

PART ONE

MICROSCOPY
AND
TISSUE
PREPARATION

CHAPTER 1

Introduction

Ken Gross and Howard K. Steinman

MOHS SURGERY will remain the gold standard for the treatment of skin cancer until immunotherapy or other nondestructive modalities replace current surgical treatments. It allows skin cancer removal with higher cure rates and greater sparing of normal tissues than other excisional techniques. Mohs surgery accomplishes this in an office setting and at reasonable cost when practiced in an optimal fashion.

There are some common misconceptions about Mohs surgery that may stand in the way of optimizing the technique and that may unnecessarily increase its cost.

MISCONCEPTION 1

Mohs surgery is first and foremost about tissue sparing. It is not; Mohs surgery’s first goal is complete cancer removal. A focus on tissue sparing leads some Mohs surgeons to excise specimens with very narrow surgical margins even from areas where taking wider margins would not compromise function or closure. There are clearly many situations where excising wider margins would allow fewer stages of surgery, lower surgical costs, and would not substantively change the type of closure or lead to any cosmetic or functional degradation. In addition, if tissue sparing were the main goal of treatment, other modalities such as cryotherapy, radiation, and in selected cancers, presently available immunotherapy such as interferon alfa-2b and imiquimod would spare tissue to a greater extent while compromising cure rates by less than 5–10%.

MISCONCEPTION 2

Specimens need to be divided into many subsections (blocks) to allow optimal processing. This leads to relatively small specimens being divided into more than five tissue blocks for processing, which increases the cost and complexity of the procedure and the potential for processing and interpretation errors. It is more beneficial to process the largest blocks your technician and equipment allow

and produce more high-quality wafers from each block, thus allowing easier orientation and interpretation at lower cost.

MISCONCEPTION 3

Because Mohs surgery presumably allows for precise examination of approximately 100% of the tissue margins and precise localization of tumor foci, only minimal overlap of areas adjacent to positive findings needs to be excised. While Mohs surgery has more built-in precision than other cancer surgical modalities, there is plenty of room for small errors, which can be additive. For this reason, a wider margin around positive foci is sometimes rewarding to the surgeon and beneficial to the patient.

MISCONCEPTION 4

“Good enough is good enough.” The strength of Mohs surgery is that it examines approximately 100% of the true surgical margin. If complete base and epithelium (when available) are not represented on the slides, then 100% margin assessment has not been done and more sectioning through the block or obtaining more tissue from the patient is necessary. This is also an argument against doing vigorous curettement prior to the first stage of Mohs surgery: it may disrupt the peripheral epidermis, leading to incomplete peripheral margin on the slides.

MISCONCEPTION 5

Mohs surgery is difficult to perform and requires extensive training. Mohs surgery differs from other types of skin cancer excisional surgery only in a few aspects:

- 1. It is highly organized and system dependent, requiring excision of tissue to allow optimal processing of the complete, contiguous surgical margin and a highly skilled technician to prepare quality slides.

4 MOHS SURGERY AND HISTOPATHOLOGY

- 2. It requires accurate pathologic interpretation of horizontally cut tissue (see Chapter 11) in contrast to vertically oriented tissue normally reviewed on pathologic and dermatopathologic slides.
- 3. It requires the surgeon to assess the pathologic slides as well as perform the surgery.

The greatly advanced surgical skill set of many dermatologists, improved surgical training opportunities, modern surgical instruments, better local anesthetics, improved agents and devices for hemostasis, and better patient monitoring and resuscitation equipment make the surgeon’s job easier and faster. Modern cryostats, chromacoding inks and stains, improved automated tissue processing, and better microscopes have all combined to make slide preparation faster, quality better, and interpretation easier than ever before.

It is therefore reasonable to assume that a higher degree of accuracy is possible in utilizing the Mohs technique today than has been in the past. In assessing the relative value of more surgical training versus more pathology training for a dermatologist (or any other physician) wishing to perform Mohs surgery, an additional year of pathology study might be of more value than an additional year of surgical training – although this is viewed with reservations, as one of the justifications for a Mohs fellowship is to allow the pathologic and clinical review of hundreds of Mohs cases. In either case, additional training is of value. We would argue that dermatologic residency training usually does, and always should, provide the training in skin surgery and skin pathology that allows the performance of a basic level of Mohs surgery within a dermatologic practice.

Mohs surgery is a cancer removal modality and defect repair may be done by the Mohs surgeon or another surgeon, and wounds may be allowed to heal by second intention.

MISCONCEPTION 6

Mohs surgery is like a religion with inviolable precepts dictating who may perform it and how it must be performed.* The only absolute precept is that, as closely as possible, 100% of the true surgical margin must be accurately assessed to ensure complete removal of the cancer. There is no specific way in which the technique must be performed. Mohs surgery obviously cannot deal with discontinuous tumors, cancer cells that have already left the surgical field, or occult satellite and/or in-transit metastasis. Whichever reliable and reproducible method the Mohs surgeon selects to accomplish the goal of 100% margin assessment and complete cancer removal should be accepted, politics aside. Although sparing normal tissue is an important virtue of Mohs surgery, the one true goal of Mohs surgery is curing the cancer. It is the ability of properly performed Mohs surgery to achieve a high cure rate that allows it to be the premier technique for the removal of skin cancer. How exactly to optimize Mohs surgery techniques that allow the Mohs surgeon to reproducibly and consistently produce these high cure rates is just detail. These details are what this book is about. We assume that the reader of these pages has a basic understanding of Mohs surgery or has read *Mohs Surgery: Fundamentals and Techniques*.¹

REFERENCE

1. Gross KG, Steinman HK, Rapini RP. *Mohs Surgery: Fundamentals and Techniques*. St. Louis, Mo: CV Mosby Co; 1998. (The book may be ordered from ASMS (800) 616-ASMS.)

* The requirement that the Mohs surgeon must also be the Mohs pathologist is arbitrarily based only on CPT regulations. No other cancer surgery excludes the participation of pathologists or dermatopathologists from helping interpret pathology specimens.

CHAPTER 2

How to Excise Tissue
for Optimal Sectioning

Ken Gross

THE GOAL OF MOHS surgery is to cure skin cancer. Optimization of Mohs surgery ensures that the high cure rates available with this technique are achieved in practice. Production of the highest-quality Mohs slides makes possible the most accurate interpretation of the surgical margins represented on those slides. The Mohs surgeon, by optimizing tissue excision at the operative table, allows the Mohs technician to produce high-quality slides that present complete surgical margins of all excised tissue.

A masterful Mohs technician may be able to salvage tissue excised with poor surgical technique, and a poor technician can make garbage from an exquisite surgical specimen. In this chapter we focus on issues of surgical technique that will help the competent Mohs technician prepare better slides and allow faster and more cost-effective Mohs surgery. Optimizing surgical technique allows for the most favorable slide preparation. The Mohs surgeon, when switching hats and becoming the Mohs pathologist, will then have the best chance of making accurate surgical margin assessments.

HOW TO EXCISE TISSUE FOR OPTIMAL
SECTIONING

Even before making the first incision, the Mohs surgeon can increase the chance for complete cancer removal in as few stages as possible. The clinical margins of the tumor should be assessed with bright light and magnification. Use of an episcope and Wood’s light may help define the margins of some cancers, especially pigmented lesions. Re-assess the clinical margins after injection of anesthesia, as tumor margins may become more distinct after injection. Small ink dots can be drawn on the skin around the cancer to define the clinical cancer margins. Three decisions must then be made:

- 1. Should curettage be done to help further define the margins?
- 2. Should the cancer be debulked?

- 3. How much surgical margin past the clinical tumor should be removed?

Curettage prior to stage I cancer removal may help define the tumor margin and also debulks the cancer, but the downside of curettage is possible removal of some of the epithelial edge, especially in older patients with fragile skin. This makes margin assessment more difficult and may require a wider surgical margin around the disrupted epithelium.

Debulking is primarily done in two situations:

- 1. When there is a large bulky exophytic tumor, debulking the tumor makes tissue processing easier. As this is done sharply and should not extend to the specimen margins, there should be no loss of epithelial edges. If the Mohs surgeon debulks using a surgical scalpel, the blade must be wiped thoroughly free of tumor fragments, or changed, before excising stage I tissue. The Mohs technician may also debulk tissue in the laboratory, also thoroughly wiping the blade or using a separate blade before processing the specimen, to prevent artifactual tumor “floaters” from appearing on the slides.
- 2. To produce a vertically oriented slide of the tumor pathology when a previous biopsy is unavailable or has not been done. Having the tumor pathology available is very helpful for accurate slide interpretation.

Since Mohs surgery is most often performed following a diagnostic biopsy, there is frequently a “scab” on the cancer site. In large or deep cancers this scab is of no consequence, but in small and relatively shallow tumors it will interfere with the production of high-quality slides. It should be removed by the surgeon or technician prior to processing the tissue.

The issue of how much margin to take past clinically evident tumor is influenced by several factors:

- 1. Will removal of margin past the clinically clear margin cause a functional defect?

6 MOHS SURGERY AND HISTOPATHOLOGY

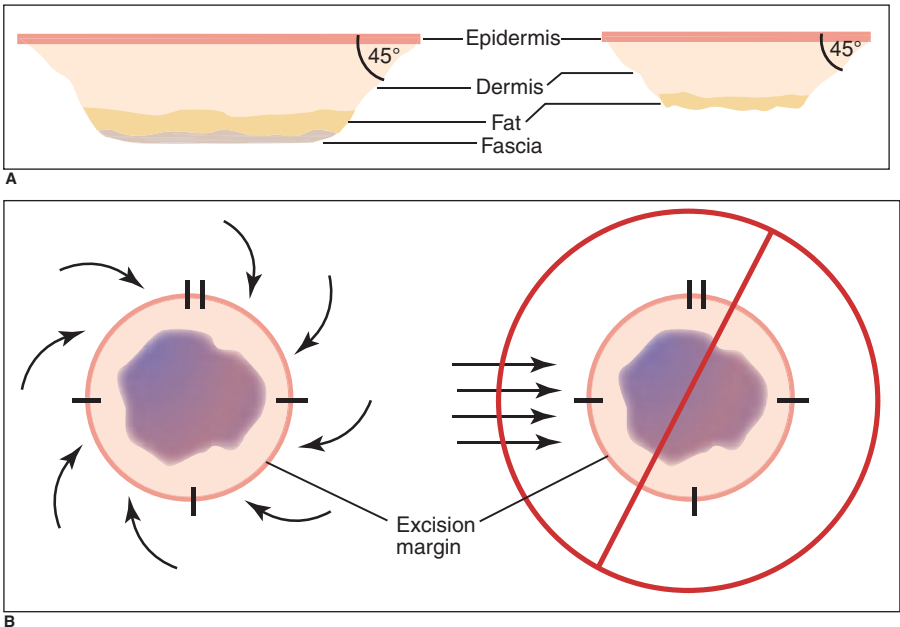


FIGURE 2.1: (A) The bevel is continued to the level at which the horizontal base of the specimen is to be cut, or to the level at which closure will be done, if that is deeper. On the arm, the Mohs surgeon would carry the bevel to the level of the fascia, as undermining and repair will usually be done at that level (left). On most areas of the face, the bevel would be cut to superficial fat (right), unless it is clinically obvious that the tumor is deeper, because that is the level at which undermining and closure will be done. (B) The specimen should be undermined from all edges toward the center and not from one edge through to the other side. Curved arrows (left) indicate that the Mohs surgeon is correctly cutting the specimen from all sides toward the center. Straight arrows (right) indicate that the Mohs surgeon is incorrectly cutting the specimen from the 9 o'clock side straight through to the other side. This is likely to lead to an unevenly cut specimen base and an irregularly beveled peripheral margin.

- 2. Will removal of additional peripheral margin increase the difficulty or morbidity of the closure?
- 3. Will removal of additional deep tissue margin compromise the function of a motor nerve or other important underlying structure?

If a smaller margin is taken around the tumor for any of these reasons, the cure rate is not compromised because any positive margin will be removed in a subsequent stage.

The primary purpose of Mohs surgery is to achieve a high cancer cure rate. When necessary, it also has the ability to spare tissue. But this ability is subject to abuse. A small but poorly clinically demarcated sclerosing basal cell in the mid-to-lateral cheek can be removed to below mid-fat with little risk of damage to underlying structures; and a peripheral surgical margin of 5 mm or more, as opposed to 1–2 mm, will be unlikely to cause closure or cosmetic problems. This would not be true for the excision of the same tumor on the lip or eyelid. Here, the ability of Mohs surgery to spare tissue shares equal importance with its ability to achieve a high cure rate.

The Mohs technique usually requires that the edges of the specimen(s), which in stage I are epidermal or mucosal,

be flattened into the same plane as the base during processing (see Chapter 6 through 8). This allows the entire deep and peripheral margins to be represented contiguously within the tissue wafers. To allow the technician to more easily flatten the tissue into a single plane for sectioning, the Mohs surgeon generally excises specimens at an approximately 45-degree angle (bevel). In the excision of large specimens, this angled cut continues only to the deepest plane of excision. At the deepest plane, the rest of the excision is horizontal (Figure 2.1A). To ensure as uniform a bevel and as flat a base as possible, the specimen should be cut from all sides toward the center and not from one edge continuously through to the other side (Figure 2.1B).

Although a 45-degree bevel is often stated to be ideal, many thin tissue areas such as the eyelid, genitalia, neck, and mucosa require little or no beveling. Thick, stiffer tissue areas, such as the back, may require more of a bevel (an angle of 30 to 40 degrees). As excisions progress deeper, the scalpel traverses first the epithelium and dermis, then fat, fascia, muscle, and periosteum. These tissues have differing abilities to flatten during tissue processing; thus, the amount of bevel needed to produce optimal slides will also change. Specimens of smaller diameter may be more difficult for the technician to flatten than larger specimens

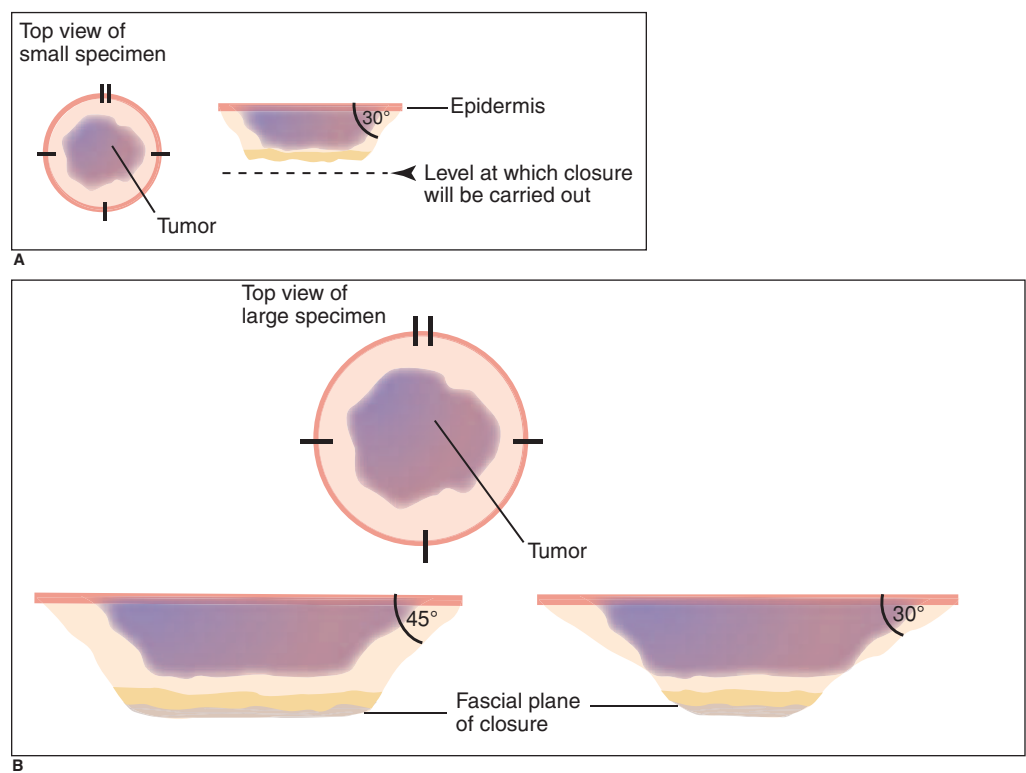


FIGURE 2.2: (A) Because the specimen is small, a beveling angle of 30 degrees instead of 45 degrees may be necessary to allow the technician to flatten the edges and base of the specimen into a single plane for processing, and the excision may be carried down only to a level above the level at which the defect would be undermined for closure. (B) The excision of a larger-diameter cancer may allow a steeper bevel that can be extended all the way down to the plane of eventual closure. If the cancer is obviously deeper than this plane, the excision may be carried even deeper. “Standard” surgical technique of cancer uses vertically cut specimens. Mohs surgery uses cuts made at a bevel; this bevel allows the technician to flatten the edges of the specimen into the same plane as the base so that the entire peripheral and deep margin can be completely assessed pathologically. The usual bevel is 45 degrees from the standard vertical cut and yields a specimen whose sides are cut at 45 degrees from the horizontal surface of the specimen (left). A larger bevel would produce a specimen whose sides are cut at a smaller number of degrees from the flat surface of the specimen (right). The 30-degree bevel makes it easier for the Mohs technician to flatten the specimen into a single plane for processing but also increases the chances that the bevel cut will transect cancer.

and may require more of a bevel; either a wider surgical margin is required or they may not be able to be excised as deeply (Figure 2.2). This is a small disadvantage of Mohs, as opposed to standard excision technique.

When possible, the first stage in Mohs surgery should be cut to the depth of eventual closure (Figure 2.1A). This is limited by the surface dimensions of the excised specimen. A small-diameter specimen cut at a bevel of 45 degrees may not be able to be adequately flattened into a single plane by the technician and would likely reach a depth less than the probable final plane of wound closure before the base of the specimen is completely cut. To initially cut a large specimen above the depth that will be utilized for closure increases the chance of leaving cancer at the deep margin, increases surgical time and cost, and does not spare tissue, as undermining and closure in the proper plane will likely require removal of this deep tissue that was “spared” during

the Mohs procedure. Thus, for example: scalp excisions should be carried to subgalea, and extremity excisions to muscle fascia.

In some situations, a sufficient bevel to allow optimal processing of the specimen may not be achievable. This is frequently true of deep but narrow alar crease tumors. If specimens require deep tissue removal, but the specimen is too narrow to allow an adequate bevel, the specimen can be taken with little or no bevel and instead prepped by the technician using the “open book” technique (Figures 2.3 and 2.4A–C).

This technique may also be used when re-excising a surgical scar after permanent section pathology has shown a positive margin. The entire scar needs to be removed to a deeper plane than the previous surgery and the peripheral margin needs to be cut as widely as the area was previously undermined. The open book technique

8 MOHS SURGERY AND HISTOPATHOLOGY

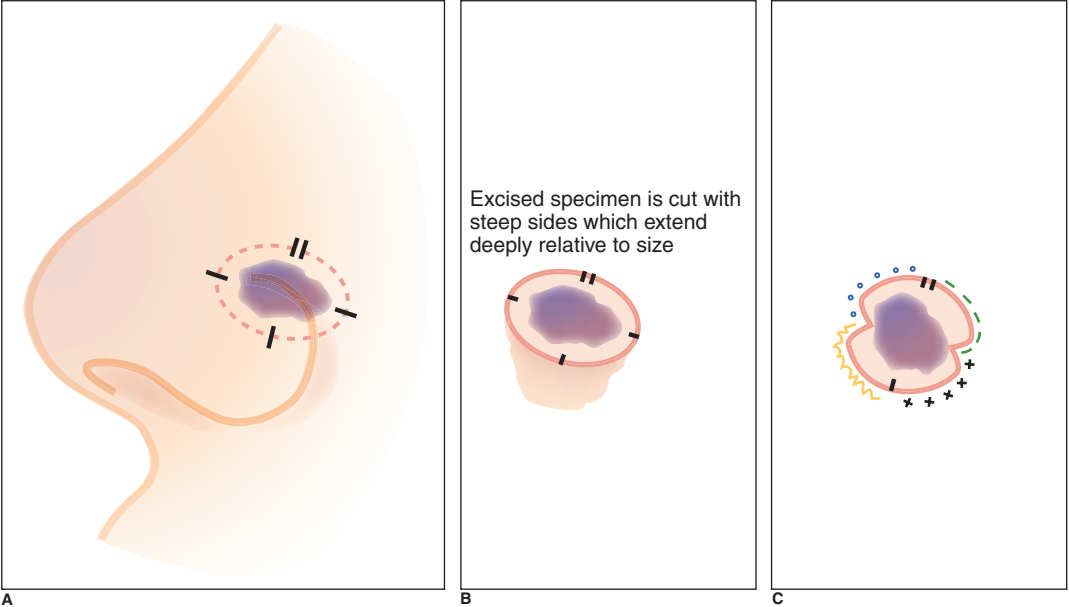
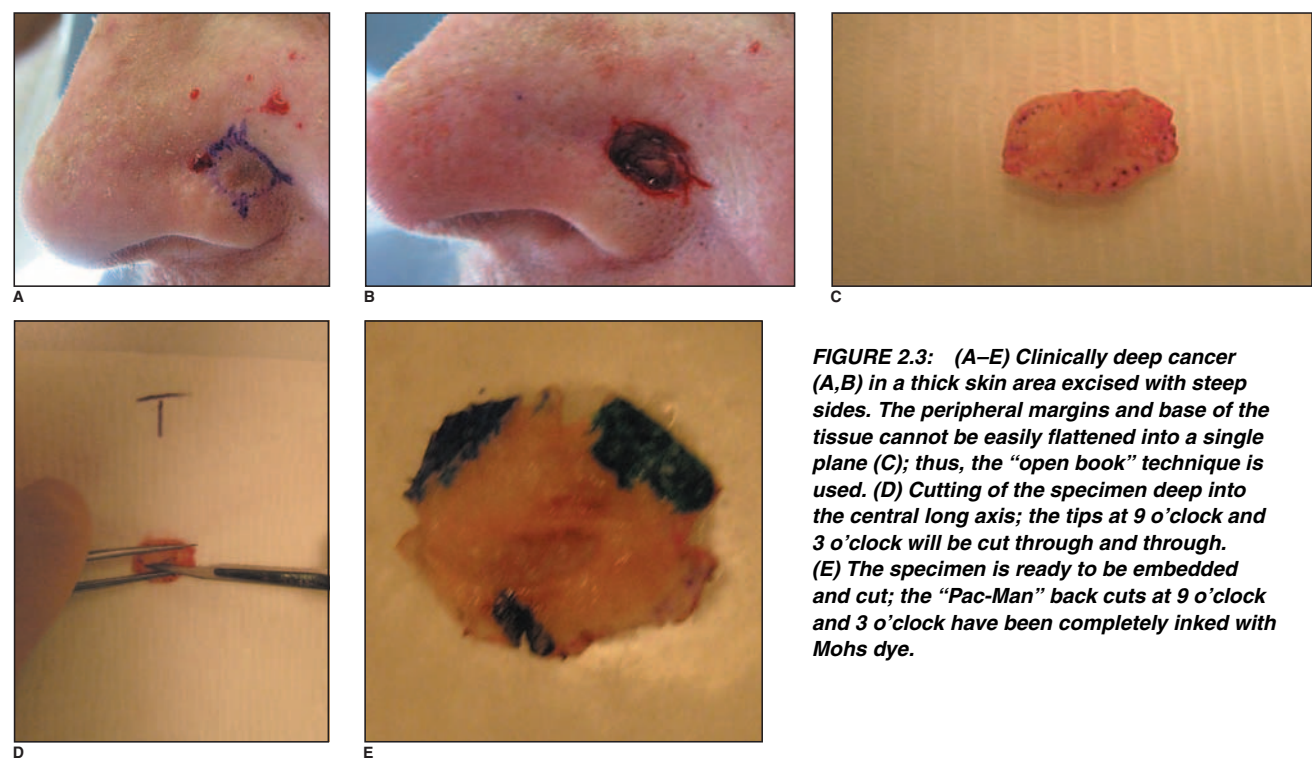


FIGURE 2.4: Diagrammatic illustration of the same type of excision as in Figure 2.3. The inking diagram shows complete inking of the ends of the specimen, opened to allow flattening using the open book technique. (A) Diagrammatic illustration for a cancer similar in type and location as in Figure 2.3. Excision for this cancer cannot be easily done with standard beveling technique because, although clinically small, the cancer is deep and located in a thick skin area. (B) The excised tissue has very steep edges and cannot be flattened into a single plane using standard tissue relaxing incisions. Standard relaxing incisions would not be enough to get the edges and base to lie in a single plane for sectioning. (C) “Pac-Man” cuts along the long axis of the specimen (at 3 o’clock to 9 o’clock in this example) allow the specimen to “open like a book” and all the edges to be flattened into a single plane for processing. The tips are cut all the way through to further allow the specimen to lay flat. Tumor extending to the edges of the Pac-Man cuts is not at a margin because this is an artificial edge produced by the technician. The true margins include the entire base and the peripheral epithelial edges. The entire cut tips must be chromacoded so that the Mohs surgeon-pathologist has a way to ensure that the tissue at these cut tips is completely represented.

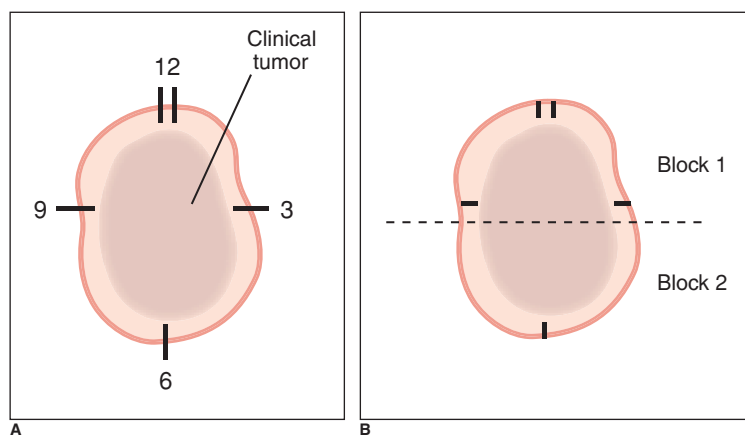


FIGURE 2.5: (A) A large specimen is cut. The Mohs surgeon should have but did not place the 3 o'clock and 9 o'clock reference nicks at the midpoint of the long axis of the specimen. (B) The technician has subdivided the specimen into two equally sized blocks but not at the 3 o'clock and 9 o'clock marks cut by the surgeon. This makes it harder for the Mohs surgeon-pathologist interpreting the findings on the slides to transfer them to the proper location within the excision defect on the patient because the 3 o'clock and 9 o'clock reference nicks on the patient do not correspond to where the technician subdivided the specimen into two blocks.

works well for preparation of first-stage excisions of these specimens.

The open book technique requires special care on the part of the technician to ensure that in making cuts through the specimen, tumor is not inadvertently carried by the blade into the margins. Both ends of the specimen must also be completely inked so that the Mohs surgeon-pathologist is certain that complete surgical margins are represented on the slides.

The surgeon should choose distances between reference nicks (hatch marks) that attempt to correspond to the size of the blocks the technician is able to process in a microtome (Figure 2.5); this allows easier translation of the exact location of the tumor at a margin from the microscope to the patient. The technician should attempt to subdivide the surgical specimen at the hatch marks placed in the tissue by the surgeon, even if this results in not dividing the specimen into blocks of equal dimensions (Figures 2.5 and 2.6).

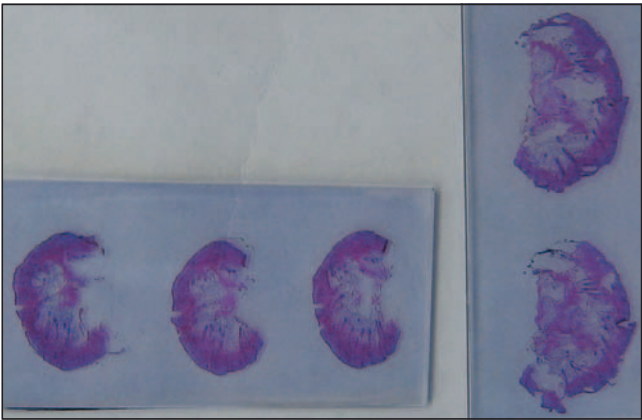


FIGURE 2.6: This photo illustrates how the technician actually processed the two blocks. The blocks are not of equal size; but were subdivided by the technician at the reference nicks placed on the specimen and on the patient by the surgeon. This will make it easier for the Mohs surgeon-pathologist to translate findings from the slides to the patient's wound.

When dividing the specimen into multiple blocks, the technician must ensure that tumor is not artifactually carried to a margin where it could be misread as a positive margin. This probability may be reduced by debulking obvious tumor from the top of the specimen, by cutting from the specimen edges toward the center, and by wiping the blade after each cut (see Chapters 6, 7, and 8). Likewise, the surgeon should wipe the scalpel blade frequently when making excisions to preclude carrying cancer from a clinically occult area of the tumor to a tumor-free area. In Mohs surgery, the surgeon only knows in retrospect that the area being cut is cancer-free.

The Mohs surgeon must determine where to designate “12 o'clock” on the excised specimen. How each surgeon does this is not as important as consistency in a chosen method, so that even if a reference nick is not seen at the edges of the wound when the patient is brought back for additional cancer excision stages, the Mohs surgeon still has at least a general idea of where the 12 o'clock reference nick might have been. This may allow excision of that Mohs stage with only slightly greater than usual overlap (Figure 2.7). In some practices, 12 o'clock may always point superiorly, medially, and/or posteriorly. Other surgeons may always orient 12 o'clock toward the tip of the ear lobule on the side of the body where the cancer is located. Many other methods are equally valid. A digital photo (Figure 2.8) taken of the ink outline and reference nicks of the area to be excised can be viewed later on the camera, or printed, if confusion exists in the surgeon's mind.

As well as choosing where 12 o'clock will be on the tissue specimen, the Mohs surgeon must also ensure that clear orientation of the specimen is maintained from the operative table to the technician's inking station. This can be done in a number of ways:

1. Prior to making the final cut of the base of the specimen, the surgeon-pathologist should visually double check that the reference nicks can be clearly seen on the specimen and corresponding wound edges.

10 MOHS SURGERY AND HISTOPATHOLOGY

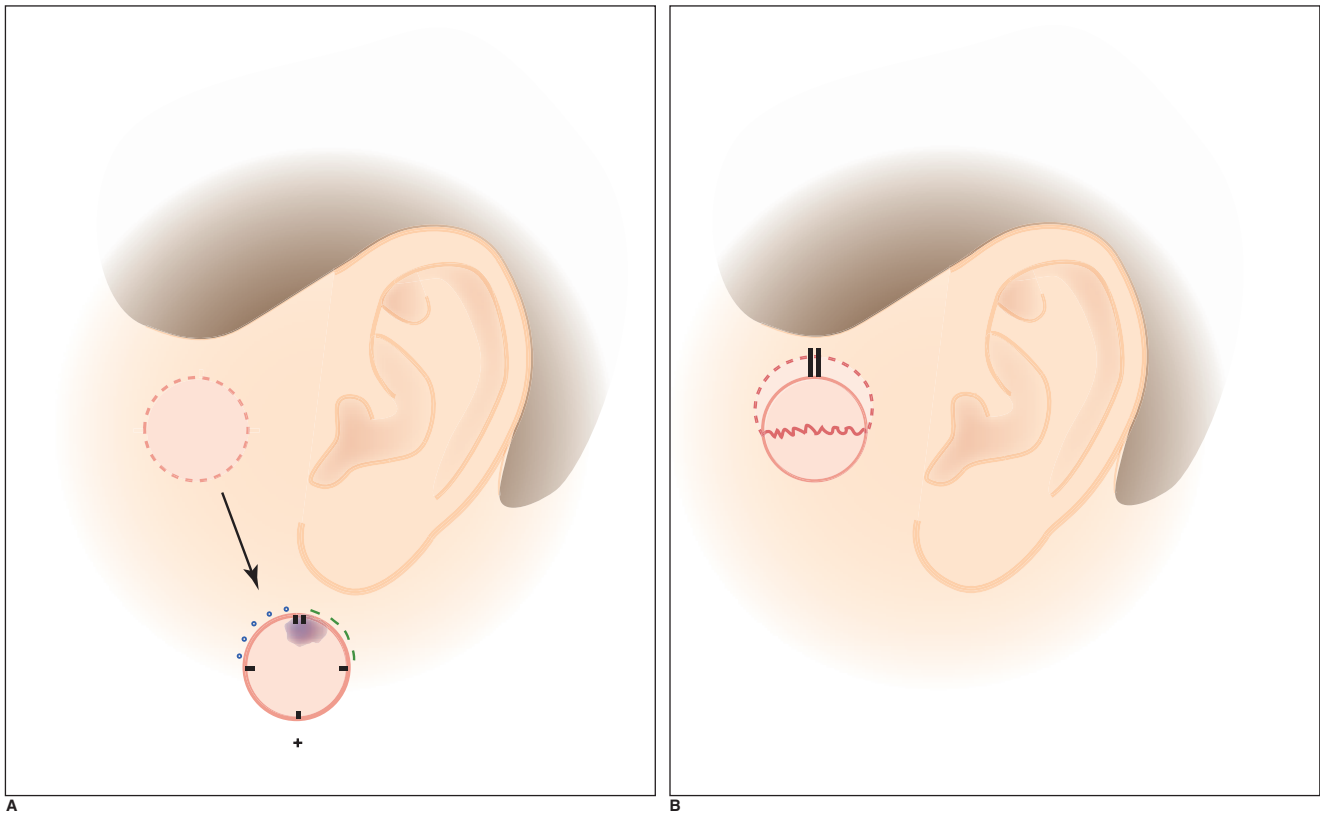


FIGURE 2.7: (A) Mohs specimen cut with a 12 o'clock superior orientation. Cancer is noted at the 12 o'clock margin on the Mohs slide from this excision, but, unknown to the surgeon, the reference nicks did not "show" on the patient's skin after the specimen was removed. (B) No reference marks were visible when the surgeon-pathologist returned to the operative table and viewed the patient's wound. This surgeon always uses

12 o'clock as superior orientation and has a preop digital photo of the area with the tumor outlined and reference nicks demarcated with ink; therefore, in this example, the correct margins are easily ascertained by the surgeon-pathologist and a correct stage II excision easily determined. It is not always this easy.

2. An arrow from the sterilization indicator strip can be cut and the specimen placed on the arrow so that 12 o'clock lies in the same direction as the tip of the arrow (Figure 2.9).



FIGURE 2.8: Photo of a large cancer with the reference nicks drawn; the subdivision of the tissue into smaller blocks for processing will be done at these hatch marks.

3. A blood and/or ink dot on a corner of the transfer gauze can be used to designate the 12 o'clock margin.

4. For large and/or complex specimens, digital photographs should be taken and printed of the specimen before lifting it from the wound, and again with the specimen removed but placed next to the defect (Figure 2.10); this will show the new shape of both the specimen and defect, both of which change shape after the specimen is removed. Because the Mohs map is intended to depict the patient's wound after the removal of the tissue specimen, some Mohs surgeons don't draw their maps until after the specimen is excised.

When excising cartilage, including some noncartilaginous tissue at one or more of the specimen edges will help the technician prevent the cartilage from "floating" off the slides during processing. Even if this results in a slightly larger defect, it can be so helpful in producing better-quality slides that the net result is usually worthwhile. (See also Chapter 6 for techniques for slide preparation of tissue containing cartilage.)

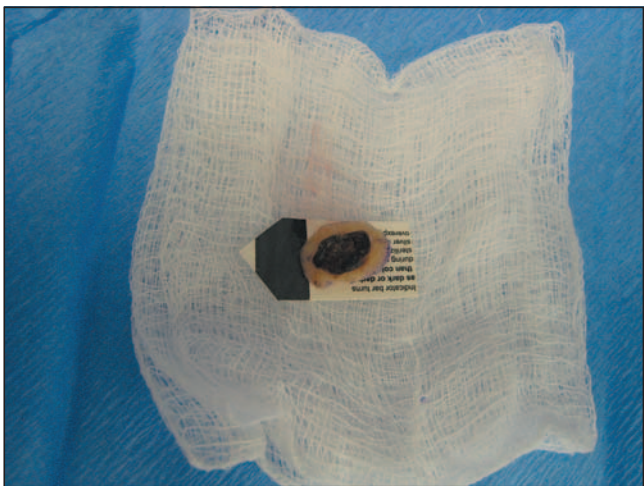


FIGURE 2.9: A piece of sterilization indicator strip cut as an arrow with the specimen is laid upon the “arrow” so that the arrow point and the specimen’s 12 o’clock reference nick are oriented in the same direction.

When excising large specimens from the vermillion or helix, it is sometimes best not to bevel. Cutting straight through the tissue without beveling allows these cut ends to be processed more easily (Figure 2.11).

When excising a positive margin, the surgeon should significantly overlap beyond the diagrammed extent of the positive margin unless this will cause significant functional postoperative problems.

It is critical to understand that sometimes the location of a deep positive tumor margin noted within a Mohs wafer on a slide, and then depicted on the two-dimensional (2D) Mohs map may be incorrectly located within the three-dimensional (3D) wound when the Mohs surgeon-pathologist returns to the surgical table. A deep positive margin in a 2D flattened specimen may actually lie on or partially on the wall of the 3D wound, not only at its base. This will require the removal of both additional peripheral



FIGURE 2.10: A large specimen is photographed next to the wound from which it was cut. This allows the Mohs surgeon-pathologist to easily see the relationship between the specimen and the wound, both of which change shape after excision.

and deep margins within the patient’s wound (see Chapter 9).

Tumor in a nerve at the deep/central margin of a specimen requires excision of additional peripheral margin as well as deep margin because nerves may run in any direction and at any angle from vertical to horizontal. Furthermore, additional nerve must be seen on slides from the next Mohs stage (and be assessed as free of cancer) for the margins to be interpreted as clear. If no nerve tissue is present on the slides, that stage of surgery cannot be considered clear even

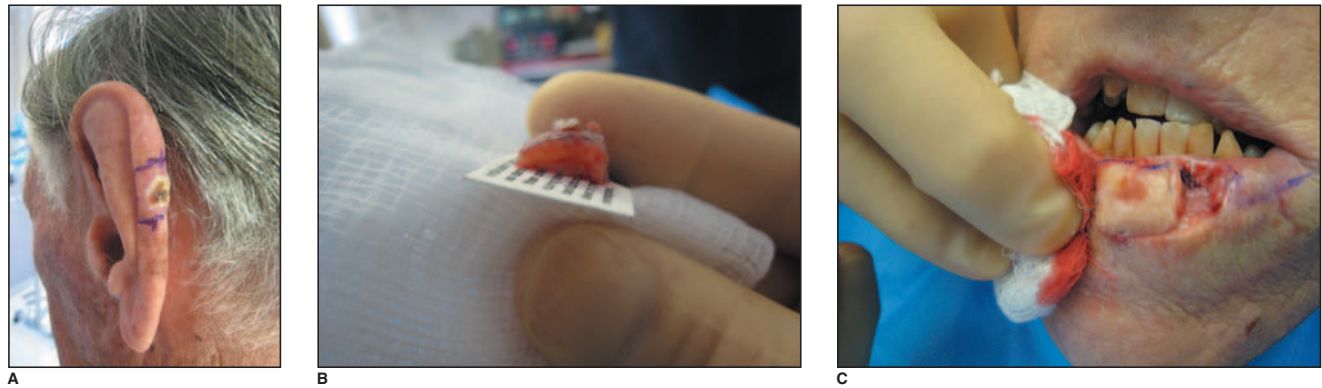


FIGURE 2.11: This specimen from the helix of the ear has been cut at both ends without a bevel. The technician will have to cut both ends across their short axes in a partial “bread loaf”

technique to allow the ends to lie in the same plane as the base. The specimen from the lip has been excised similarly.