Letter

Cytokeratin 10/13, 14, 7, 8, and 18 in invasive squamous cell carcinoma and adenocarcinoma of the uterine cervix

Maddox *et al* are to be congratulated for their thorough study of cytokeratin expression in the normal cervix, cervical intraepithelial neoplasia (CIN), and cervical carcinoma.[1] I was involved in a study of cytokeratin intermediate filament protein expression in cervical squamous carcinomas and adenocarcinomas.[2] We did not include details of the intensity of staining or the proportion of cells reacting with the antibodies in our cases and would like to take the opportunity of doing so.

Eleven patients with squamous cell carcinoma (five well differentiated, six moderately differentiated) and 20 patients with adenocarcinoma (14 with well differentiated and six with moderately differentiated tumours) were studied. The patients had been treated at the Jessop Hospital for Women in Sheffield and the presence of mucin elaboration was confirmed using the periodic acid-Schiff/Alcian blue technique with and without diastase digestion. Following routine fixation and processing and microwaving in 0.01 M trisodium citrate for 2 × 5 minutes at high power (600 W microwave), the sections were incubated in primary antibodies to cytokeratins 10/13 (NCL-DEK13), 14 (NCL-LL002), 8 (NCL-CK8), 18 (NCL-CK18), and 7 (NCL-CK7), all supplied by Novocastra and used at a dilution of 1:50. The reaction was detected using a three step peroxidase technique (Vectrastain Elite ABC kit, Vector Laboratories) and diaminobenzidine with H_2O_2. The slides were counterstained with haematoxylin. A positive reaction with each of the antibodies was noted when there was distinct brown staining in the epithelial cells. The percentage of cells reacting in each antibody, assessed semiquantitatively by reviewing all the sections available, and the intensity of the staining as demonstrated by the depth of colour in the cell cytoplasm were noted.

The results are presented in the table. There is no evidence of a statistically significant difference in either the intensity of the reaction or the proportion of cells reacting with any of the antibodies when well differentiated tumours are compared with moderately differentiated tumours in each of the cell types.

Although our findings differ from those of Maddox *et al*, it should be noted that the specificities of the antibodies used to detect cytokeratin 10 in the two studies differed, as the one we used also detects cytokeratin 13. Whereas Maddox *et al* used a quantitative method for assessing the proportion of cells reacting, we used a semiquantitative assessment. Finally, the number of carcinomas studied in each series was small. We are, however, grateful for the opportunity to present data regarding the intensity and proportion of cells reacting in our cases.

M K HEATLEY
Department of Pathology, Royal Liverpool University Hospital, Prescot St, Liverpool L7 8XP, UK

1 Maddox P, Sasieni P, Szarewski A, *et al.* Differential expression of keratins 10, 17, and 19 in normal cervical epithelium, cervical intraepithelial neoplasia, and cervical carcinoma. *J Clin Pathol* 1999;**52**:41–6.
2 Heatley MK, Cork K. Involucrin and cytokeratin intermediate filament protein expression in cervical carcinoma. *Int J Gynaecol Cancer* 1998;**8**:37–40.

Book reviews

Introduction to Oncogenes and Molecular Cancer Medicine. By D W Ross. (£22.50.) Springer-Verlag, 1998. ISBN: 0 387 98392 9.

This book aims to explore oncogenes and the current view of cancer as a molecular disease. The author assumes that the reader has no specialised knowledge and in the first five chapters he builds up a plausible story about the multistep process of carcinogenesis, somewhat spoilt by some inaccuracies in the detail. Perhaps this does not matter in an introduction, but to overcome this the book should be read from cover to cover. Part 1 is devoted to the principles of molecular biology of cells and is generally good. There are useful tables of fact and informative line diagrams. While reading, I noted facts that I thought had been left out, but in most cases these were covered in later chapters and the footnotes tell you what is to follow. At first this was annoying but in the end it does give one a good idea of the complexities of the subject. The second part is devoted to clinical examples of molecular oncology, starting with molecular diagnostics. Surprisingly there is no mention of comparative genomic hybridisation (CGH), a technique which is used with increasing frequency and has become an informative procedure, especially if similar types of tumour are compared. Chapters on leukaemia and lymphoma, colon cancer, cervical cancer, and breast cancers follow. Again the diagrams used to explain the text are good but the photomicrographs are of poor quality, particularly for a book which costs £22.50.

DIANA M BARNES

Atherosclerosis. Pathology of The Vasculature in Live Patients. Edited by J M Isner and M Kearney. (£60.00.) W B Saunders, 1999. ISBN 0-7020-1927-5.

In this book, studies of an impressive tissue bank of specimens obtained by directional atherectomy are described. Indeed, the amount and quality of these "in vivo" atherosclerotic specimens are unique.

In most chapters specimens from coronary, saphenous graft, peripheral, and dialysis fistula lesions are compared. Furthermore, primary, restenosis, and multiple restenosis

Reaction pattern of anticytokeratin antibodies in squamous cell carcinoma and adenocarcinoma of the cervix

Percentage of cells reacting with each antibody

	Cytokeratin 1/10						*Cytokeratin 14*						*Cytokeratin 7*						*Cytokeratin 8*						*Cytokeratin 18*					
	0	*1—25*	*26—50*	*51—75*	*76—99*	*100*	*0*	*1—25*	*26—50*	*51—75*	*76—99*	*100*	*0*	*1—25*	*26—50*	*51—75*	*76—99*	*100*	*0*	*1—25*	*26—50*	*51—75*	*76—99*	*100*	*0*	*1—25*	*26—50*	*51—75*	*76—99*	*100*
Squamous cell carcinoma																														
Well differentiated	0	1	0	0	1	1	1	1	1	0	1	0	1	2	0	0	1	0	2	0	1	0	0	0	1	1	2	0	1	0
Moderately differentiated	2	0	0	0	1	1	1	3	0	0	1	0	2	0	0	2	1	0	2	0	0	0	1	1	2	1	1	0	1	0
Adenocarcinoma																														
Well differentiated	2	1	1	0	2	7	11	1	0	0	1	0	4	0	4	3	1	1	1	4	1	0	4	0	0	1	1	2	7	2
Moderately differentiated	2	0	2	0	1	1	4	2	0	0	0	0	1	3	0	1	1	0	1	2	0	1	1	0	0	2	0	0	3	0

Intensity of intermediate filament expression in squamous cell carcinoma and adenocarcinoma of the cervix

	Cytokeratin 1/10					*Cytokeratin 14*					*Cytokeratin 7*					*Cytokeratin 8*					*Cytokeratin 18*					
	0	*II*	*III*	*IV*	*V*	*0*	*II*	*III*	*IV*	*V*	*0*	*II*	*III*	*IV*	*V*	*0*	*II*	*III*	*IV*	*V*	*0*	*II*	*III*	*IV*	*V*	*Total*
Squamous cell carcinoma																										
Well differentiated	3	0	0	1	1	2	0	0	1	2	1	1	1	0	1	4	0	0	0	1	1	0	3	0	1	5
Moderately differentiated	3	0	1	2	0	2	0	0	1	3	3	0	0	1	2	4	0	1	1	0	3	1	0	1	1	6
Adenocarcinoma																										
Well differentiated	3	0	3	6	2	12	0	0	0	2	5	0	0	2	7	5	0	2	1	6	1	0	0	2	11	14
Moderately differentiated	2	0	2	0	2	4	0	0	0	2	1	0	0	2	3	2	0	0	1	3	1	0	1	1	3	6

lesions are described. Besides this, more recent development in atherosclerosis, such as apoptosis, gene expression, cell culture, and so on, are also studied in separate chapters. The chapters are based on studies by Isner and Kearney's group. Parts of these studies have been published recently in high ranking journals such as *Circulation*, *Journal of Clinical Investigation*, and the *American Journal of Pathology*.

Each chapter is well illustrated with coloured figures. However, most of the chapters lack a concise and clear summary. Furthermore, a general discussion at the end, where the results of all the chapters are combined to give a more general idea of the pathogenesis of atherosclerosis through this unique in vivo material is also missing.

This implies that it is difficult to see where the appeal of this book lies. I believe it is not particularly suitable for the general histopathologist. It is of more interest for pathologists, especially those interested in vascular pathology, or for the researcher in this field.

Personally, I enjoyed reading this well illustrated book.

H W M NIESSEN

Confocal Microscopy: Methods and Protocols. Edited by S W Paddock. (£99.50.) The Humana Press, 1998. ISBN 0 896 03526 3.

Confocal microscopy allows the imaging of discrete regions of tissues virtually free of out of focus fluorescence and is now used in many areas of biological research. This book is volume 122 in the long running *Methods in molecular biology* series and is written with the aim of leading the researcher using confocal techniques from the bench top, through the imaging process, to the journal page. It is, as its title makes clear, very much a book about methods and protocols.

After some introductory chapters covering practical considerations for collecting images and fluorescent probes, the bulk of the book is made up of specific methods for most of the commonly used model organisms: worms, sea urchins, flies, plants, yeast, frogs, and zebrafish. The protocols have been chosen with the novice user of a laser scanning confocal microscope in mind and include considerable detail. There follow chapters on live cell analysis including methods of imaging various ions and green fluorescent protein. The final chapters deal with the analysis and presentation of confocal images. Many black and white photographs and diagrams accompany the text and all seem appropriate and clear. There are four stunning colour plates (*Drosophilia* embryos, chondrocytes, intracellular calcium dynamics, and some three dimensional illustrations).

While most pathologists are unlikely to be using the model organisms specifically described, the protocols are readily adaptable, with a little bit of ingenuity, to human or other animal tissues. For pathologists who have access to a (usually multiuser) confocal microscope facility, this book would be a good buy, crammed full as it is with useful tips and hints.

J R SALISBURY

CD-ROM review

Atlas of Pathology: Urological Pathology. By A Viellefond, H Bastien, C Billerey, N Berger, R Bouvier, B Cochand-Priollet, M-C Dauge-Geffroy, and B Fontanière. (£116.04.) Springer, 1999. ISBN 3 540 14657 1.

As this newest electronic medium lends itself so readily to storing nearly uncountable numbers of photographs, it is hardly surprising that a bevy of pathology atlases has recently appeared on CD-ROM. Unfortunately, the photographic quality still compares unfavourably with that of the printed page; for this the authors bear no blame, of course, and will benefit from improved technology in the near future. The editing, however, is their purview: the reproductions of only a few gross specimens can be tolerated, but the histological photographs, especially those at low magnification, are of no use whatsoever. The quality of the schematic diagrams and written slides is exceedingly poor, having been subjected to only the most primitive techniques. It is hard to believe that such renowned publishers did not have more sophisticated techniques available.

The authors themselves also bear responsibility for the arbitrary use of terminology, as well as the chaotic contents. I made every effort to comprehend the principles on which the individual chapters were organised, but in vain. The separate topics, for example prostate cancer, start with microphotographs, then somewhere later there appear some diagrams, a bit later gross surgical specimens, accompanied by more microphotographs. Pictures representing high grade PIN are captioned "Dysplasia or glandular hyperplasia with severe atypia"—highly misleading terminology, quite unacceptable in international pathology. The 150 pictures of testicular tumours are not accompanied by any classifications or explanatory notes. Under the heading "transplanted kidney," only one picture depicting a "malakoplakia" of the testis could be found—an utterly insignificant complication observed only once by one of the authors of the CD-ROM. Histology entitled "mild hypospermatogenesis" displays a severe maturation arrest. One could go on and on listing further examples of sloppy terminology combined with pictures of poor quality. Needless to say, the spelling errors (the entry "Fuhrman"—grading of renal cancer—has changed to a German looking "Führman") only round off the disastrous impression made by the work.

G MIKUZ

Notices

Postgraduate Course on Gynecologic Pathology, San Diego, 15–17 January 2000

At the San Diego Hilton Beach and Tennis Resort, San Diego, California, USA.
Presented by the department of pathology, Massachusetts General hospital, Harvard Medical School under the direction of Dr Robert H Young.
For further information, contact: Department of Continuing Education, Harvard Medical School, 25 Shattock St, Boston, MA 02115, USA; tel +1 617 432 1525; fax +1 617 432 1562; email: hms-cme@ hms.harvard.edu

Third International Surgical Pathology Symposium, Liverpool, 14–16 June 2000

Modern Molecular Diagnostics

An introduction to basic molecular biological techniques in diagnostic surgical pathology. Evening workshops will include an introduction to gene-chip technology and gene amplification techniques.

Further details from: Professor C S Foster, Department of Cellular and Molecular Pathology, University of Liverpool, Duncan Building, Daulby St, Liverpool L69 3GA, UK; tel +44 (0)151 706 4480; fax +44 (0)151 706 5883; email: csfoster@liv.ac.uk

Correction

In the paper by Jacobs *et al* in the July issue (Reliable high risk HPV DNA testing by polymerase chain reaction: an intermethod and intramethod comparison; volume 52, pp 498–503), the following acknowledgement should be included:

Financial support was given by Europe against Cancer SOC 96 201748.

Instructions for Authors

Papers for publication should be sent to the Editor, *Journal of Clinical Pathology*, BMA House, Tavistock Square, London WC1H 9JR (tel: 0171 383 6209/6154; fax: 0171 383 6668; email: ndavies@bmjgroup.com).
Receipt of manuscripts will be acknowledged by the editorial office.

Submission of a paper will be held to imply that it contains original work not being offered elsewhere or published previously. Manuscripts should be prepared in accordance with the Vancouver style.[1] The Editor retains the right to shorten the article or make changes to conform with style and to improve clarity. All authors must sign the copyright form after acceptance.

Failure to adhere to any of these instructions may result in delay in processing the manuscript and it may be returned to the authors for correction before being submitted to a referee.

General

- Authors must submit four copies of the original manuscript typed in double line spacing. The journal is now produced electronically and revised manuscripts should be submitted as printed copy and on disk. A guide to submitting an article on disk will be sent when requesting a revision or on notification of an acceptance. Authors should not submit the original paper on disk.
- The names of the authors, with initials, should be followed by the name of the institution where the work was carried out. An indication of the position held by each author should be given in an accompanying letter to the Editor, and manuscripts should bear the name of one author to whom correspondence should be addressed. If available, a fax number and an email address should be supplied.
- Identifying information should not be published in written descriptions, photographs, or pedigrees unless the information is essential for scientific purposes and the patient (or parent or guardian) gives written informed consent for publication; but patient data should never be altered or falsified in an attempt to attain anonymity. Informed consent should be obtained if there is any doubt. Masking the eyes in photographs of patients is inadequate protection of anonymity (for the full statement see the *BMJ*[2]).
- Authors should include the names and addresses of four experts whom the authors consider suitable to peer review their work.
- When submitting original manuscripts authors should send a copy of any of their other papers on a similar subject to assure the editors that there is no risk of duplicate publication.
- If requested, authors should produce the data upon which the manuscript is based for examination by the Editor.
- The number of authors should be kept to a minimum and should include only those who have made a contribution to the research: justification should be made for more than five authors. Acknowledgments should be limited to workers whose courtesy or assistance has extended beyond their paid work, and to supporting organisations.
- Sponsors of research must be declared.
- Authors should provide up to four keywords/phrases for the index.
- All measurements must be in SI units apart from blood pressure measurements, which should be in mm Hg, and drugs in metric units.
- Abbreviations should be used rarely and should be preceded by the words in full before the first appearance.
- In the statistical analysis of data 95% confidence intervals should be used wherever appropriate.
- Any article may be submitted to outside peer review and for statistical assessment.
- No free offprints will be provided; reprints may be ordered when the proof is returned.

Original articles

- Papers should be no more than 2000 words long and should report original research of relevance to the understanding and practice of clinical pathology. They should be written in the standard form: abstract; introduction; methods; results; and discussion.
- The journal uses a structured form of abstract in the interests of clarity. This should be short (no more than 250 words) and include four headings: *Aims*—the main purpose of the study; *Methods*—what was done, and with what material; *Results*—the most important results illustrated by numerical data not p values; and *Conclusions*—the implications and relevance of the results.

Leaders/Editorials

- Leaders and Editorials are published by editorial invitation; unsolicited reviews or commentaries are unlikely to be accepted, though the Editor is always pleased to receive suggestions.

Short reports

- Single case reports of outstanding interest or clinical relevance, short technical notes, and brief investigative studies are welcomed and usually published in the form of a Short/Technical report.
- Length must not exceed 1500 words, including an unstructured abstract of less than 150 words, up to two figures or tables (or one of each) and up to 10 references. If more illustrations are required the text must be reduced accordingly.

Letters

- Letters must be typed in double line spacing, should normally be no more than 500 words, have no more than five references, and must be signed by all authors. Two copies should be provided.

Tables and illustrations

Tables should be presented separately in double line spacing without ruled lines; when presented on disk they should be in a separate file from the text.

- Letters and other marks which are to appear on the face of a photomicrograph should be made on a photocopy: they will be added in the Journal style in the editorial office when the manuscript is accepted.
- Legends for illustrations should be typed with double spacing on a separate sheet. The staining technique used should be stated. Magnifications should be given for electron micrographs but not for light micrographs except in cases where this is important.
- Photographs and photomicrographs should be on glossy paper for half tone reproduction. The printing process requires that prints are unmounted and unbacked, and of high quality, with full tonal scale. Illustrations that will not reproduce well will be returned and this may delay publication. Areas in which tissue does not appear ("background") should be as near white as possible. Three sets of prints must be supplied with each manuscript. Only salient features should be included to preserve detail.
- Colour reproduction of figures in papers is encouraged and is heavily subsidised by the Journal. Advice on costs and material to be submitted for colour work should be sought from the editorial office. The journal can accept colour images as TIFF files in the following media: zipped or unzipped files on floppy disks, compact disks, or optical disks. A hard copy of the image should be provided.
- If any tables or illustrations submitted have been published elsewhere, written consent to republication should be obtained by the author from the copyright holder (usually the publisher) and the authors. A copy of the letter giving consent must be included.

Descriptions of laboratory methods

- When a manufacturer's method is used in a study with a particular item of equipment or kit of reagents, the source of this method and reference to the scientific literature on which it was based should be given. Authors might consider it courteous to inform manufacturers that an article assessing their product has been submitted for publication.
- For quantitative methods, information on the sensitivity, precision, and accuracy in the hands of the authors should always be provided. When a well recognised method is used, these requirements could be met simply by providing the references to the methodology and discussing the performance in a recognised current quality assurance scheme. Modifications to methods that have not been previously published should be detailed in the text and supported by evidence of their efficacy.
- It is useful to indicate, either from personal observations or by reference, the working range of an assay and the normal reference range when it is used on samples from humans. When information is expressed as mean ± 2SD, the distribution of the range (normal, skew, or logarithmic) should be stated.

References

- References must be numbered in the order they appear in the text and include all information (Vancouver style; references with more than three authors should give only the first three followed by *et al*):

1 Fletcher CDM, McKee H. Sarcomas - a clinicopathological guide with particular reference to cutaneous manifestations. I. Dermatofibrosarcoma protuberans, malignant fibrous histiocytoma and the epithelial sarcoma of Enzinger. *Clin Exp Dermatol* 1984;**9:**451–65.
2 Washington JA. Conventional approaches to blood culture. In: Washington JA, ed. *The detection of septicemia*. West Palm Beach, Florida: CRP Press, 1978:41–87.

- References in the text should be identified by arabic numerals in brackets—for example, [1] [2].
- Information from manuscripts not yet accepted, or personal communications may be cited only in the text and not included in the references. References are not checked by us; authors must verify references against the original documents before submitting the article.

1 International Committee of Medical Journal Editors. Uniform requirements for manuscripts submitted to biomedical journals. *BMJ* 1991;**302:** 338–41.
2 International Committee of Medical Journal Editors. Protection of patients' rights to privacy. *BMJ* 1995;**311:**1272.

Manuscript checklist:
- Is there an abstract?
- Are the abbreviations spelt out?
- Are the measurements in SI units?
- Are the references in Vancouver style?

Revised January 1999

Best Practice articles (formerly "Broadsheets") prepared by the Association of Clinical Pathologists

Just published

154 *Helicobacter pylori* 1999 CAM MCNULTY, JI WYATT (with correction in June issue)

Recent Publications

153 The laboratory investigation of vaginal discharge 1998 KF MACSWEEN, GL RIDGWAY

152 Clinical implications of plasma homocysteine measurement in cardiovascular disease 1998 RA STILL, IFW MCDOWELL

Other Best Practice articles are still available for purchase

151 Investigation of dyslipidaemias 1997 AF WINDER, W RICHMOND, DT VALLANCE

150 Antenatal serological testing and prevention of haemolytic disease of the newborn 1997 JKM DUGUID

149 Serological diagnosis of gluten sensitive enteropathy 1996 DJ UNSWORTH

148 Laboratory diagnosis of malaria 1996 DC WARHURST, JE WILLIAMS

147 Mycological techniques 1996 KG DAVEY, CK CAMPBELL, DW WARNOCK

146 Macroscopic examination of prostatic specimens 1995 P HARNDEN, MC PARKINSON

145 Investigation of patients with autoimmune haemolytic anaemia and provision of blood for transfusion 1995 RJ SOKOL, DJ BOOKER, R STAMPS

144 The investigation of hypercalcaemia 1994 PL SELBY, PH ADAMS

143 Detection of autoantibodies to neutrophil cytoplasmic antigens 1994 RJ LOCK

142 Measurement of carbon monoxide and cyanide in blood 1993 RW MAYES

141 Role of endocrine biochemistry laboratories in the investigation of infertility 1993 GH BEASTALL

140 Techniques in pulmonary cytopathology 1993 JA YOUNG

139 Post mortem techniques in the evaluation of neck injury 1993 P VANEZIS

138 Gross examination of uterine specimens 1993 J SCURRY, K PATEL, M WELLS

137 Obtaining samples at post mortem examination for toxicological and biochemical analyses 1993 ARW FORREST

136 Detection and importance of anticardiolipin antibodies 1993 MA KHAMASHTA, GRV HUGHES

Earlier Broadsheets may still be available from the author. A full list can be obtained from the Publications Secretary, Association of Clinical Pathologists, 189 Dyke Road, Hove, East Sussex BN3 1TL.

Prices

INLAND One copy, £2.50; 2–10 copies (of any one broadsheet or reprint), £2.00 each; 11–100 copies (of any one), £1.75 each; 101 plus copies (of any one), price to be agreed; authors (over 50 free copies), £1.25 each.
OVERSEAS One copy, $6.75; 2–10 copies (of any one broadsheet or reprint), $5.25; 11–100 copies (of any one), $3.75; 101 plus copies (of any one), price to be agreed.
Authors $2.25. Prices include postage but air mail will be charged extra. Trade discount 10%. All orders (and all changes of address of regular subscribers) should be sent to the Publishing Manager • *Journal of Clinical Pathology*, BMJ Publishing Group, BMA House, Tavistock Square, London WC1H 9JR.