# Innovative Laboratory Techniques to Facilitate Processing of Large Mohs Cases

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BACKGROUND. **Processing multiple tissue sections in large Mohs cases is time consuming and labor intensive.**
OBJECTIVE. **To present innovative laboratory techniques to facilitate processing of large Mohs cases.**
METHODS. **A method for processing a large dermatofibrosarcoma protuberans Mohs case is outlined.**
RESULTS. **Modifications in tissue processing and equipment employed in a large Mohs case are presented.**
CONCLUSION. **Innovative modifications to the standard Mohs laboratory technique can facilitate processing of large Mohs cases, resulting in high-quality, rapid frozen sections while optimizing efficiency.**

*SEAN A. SUKAL, MD, PHD, MARIE TUDISCO, HT (ASCP), BARBARA STRIPPOLI, HT (ASCP), AND KISHWER S. NEHAL, MD, HAVE INDICATED NO SIGNIFICANT INTEREST WITH COMMERCIAL SUPPORTERS.*

MOHS SURGERY for large tumors, such as dermatofibrosarcoma protuberans (DFSP), requires substantial planning of time and staffing for laboratory processing and microscopic evaluation of multiple tissue sections. A rapid turnaround time with high-quality frozen sections is critical when treating large tumors with the Mohs technique. Multiple methods have been described to optimize efficiency in Mohs surgery. One method of improving efficiency entails the histologic preparation of multiple tissue specimens on a single glass slide.[1] This technique is generally limited to small specimens and can introduce errors caused by inadvertent tissue mislabeling.

The single-section method was described in 1993 for the histologic processing of Mohs specimens.[2] This method saves time because the tissue specimen is not subdivided into smaller sections. The method was initially limited to small specimens owing to the size restrictions imposed by the cryostat chuck and slide. Although processing of large cases using the large single-section method has been reported,[3] several factors still exist that pose limitations to its practical application to large specimens.

Our purpose is to present a comprehensive approach for processing large Mohs sections with innovative but simple modifications in tissue processing and use of commercially available equipment to optimize efficiency while maintaining the accuracy of Mohs margin evaluation.

## Mohs Laboratory Technique

A DFSP located on the back is presented. The 2.3 × 2.0 cm clinical lesion was excised with 1 cm beveled circumferential margins to the deep subcutaneous plane as a single saucerized specimen. Eight scores were placed along the tissue margins in clockwise fashion to preserve orientation and were drawn on a reference Mohs map.

In the Mohs laboratory, the specimen was left dry to prevent tissue swelling. The specimen edges were teased down with deep relaxing nicks through the scores, allowing the tissue to lie flat. Davidson's marking dyes (Bradley Products Inc, Bloomington, MN, USA) were used to uniquely identify the eight scores and deep margins corresponding to the Mohs map. The specimen was then dissected horizontally through the 9 and 3 o' clock scores with a #10 scalpel, and the pieces were labeled 1 and 2.

Each of the two large pieces was frozen, undersurface down, onto the cryostat (Leica 1510) knife holder plate with optimal cutting temperature (OCT) embedding compound. This provides a hard matrix for fat and prevents chipping or specimen or chuck disengagement during trimming and cutting. After freezing, the specimen was pried off and inverted onto a 55 mm room temperature specimen disk (chuck) coated with OCT compound. The cryostat was set at −25°C. Liquid nitrogen spray was used to

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facilitate freezing. The block was secured to the holder and cut at 6 μ on a high-profile Extremus blade to reduce resistance. The Leica 1510 allowed a large cutting surface clearance angle from the block to the blade with good block holder stability. This large clearance angle is crucial for optimally cutting large specimens.

The sections were retrieved onto large 50 × 75 mm glass slides. As these commercially available slides lack a labeled end for pen marking, a diamond-tipped pen was used to permanently etch patient data onto the slide. For staining, the slides were placed into a 25-capacity acrylic rack with a side handle. This basket was able to accommodate the height of the larger slides. It was modified to fit into the Tissue Tek II Staining Dish by cutting it in half down the center. The slides were stained with routine rapid hematoxylin-eosin stain for frozen sections and were permanently mounted with large 45 × 60 mm coverslips (Figure 1). To accommodate the larger slides, the slide folders were modified by alternately removing the cardboard dividers. The standard double-row slide file drawer was modified by removing the slide divider to accommodate the larger glass slides.

The slides were initially scanned by the Mohs surgeon with a 1.25× objective lens and were then examined in detail under higher power as necessary. The 1.25× objective provides a wider field of view than the standard 2× or 4× objective and allows more rapid specimen scanning with greater ease.

## Discussion

The processing of multiple tissue sections of a large Mohs case poses a number of technical difficulties. Handling a large number of Mohs tissue sections can increase the probability of orientation and labeling errors. The presence of many tissue sections can also result in tissue sitting unprocessed for a longer period of time, thereby increasing the risk of tissue autolysis and swelling. False-positive margins can occur with the standard Mohs technique when sectioning inadvertently inoculates peripheral malignant tumor into the deep plane.

In our experience, the rapid processing of a large DFSP case by the method presented avoids all of these potential pitfalls. Tissue orientation and labeling are simplified by keeping excised tissue as large sections. In addition, there is minimal delay between excision and freezing of tissue, reducing the risk of tissue swelling and autolysis. Finally, fewer sectioning steps are needed, minimizing the risk of false-positive margins. The maximum size of each large section is determined by the largest specimen disk diameter of 55 mm and the glass slide measuring 50 × 75 mm.

With the method described, we have also been able to achieve efficiency with a rapid turnaround time in tissue processing of large DFSP cases. The enhanced efficiency allows scheduling of a routine Mohs case load while avoiding staff fatigue. There is also benefit to the patient because there is a shorter waiting time between Mohs

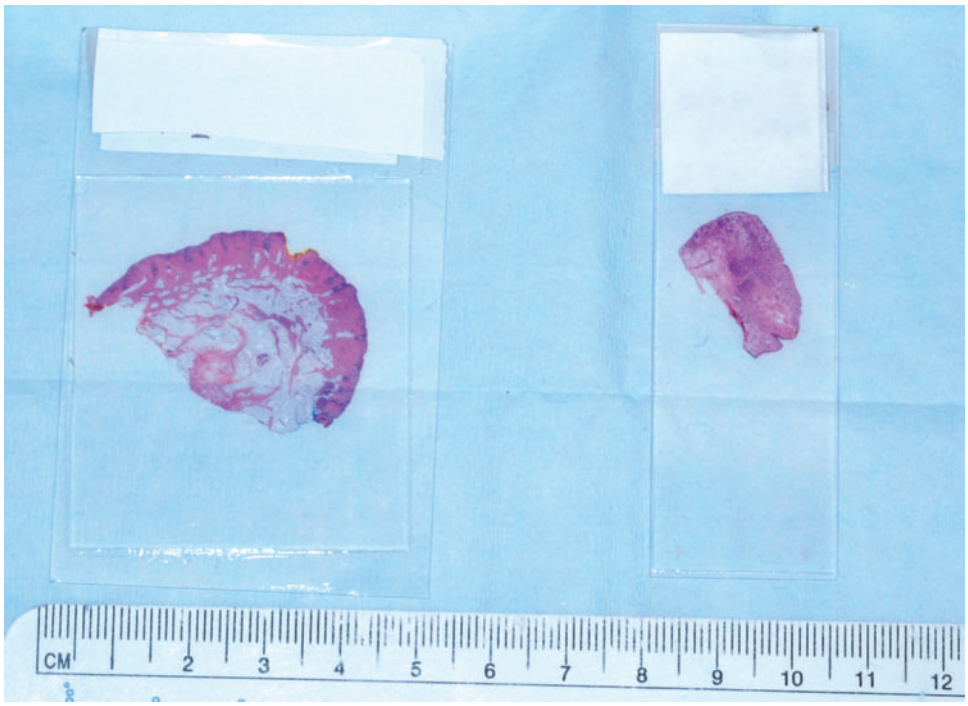


**Figure 1.** Excised dermatofibrosarcoma protuberans tissue divided into large sections, processed into Mohs frozen sections, stained with hematoxylin and eosin, and mounted onto a large glass slide/coverslip compared with a small Mohs tissue mounted onto a standard-size glass slide.

stages. Finally, the advantage for the Mohs surgeon is a reduction in microscopic viewing and interpretation time owing to the presence of fewer sections.

There are several disadvantages to the modified Mohs technique described. Skilled histotechnicians are needed for the processing of large tissue sections, but with adequate training, the skill can be acquired. Some of the steps require specialized laboratory equipment and supplies, which must be acquired and stocked for the day of surgery. In addition, certain cryostat models do not have the necessary block to blade clearance for a large cutting surface or may exhibit poor chuck holder stability. Finally, the use of a 1.25× objective lens for histologic interpretation significantly enhances the ability to scan large tissue sections with ease owing to a wider field of view. The wider field of view, however, requires the Mohs surgeon to more carefully scan the tissue to prevent missing a tumor focus within a large tissue section. With repeated use of the 1.25× objective, the Mohs surgeon can easily become accustomed to viewing tissue at this magnification.

In conclusion, we described a modified single-section method for the processing of a large Mohs case. Innovative but simple modifications were presented that make the processing of large cases efficient, facile, and cost-effective. This report demonstrates methods for avoiding previously reported limiting factors associated with the single-section method.[2,3] The rapid tissue processing time not only preserves tissue integrity but also has the added benefits of ensuring a smooth work flow and reducing staff fatigue while maintaining a routine Mohs case load.

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