15 The Antibody Revolution: How 'Immuno' Changed Pathology

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"*You are young still, pathologist*", *the old professor said*,
"*And your slides aren't just purple and pink*,
They have staining in brown, of both nuclei and membranes,
Pray, is this a good thing, do you think?"

Rather dramatically adapted from 'Father William' by Lewis Carroll.

INTRODUCTION

What we Mean by the Antibody Revolution

In the early 1980s one of our senior consultants in Oxford was asked to review a case of head and neck lymphoma that had been misdiagnosed as carcinoma. He could not find the H&E because this was out with us 'young thrusters' alongside the frozen section immunostains. An immediate letter was sent to the ENT consultant with copies to all and sundry. The gist was 'I don't mind these young chaps playing around in the lab, but when they get in the way of my diagnostic practice I really do draw the line'. He was actually a kindly old stager and we (David Mason and K. C. G.) portrayed him as such in a cartoon illustrating an early review of ours on the diagnostic value of immunohistochemistry (Fig. 15.1) (Mason and Gatter, 1987).

The whole thing seems ludicrous now. There is no pathologist in this country, or possibly the world, who does not use some immunocytochemistry regularly for diagnosis. You would certainly be up before 'his lordship' if you made wrong diagnoses through resistance to incorporate antibodies. That is probably not true for any other technique in histopathology and is one way of introducing this as a revolution. It has completely and permanently changed our routine practice.

Another way of assessing the revolution is to look at the impact of immunocytochemistry on the laboratory workload. In 1980 in Oxford this was, to all intents and purposes, zero. By 1992 it had moved to form nearly 3% of our workload, whereas now it is almost 15%. In 2004 we produced 24 798 immunostained glass slides against an overall output of 166 332 slides for all purposes. Four boxes of glass slides with 50 slides to a box weighs 1 kg which means that we imported 124 kg of glass into the department just to perform immunostains. Multiplied around the country and to other developed nations the 'immuno' revolution is, if nothing else, a significant contributor to road transport use.

When was the Revolution?

We stated previously (Mason and Gatter, 1987) that although there were forays into immunocytochemical methods in the 1960s the game as we know it today started in 1974 with the work of Taylor and Burns demonstrating immunoglobulin in plasma cells in paraffin sections (Taylor and

Figure 15.1 Cartoon showing how we thought of the situation in histopathology with regard to immunocytochemistry in 1987, reproduced from a previous review (Mason and Gatter, 1987).

Burns, 1974). This was soon confirmed by a number of laboratories but remained largely a laboratory oddity due to the lack of markers of real diagnostic value other than light chains. In addition, the techniques used at the time were relatively insensitive so antigen detection was a variable and unreliable endeavour. Indeed, as a number of workers have stated, it was lucky that Taylor and Burns chose immunoglobulin for their experiments because it is so abundant in plasma cells that it allows easy detection.

Enhanced Methods

The next significant advance made in the introduction of diagnostic immunocytochemistry was the discovery that proteolytic digestion of paraffin sections considerably enhanced both the intensity and reliability of antigen detection (Huang *et al*., 1976; Reading, 1977; Mepham *et al*., 1979). Of course this early example of importing kitchen technology into laboratory science has been supplemented by many others, most notably recent methods using microwave ovens and pressure cooking in a variety of different pH buffers (Leong *et al*., 2003) – if only we had thought to do it the other way round and we would have beaten a certain well-known chef to best restaurant in the world and become millionaires!

The Repertoire of Reagents

With all of these wonderful technical advances in the 1970s, why did it take so long for immunocytochemistry to become a standard technique in routine diagnosis? The answer is simple in retrospect. There were just no useful antibodies. This hurdle was solved immediately when pathologists realised that the invention of monoclonal antibodies by Kohler and Milstein (Kohler and Milstein, 1975) enabled a continuous production of new markers for evaluation. At first many of these needed frozen sections to detect their antigens, but as antigen retrieval methods improved and further antibodies were produced, an extensive range of diagnostic markers emerged.

One measure of this progress is to look at the work of the various conferences set up to characterise and classify monoclonal antibodies against human leucocyte differentiation antigens. These are the so-called CD conferences from which the CD numbers used mainly in haematopathology have come. CD stands for 'Cluster of Differentiation' and represents an internationally agreed system for classifying antigens and their respective monoclonal antibodies. A CD group is a cluster of antibodies recognising the same antigen. Where there is a series of related genes giving rise to antigenic variants, the CD groups have been subdivided, e.g. CD1 a, b, c or CD11 a and b. These groupings are defined at International Workshops on Human Leucocyte Differentiation Antigens. The first of these was held in Paris in 1982, when 15 CD groups were defined. To date there have been eight workshops and the number of clusters has increased to 247. Although this seems a large number it is nothing compared to the thousands of antibodies, each with their own 'laboratory' names, that have been allocated to the clusters. Interested readers can discover more about the CD system from the relevant workshop reports or about many of the antibodies from a number of antibody companies that have information fact sheets available, e.g. R&D Systems, DAKO or Visionbiosystems – all of these have websites and in addition there are many antibody search and review sites available on the web.

The anti-CD antibodies are of course only the tip of the immunocytochemical iceberg. The number of reagents against other markers of diagnostic usage, only some of which will be mentioned here, is now very extensive. To detail them all would take several times more space than is available for this review. An excellent starting point for the novice is the recent laboratory manual on diagnostic antibodies by Leong and colleagues (Leong *et al*., 2003).

WHAT DO WE USE ANTIBODIES FOR?

Today a formidable range of antibodies, monoclonal and polyclonal, from a variety of animals and increasingly from gene expression libraries, is available to the pathologist. So what do we actually use them for? In our view there are three major areas of practical value at present. These are:

1. To make or confirm a diagnosis.
2. To provide prognostic information.
3. To determine treatment.

The remainder of this short overview will attempt to summarise these three areas, mainly in the form of tables, with some comments, specific and general, where we feel that they will be useful. In order to make the tables reasonably aesthetic, we have used frequent abbreviations, which may not be familiar to all readers. These abbreviations are explained in the footnotes to the tables, in order of appearance.

Table 15.1 Differentiating major tumour types

Class of neoplasm	Antibodies	Comments
Carcinoma	Cytokeratin, epithelial membrane antigen (EMA)	EMA is not epithelial-specific (vascular lesions, plasma cells, meningioma and some lymphomas (Theaker *et al*., 1986; Gatter and Delsol, 2002). It has a particular use in some poorly differentiated carcinomas that express little or no cytokeratin, such as renal cancers (Langner *et al*., 2004)
Sarcoma	Vimentin and markers of lineage (see Table 15.3)	Some overlap with carcinoma and melanoma, depending on the type of sarcoma (Fletcher *et al*., 2002)
Melanoma	S100, melan-A (Mart-1), HMB-45, MITF, PNL2 (Rochaix *et al*., 2003)	S100 is sensitive but not specific, so a combination of these markers is needed (Fletcher, 2000)
Lymphoma	CD45 and lineage markers (see Table 15.5 and 15.6)	Hodgkin lymphoma and myeloma are typically negative (Jaffe *et al*., 2001)

To Make or Confirm a Diagnosis

The first area of diagnostic impact was in differentiating the major groups of malignant tumours when they were too poorly differentiated histologically (Gatter *et al*., 1982, 1984; Warnke *et al*., 1983). There was some grumbling at the time that we needed to do this because we, the new generation of pathologists, were not up to scratch morphologically. We countered this with a wicked little study that dug out cases of poorly differentiated tumours from the Oxford archives and showed that there was a 40% error rate in assigning these to their major tumour types (Hales *et al*., 1989). Initially these studies were heavily biased towards the commoner and crucial distinction of carcinoma from lymphoma (Gatter *et al*., 1985) but today, as shown in the tables below, the range of uses is much larger.

Table 15.1 shows a current plan of action for differentiating so-called anaplastic tumours. This has not changed much in the last ten years and most modern pathologists probably think much more of immunostaining for subdividing or classifying tumour types. Indeed, the act of classifying will usually give a robust assignation, e.g. a B-cell phenotype is a lymphoma. Nevertheless, the correct identification of a tumour's origin is crucial and the role of immunostaining in this should not be overlooked.

Table 15.2 is similar to Table 15.1 but deals with tumour types confronted less commonly as unknowns or anaplastic lesions. Nevertheless, a few poorly differentiated tumours are met in practice that fail to be categorised by the antibodies of Table 15.1 – they have no clinical or morphological clues or do not make sense clinically or morphologically. In these cases in adults the tumours in Table 15.2 are worth considering, and in children and adolescents those in Table 15.3. An obvious difference from Table 15.1 is the much greater number of antibodies recommended by various experts. This reflects the heterogeneity of many of these tumours, especially those of germ cell or paediatric type.

The phenotypes of paediatric tumours given in Table 15.3 are an amalgamation of several detailed reference works and should not be considered in any way definitive. Paediatric tumours that are classified only on immunophenotype are a peculiarly primitive group of tumours and great overlap in marker expression does exist (Kleihues and Cavenee, 2000; Jaffe *et al*., 2001; Fletcher *et al*., 2002, Mills *et al*., 2004; Sebire *et al*., 2005).

Table 15.2 Differentiating less common tumour types

Class of neoplasm	Antibodies giving positive immunostaining	Comments
Germ cell tumour	PLAP, AFP, HCG, CD30, CD117, CK	All of these markers appear on other tumour types (Mills *et al*., 2004)
Mesothelioma	CK5/6, CK7, WT-1, calretinin, mesothelin, thrombomodulin, EMA, HBME-1	Expression of many of these antigens is variable (Fletcher, 2000; Mills *et al*., 2004; Politi *et al*., 2005)
Central nervous system (CNS) tumours	GFAP, neurofilaments, S100	CNS tumours show a wide range of different immunophenotypes (Kleihues and Cavenee, 2000)

PLAP, placental alkaline phosphatase; AFP, alpha fetoprotein; HCG, human chorionic gonadotrophin; CK, cytokeratin; WT-1, Wilon's tumour protein 1; EMA, epithelial membrane antigen; GFAP, glial fibrillary acidic protein.

Quite frequently poorly differentiated tumours are clearly of haematological origin, perhaps because of a distinctive morphology or after a first round of immunostaining. However, it may still be unclear what type of tumour one is dealing with. Before launching out on detailed immunophenotyping it may be sensible to undertake a further small study such as is illustrated in Table 15.4. Here it can be seen that with a limited range of markers it is usually possible to identify the major tumour group that one is dealing with, and then a more detailed classification can be undertaken (Jaffe *et al*., 2001).

Tables 15.5 and 15.6 feature B-cell lymphomas classified into low and high grade. Strictly speaking the terms low and high grade for lymphomas were abolished by the WHO classification in 2002 (Jaffe *et al*., 2001) but they have lingered on as pathologists have struggled to find a better term for their comparison and differential diagnosis. A few comments may be helpful. Cyclin D1 staining has revolutionised the recognition of the important entity of mantle cell lymphoma. Initially this was a difficult marker for routine laboratories but the introduction of new rabbit monoclonal antibodies has changed all of this. Most centres now use these but caution is needed when interpreting focal or weak staining. Some normal cells such as endothelium and macrophages express cyclin D1 and with enhanced staining techniques the rabbit antibodies are starting to show some weak positivity in other lymphoma types, especially follicular lymphomas. Follicular lymphomas are clearly different from the other tumours here but are easily confused with reactive lymph nodes. Here the key immunostains are CD10 and bcl2: CD10 positivity in interfollicular lymphocytes is diagnostic of follicular lymphoma, as is bcl2 positivity in their neoplastic germinal centres. Immunostain bcl2 does not appear in Table 15.5 because all of these tumours are positive. Sometimes nodular lymphocyte predominant Hodgkin lymphoma looks like follicular lymphoma but here the large abnormal cells, although bcl6 positive, are CD10 negative (Jaffe *et al*., 2001; Gatter and Delsol, 2002).

At present the important differential to make among the high-grade B-cell lymphomas is to recognise Burkitt and lymphoblastic lymphoma. Burkitt lymphoma is the entity that usually causes most problems but if strict immunocytochemical criteria are applied then the diagnosis is usually pretty robust and consistent. If in doubt it is better to call it a diffuse large cell lymphoma. Lymphoblastic lymphoma is generally more obvious, especially in a younger patient, when one is thinking of this entity. Care should be exercised in older patients not to confuse mediastinal T lymphoblastic lymphoma with a thymoma (see Table 15.4).

Although they took some time to catch up with lymphomas, epithelial tumours now have a useful panel of markers for helping to assess their type and origin, as shown in Table 15.7 for

Table 15.3 Paediatric tumour differential diagnosis

Tumour/ antibody	Vimentin	Cytokeratin	EMA	NSE	Myogenin	Desmin	MyoD1	Muscle – specific actin	CD99	TdT	WT-1	NB84	NF
Neuroblastoma	−	−	−	+	−	−	−	−	−	−	−	+	+
Ewing's sarcoma/ PNET	+	−	−	−/+	−	−	−	−	+	−	−	−/+	−
Rhabdomyosarcoma	+	−	−	−	+	+	+	+	−	−	−	−	−
(Intra-abdominal) desmoplastic small round cell tumour	+	+	+	+	−	+	−	−	−	−	+	−	−
Congenital rhabdoid tumour	+	+	+	+	?	+/−	?	−	−/+	−	−	−	−
Lymphoblastic lymphoma	+	−	−	−	−	−	−	−	+	+	−	−	−
Synovial sarcoma	+	+	+	−	−	−	−	−	−/+	−	−	−	−
Wilm's tumour	+/−	+/−	−	−	−	−	−	−	−	−	+/−	−	−

PNET, peripheral neuroectodermal tumour.

Table 15.4 Haematological tumours

Haematolymphoid tumour	Immunostaining panel	Comments
B-Cell lymphoma	CD20, CD79a, Pax-5 (Torlakovic *et al*., 2002)	Anti-CD20 antibody therapy may alter the staining in relapse
T-Cell lymphoma	CD3, CD2, CD5, CD7, CD4/8	Frequently have abnormal T-Cell antigen patterns
Hodgkin lymphoma	CD15, CD30, MUM-1 (Carbone *et al*., 2002)	CD15 may be focal or absent
Myeloma	CD38, CD138, VS38c, kappa, lambda, CD79a, CD56, MUM-1	About a third of cases are also CD20+, which can be confusing
Histiocytic lymphoma	CD68, S100, CD1a, lysozyme	Covers a wide range of tumour types
Granulocytic sarcoma or other myeloid neoplasm	MPO, lysozyme, CD43	CD43 can cause confusion with T-cell lymphoma
Thymoma	CK (for epithelium) CD1a, TdT, CD99 (for lymphocytes) (Travis *et al*., 2004)	Easily misdiagnosed as lymphoblastic lymphoma if the CK is overlooked

CK, cytokeratin; TdT, terminal deoxynucleotidyl transferase.

Table 15.5 Classification of low-grade B-cell lymphomas

Lymphoma type	IgM	IgD	CD5	CD10	CD23	CD43	bcl6	Cyclin D1
Chronic lymphocytic leukaemia	(+)	(+)	+	−	+	+	−	−
Lymphoplasmacytic lymphoma	+	−	−	−	−/+	−/+	−	−
Marginal zone lymphoma	+	−	−	−	−	+/−	−	−
Splenic marginal zone lymphoma	+	+	−	−	−	−	−	−
Follicular lymphoma	+	−/+	−	+	−/+	−	+	−
Mantle cell lymphoma	+	+/−	+	−	−	+	−	+

Table 15.6 Classification of high-grade B-cell lymphomas

Lymphoma type	CD5	CD10	CD23	Ki67	TdT	bcl2	Cyclin D1
Burkitt lymphoma	−	+	−	>95%	−	−	−
Diffuse large B cell	−/+	−/+	−	<90%	−	+/−	−
Mantle cell blastic type	+	−	−	<90%	−	+/−	+
CLL Richter's transformation	+	−	+	<90%	−	+/−	−
B lymphoblastic	+	+	−	<90%	+	−	−

TdT, terminal deoxynucleotidyl transferase; CLL, chronic lymphocytic leukaemia.

Table 15.7 The differentiation of epithelial tumours with cytokeratins 7 and 20

CK7+ CK20+	CK7+ CK20–	CK7– CK20–	CK7– CK20+
Transitional cell	Breast	Hepatocellular	Colorectal
Pancreatic mucinous	Non-small-cell lung	Renal cell	
Ovarian mucinous	Ovarian serous	Prostate	
	Mesothelioma	Squamous	
	Endometrial	Neuroendocrine	
	Pancreatic		
	Thyroid		

Table 15.8 The differentiation of epithelial tumours with other markers

Carcinoma	Cytokeratin, epithelial membrane antigen (EMA), keratin subtyping (see Table 15.7)
Thyroid	Thyroglobulin, TTF-1, calcitonin (medullary)
Prostate	Prostatic acid phosphatase or prostate – specific antigen
Breast	Oestrogen receptor, progesterone receptor, c-Erb B2
Lung	TTF-1
Liver	CK8, CK18, hep-par-1, AFP
Pancreas	Ca19.9
Endometrium/ovary	CA125
Neuroendocrine	NSE, pgp9.5, NCAM (CD56), synaptophysin, chromogranin A

TTF-1, thyroid transcription factor 1; NSE, neuron-specific enolase; NCAM, neural cell adhesion molecule.

cytokeratin subtyping (Chu and Weiss, 2002) and Table 15.8 for a range of other antigens (Mills *et al*., 2004).

We could continue with many more tables of differential diagnoses but space dictates we call a halt. Just to show that not everything is tissue or tumour in pathology, we shall finish with renal disease, cytology and infection. Differentiating the different types of renal glomerular disease is a highly specialised area, but we hope that Table 15.9 gives a flavour of how immunostaining, still predominantly by immunofluorescence, may assist.

Differentiating mesothelioma from carcinoma in serous effusions has long been extremely difficult. Indeed, reactive mesothelial cells can also look very malignant at times so care is still needed in this differentiation. Nevertheless, the panel of markers available (shown in Table 15.10), which continues to grow, is of great assistance to cytologists today (Fletcher, 2000; Mills *et al*., 2004; Politi *et al*., 2005).

The number of infectious agents (whether viruses or bacteria) that can be identified in routine tissues continues to expand regularly. Table 15.11 outlines some of the commoner and more reliably identified agents in current practice.

Table 15.9 Immunostaining in renal disease with glomerular crescent formation

Diagnosis	Common immunofluorescent staining pattern
Antiglomerular basement membrane antibody disease (Goodpasture's Syndrome)	Linear GBM staining for IgG and C3 in majority, with fibrin/ fibrinogen in crescents
Immune complex crescentic glomerulonephritis related to infection	Coarse granular capillary wall staining with C3 +/− IgG, with fibrin/ fibrinogen in crescents
Immune complex crescentic glomerulonephritis related to lupus	Granular capillary wall and mesangial staining for C3, C1q (and C4), IgG, IgM and IgA, with fibrin/ fibrinogen in crescents
Pauci-immune necrotising glomerulonephritis (ANCA-related)	Negative immunofluorescence for complement and immunoglobulin in majority, with fibrin/ fibrinogen in crescents
IgA nephropathy/ Henich-Shönlein purpura	IgA and often C3 positive immunofluorescence, with fibrin/ fibrinogen in crescents

ANCA, anti-neutrophilic cytoplasmic antibody.

Table 15.10 Identification of malignant cells in serous effusion

Antibody/condition	Adenocarcinoma	Mesothelioma
EMA	+ cytoplasm and membrane	+ membrane
CEA	+ (usually)	−
B72.3	+	−
CK7	+ (variable)	+
CK20	+/−	−
LeuM1 (CD15)	+	−
MOC-31	+	−
CK5/6	−	+
Thrombomodulin	−	+
HBME-1	−	+
WT-1	−	+
Calretinin	−	+
Vimentin	−	+

EMA, epithelial membrane antigen; CK, cytokeratin; WT-1, Wilm's tumour protein 1.

To Predict Prognosis

Almost from the start as the early studies of tumour typing with antibodies were emerging, there was a dawn chorus from our clinicians of 'but does it tell us anything about prognosis?'. Sadly 25 years later immunostaining is still not a very reliable or helpful means of predicting prognosis. Nevertheless, there are a few tried and tested stains that have stood up to the unpredictability of human cancer. Some of these are summarised in Table 15.12.

For most other tumours, in spite of a huge amount of effort there just has not been enough data to substantiate any markers as truly meaningful for prognosis (Compton *et al*., 2000; Compton, 2003; Altman and Riley, 2005). Three antigens p53, bcl2 and Ki67 are (and have been for some time) front-runners as generic markers of prognosis in many, if not all, tumours. There is evidence (though not conclusive) that positivity for p53 and a high proliferative index with Ki67 are associated with a more aggressive tumour and hence a poorer prognosis (Steele *et al*., 1998; Brown and Gatter, 2002). Bcl2 is more problematic, with some tumours showing positivity in aggressive

Table 15.11 Infectious agents identifiable by immunohistochemistry

Infectious agent	Site
Helicobacter	Stomach
Polyoma	Urogenital tract (including positive immunostaining in urine cytology)
Herpes simplex virus	Epithelia: skin, orogenital, oesophagus
Cytomegalovirus	Endothelial cells and macrophages in many sites (especially immunosuppressed patients)
Hepatitis B virus	Liver
Human immunodeficiency virus p24	Lymphoid tissue
Epstein Barr virus	Lymphoid tissue, nasopharyngeal carcinoma
HHV-8	Human herpes virus 8, involved in the pathogenesis of Kaposi's sarcoma, primary effusion lymphoma and the plasma cell variant of Castleman's disease
Toxoplasma	Central nervous system, placenta

Table 15.12 Use of immunohistochemistry in the prediction of prognosis

	Prognosis	
Tumour type	Good	Bad
Breast	ER, PR	Her2
Colon	bcl2, beta-catenin	p53
Chronic Lymphocytic leukaemia		ZAP70, CD38
Anaplastic large cell lymphoma	Alk-1	
Diffuse large B-cell lymphoma	CD10, bcl6	p53, bcl2
Neuroblastoma	Trk-A	

ER, oestrogen receptor; PR, progesterone receptor; Alk-1, anaplastic lymphoma kinase 1.

tumours and others negativity. Many studies show complete conflict of results in the same tumour type. An example of this is diffuse large B-cell lymphoma. Here, a variety of studies have shown bcl2 to be either good or bad. It looks as though the resolution is that bcl2 is a poorer prognostic marker in non-germinal centre type tumours, which basically means, in antibody terms, when CD10 and bcl6 are negative (See Table 15.11) (Berglund *et al*., 2005; Biasoli *et al*., 2005).

To Determine Treatment

The most striking (and also in some cases expensive) change in immunocytochemical practice for pathologists has been the introduction of immunocytochemical testing as a precursor to the selection of therapy. This has been most dramatically demonstrated by the use of antibodies against the Her-2 oncogene on cases of breast cancer selected by immunostaining for a high level of expression of it on the tumour cells (Slamon *et al*., 2001; Vogel *et al*., 2002). There are now a number of other examples of immunohistochemical testing to determine or at least strongly guide treatment, as indicated in Table 15.13.

IN SUMMARY: THE PROS AND CONS OF THE ANTIBODY REVOLUTION

Pros

Any list of benefits or deficiencies in diagnostic immunocytochemistry must inevitably be subjective. Nevertheless, it seems to us that the following are definite benefits.

Table 15.13 The use of immunohistochemistry to determine treatment

Disease type	Relevant antibodies	Treatment
Breast cancer	ER, PR	Tamoxifen and analogues
Breast cancer	HER-2	Herceptin
Gastrointestinal stromal tumour	CD117	Imatinib mesylate/ STI571/ Glivec
B-Cell lymphoma	CD20	Rituximab
Chronic lymphocytic leukaemia	ZAP-70, CD38	Consideration of autologous stem cell transplantation
Infections	Identification of any infection agent (see Table 15.11)	Appropriate chemotherapy

1. Improved accuracy of diagnosis.
2. Increased reliability of diagnosis (fewer sleepless nights).
3. More objectivity in classification (e.g. lymphomas).
4. Preservation of morphology when stained (advantage over DNA technology).

Cons

1. Significant increase in laboratory workforce and consumables (see Introduction).
2. Increased temptation to ignore morphology, with consequent deskilling.
3. Increased delay in diagnosis, especially if repeating or ordering more immunostains.
4. Overlap of immunophenotypic profiles causes confusion ('tumours haven't read the textbooks' syndrome).
5. Now almost a legal requirement to do certain immunostains when they are possibly unnecessary, e.g. cyclin D1 on every lymphoma in case a mantle cell is missed.

CONCLUSION

Basically antibodies in the routine laboratory are here to stay. The public and governments are demanding the introduction of modern methods into diagnosis to prevent avoidable errors. Any costs will just need to be subsumed into our practice somehow. It is often suggested that the days of immunostaining in diagnosis are numbered. But will antibody technology ever be superseded by genomic methods, e.g. DNA microarray technology? We think probably not, because antibody technology is relatively cheap, very sensitive and now highly reliable. In addition, it can be readily automated. Finally, the thrust of modern biological research is moving the action towards proteins and not DNA or RNA, and that is exactly where immunostaining is targeted.

Antibodies are here to stay!

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