Immunohistochemical detection of Ki67 antigen in formalin fixed, paraffin wax embedded sections

We read with interest the recent paper by Torp et al. reporting the detection of the Ki67 antigen in formalin fixed, paraffin wax embedded glioblastoma tissue sections using a monoclonal antibody directed against Ki67, purchased from Dako, Glostrup, Denmark. The authors did not recommend the use of this antibody on paraffin wax sections. We have also tested the antibody on sections from routine blocks from human gingiva, tonsils and lymph nodes using different time schedules for microwave pretreatment and application of the primary antibody. Two crucial procedures for detecting Ki67 antigen successfully were identified. Firstly, slides should be pretreated in a microwave oven in citrate buffer (pH 6.0) for over 20 minutes (4 x 5 minutes). Secondly, Ki67 antibody (diluted 1 in 100–200) should be incubated overnight at 4°C to produce strong staining. Torp et al. treated sections by microwaving for only 10 minutes (2 x 5 minutes) and incubated the Ki67 antibody for only one hour. Under these conditions, we could not achieve satisfactory staining on sections of the three tissues described above. Bankfalvi et al. have reported that almost identical results were found with the Ki67 and MIB1 antibodies. In their method, microwave heating was carried out for 35 minutes (7 x 5 minutes) and the Ki67 monoclonal antibody was applied overnight.

However, it should be noted that microwave treatment has certain drawbacks compared with autoclave treatment. Human gingival tissue sections were sometimes disrupted and detached from slides during microwave heating, presumably because of the violent boiling procedure, whereas tonsil and lymph node sections were resistant to this procedure even after one hour in our laboratory. Furthermore, another disadvantage of the Ki67 antibody was background staining on epithelium and various cell types except polymorphs, although these were easily distinguishable from genuinely reactive structures. Thus, Ki67 monoclonal antibody is as useful as MIB1 even in formalin fixed, paraffin wax sections, if the tissue sections to be examined can withstand more than 20 minutes in a microwave oven.

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This softback edition sets out to offer a practical introduction to undergraduates, postgraduate scientists and technicians in the field of electrophoretic protein separation methods. Overall, I found this book to be clearly written, well laid out, amply illustrated, and well referenced. I found the concept of summarising transcription, translation and protein structure in two and a half pages of text quite inspired, in that it summarised a lot of information in a little space. The book concentrates primarily on polycrylamide gel electrophoresis (PAGE) and gives little space to any other forms of protein electrophoresis.

R A STOCKLEY
