Article:
Goldsworthy, M, Tinkler-Hundal, E, Maisey, T et al. (2016) 5-Aminolevulinic acid-mediated fluorescence diagnosis of colon cancer: A histopathological comparison of fluorescent and non-fluorescent tumours. European Journal of Surgical Oncology, 42 (11). S240-S240. ISSN: 0748-7983

https://doi.org/10.1016/j.ejso.2016.07.091

5-aminolevulinic acid-mediated fluorescence diagnosis of colon cancer: a histopathological comparison of fluorescent and non-fluorescent tumours

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Background

5-aminolevulinic acid (5-ALA) selectively accumulates in cancer cells and is metabolised in the mitochondria to the fluorophore protoporphyrin IX. The GLiSten trial evaluated 5-ALA as a fluorescent probe for intraoperative detection of colon cancer and lymph node metastases. Only 13 of 40 cases showed fluorescence, suggesting a fundamental difference between fluorescent and non-fluorescent cancers. The aim of this study was to investigate whether differences in fluorescence were due to tumour cellularity, in particular T cell infiltration, which may be of prognostic significance.

Methods

Primary tumour tissue was available from 30 patients. The density of tumour cells, vascularity and stromal compartment size were quantified using digitally scanned tissue sections stained with haematoxylin and eosin. A set of 300 random points was superimposed onto each tumour image. The structure indicated by each point was then categorised as tumour, stroma, vessel or other. The proportions of tumour and vessel points gave the tumour cell density and vessel density respectively. The relative size of the stromal compartment was given by the tumour to stroma ratio. A tissue section was also stained for the T cell marker CD3 by immunohistochemistry. Percentage staining was quantified in three high-density fields using the Nuance imaging system.

Results

We were unable to detect any difference between fluorescent and non-fluorescent cancers in terms of tumour cell density (difference in means 3.7%; P=0.452), vessel density (difference in means 0.17%; P=0.684), tumour-stroma ratio (difference in mean ratios 0.12; P=0.934), or T cell count (difference in means 0.92%; P=0.726). Furthermore, comparisons of the distributions