DNA Abnormalities in Benign Barrett's Esophagus (BE) are associated with Subsequent Progression to Esophageal Adenocarcinoma (EAC)

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Introduction

- Clinical management of Barrett's esophagus (BE) with no dysplasia or low-grade dysplasia is hindered by an absence of accurate predictors of progression
- Because of inability to stratify risk, subjects with benign disease may receive unduly aggressive intervention. Conversely, the window of opportunity for endoscopic intervention in those destined for cancer may be missed
- Cell biologic markers may help stratify risk of progression to cancer in patients with BE, allowing for more aggressive therapy to be directed at patients with highest risk for progression
- We performed a nested case-control study to assess whether the presence of DNA abnormalities as detected by fluorescence in-situ hydridization (FISH) in non-dysplastic BE or BE with low-grade dysplasia (LGD) predicted the subsequent occurrence of high-grade dysplasia (HGD) or esophageal adenocarcinoma (EAC).

Methods

- **Study Design**: A four-probe fluorescence in situ hybridization (FISH) assay was performed on esophageal biopsy specimens of subjects with and without progression to HGD or EAC
- **FISH Assay**: Assessed for DNA abnormalities in MYC[8q24], p16[9p21.3], HER2[17q11.2],and ZNF217[20q13.2]. Assays results quantified by the percentage of cells with:
  - **Multiple probe gains**
  - **Single probe gains**
  - **Homozygous 9p21 loss**
  - **Any probe gains with any 9p21 loss**

Methods, cont.

- **Esophageal biopsy specimens**: Formalin-fixed, paraffin-embedded, archived samples from large pathology database
- **Non-progressor population:** 33 subjects with NDBE at baseline
  - All subjects with NDBE on follow up biopsies at ≥ 6 mos (median f/u 25 months, range: 9 – 76 mos)
  - Median age 63 years, 100% male (33/33)
- **Progressor population**: 28 subjects with NDBE (n = 20) or LGD (n = 8) at baseline
  - All subjects with HGD (n = 7) or EAC (n = 21) on follow up biopsies at ≥6 mos (median f/u 21 mos, range: 6 – 37 mos)
  - Median age 63 years, 86% male (24/28)
- **Statistical analysis**:
  - Assay results were compared among groups using non-parametric tests (Wilcoxon Rank-Sum Test for continuous data, Fisher's Exact Test for categorical data)
  - The following thresholds previously established for detection of EAC were used to evaluate risk of progression:

•Multiple probe gains ≥4%
•Single probe gains ≥16%
•Homozygous 9p21 loss ≥9%
•Any probe gains with any 9p21 loss ≥4%

  - Area under the ROC curve, sensitivity, and specificity for each dichotomous test measurement

Results

FISH Probe Assay Results (Continuous)

Median % cells (IQR)	Non-progressors (n=33)	Progressors (n=28)	P-value
Multiple probe gains	1 (0 – 3)	2 (1 – 6)	0.01
Single probe gains	4 (1 – 12)	7 (3 – 15)	0.001
Homozygous 9p21 loss	0 (0 – 4)	1 (0 – 2)	0.47
Any gain plus any 9p21 loss	0 (0 – 2)	1 (0 – 3)	0.008

FISH Probe Assay Results (Dichotomous)

% Positive (N)	Non-progressors (n=33)	Progressors (n=28)	P-value
Multiple probe gains ≥4%	9% (3)	36% (10)	0.01
Single probe gains ≥16%	9% (3)	29% (8)	0.09
Homozygous 9p21 loss ≥9%	3% (1)	0	1.0
Any gain plus any 9p21 loss ≥4%	6% (2)	11% (3)	0.65
Any positive test	**15% (5)**	**39% (11)**	**0.04**

Results, cont.

FISH Probe Performance Characteristics

	Area under ROC curve	Sensitivity	Specificity
Multiple probe gains	0.63	36%	91%
Single probe gains	0.60	29%	91%
Homozygous 9p21 loss	0.48	0%	97%
Any gain plus any 9p21 loss	0.52	11%	94%
Any positive test	0.62	**39%**	**85%**

Conclusions

- DNA abnormalities in subjects with benign Barrett's histology were associated with subsequent progression to cancer
- These markers, perhaps in combination with other demographic and disease-specific predictors of progression, may allow for more accurate risk stratification of subjects under endoscopic surveillance for BE
- Future study is necessary to determine if analysis of genetic abnormalities in benign BE can reliably predict patients at risk for progression

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