Comparison of Image Cytometry and Flow cytometry for detection of DNA ploidy abnormalities in Barrett’s oesophagus

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Introduction: DNA ploidy abnormalities (aneuploidy/tetraploidy) are strong predictors of future cancer risk in Barrett’s oesophagus (BE) as measured by flow cytometry (FC). Diagnostic accuracy varies between laboratories, perhaps explained by technical and sampling errors. Image cytometry (IC) is an optical technique allowing visualisation of abnormal nuclei and can be undertaken on archival tissue. Aim: To compare the accuracy of IC versus FC to detect DNA ploidy abnormalities. Methods: 48 paraffin embedded blocks from 35 patients were retrieved. 16 endoscopic mucosal resection specimens showed high grade dysplasia or intramucosal cancer. Of 32 oesophagectomy specimens, 25 showed invasive adenocarcinoma and 7 blocks were cancer free margins used as controls. 40 µm sections were cut and deparaffinised with xylene, rehydrated through graduated ethanol, and denatured by protease XXIV. After filtration the nuclear suspension was separated for IC analysis at UCL and FC at UW. Results: 44 samples were analysed. 91% (40/44) were classified identically. 4 were not concordant; 2 near diploid aneuploid at UW/diploid at UCL; 1 aneuploid at UW/tetraploid at UCL; 1 diploid UW/aneuploid UCL. All controls were diploid at both centres. Of the matched cases 67% were aneuploid, 9% tetraploid, 24% diploid. There were no significant differences in the DNA index of aneuploid peaks. Conclusion: These data demonstrate IC is highly accurate for the detection of DNA ploidy abnormalities against the current gold standard of FC.