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ANATOMIC PATHOLOGY

Lymph Node Evaluation as a Colon Cancer Quality Measure: A National Hospital Report Card

Karl Y. Bilimoria; David J. Bentrem; Andrew K et al

Examination of 12 or more regional lymph nodes for colon cancer is associated with improved staging and survival, and the National Quality Forum recently endorsed lymph node examination for colon cancer as a quality surveillance measure. However, information regarding the extent of hospital compliance with the 12-node measure in the United States is lacking.

Methods: From the National Cancer Data Base, 1296 hospitals that performed 156 789 colectomies in 1996-1997 and 2004-2005 were identified, and rates of hospital-level compliance (defined as examination of 12 nodes in 75% of patients) in these two time periods were compared. Multivariable models were developed to determine if hospital type, volume, or differences in case mix were associated with 12-node measure compliance. All statistical tests were two-sided.

Results: In 1996-1997, 15% of hospitals were compliant with the 12-node measure; in 2004-2005 the percentage of compliant hospitals had increased to 38%. From 1996-1997 to 2004-2005, 12-node measure compliance increased at 980 hospitals, remained unchanged at 6 hospitals, and decreased at 310 hospitals. In 2004-2005, National Cancer Institute-designated Comprehensive Cancer Centers were more frequently compliant with the 12-node measure than other academic hospitals, Veterans' Administration hospitals, or community hospitals (78.1% versus 52.4%, 53.1%, and 33.7%, respectively, all $P < .001$), even after adjustment for differences in characteristics of the colon cancer patients at these hospitals.

Conclusions: This study provides a national report card of nearly 1300 hospitals showing that more than 60% of institutions failed to achieve a compliance benchmark for the 12-node measure. Considerable improvement is needed in colon cancer nodal evaluation in the United States.

J Natl Cancer Inst. 2008;100(18):1310-1317.

Methylene Blue-Assisted Lymph Node Dissection in Colon Specimens: A Prospective, Randomized Study

Märkl B, Kerwel TG, Jähnig HG, et al

Recently, we introduced ex vivo intra-arterial methylene blue injection into the inferior mesenteric artery as a novel method to improve lymph node (LN) harvest in rectal cancer. We have now adapted this method to the other segments of the colon. A total of 60 cases were enrolled. Primary LN dissection was followed by fat clearance and a secondary dissection. The mean \pm SD primary LN harvest differed highly significantly with 35 \pm 18 and 17 \pm 10 LNs in the methylene blue-stained and unstained groups,

respectively. Primary insufficient LN harvest occurred in 8 cases of the unstained group and in only 1 case of the methylene blue-stained group ($P = .0226$). After secondary dissection, upstaging was seen exclusively in the unstained group. The time/LN ratio differed significantly with 0.9 and 0.6 min/LN in the unstained and methylene blue-stained groups, respectively. Intraarterial methylene blue injection is recommended as a routine technique in the histopathologic study of colon cancer.

Am J Clin Pathol. 2008 Dec 1; 130(6):913-919.

New Scoring System Rules out Advanced Liver Fibrosis

NEW YORK (Reuters Health) Oct 17 - A simple scoring system accurately identifies patients with nonalcoholic fatty liver disease (NAFLD) who don't have advanced disease, according to a report in the October issue of Gut.

The "BARD" score was developed to incorporate 1 point for a BMI of at least 28, 2 points for AST/ALT ratio of 0.80 or greater, and 1 point for diabetes.

A BARD score of 2-4 has a positive predictive value of 43% and a negative predictive value of 96% for detecting stage 3-4 fibrosis, the researchers note.

"If the BARD score is < 2 , physicians can be reassured the patient does not have advanced liver disease, and if the patient has a BARD score of 2 or greater, they really need to be concerned and take action," Dr. Stephen A. Harrison from Brooke Army Medical Center, San Antonio, Texas, told Reuters Health.

Dr. Harrison and colleagues defined the clinical, laboratory, and histopathological characteristics of 827 patients with biopsy-proven NAFLD and developed a simple clinical scoring system for identifying patients with advanced fibrosis.

Compared with patients having fatty liver alone, patients with nonalcoholic steatohepatitis (NASH) were older, heavier, predominantly female, hypertensive, diabetic, insulin resistant and had a higher ALT and AST, the authors report.

Patients with stage 3-4 fibrosis were also likelier than patients without fibrosis to be older, female, diabetic, insulin resistant, and to have higher AST/ALT ratios.

Based on additional analyses, the investigators found that the BARD score was at least equivalent to the more complex NAFLD fibrosis score in excluding patients with advanced fibrosis.

A simple scoring system that relies on only three clinical features was validated in a large group of patients. BARD "may assist clinicians in targeting those patients in need of liver biopsies to confirm the diagnosis and guide management, particularly among patients with diabetes," the authors conclude.

Gut 2008;57:1441-1447.

How do surgical pathologists evaluate critical diagnoses (critical values)?

Masoud Mireskandari

After introduction of the concept of critical value (CV) in laboratory medicine, some efforts were performed to define possible critical values in surgical pathology. Critical diagnosis (critical value) is a concept recently established in surgical pathology and is a challenging issue among pathologists and clinical specialists. The concept may be the subject of variation according to the geographical or work setting differences. The current study was performed to bring the contribution of the Iranian pathologists to the evolving concept of critical diagnoses (critical values) in surgical pathology.

Materials and methods: During annual meeting of Iranian Pathologist Society, November 2006, Tehran, Iran, anonymous questionnaires were distributed among participants. They were requested to openly name conditions in which a pathologist should communicate the results immediately with clinicians.

Results: 147 pathologists completed the questionnaire. They were varied in their level of experience and setting of workplace. Each participant referred to 1–7 (mean 3) conditions as CV. About 90 different conditions which were considered as CV by participants were extracted from the questionnaires.

Discussion: The list of conditions obtained through this survey as CVs in surgical pathology covered most items previously described in literature. Major differences are low number (or lack) of refers to some relatively routine and potentially important conditions and considering many unimportant conditions as CV by participants of present survey. Almost all conducted surveys have been performed on this issue so far (including the present survey) suffer from lack of supportive scientific evidences and based mainly on experience and common sense of participants in survey. Potential problems with application of CV concept in daily routine work flow of pathology, particularly in developing countries like Iran, were discussed.

In each questionnaire from 1 to 7 (mean 3) conditions were notified by participants as CVs. About 90 different conditions were named by participants as critical values in surgical pathology (table 1). The number of times each condition was referred was ranged from 1 to 52. The most frequently referred situation was acute leukemia in bone marrow sampling (52) followed in decreasing order by reports of intraoperative consultation (26), unexpected malignancy in any specimens (23), and mucor mycosis (12). High grade lymphomas and insufficient or absent specimen were referred 9 times. Breast malignancies (with no further qualification) were referred as a critical value 7 times. Malignancy in endoscopic biopsy specimens, malignancy in fine needle aspiration of tissues, and malignant melanoma were referred 6 times. Among the conditions which were referred more than one time but less than 6 times, noticeable conditions include malignant cells in cytology of fluids (5), unexpected benignancy (5), any difference between interpretation of present evaluation and previous pathologic diagnosis (4),

necrotizing vasculitis (4), renal needle biopsy in transplanted kidney (3), mediastinal lymphoblastic lymphoma (3), septic arthritis (3), any mediastinal mass in patients with superior vena cava syndrome (3), Burkitt's lymphoma (2), diagnostic curettage in suspected ectopic pregnancy cases (2), margins involvement in resection specimens of malignant tumors (2), rapidly progressive glomerulonephritis (2), and infectious agents in immunocompromised patients (2). About 45 conditions were referred only once by participants. Some noticeable conditions among them are: diagnostic curettage in patients suspected to malignancy, discordance between clinical and pathologic diagnosis, false sampling in confirming biopsies (tubal ligation, vagotomy, vasectomy, etc), temporal arteritis, and transplant rejection.

Table 1

Most common conditions considered as CV by participants of present survey.

Conditions	Number of Refers
Acute leukemia in bone marrow specimens	52
FS reports	26
Unexpected Malignancy	23
Mucor mycosis	12
All malignancies	9
High grade lymphomas	9
Unsuitable or absent specimen	9
Breast malignancy	7
Malignancy in endoscopic specimens	6
Malignancy in FNA	6
Malignant melanoma	6
Brain Tumors	5
CIN II and III	5
Malignant cells in cytology of fluids	5
Tuberculosis	5
Unexpected Benignancy	5
Bone marrow biopsy for thrombocytopenia	4
Difference with previous pathologic diagnosis	4
Necrotizing vasculitis	4
Renal needle biopsy in transplanted kidney	4
CSF cytology with malignant or leukemic cells	3
Cutaneous vesiculo-bullous diseases	3
Fluid cytology	3

Mediastinal lymphoblastic lymphoma	3
Metastatic tumors	3
Septic arthritis	3
Wilm's tumor	3
Mediastinal mass in SVC syndrome	3

Second column numbers depict how many times the condition was referred by participants.

"Critical value" is a well known concept in clinical pathology laboratory. It has been defined as quantitative levels of analytes in any body fluids (particularly in serum) which impose patients directly or indirectly to life threatening consequences, and hence needs rapid communication with physician and immediate intervention. The most usual examples in daily practice are very high or very low levels of serum potassium.

In surgical pathology the situation is very different from clinical laboratory setting. Surgical pathology reports' contents are usually in the form of "interpretations". Very rarely "numerical data" can be found in surgical pathology report and if they can, most often are indicators of grading or staging of a benign inflammatory (viral hepatitis) or malignant neoplastic process. Hence critical values in surgical pathology cannot be defined by "cutoffs". In addition most critical values in surgical pathology are those which depend on the clinical variables and condition of patient. For instance absence of chorionic villi, trophoblastic cells and embryonal tissue in an endometrial curettage can be considered critical only when the patient was suspected to be pregnant. Accordingly the more appropriate term might be "critical diagnoses" rather than "critical values". Such limitations hampered precise definition of CV in surgical pathology. On the other hand if in a clinical situation the problem of time plays a critical role, one of routine approaches is intraoperative consultation and frozen section examination. One can postulate that CVs in surgical pathology are those conditions which cannot be managed by frozen section examination. These are among the reasons why defining CV in surgical pathology is not straightforward and why the pathology literature including pathology textbooks are very poor on this issue.

One of the rare studies performed to define CVs in surgical pathology is a survey performed by Pereira and colleagues. According to their personal experience they provided a list of 11 conditions which considered being critical values in surgical pathology. Then they presented the list to a group of 11 pathologists and 5 clinical specialists and asked them to grade the situations as a CV in a four tier scale according to the level of urgency for communication with clinicians. The situations included crescents in kidney biopsy specimen, vasculitis, bacteria in heart valve or bone marrow, organisms in an immunocompromised patient, fat in an endometrial curettage specimen, uterine contents without villi or trophoblasts in the workup of a patient suspected to be pregnant, mesothelial cells in a heart biopsy specimen, transplant rejection, malignancy in superior vena cava syndrome, neoplasms potentially causing paralysis, and large vessels in a core

biopsy specimen. Additional situations were added by participants to the above list. Participating pathologists added unexpected malignancy, disagreement between frozen section and permanent diagnoses, all fine-needle aspirations performed by a pathologist, fat in snare of biopsies of a colon polyp, polyomavirus in urine cytologic specimen, hydatidiform mole, hemophagocytic syndrome, necrotizing fasciitis, staphylococcal scalded skin syndrome, and various hematologic malignant neoplasms such as acute leukemia (also listed specifically as acute myelogenous leukemia, French-American-British type M3), Burkitt lymphoma, and leukemia cutis. The additional diagnoses listed by clinicians were unexpected malignancy, change of diagnosis in inflammatory bowel disease (from Crohn disease to ulcerative colitis or vice versa), acid-fast bacilli in a tissue biopsy specimen (eg, lymph nodes), and invasive aspergillosis or fungi in the nasal sinus or lung.

The same group of scientists later conducted another survey for evaluation of critical values in cytology. Critical value conditions which presented for scaling to participants were unexpected malignancy, disagreement between preliminary and final fine-needle aspiration diagnoses, and organisms in nongynecologic and fine-needle aspiration specimens. Additional CV cases suggested by the survey participants included herpes in a Pap smear in a pregnant patient, atypical glandular cells of uncertain significance in Pap smears, amended reports, very unusual tumors, disagreement with outside slide interpretation, infection or malignancy in orbital fine-needle aspiration samples, discrepancy between clinical expression and pathologic interpretation, and delay in signing out the cytology report.

Considering the long list of conditions mentioned by participants in the present survey (about 90 conditions), it is not surprising that many of conditions which has been discussed in two surveys of Pereira et al were mentioned by participants of present survey, too. But there are some concerns about the viewpoint of present survey participants. The first is the large number of refers to conditions which they are not included in the Pereira studies and rationally cannot be considered as a CV. Such conditions include breast malignancy (9), malignancy in all endoscopic specimens (6), malignant melanoma (6), brain tumors (6), and CIN II and III (5). The second is very low number of refers to the conditions which are considered as CV in the Pereira studies. Such conditions include diagnostic curettage in suspected ectopic pregnancy (2) and presence of crescents in kidney biopsy specimens (2). There are some important conditions in the Pereira studies which were never mentioned by participants in present survey. These include bacteria in heart valve or bone marrow, organisms in an immunocompromised patient, fat in an endometrial curettage specimen, mesothelial cells in a heart biopsy specimen, large vessels in a core biopsy specimen, fat in snare of biopsies of a colon polyp, and acid-fast bacilli in a tissue biopsy specimen. Although many of these conditions are rare in general pathology laboratories and are only encountered in specific situations (e.g. mesothelial cells in heart muscle biopsy), many of them are among the conditions that may happen in every general hospital pathology laboratory (e.g. fat in endometrial curettage, fat in the snare of biopsies of colonic polyps, or large vessels in needle biopsy specimens). This comparison shows that the general knowledge about the critical values in surgical pathology is poor among pathologists

community. One of the reasons may be the rarity of documents covering this important issue in pathology literature. Dedication of a book chapter or appendices of textbooks to this topic can definitely attract attention of under or post graduate pathologist to this important topic.

To fill this gap in our knowledge, in 2006 the Association of Directors of Anatomical and Surgical Pathology (ADASP) provided a list of CVs in surgical pathology. Based on the survey among ADASP members, the following conditions were extracted and categorized under three different headings. Group one is conditions that have immediate clinical consequences. This group includes crescents in greater than 50% of glomeruli in a kidney biopsy, leukocytoclastic vasculitis, uterine contents without villi or trophoblast, fat in an endometrial curettage, mesothelial cells in a heart biopsy, fat in colonic endoscopic polypectomies, transplant rejection, malignancy in superior vena cava syndrome, and neoplasms causing paralysis. The second group is composed of conditions in which there are unexpected or discrepant findings. This group includes significant disagreement between frozen section and final diagnosis, significant disagreement between immediate interpretation and final FNA diagnosis, unexpected malignancy, and significant disagreement and/or change between primary pathologist and outside pathologist consultation (at either the original or consulting institution). The third group is constituted by infectious conditions. They include bacteria or fungi in CSF cytology in immunocompromised or immunocompetent patients, pneumocystis, fungi or viral cytopathic changes in bronchoalveolar lavage (BAL), bronchial washing or brushing cytology specimens in immunocompromised or immunocompetent patients, acid-fast bacilli in immunocompromised or immunocompetent patients, fungi in FNA of immunocompromised patients, bacteria in heart valve or bone marrow, herpes in Pap smears of near term pregnant patients, and any invasive organism in surgical pathology specimens of immunocompromised patients. ADASP committee has emphasized that the above list would be considered only as a template and each institute should define their list individually and by cooperation with clinical colleagues. They also commented to avoid overuse of this terminology.

Diagnostic Pathology 2008, 3:30

What are the Features of an Expert Haematological Microscopist?

Galvani, D.

Aims: To determine what skills make a good consultant haematological microscopist; to explore how these skills develop during training and to determine whether these skills are maintained following training.

Methods: Twenty consultant haematologists underwent a semistructured interview to explore these issues. The interviews were transcribed and analysed for common themes using the N-vivo analytic package. This provides rich subjective qualitative data as opposed to hard objective quantitative data.

Results: Experience, methodicity and interest were the commonest skills mentioned. However, 25% of interviewees admitted they no longer followed a format when reporting. Interviewees agreed they had passed from a hypothetico-deductive to a scheme-inductive diagnostic reasoning model during acquisition of expertise. Only 20% had undertaken refresher training since becoming consultants, but the majority undertake some peer review of their work.

Conclusions: These skills could form the basis of vocational and revalidation assessments in the practice of haematological microscopy. The elucidation of such skill development can help refine standards and remedial training through the process of "deliberate practice." Finally, the low uptake of refresher courses for established consultants needs serious consideration.

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Fine-Needle Aspiration Cytology Identifies Noncompliance in Children Taking Antituberculous Medication: Presented at ASCP

Maggie Schwarz

BALTIMORE, Md -- October 17, 2008 -- Fine-needle aspiration cytology offers an effective way to diagnose noncompliance with antituberculous medication, is inexpensive, and is readily available, according to a study presented here at the American Society for Clinical Pathology (ASCP) 2008 Annual Meeting.

"In developing countries, polymerase chain reaction -- which gives immediate results -- is not available for diagnosing noncompliance with antituberculous medication," asserted researcher Sufian Zaheer, MD, Department of Pathology, Jawaharlal Nehru Medical College and Hospital, Aligarh, India.

Therapeutic failures in antituberculous therapy are common in children and need to be identified so that medications can be restarted. Dr. Zaheer and his colleagues set out to determine whether fine-needle aspiration cytology can accurately diagnose untreated tuberculosis or tuberculosis that has undergone a lapse in treatment.

He presented the study's findings on October 16.

Tuberculosis remains a major health problem in the less developed nations and is now also a major health problem in developed countries, owing to the emergence of HIV infection and AIDS, Dr. Zaheer explained. Despite the availability of effective chemotherapies, however, patient noncompliance with regimens is the most common reason for therapeutic failures. Compliance is particularly difficult to achieve in children because they are totally dependent on their parents.

A total of 305 children diagnosed with tuberculosis on the basis of x-ray, fine-needle

aspiration cytology, and acid-fast bacilli staining and/or culture were assessed for drug compliance by cervical lymph node fine-needle aspiration cytology.

Fifteen cases were found to be drug defaulters on detailed and meticulous follow-up.

Patients receiving inadequate chemotherapy were found to have splintered epithelioid cell granuloma with focal increase in fibrous elements, the researchers found.

Dr. Zaheer and the investigators concluded that fine-needle aspiration cytology was effective for identification of patients who were noncompliant with therapy.

MICROBIOLOGY

Identification of Four Species of Human Malaria Parasites by Fast PCR

[Article in Japanese]

Sato K, Shinomiya N.

Malaria is one of the most prevalent infectious diseases in the world. Accurate identification of four species of human malaria parasite is essential for appropriate treatment. Here, we developed a simple and rapid method of identifying Plasmodium species using a fast polymerase chain reaction (PCR) assay. Based on the previous literature, we amplified small subunit ribosomal RNA genes of four human malaria parasites. To establish a minimum detection limit, a blood sample with a known number of *P. falciparum* parasites (parasitemia: 3%) was diluted serially (from 0.03% to 0.000003%). We compared the detection limits between single (one-step) PCR and nested (two-step) PCR. Other clinical blood samples, which were infected with *P. falciparum* (parasitemia: 2.8%), *P. vivax* (parasitemia: 0.13%), *P. ovale* (parasitemia: 0.04%), respectively, were also tested by our PCR system. The PCR findings were compared to those of blood film Giemsa staining and rapid diagnostic tests (RDT). The sensitivity of our method is less than one parasite in 1 microl of blood (estimated parasitemia: 0.000003%) for both single PCR and nested PCR, though an increased number of cycles (40 cycles) was required for single PCR. Using clinical samples, it was proven that amplified products by single PCR could clearly distinguish between *P. falciparum*, *P. vivax*, and *P. ovale*. To detect *P. vivax* and *P. ovale*, the PCR system was more sensitive than RDT. The total required time for our method was within three to four hours from DNA extraction to PCR detection. Taken together, our method is easier and faster than the previously reported PCR-based malaria parasite identification systems, and is also useful for cases in which diagnosis by Giemsa staining and RDT is difficult.

Rinsho Byori. 2008 Aug; 56(8):657-61.

BOTTOM LINE

The Modern Histopathologist: In the Changing Face of Time

Biman Saikia, Kirti Gupta, Uma N Saikia

The molecular age histopathologist of today is practicing pathology in a totally different scenario than the preceding generations did. Histopathologists stand, as of now, on the cross roads of a traditional 'visible' morphological science and an 'invisible' molecular science. As molecular diagnosis finds more and more applicability in histopathological diagnosis, it is time for the policy makers to reframe the process of accreditation and re-accreditation of the modern histopathologist in context to the rapid changes taking place in this science. Incorporation of such 'molecular' training viv-a-vis information communication technology skills viz. telemedicine and telepathology, digital imaging techniques and photography and a sound knowledge of the economy that the fresh entrant would ultimately become a part of would go a long way to produce the Modern Histopathologist. This review attempts to look at some of these aspects of this rapidly advancing 'art of science.'

Introduction

In an era when we are talking of molecular classification of traditional histology and immunotherapy for cancers, the role of the histo-morphologist seems to be exploring areas which one could never have imagined a few decades ago. With the rapidly advancing field of biotechnology and molecular biology, a modern histopathologist is expected to be well versed not only in the traditional histopathological techniques but also keep pace with the ever expanding frontiers of science and technology. With molecular diagnosis threatening to overrule the histopathological diagnosis with every new discovery, it is time for the histopathologist to embrace and incorporate the recent advancements of practicing pathology in its present modern context.

With the fast changing scenario, it is but obvious that the accreditation of the histopathologist needs to undergo a drastic change, and re-accreditation of those already in the field a necessity. The current accreditation process prevalent across most of the countries is to train a student with varying degrees of vigorousness and at the end of the training period, assess the candidate through an examination which gives the license to practice pathology throughout life. In this scenario, it is totally up to the pathologist's own efforts and interest to keep up all the enthusiasm of keeping abreast with the newer technologies and new diagnostic trends. Whereas primary accreditation looks at what the new practitioner can do, re-accreditation looks at what established practitioners actually do. The accreditation and re-accreditation procedure hence needs to incorporate not only the traditional histopathological techniques but also the fields of biotechnology, telecommunication, information communication technology, professional photography and a bit economics. To this list could be added a number of other entities such as socio-psychological factors, hospital management, quality control and host of others which is

beyond the scope of this discussion. The subject of accreditation and re-accreditation however has been a topic of constant reviews where authors have tried to devise list of areas where a histopathologist should be able to demonstrate continuing fitness to practice, and activities like recording educational activity, testing a pathologist's knowledge and interpretative skills, testing his diligence and peer review/appraisal has been proposed as various ways and tools to assess a practicing pathologist.

Discussion

There was a time when Pathology was considered to be the mother of medical science. Histopathology, over the years has witnessed its evolution from a mainly autopsy based pathology to the current molecular histopathology. Recent advances in various fields of science and technology and the incorporation of these to histopathological practice and acceptance of some philosophical concepts, particularly the functional correlation of morphological studies, has changed the outlook of both histopathology and the histopathologist. An elaborate review on the topic at the turn of the century by Mapstone and Quirke on the pathologist in context to the 21st century reveals that the ideas and concepts that were envisioned then, has to a large extent, already found widespread acceptance in the pathology community, if not moved further into it. This is particularly true for the application of information technology, and to a lesser extent to molecular biology advances.

It's been sometime now since the human genome has been sequenced. The initial enthusiasm of reading the blueprint of human biology however seems nowhere in sight and it is likely to be a number of decades before many of the expected benefits of the genomics revolution are actually realized to the extent initially expected. The human genome project seems to have opened the Pandora's Genomic Box resulting in more questions than answers.

The first and most widespread application of molecular techniques in histopathology perhaps came in the form of immunohistochemistry (IHC), though IHC perhaps cannot be called a molecular technique in its true sense. Albert H. Coons and his colleagues were the first to use fluorescent dye labeled antibodies to identify antigens in tissue sections. Subsequently, enzyme labels such as peroxidase (Nakane and Pierce 1966), alkaline phosphatase (Mason and Sammons 1978) and Colloidal gold (Faulk and Taylor 1971) were introduced. Within a short time, histopathology literature was flooded with immunohistochemistry-based studies, expanding both the list of markers as well as the list of confusion. A pubmed search as on 31st March 2008 for "immunohistochemistry" yielded a total of 415067 papers with 12,426 reviews reflecting the impact immunohistochemistry have had on the scientific community. The impact of the scientific breakthroughs has however started becoming more apparent gradually. This can be said more confidently when we talk of the contribution of the gene array technology, which allows gene expression measurements of thousands of genes in parallel, providing a powerful tool for pathologists seeking new markers for diagnosis. With such kind of data accumulating rapidly, it won't be long when molecular signatures would be assigned to each and every pathological lesion and a molecular diagnosis would come even before

the paraffin sections are ready, and the histopathologist would already know what to look for in the sections.

Various studies have demonstrated that micro-array based gene expression profiling enables accurate tumor classification and can be very helpful diagnostic tool for cancers with unknown primaries and histologically undifferentiated tumors. Molecular profiling has also given way to the histo-clinical classifications since such a classification would be expected to corroborate more accurately with the functional behavior of a tumor. Molecular classification systems have been attempted extensively in organ systems like the breast and the kidney and to a lesser extent in lungs, thyroid, endometrium, ovarian, testicular, and sarcomas. Molecular classification systems in breast carcinoma and renal cell carcinoma have been shown to be of relevance in not only classification and diagnosis but also in assessing response to therapy, and as a predictor of survival. Whereas genomic studies are establishing new molecular classifications, genetic alterations are also being identified and characterized, generating new targets for therapy and new tools to predict disease recurrence and response to therapy. This combined molecular approach is expected to have an impact on individual 'tailored' therapy for cancer patients.

However, with the concept of "tumor heterogeneity" being increasingly recognized, it has become imperative to define and analyze pure population of cells separately within the neoplasm before assigning a molecular signature to a particular neoplasm. The advent of Laser capture microdissection (LCM) technique has brought about this reliable procurement of pure population of cells from tissue sections under direct microscopic visualization and bridged a very significant technical gap between the histopathologist's microscope and the molecular biologist's work bench. This has now opened the doors to enhancing our understanding of molecular mechanisms regulating cellular developments and its functioning both in normal and diseased states.

Whereas techniques like immunohistochemistry are relatively simple and can be performed by the technical staff, more sophisticated techniques involving handling of cell culture for LCM and other molecular methods require a more intricate knowledge on the part of the pathologist for meaningful interpretation. It is thus important that a student is given hands-on experience on performing these techniques, designing of such experiments and more importantly interpreting the data, during the initial training.

The initial excitement surrounding the development of DNA micro array analysis and proteomics has also raised questions about the role of these techniques in actual clinical practice and patient management. Though it is theoretically possible to build a comprehensive gene expression database for each of the organ systems/tumor type and use it as a clinical diagnostic and prognostic tool, microarray technology remains complex and time consuming and hence till date remains largely a research tool. It is prudent thus that the histopathologist takes this molecular approach with all his background knowledge of morphology rather than leave tumor diagnosis entirely in the hands of the molecular biologist to have a better and long lasting impact.

Nature however has never been too kind to the scientist. The story does not end with mere expression or non-expression of a gene and production of the corresponding mRNA. What matters at the end is the functionality of the end product which is the 'functional' protein. The story only begins with the production of the mRNA and the probability of the mRNA ultimately landing up in a functional protein would depend on the smooth co-ordination of an even more complicated array of biological functions like splicing, post-translational modifications, DNA methylation, acetylation and a host of other epigenetic factors. What needs to be assayed is therefore the functional aspect of the gene and the focus hence shifts from genomics to proteomics and more recently to glycomics. The latter, scientists believe, will have an equally dramatic effect as genomics have had. Given that a single protein can come with 10 or more different forms of sugars attached to it, giving the protein subtle differences in function, which cannot be detected by the current techniques in proteomics, leaves us with the possibility of modulating the function of the protein by modulating the activity of the enzymes that make the sugars on the protein. With the advent of the "glyco-chip" it is now possible that histopathologists too will be hunting for the sugars on these proteins and possibly do "Immuno-glyco-histochemistry" to look at the functional aspect of the protein they currently detect with immunohistochemistry. The scope is ever expanding and unlimited.

Information Communication Technology (ICT) and the histopathologist

Working in isolated environments where access to peers, education and information is limited, is one of the highest risk factors for physicians' loss of medical competence. This can't be less true for the histopathologist as well. So it's evident that for keeping pace with all the advances taking place at an outstanding pace, dissemination of information becomes a major issue. It is not surprising therefore that medical science has found its place in the electronic media or *vice versa*. It is also not surprising that most if not all prominent journals in life sciences are available on-line, though with a price tag and majority of them has options for online-submission and peer review. Information Communication Technology has also paved way for telemedicine and a science which, a few years back was a mere concept is rapidly gaining recognition as a well defined, accepted and effective application and is already showing its impact in clinical practice and modern healthcare. While telemedicine is a wider concept, Telepathology is the sub-discipline of telemedicine that deals with the capture, transmission, and viewing of pathological and histological images via telecommunication channels such as the internet, dedicated satellite or telephone, as opposed to the conventional methods of microscopy. Offering a host of innovative benefits and applications, modern telepathology systems now help to deliver more accurate remote diagnoses and are tending to replace traditional consultancy methods using light microscopy.

Telepathology over the years has undergone drastic evolution from static telepathology, which is a store-and-forward concept to dynamic, real time telepathology, using fully motorized robotic systems. A concordance rate of as high as 99–100% has been reported between telepathology and light microscopic diagnosis using such technologies. What has however revolutionized telepathology applications and has to a great extent removed

the most fundamental drawback of telepathology, that is the limitation of the available image for diagnosis, is the concept of the "virtual slide."

Virtual slides are digitized images where the entire slide is scanned at a very high resolution, acquiring the entire image of the histopathology section at all magnifications available on the microscope. Software driven motorized stage helps acquire all the fields of view and digitally stitch all the images into one single image which can be viewed by multiple pathologists.

The novel array microscope for the first ultra-rapid virtual slide processor (D Metrix DX-40) digital slide scanner is an example. The optics consists of a stack of three 80-element 10×8 -lenslet array. Uniquely shaped lenses in each of the lenslet arrays constitute a single "miniaturized microscope" constituting a set of 80 microscopes. Scanning a glass slide with the array microscope produces seamless two-dimensional image data of the entire slide i.e. a virtual slide. Image acquisition can be as rapid as 58 secs for a 2.25 cm^2 tissue section and 40 slides can be scanned per hour. The only limitation would be the extremely large file size of the images generated, sometimes exceeding 1.5 Gb and hence the limitation with the bandwidth available for transmitting these images. It is however not unachievable considering the fact that investing in such a facility with a high bandwidth capacity is a one time effort, the fruit of which can be reaped for a long time. So, with the advent of virtual microscopy, it may be possible that an expert referral pathologist in the near future will be interpreting virtual images on LCD screens rather than glass slides. What needs to be emphasized is incorporation of the concept right at the inception of the pathology training with hands-on experience on these concepts rather than giving a theoretical knowledge for the simple reason that the factor which has till now hampered the growth and wide-spread application of telepathology is the mindset of the traditional pathologist, despite scientific evidence of efficacy of the technique. A technology can improve only when it finds its place in routine practice, and it is only through trial and error that a desired perfection is achieved. The situation however is not that grim, and excellent examples of telepathology applications have been set even in a country like India which is fast shedding its third world image. With the opening of the School of Telemedicine at the Sanjay Gandhi Post Graduate Institute of Medical Sciences at Lucknow, the first of its kind in the country, telepathology promises a bright future in the times to come.

Histopathologist as a professional photographer

A morphologist's job is to play with images, be it live or captured. Photography hence has been an integral part of pathology practice since its inception. But chemical photography now has largely been replaced by digital photography for obvious reasons. Digital cameras utilize charge-coupled devices (CCD) or Complementary Metal Oxide Semiconductor (CMOS) image sensors to measure light energy and their circuitry to convert the measured information into a digital signal. Digital photography has the advantage of lower running cost, instant availability without any processing, easy archiving and transmission through the electronic media. It allows for adjusting, enhancing, and annotating images. Moreover it allows for post acquisition image

optimization and modification to suit one's satisfaction of image quality using softwares like Adobe Photoshop. This however also carries the disadvantage and the fear of falsification of images. But as is true for every technological advance distraught with some side-effects, the advantages of digital photography in histopathology practice far outweighs its disadvantages. Patient care is enhanced by the transmission of digital images to other individuals for consultation and education, and by the inclusion of these images in patient care documents. In research laboratories, digital cameras are widely used to document experimental results and to obtain experimental data.

Since pathology is a visual science, the inclusion of quality digital images into lectures, teaching handouts and electronic documents is as essential as taking lectures. A few institutions have gone beyond the basic application of digital images to developing large electronic atlases, animated, audio-enhanced learning experiences, multidisciplinary internet conferences, and other innovative applications. So isn't it wise for a postgraduate student to handle a short course on photography at the beginning of the training as a pathologist?

Conclusion

So, are we looking at a histopathologist, who is not only a good traditional morphologist, but also an IT savvy scientist, with drops of Leonardo Da Vinci and Picasso in blood, who is ready to take technology head-on, without the inhibitions the current generation of pathologists face and who would look at the histopathology market with a much wider perspective of the current economy? The making of this Modern Histopathologist would require a drastic change in the accreditation procedure and a major initiative on the part of the policy makers and the teachers of Pathology. The transition from a field of visual interpretations to a largely invisible molecular science will take some time to firm foot but will eventually be there.

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