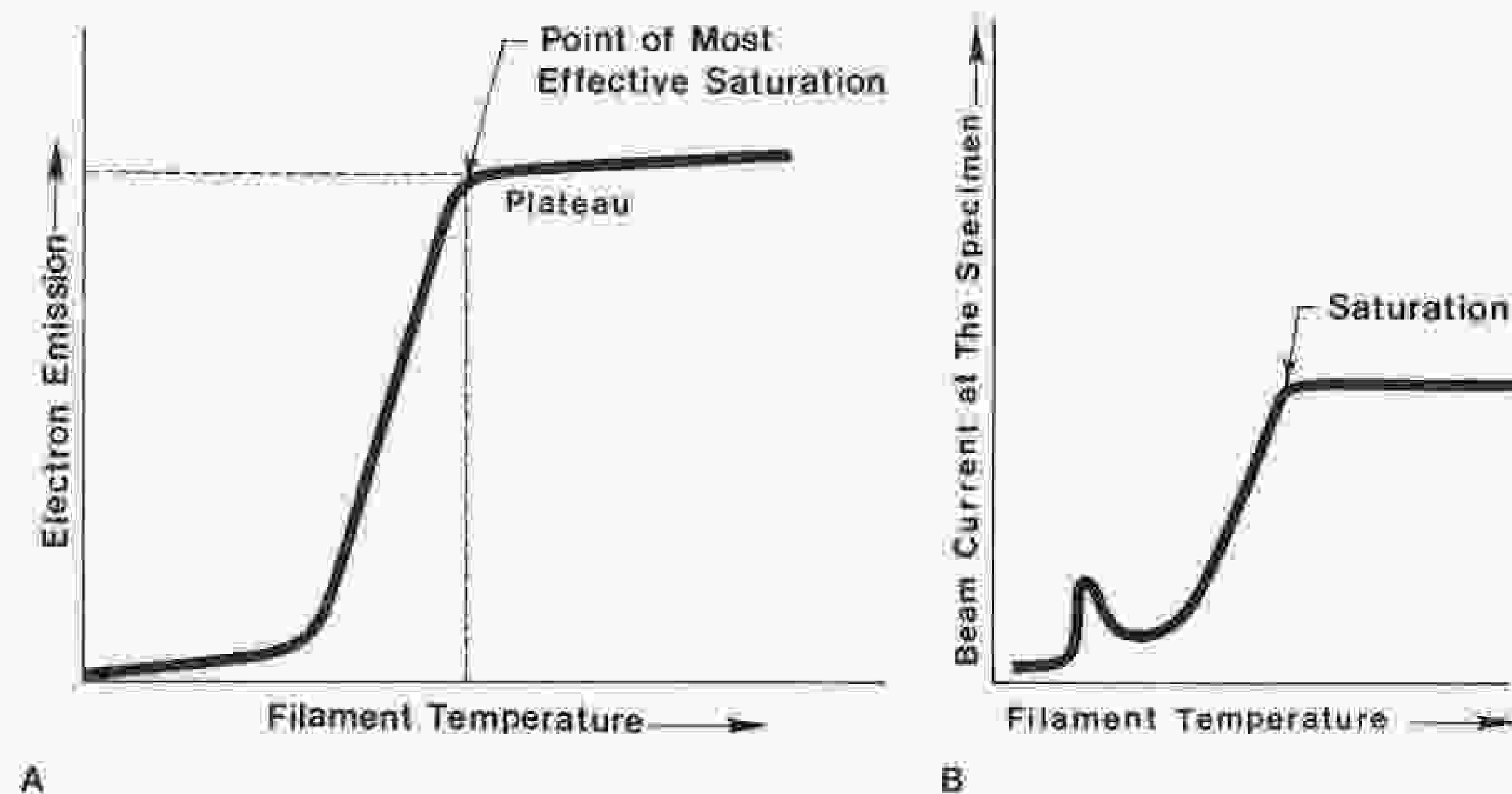
The positioning of the filament within the gridcap is important in creating a focused beam of electrons at the specimen. The correct configuration optimizes a controlled flow of electrons down the column to the specimen. When the best configuration is achieved, less heating of the filament is required and filament life is extended.

As current flowing through the filament increases, there is a significant increase in the number of electrons emitted up to a specific point referred to as saturation (Fig. 2-4). An increase in current beyond this point only slightly increases further electron emission. At the point of most effective saturation, the highest quantity of electrons are generated for the least amount of current.

The tungsten wire that composes the filament will wear out by evaporation and ion bombardment after a period of time. The life of a filament is dependent upon several factors: maintenance of high vacuum, cleanliness of the gun, and filament current. One of the major causes for the depreciation of filament life is its overheating by using currents higher than saturation (Fig. 2-5).

The electrons generated by the heating of a tungsten filament, as indicated above, are drawn toward the anode by an accelerating voltage. The accelerating voltage is variable on most scanning electron miscroscopes from approximately 1,000 to 50,000 volts. Each electron accelerating from the filament to the anode, acquires the energy of the accelerating voltage. Thus, when an accelerating voltage of 25,000 volts is used, the electrons in the beam acquire an energy of 25,000 electron volts. For the majority of the scanning electron microscopes and most specimens, 15,000 eV (15 KeV) to 25,000 eV (25 KeV) is optimal, but this may vary greatly with the specimen and particular application.

The accelerated electrons pass through a hole in the anode which acts as a crude aperture. This aperture allows some of the electrons to continue down the column while blocking the peripheral electrons. The smaller, more



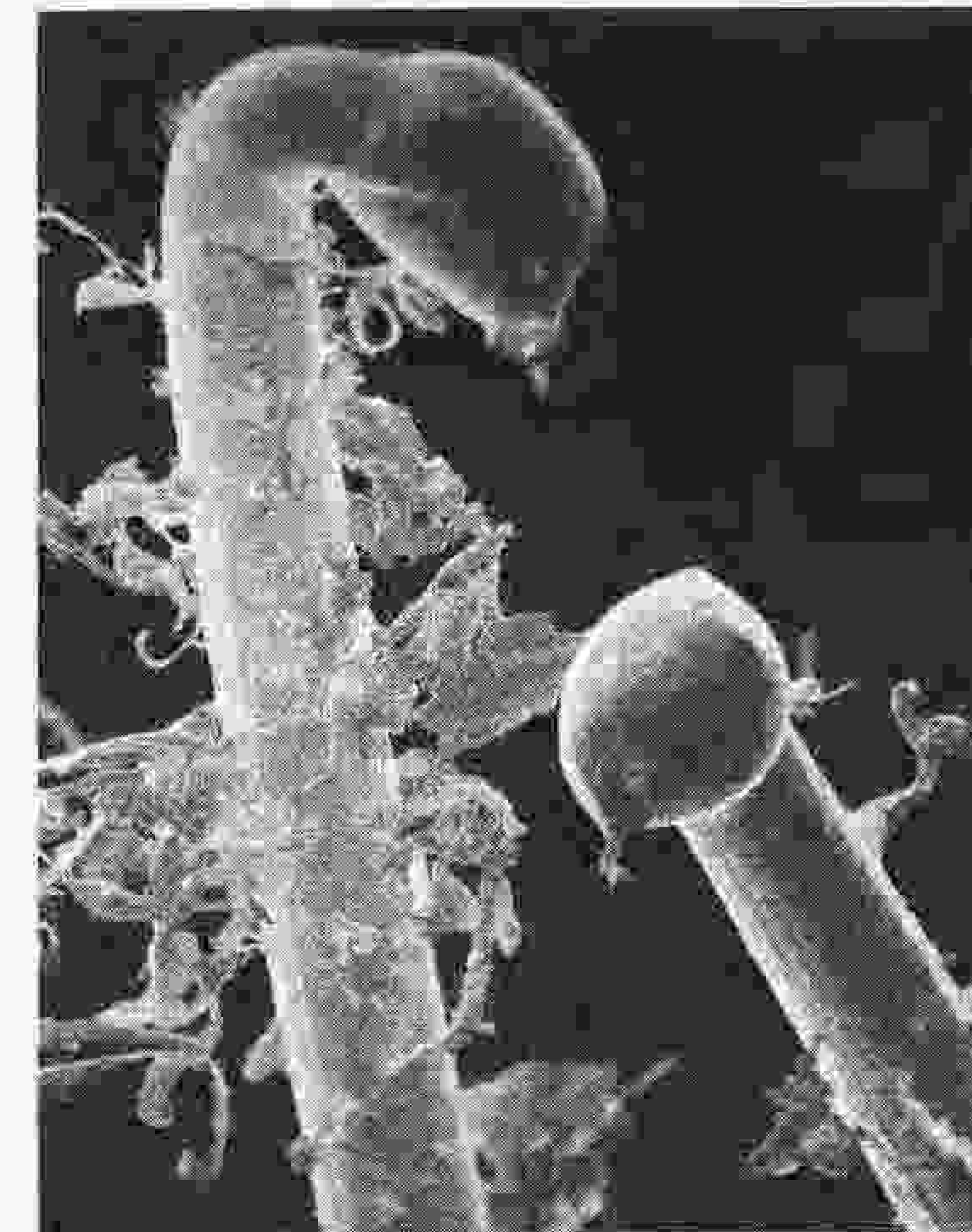
2-4. Saturation curve of a tungsten filament as measured by (A) increased current applied to the filament versus (B) the observed current at the specimen.

cohesive electron beam, continuing down the microscope column, tends to naturally diverge below this point. This spread outward is now influenced and converged by electromagnetic lenses.

ELECTROMAGNETIC LENSES. Electrons are charged particles. Thus, their paths may be influenced as they move through magnetic fields. An electromagnetic lens is essentially a length of wire coiled around a metal cylinder. Within the cylinder is found a soft iron pole piece. As current applied to the wire is increased, the magnetic field in the bore of the pole piece also increases in strength. The degree to which the path of an electron traveling through the pole piece is altered depends on the strength of this field, the velocity of the electrons, and the relative angle between the electron path and lines of force. Under the influence of such a magnetic field, electrons assume a helical path, spiraling toward the center of the pole piece. An excellent source for further information on the theory of electromagnetic lenses is available in Wischnitzer, 1962 (44) and Wells, 1974 (40).

The first lens that influences the electron beam is the condenser lens. This lens causes the electron beam to converge and pass through a focal point. As the condenser lens current is changed, the position of the focal point in the column (focal length) is altered, moving up or down the column, depending upon the current applied. A condenser lens setting is generally used that produces a focal point above a condenser aperture (Fig. 2-6). Although only a small part of the beam passes through the aperture, it is far more compact with many of the nonhomogeneous and scattered electrons having been excluded. Many modern microscopes use a second condenser lens below the first which provides more control over the electron beam.

The condenser lens, in conjunction with the chosen accelerating voltage, is primarily responsible for determining the intensity of the electron beam when it strikes a given specimen. As a result, it directly affects the brightness of the image

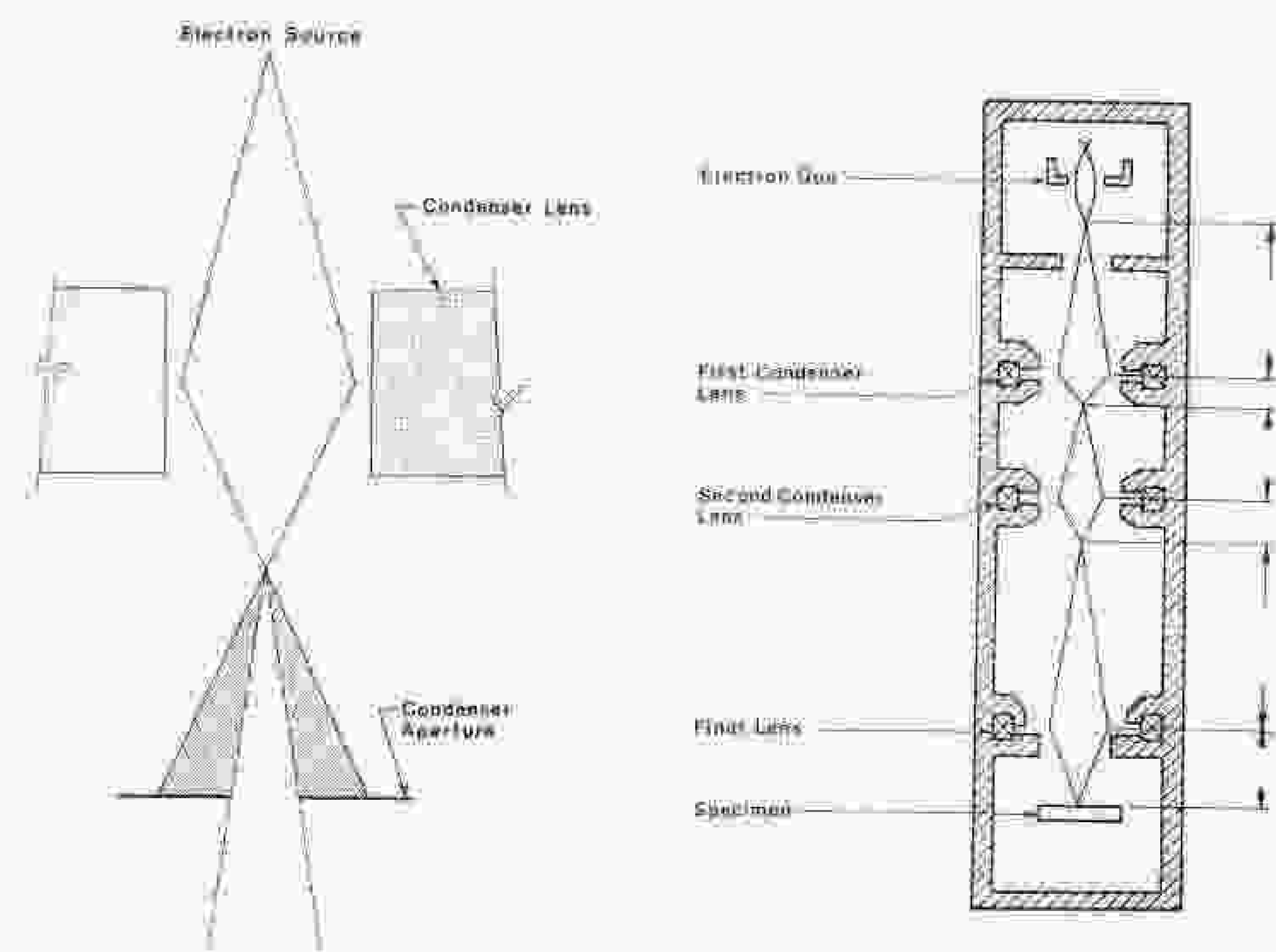


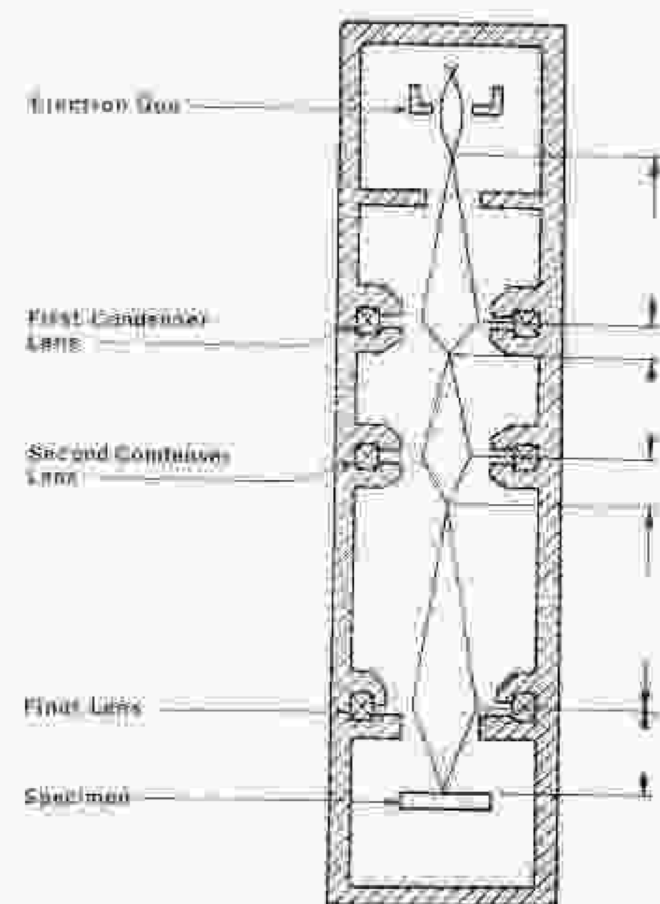
2-5 A scanning electron micrograph of a tungsten filament that has been "blown-out" due to gross overheating. Note the melted wire and the presence of the tungsten particles.

signal from that specimen. It is important to keep in mind that a high intensity beam may be harmful to a delicate specimen. Likewise, a high intensity specimen signal may damage the scintillation component of the detector system (to be discussed below). Therefore, caution should be exercised with respect to the condenser lens setting so that the safe operating limits of the specimen and scintillator are not exceeded.

The beam will diverge again below the condenser lens aperture. A final lens is used to bring the beam into focus at the specimen by demagnifying (converging) it to a focal point at the specimen surface (Fig. 2-7). The final lens demagnification determines the diameter or spot size of the electron beam at the specimen (17). Specimen resolution is determined by this beam diameter (37). Spot size reduction for optimum results is shared equally by all lenses in the column (25). Reduction is achieved by the appropriate combination of lens demagnification and aperture sizes.

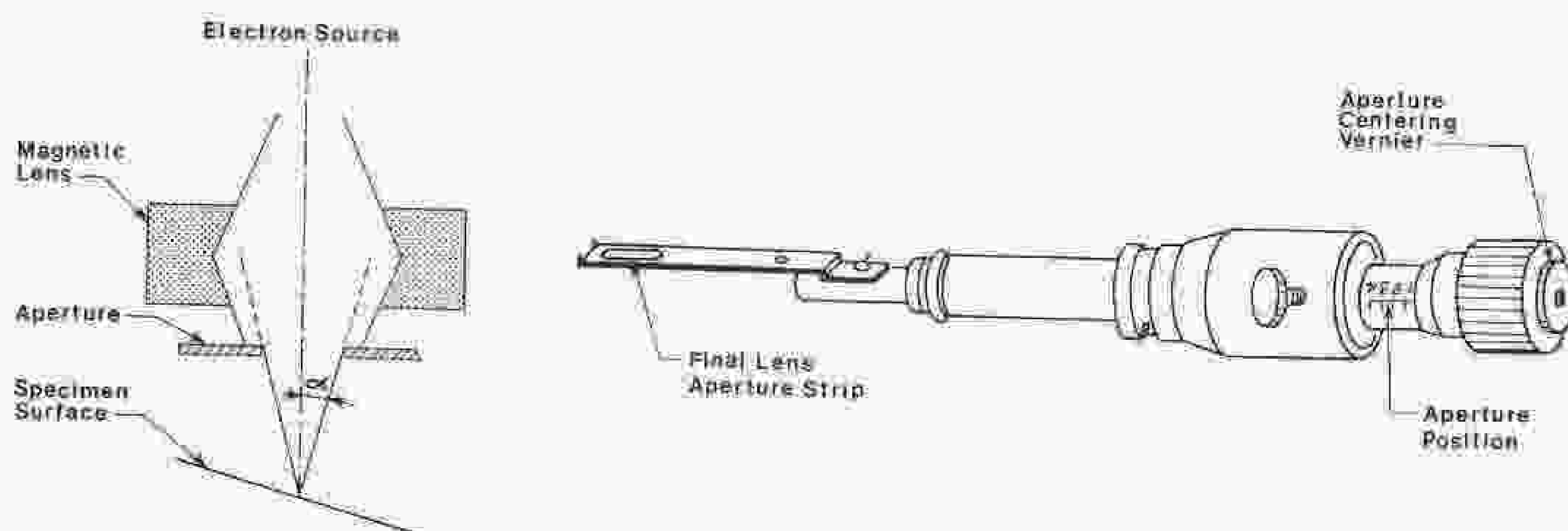
Apertures. Depending on the design of the SEM, one or more apertures may be present in the microscope column. "Spray-type" apertures may be used to reduce and exclude extraneous electrons in the lenses. In addition, an aperture may be used to reduce spherical aberrations in the final lens (35). This final lens aperture also affects the depth of field by determining the angle aperture (Fig. 2-8). Final lens apertures vary in diameter from about 100 to 400 μm, depending on the microscope. In many scanning electron microscopes, the operator may select one of several apertures for use. Decreasing the aperture size will result in an increase in depth of field with, however, a loss in relative brightness (Fig. 2-9).





2-6. A ray diagram of the electron beam as it is affected by the condenser lens and condenser lens aperture.

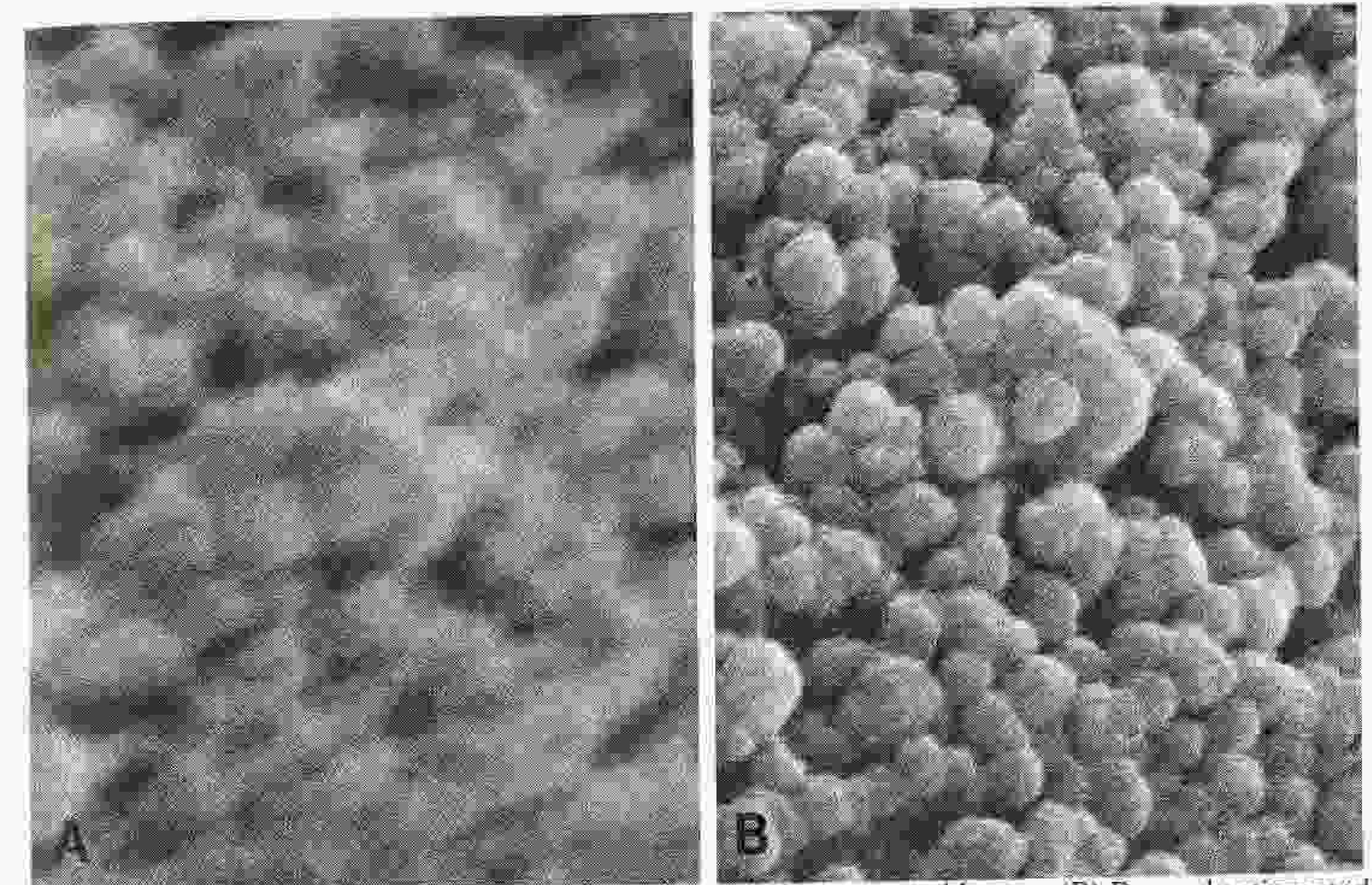
2-7. Diagram of the lens system of a scanning electron microscope column (Courtesy of Hitachi, Ltd.).



2-8. Angle aperture (α) as determined by the final lens aperture.

2-9. Selectable final lens aperture. Diagrammatic representation showing the various components. The aperture strip is composed of apertures of several sizes in a thin metal sheet. Note: To facilitate understanding, it has been drawn upside down (Courtesy of Hitachi, Ltd.).

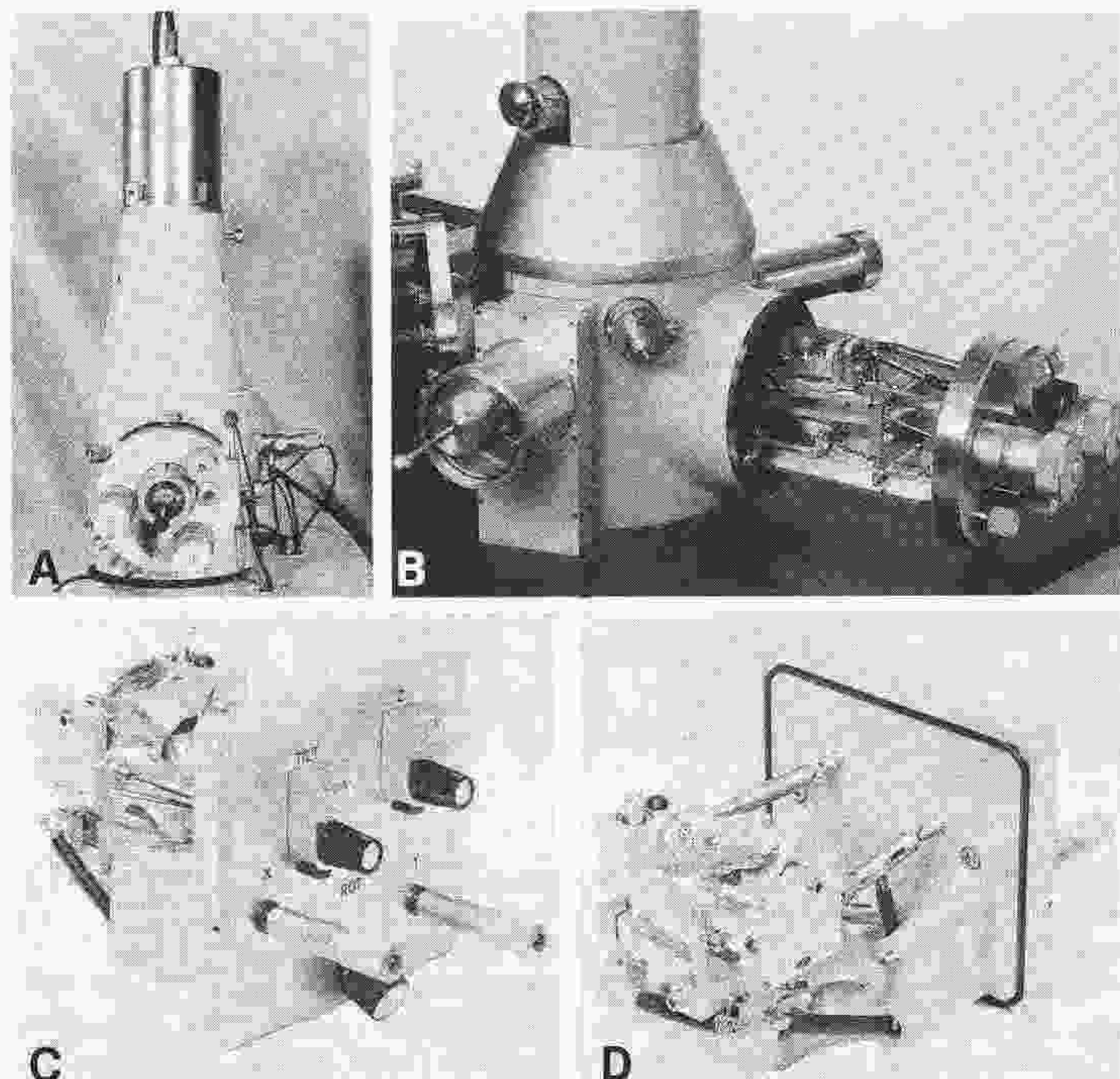
Stigmation. The electron beam should be circular in cross-section when it reaches the specimen. Because of lens defects (aberrations), contamination on the apertures, or contaminating particles in the column, the cross-section of the beam can vary in shape, for example, forming an ellipse. To compensate for such variations, the beam shape can be adjusted by a stigmator. A stigmator is a series of coils surrounding the beam below one of the lenses. By applying a directed magnetic field across the path of the electron beam, the stigmator induces the desired circular cross-section. This field is varied in orientation and magnitude by the operator of the microscope. Figure 2-10 demonstrates how such a stigmation process results in a higher resolution image.



2-10. A specimen showing extreme astigmatism. (A) Uncorrected image. (B) Properly stigmated image (Courtesy of AMRAY).

THE SPECIMEN STAGE. The specimen stage is the platform upon which the specimen rests in the column of the scanning electron microscope. The stage is located directly below the final lens and represents a significant portion of the specimen chamber. The design of the specimen stage and, for that matter, the specimen chamber, varies greatly from one microscope to another. Two basic design trends are foremost: pre-pump and drawer-type chambers. In the pre-pump chamber design, the major mechanical parts of the stage assembly are always within the vacuum chamber. The specimen, in such a system, is inserted through an air-lock from an evacuated pre-pump chamber (Fig. 2-11a). In the second design trend, the entire specimen chamber is vented at the time of specimen exchange, thus permitting the entire specimen stage to be removed in a drawer-like fashion (Fig. 2-11b). Independent of the manner in which the specimen is exchanged, these stages (commonly called goniometer stages) are designed so that the specimen can be oriented in nearly any direction under the electron beam. Specimen manipulation is accomplished by delicate micrometers which can move the specimen in the x, y or z directions. Further, the specimen may be tilted at least 55° or rotated 360° (Fig. 2-11c and d). All manipulations may be done while observing the specimen under the electron beam, thus allowing it to be photographed from any desired angle. Many stage modifications are available for specific applications such as large specimen observation, eucentric manipulation (rotation and tilt about a single point), and specimen temperature control. Another modification, the biological freezing stage, is discussed in Chapter 5.

Depth of field. Basically, "depth of field" is the extent of the zone on a



2-11. Basic specimen stage designs. (A) Pre-pump chamber type (courtesy of JEOL, USA); (B) Drawer type (courtesy of AMRAY). (C) and (D) Front and rear views of removable drawer (Courtesy of AMRAY).

specimen which appears acceptably in focus. When lenses are set to give the sharpest focus at an object, it is possible for the eye to observe an area above and below (or beyond) the focal plane which also appears to be in focus. This range over which the structures appear to be acceptably in focus is referred to as the depth of field. This concept has been defined for the SEM by Oatley, 1972 (35) and Goldstein and Yakowitz, 1975 (17).

In contrast to the light microscope, the scanning electron mischroscope has demonstrated, as one of its characteristics, a great depth of field at all magnifications. This is reported to be more than 100-500 times greater than the light microscope (31). Such an improvement is a consequence of the fact that the final lens aperture of the scanning electron microscope is small and, thus, the angle aperture of the focused electron beam scanning the specimen is also very small.

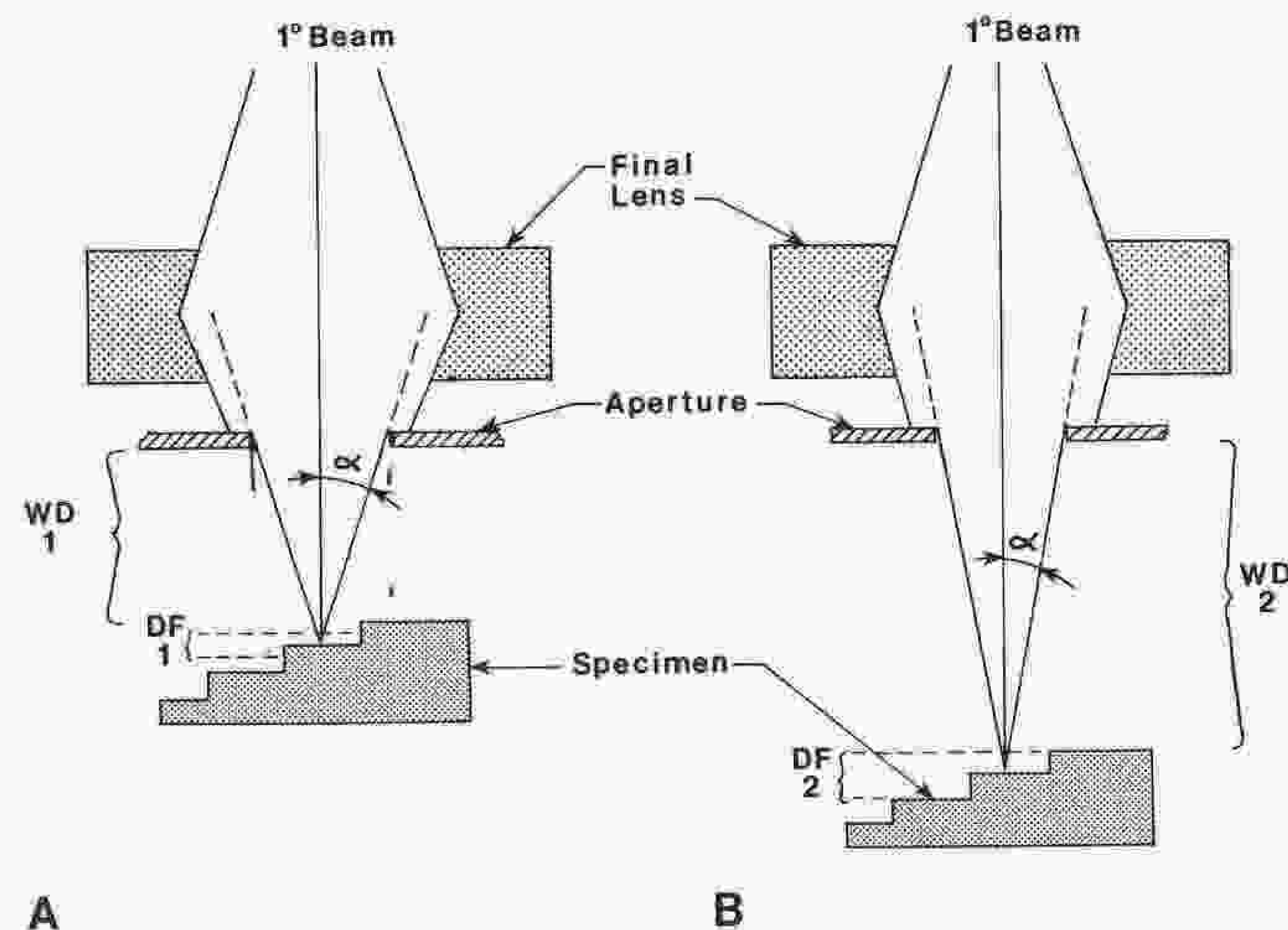
Depth of focus is a term which has been used interchangeably with depth of

field in regard to the scanning electron microscope. The term "depth of focus" corresponds to the depth of field in *image* space (44).

This confusion of terms stems from the design of the scanning electron microscope where the specimen is found below the final lens. Therefore, it is not analogous to either the light microscope or transmission electron microscope. In essence, either term could be and both have been used interchangeably for the scanning electron microscope. Since depth of field is defined using an area *on the specimen* itself and depth of focus refers to an image, in this book, we will use depth of field to refer to this characteristic of the electron beam-specimen interaction.

Enhancement of depth of field. One method for enhancing the depth of field in the scanning electron microscope is to lower the specimen from the final lens, thus increasing the *working distance,* and effectively decreasing the aperture angle (Fig. 2-12). The working distance is defined as the distance between the final lens pole piece to the uppermost portion of the specimen. The effective change in working distance on depth of field is summarized in Table 2-1.

A second method for increasing depth of field in the conventional scanning electron microscope can be accomplished by a reduction in the size of the final lens aperture. The smaller the aperture, the greater the depth of field, and conversely, the larger the aperture, the smaller the depth of field (Fig. 2-13).



2-12. Enhancement of depth of field by increase in the working distance. (A) Short working distance (WD). (B) Longer working distance showing increased depth of field (DF).