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Book Review

Techniques in Apoptosis: A User's Guide

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Edited by T.G. Cotter and S.J. Martin. (Portland Press, London, 1996)

Over the last five years, the field of programmed cell death (PCD) has expanded enormously. New methods to study apoptotic cells have been developed and older ones refined, leaving newcomers to the field without much practical guidance unless he consults colleagues or burrows through the literature. Now, however, there is an easier way to get started: a new methods book, 'Techniques in Apoptosis: a user's guide', edited by Tom Cotter and Seamus Martin, serves as a practical guide to apoptosis methods. Although targeted mainly to those researchers new to the field, it also provides a valuable reference for so-called 'Death Professionals'.

The style and format of 'Techniques in Apoptosis' is somewhat between that of one of the popular molecular biology protocol books, such as Sambrook *et al*. (Molecular Cloning: A Laboratory Manual), and an issue of Methods in Enzymology. The first half of the book is divided into chapters each devoted to a specific type of measurement or technique for the characterisation of apoptotic cells: morphology, transglutaminase assay, DNA fragmentation, flow cytometry, annexin-V staining, preparation of *in vitro* extracts, measuring calcium flux, and phagocytosis. The latter half of the book is concerned with specific PCD systems: cytotoxic lymphocytes, thymus cultures, *Drosophila*, transgenic mice, intestinal crypts, and plants. This arrangement is logical and useful, and most of the specialised chapters are by experts in their respective fields. References at the end of each chapter allow users to explore the background and application of a particular method in more detail. Unfortunately, the format also makes it difficult to jump quickly to specific protocols, except via the index: one has to repeatedly flick between the main table of contents and the header page for a specific chapter and then find the method desired, which is in some cases difficult to do.

Not all of the chapters are of the same standard, although, the majority of chapters are very good and provide a healthy balance of both practical direction and theoretical perspective: many of the specialized chapters, and those on phagocytosis, annexin-V staining, calcium fluxes, and flow cytometry are excellent. From this book one can learn, for example, how to kill cells with cytotoxic lymphocytes, measure changes in intracellular calcium concentration, prepare macrophages, stain *Drosophila* embryos, and prepare carrot cells for culture. For specialists already familiar with such techniques there are even some helpful variations, such as the use of calcein-AM instead of ^{51}Cr-release in target-cell killing assays. Although the overall structure is consistent between chapters, the level of detail, background explanation, and discussion vaires considerably in both quantity and quality: some chapters read like a speculative review, lacking technical perspective or practical advice; other describe methods without providing sufficient explanation of the rationale behind them.

There are also other inconsistencies. For example, recipes in some chapters list ingredients in molar units, whereas others list them in weights (cataloging both would be most useful). Splitting the two DNA-nick-end-labelling techniques (TUNEL and ISEL) and burying them in two different chapters is also frustrating and precludes a ready comparison of the two methods. In this particular case there is serious confusion between the two methods in the chapter on apoptosis in intestinal crypts. Here, the ISEL (*in situ* nick-end-labelling) technique, which uses Klenow-fragment DNA-polymerases to label DNA ends, is described, but the discussion refers to the terminal deoxynucleotidyl transferase (TdT) enzyme, used in the TUNEL method. Although there is a useful discussion of some of the pitfalls of these *in situ* methods, there is a noticable absence of any attempt to compare and contrast their relative merits. The chapter on transgenic mouse methods, although an excellent primer on the technology, seems somewhat unnecessary in a book aimed primarily at cell-death novices. Anyone contemplating the generation of transgenic mouse lines should probably consult more detailed manuals specifically written on the subject and consult experts.

Not surprisingly for a book describing methods in a new and rapidly changing field, there are some noticeable gaps: for example, some widely used assays, such as MTT hydrolysis, LDH release and histone release, are not included at all. There is also little general discussion on the ICE-family proteases or their inhibitors or substrates. Specialized sections on specific organs (thymus, intestine) are included, but there is no mention of apoptosis in the nervous system. A conspicuous weakness of this book is the paucity of figures and many of the figures that are included are poor or are badly reproduced. This is especially noticeable where they are most needed, such as in the first chapter on morphological methods. For a manual aimed primarily at those unfamiliar with cell death, many more illustrations of apoptotic cells viewed by different methods, and plenty of explanatory and concept diagrams would have been extremely useful.

Nevertheless, this book provides a good compilation and explanation of many of the methods used by

researchers in the broad field of apoptosis. It is small enough to fit on a laboratory bench, and its spiral binding allows it to open flat, though the cover is not firmly attached, so it may not hold up well to heavy use. Most importantly, it is – despite its inconsistencies – useful: it has been in our laboratory for only a few months and has already provided some good protocols that are difficult to extract from the literature. However, at £75 per copy, the price may be too steep for anyone other than committed cell-death aficionados.