

## DETERMINATION OF PLOIDY IN COLON TUMOURS USING A VIDEO IMAGE ANALYSIS SYSTEM

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There is evidence that abnormalities of nuclear DNA content (ploidy) may be related to biological behaviour in colonic tumours. Methods have been devised to recover cells from paraffin embedded tissues.

In brief, the blocks are dewaxed in xylol and cells separated using a combination of mechanical and enzyme disaggregation. "Cytospin" preparations are made and stained by the Feulgen method for DNA in a staining bath. The nuclear DNA content is then measured using the video imaging system devised by Jarvis (1985).

We have examined cells from 32 cancers, and 16 tubulo-villous adenomas (TVA's). As controls cells from 10 morphologically normal large bowel mucosas, taken from cases of non-neoplastic disease were examined.

15 of the cases were also examined using the FACS IV cell sorter/analyser, the cells being stained with the fluorescence dye DAPI, to compare this established methodology with the results for DNA histograms obtained by the video image system.

The DNA content histograms for both systems were similar. Furthermore, only 300 randomly selected epithelial cells were examined using the video system, whereas at least  $2 \times 10^4$  were needed for FACS analysis.

All the normals and all but one of the TVA's showed a diploid pattern.

13 cancers showed a diploid profile whilst 19 cancers and one TVA had hyperdiploid profiles with large peaks at 4N and 8N. The histograms suggest that a substantial proportion of cancers contain a clone of cells with a basic tetraploid nucleus, which is capable of division.

The ploidy data is currently being compared with more conventional histological indices of behaviour as assessed by an independent observer, using discriminant function analysis. For TVA's the degree of dysplasia is noted; mild, moderate or severe, and for cancers the cases are categorised into the prognostic subgroups A to D of the Australian Colon Cancer Prognosis Study (ACPS)>

The single TVA noted so far with hyperdiploid features on review showed an area of microinvasive carcinoma. It is suggested that a hyperdiploid profile is a characteristic of some colon cancers and can be used as one marker of malignant change in tubulovillous adenomas.

## REFERENCE

Jarvis LR. A microcomputer system for video image analysis and diagnostic microdensitometry. *Analyt Quant Cytol Histol* 1985 (in press)