

Efficient Tumor Immunohistochemistry

A Differential Diagnosis-Driven Approach



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printed in Singapore

10 09 08 07 06

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A Differential Diagnosis-Driven Approach

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Preface

Close to a quarter of a century ago, we wrote a monograph titled *Immunoperoxidase Techniques: A Practical Approach to Tumor Diagnosis*, and stated at that time that the practice of pathology has been changed by immunohistochemistry. The writing of that monograph was prompted by the assertion that the lack of a practical manual on the subject prevented pathologists from gaining full benefits of the technique in the daily assessment of surgical tissue samples. Today, immunohistochemistry is an integral part of the daily practice of pathology and there is hardly a day that goes by without the discovery of a new tumor marker. Nor is there a single issue of pathology or other specialty journals that fails to carry an article or a direct reference to immunohistochemistry. In fact, during the last 25 years, more than 165,000 articles and many textbooks have been published on that subject. Consequently, pathologists are challenged to convert the benefits from the florid array of new developments and informations in the field into the daily practice of surgical pathology. As was the case with the original monograph, our intent is to facilitate that task. Therefore, this can be considered as an update on the observations that we have made in the course of the past 25 years by adhering to practicality as the single most important criterion in the resolution of daily diagnostic problems encountered by surgical pathologists. Whenever appropriate, this approach can be complemented by referring to more exhaustive treatises such as Taylor, Clive R and Cote, Richard J, *Immunomicroscopy—A Diagnostic Tool for the Surgical Pathologist*; and Dabbs, David J, *Diagnostic Immunohistochemistry*. Updates on tumor markers may be obtained by visiting the ImmunoQuery Web site, Frisman, Dennis M., www.ipox.com.

Acknowledgments

This manual reflects the application of immunohistochemistry in the resolution of differential diagnostic problems encountered daily in the practice of surgical pathology at the University of Miami Jackson Memorial Hospital and Sylvester Comprehensive Cancer Center. Our practice encompasses cases originated from these institutions as well as those that are referred from colleagues in the United States, primarily from Florida, and also from outside the United States. We therefore acknowledge the benefits derived from more than two decades of interaction with the faculty in our department and our peers in Florida and elsewhere.

We have been most fortunate in maintaining a dedicated, responsible and most competent technical staff in our immunohistochemistry laboratory. Silvia Losa, Blanca Cuenca, Weiyu Wu, and Fay Mucha have provided their skills and commitment to the well being of our patients and to the success of our research endeavors throughout these years. We would like to extend a very special note of gratitude to Alicia Cabrera who confronted the demands of writing and many times revising the manuscript with forbearance, professionalism, and gracious attitude.

The extended efforts of Drs. Parvin Ganjei, Betina Werner and Rima Kanhoush in the preparation of this manual were essential for bringing it about. To Joshua Weikersheimer of the ASCP Press, we owe special thanks for his advice and continuous support. We also like to thank Doug Sweet, Vice President, Celerus Diagnostics, who was instrumental in encouraging us to undertake this project.

Introduction

Immunohistochemistry is now an integral part of every diagnostic pathology laboratory. Its major application is to complement histomorphology in classification of human neoplasms. With the refinement of methodologies, the availability of good quality reagents, and enhancement of the reproducibility of results by introduction of automatic stainers, problems associated with the technical aspects of immunohistochemistry are relatively uncommon. Current limitations of efficient application of immunohistochemistry in diagnostic pathology are mostly analytical, usually related to selection of appropriate markers for a diagnostic problem, as well as the correct interpretation of staining result and its incorporation in the final diagnostic decision. Factors that influence the analytical outcome include scarcity of cell lineage-specific markers, subjectivity of panel selection, lack of practical guidelines, and the experience of the observer.

It is not surprising, therefore, to witness the drastic increase in the number of antibodies used in immunohistochemical panels. However, the indiscriminate use of multiple antibodies without consideration of histomorphology and clinical history—the “shotgun” approach—is neither diagnostically efficient, nor cost-effective. Often, it entangles the pathologists in a confusing web of “positive” and “negative” staining results, leading to indeterminate or incorrect diagnostic conclusions. In practice, however, based on histologic appearance of the tumors, most differential diagnoses in tissue biopsies can be narrowed down to two or three possibilities. Consequently, the choice of antibodies in these situations can also be restricted to two or three.

In this practical handbook we recommend simple guidelines for the resolution of most common—and some uncommon—diagnostic problems by utilization of a limited number of antibodies. This is not an algorithm; it is a “tailor-made” approach that requires the pathologist’s input and necessitates his/her active participation by formulating a working differential diagnosis on the basis of clinical information and histomorphology.

Before using the book:

1. The suggested markers are those that we have found most useful. The experienced immunohistochemist may like to use the same set of markers or modify the selection according to his/her preference. The user will find that some of the most commonly utilized immunohistochemical markers are conspicuously absent. We have omitted those we have found to be of limited or no value in a given differential diagnosis, even when they are used in a panel.
2. Not all possible differential diagnostic problems are discussed. This is primarily due to the lack of reliable markers for some common differential diagnoses. For example, there are at least a dozen publications on immunohistochemical separation of renal oncocytomas from chromophobe renal cell carcinomas. None of the several different purported markers in these papers, however, has proved to be useful, at least in our experience.
3. The illustrated images are from cases seen in our own institution or referrals from colleagues in Florida and elsewhere. We have made no attempt to present the most typical examples of each diagnostic entity or the best possible histologic preparations. On the contrary, the majority of illustrations are from cases that may show crushing artifacts, knife nicks, thermal damage, or other changes that most pathologists are familiar with in their daily practice.
4. Some images exhibit positive staining as dark brown, or black, on a green background. This is notable for nuclear antigens because we intensify the color of DAB with a dip in copper sulphate and use fast green as a cytoplasmic counterstain.
5. Finally, we have only discussed and illustrated the immunohistochemistry of human solid tumors. The IHC of hematolymphoid neoplasms is not addressed except when they come into the differential diagnosis of a non-lymphoid neoplasm. The IHC of hematolymphoid neoplasms is a rather vast area and is beyond the scope of this publication..