Detection of Epstein-Barr virus in archival Hodgkin's disease specimens

The Epstein-Barr virus (EBV) is associated with several malignancies, including endemic Burkitt's lymphoma, undifferentiated nasopharyngeal carcinoma, post-transplant lymphoproliferative disease, and Hodgkin's disease. The "gold standard" for the detection of EBV infection in clinical tissues is RNA in situ hybridisation that targets the abundantly produced Epstein-Barr virus early RNAs (EBERs). This approach is effective in the detection of latent EBV infection in routinely processed, paraffin wax embedded histological material, and has been widely used to analyse the association of EBV with a variety of malignant and non-malignant diseases. In addition, a range of monoclonal antibodies directed against latent EBV proteins has enabled viral gene expression to be investigated in EBV associated diseases. For example, in Hodgkin's disease, the latent membrane protein 1 (LMP1) is consistently expressed in EBV associated cases and can be detected using the CS1-4 monoclonal antibody reagent. Although these approaches are effective on optimally processed histological material, little is known about their ability to detect EBV in tissues stored for many years, or those that may have been processed before the introduction of automated paraffin wax embedding. Therefore, we wished to investigate whether these methods could be used to detect EBV in Hodgkin's disease specimens stored for 50 years or more. In addition, these specimens were processed before the advent of improved tissue embedding procedures.

Paraffin wax embedded tissue blocks were retrieved from 14 patients diagnosed with Hodgkin's disease between 1943 and 1952. Specimens were reviewed by haematoxylin and eosin staining in all cases. All histologically confirmed Hodgkin's disease specimens were subjected to EBER in situ hybridisation and immunohistochemistry for LMP1, as described previously. LMP1 was detected in five of 14 Hodgkin's disease specimens, where staining was confined to the cytoplasm of Hodgkin-Reed-Sternberg cells (fig 1). Four of these five cases were also positive for EBER in situ hybridisation (fig 2).

These results indicate that archival specimens stored for periods in excess of 50 years, or those that were subject to less than optimal tissue processing, are still viable for the detection of EBV. These approaches will allow the collection of valuable comparative information about whether EBV prevalence in certain tumour types, such as Hodgkin's disease, has altered over time.

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All books are idiosyncratic, reflecting their authors in some way or other. This is a multi-author book, so its idiosyncrasies mirror that fact. It is intended to be a working book, not bedtime reading, and it does not fail the latter criterion. It's not something for the wee small hours. Whether it's something for the daily round, the common task, is another matter. It all depends. Depends on the task, and upon you. So what is it? It starts from a narrow view of molecular biology. Molecular biology is claimed to be nothing more than the science of DNA and RNA, which would probably come as quite a surprise to the founders of the Journal of Molecular Biology. In other words, proteins are largely ignored. However, that a working book is focused is no bad thing. Too wide a range and it would be unmanageable in a laboratory. So what is it? It's a collection of recipes and tips on how to manipulate DNA and RNA with brief introductions to the technique being considered. It covers all the basic methods that a "molecular biologist" considers important: how to extract nucleic acids, how to modify and analyse them, how to clone DNA fragments, how to make and use various sorts of DNA libraries. In addition, it covers some more specialist areas, such as chromosome analysis by fluorescent hybridisation and transgenic animals. So is it useful? Well, yes and no! I haven't tried the recipes as such in detail, but they are along the same lines as other similar books, and I'm sure they work. So is it useful? Yes, and no! It depends on what you want to do and on you. If you want to do everything from scratch, much of this book would be useful. But in my experience not many people want to do that. Commercial companies provide so many kits for so many methods with such good instructions that one hardly bothers to do it any other way. The easy ways have enormous attraction. When you can't do a gene kit, why screen a library? For the more specialist topics, I would hate to think that this was all I had.

So why did I mention idiosyncrasies? Well, there are omissions; quantitative polymerase

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**Figure 1** Immunohistochemical staining for latent membrane protein 1 in an archival Hodgkin's disease specimen, with staining confined to the cytoplasm of Hodgkin-Reed-Sternberg cells.

**Figure 2** In situ hybridisation for Epstein-Barr virus early RNA. Positive result seen in an archival Hodgkin's disease specimen.
A high throughput assay for hepatitis C virus (HCV) helicase activity is presented that might be of interest to test for anti-HCV compounds; however, no practical examples for antivirals used against HCV are given. For hepatitis B virus (HBV) several cell bound assays (Hep AD38 assay, and cell line 2.2.15 assay) are described, the importance of which cannot be overestimated. Systems have been found that allow limited replication of HBVs in vitro, in which compounds inhibiting different steps of the HBV replication cycle (be it at the transcription, DNA synthesis, or other steps) can be investigated. Some of the assays are still very research orientated, whereas others lend themselves to application in specially equipped clinical laboratories. A high throughput assay for hepatitis C virus (HCV) helicase activity is presented that might be of interest to test for anti-HCV compounds; however, no practical examples for antivirals used against HCV are given. 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pointing to inflammation and proteinase imbalance as key features of its development. The editor’s prominent role in this field clearly shows in the selection of six interesting and authoritative chapters about mouse models of emphysema, serine proteinases, elastase inhibitors, and connective tissue genes. I found the chapter about the regulation of neutrophil proteinases especially informative. Although at least three or four years have elapsed since the chapters in this section were written, judging from the reference lists, they still hold up well in 2000. Similarly favourable comments could probably be made about the other chapters of the book, considering the distinguished authors, but I am less familiar with these other topics than with emphysema.

Choices are inevitable in selecting topics for a book on such a broad field as the molecular biology of the lung from the perspective of disease. Clearly, this book includes many of the important areas, but one wonders why interstitial lung disease with its intriguing features of fibrosis and inflammation was not included. Also, I would have enjoyed an introduction by the editor so that I could read his overall perspective on the field of lung cell and molecular biology, and his rationale for the topics he chose to include in the book.

ROBERT M SENIOR

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**Notice**

**Therapeutic Applications of Leucocyte Filtration**

**Hammersmith Hospital, Imperial College, London, UK**

7 July 2000

This prestigious meeting has been organised by Professor K Taylor and Dr T Gourlay who have invited a guest panel of international speakers. Additional areas include leucocyte filtration of salvaged blood; total leucocyte control for cardiovascular surgery; and new therapeutic applications for use in sepsis patients in ICU and for patients undergoing angioplasty, etc. Speakers will include Gr W O'Neill, Professor KJ Oldhafer, Dr M Cross, Dr G Matheis, Professor S Homer-Vanniasinkam, Dr J Parker Gott, BS Allen, Dr A Fabbri, Dr L van der Waterings, Dr KA Brown, and Dr D Teacher.

Abstracts are invited for poster presentations and prizes will be awarded to the best three.

For further details, abstract forms, and registration forms please contact: Jean Bryant, Wolfson Centre, Imperial College of Science, Technology and Medicine, Hammersmith Hospital, Du Cane Road, London W12 ONN, UK; tel +44 (0) 208 383 3117; fax: +44 (0) 208 383 2428; email: wcc@rpms.ac.uk