Featured Article

[^{18}F]Fluoro-2-deoxy-D-glucose Positron Emission Tomography Detects Gastric Carcinoma in an Early Stage in an Asymptomatic E-Cadherin Mutation Carrier

Mariette C. A. van Kouwen,[1] Joost P. H. Drenth,[1,6] Wim J. G. Oyen,[2] Joyce H. F. M. de Bruin,[3] Marjolijn J. Ligtenberg,[3] J. J. (Han) Bonenkamp,[4] J. Han J. M. van Krieken,[5] and Fokko M. Nagengast[1]

Departments of [1]Gastroenterology, [2]Nuclear Medicine, [3]Human Genetics, [4]Surgery, and [5]Pathology University Medical Center St. Radboud, Nijmegen, the Netherlands; and [6]Cell Biology and Metabolism Branch, National Institute of Child Health and Human Development, NIH, Bethesda, Maryland

ABSTRACT

Purpose: **Autosomal dominant hereditary diffuse gastric cancer (HDGC) is caused by germ-line E-cadherin (*CDH1*) gene mutations. Early detection of cancer in carriers is difficult because HDGC escapes endoscopic detection. We hypothesized that the glucose metabolism is enhanced in HDGC and that this can be detected with [^{18}F]fluoro-2-deoxy-D-glucose positron emission tomography (FDG-PET).**

Experimental Design and Results: **An asymptomatic twenty-eight year-old female was seen at our outpatient clinic because of a request for screening on HDGC. Her father and younger sister died of diffuse gastric cancer, at the ages of 52 and 27, respectively. Mutational analysis of the *CDH1* gene in this patient demonstrated a novel heterozygous splice-site mutation in exon 8 (1135delACGGTA-ATinsTTAGA). Upper gastrointestinal endoscopies revealed no macroscopic abnormalities, but one of the 40 random biopsy specimens showed well-differentiated signet-cell carcinoma. A FDG-PET scan demonstrated two spots of FDG accumulation, one located in the proximal part of the stomach and the second in the region of the pylorus. A total gastrectomy was performed and microscopic examination showed focal localization of intramucosal adenocarcinoma of the signet-cell type in the cardiac and antrum area. Most notably, the localization of the FDG accumulation matched the localization of the carcinoma.**

Conclusions: **We present an asymptomatic patient from a HDGC family carrying a novel *CDH1* mutation in whom FDG-PET scanning facilitated early detection of HDGC. This calls for further investigation of the role of FDG-PET scan as a screening modality in HDGC.**

INTRODUCTION

Gastric cancer is a particularly common gastrointestinal malignant disease and has an incidence rate of 9/100,000 in the United States (1). Some 10% of the gastric cancer cases show familial clustering (2, 3), and a subgroup of these patients suffer from hereditary diffuse gastric cancer (HDGC; Mendelian inheritance in Man no. 192090; http://www.ncbi.nlm.nih.gov/entrez/dispomim.cgi?id=192090).[7] This is an autosomal dominantly inherited disorder with about 70% penetrance (4). Until now, up to 80 HDGC families have been described. (5, 6). Approximately one third of these families carry a germ-line E-cadherin (*CDH1*) gene mutation (7, 8). *CDH1* is localized on chromosome 16q22.1 and encodes for cadherin-1. This protein comprises five extracellular cadherin repeats, a transmembrane region, and a highly conserved cytoplasmic tail and functions as a calcium-dependent cell–cell adhesion glycoprotein. It is postulated that *CDH1* acts as a tumor suppressor gene, and loss of function leads to development and progression of cancer by increasing proliferation, invasion, and/or metastasis. The estimated cumulative lifetime risk of gastric cancer in *CDH1* families is very high. It has been estimated that by the age of 80 years, 67% of the males and 83% of the females will have developed gastric cancer (4). The average age of onset of gastric cancer in *CDH1* families is 38 years, but HDGC has been documented in teens as well (7, 9). The pathology of HDGC is that of a poorly differentiated isolated-cell-type carcinoma with signet cells. Clinically, it is characterized by individual cells infiltrating and thickening the gastric wall, without substantial formation of tumor mass. The infiltration results in a loss of distensibility of the wall of the stomach, and, because the individual signet cells spread submucosally, it may evade detection by endoscopy. This eliminates endoscopy as an effective screening tool in a population that would benefit most from

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Requests for reprints: Joost PH Drenth, Department of Medicine, Division of Gastroenterology and Hepatology, University Medical Center St. Radboud, P.O. Box 9101, 6500 HB Nijmegen, The Netherlands. Phone: +31 24 3614760; Fax: +31 24 3540103; E-mail: JoostPHDrenth@CS.com.

[7] Internet address for Mendelian inheritance in Man number; 192090: http://www.ncbi.nlm.nih.gov/entrez/dispomim.cgi?id=192090.

early detection of a malignant disease. However, because most malignant cells demonstrate a high glucose uptake, this opens an avenue for another screening tool: [^{18}F]fluoro-2-deoxyglucose positron emission tomography (FDG-PET; ref. 10–14). FDG-PET is a noninvasive imaging technique that visualizes the glucose metabolism in the human body. Before the PET procedure, patients fast for at least 6 hours to suppress the glucose and insulin level. FDG is injected, serum glucose level is measured, and 1 hour later, the scanning starts. FDG is a glucose analog, and it uses glucose transport receptors (GLUT-1) to enter the cell (10). Similarly to glucose, FDG, is phosphorylated by hexokinase to FDG-6-phosphate. In contrast to glucose-6-phosphate, FDG-6-phosphate, is not further metabolized; and, because it cannot transfer beyond the cell membrane, it is trapped in the cell. Most malignant cells demonstrate high FDG uptake because of increased expression of the GLUT-1 receptor on the cell membrane. In addition, a high enzymatic activity of hexokinase and low levels of glucose-6-phosphatase in tumor cells also account for an elevated FDG uptake. The great advantage of FDG-PET over conventional radiodiagnostic methods lies in its ability to visualize metabolic activity of the tumor rather than mere anatomic borders. We examined whether FDG-PET scanning is able to detect early gastric cancer in HDGC.

METHODS AND RESULTS

A twenty-eight year-old female was seen at our outpatient clinic because of a request for screening on hereditary gastric cancer. She had no physical complaints, but her family history was strongly positive for gastric cancer. Her father died, at the age of 52 years, of diffuse gastric cancer, signet-cell type. Her (younger) sister was diagnosed with metastatic gastric cancer at 27 years of age and died 10 months after diagnosis. A brother of her paternal grandfather also died at the age of 55 years of gastric cancer. In her nuclear family, two other sisters and her mother were not known to have gastric cancer.

Because we suspected hereditary gastric cancer, we proceeded to perform mutation analysis of all 16 exons of the *CDH1* gene. In view of the unsettling family history, we performed endoscopic screening in parallel. The first upper gastrointestinal endoscopy showed macroscopically normal mucosa. We took 40 (at random) biopsy specimen from all parts of the stomach. Histopathological examination revealed normal gastric mucosa, without signs of inflammation, dysplasia, or helicobacter pylori infection. Because of the possibility of sampling error, we went on to perform a second gastroscopy. Again no macroscopic abnormalities were noted, but now we detected well-differentiated adenocarcinoma of the signet-cell type in a single biopsy specimen taken from the fundus (Fig. 1). The diagnosis, diffuse gastric cancer, was made.

Preoperative examination by computed tomography of the chest and abdomen showed no signs of metastasis. FDG-PET demonstrated two hotspots, one located in the proximal part of the stomach and the second in the region of the pylorus (Fig. 2). No other hotspots were seen elsewhere in the body. A total gastrectomy with Roux-en-Y-esophagojejunostomy was performed. The gastrectomy specimen was normal in appearance and by palpation. Microscopic examination of the complete mucosa showed in the cardiac and antrum area of the stomach focal localization of intramucosal adenocarcinoma of the signet-cell type. The localization of the FDG accumulation matched the localization of the carcinoma. On immunohistochemical staining, the tumor cells showed no GLUT-1 expression Resection areas were free of tumor cells. All 19 lymph nodes examined

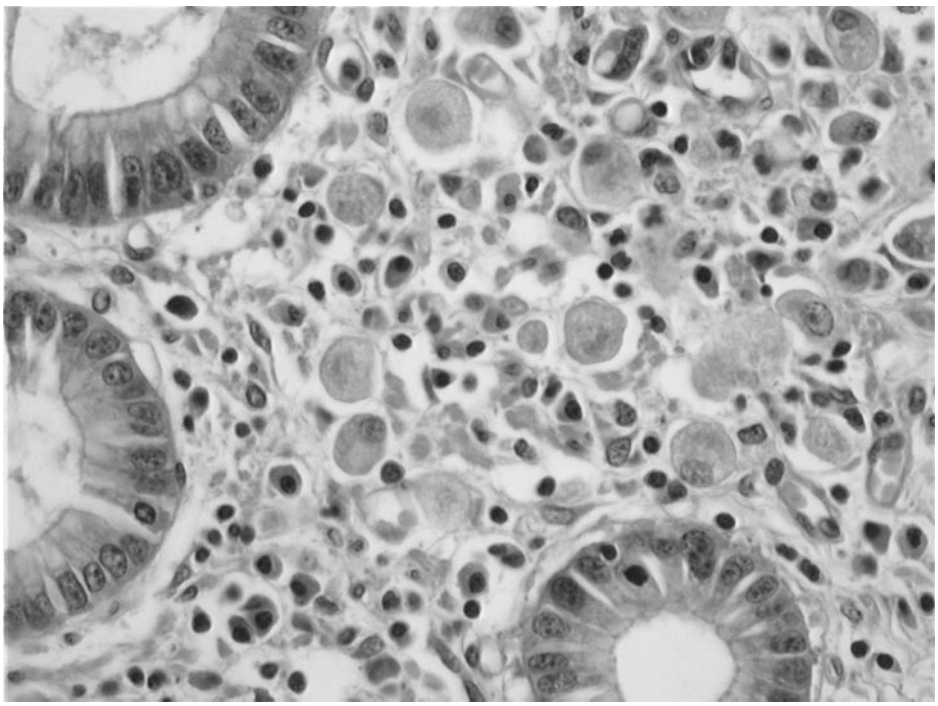

Fig. 1 High-magnification (×400) H&E-stained image of loose epithelial cells in the lamina propria of the gastric mucosa. The cells are only partially typical signet-ring cells. On immunohistochemical examination, they were proved to be epithelial (keratin-positive, CD68-negative).

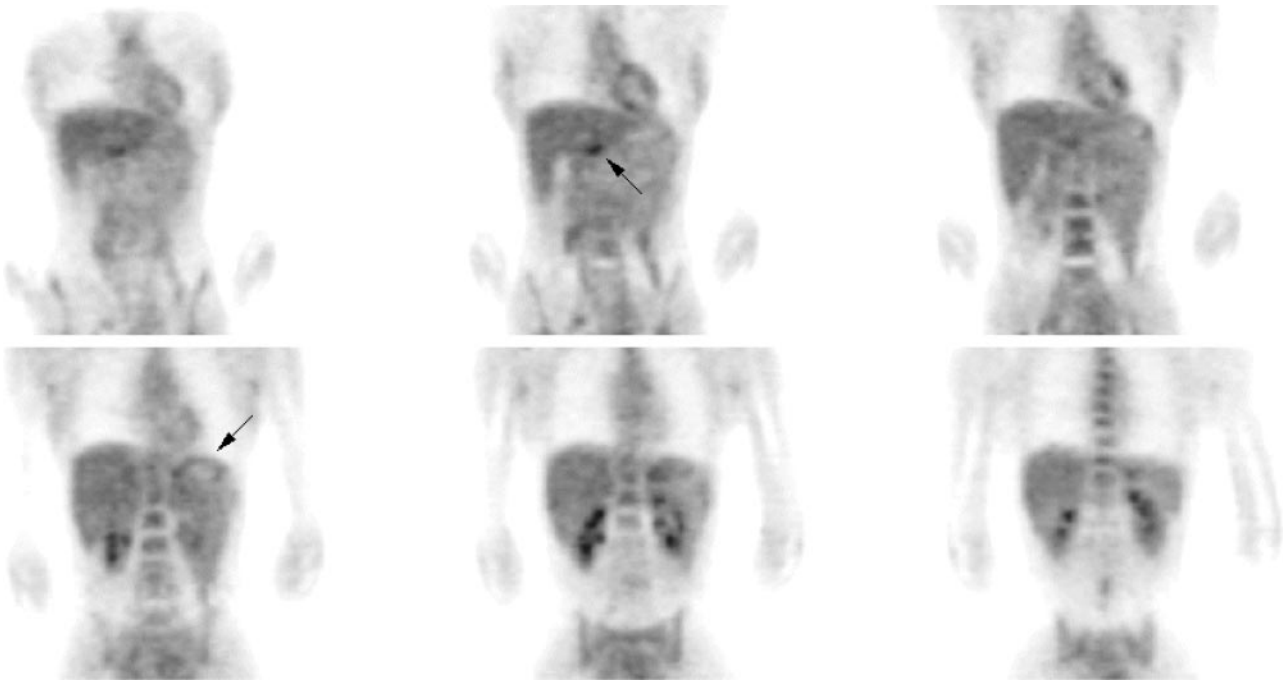

Fig. 2 Serial coronal sections (front to back) of FDG-PET in our patient; diffusely increased accumulation of FDG in the proximal part of the stomach and hotspot (*arrows*) in the region of the pylorus of the stomach.

were negative for cancer. The carcinoma was classified as $T_1N_0M_x$. There was no need for adjuvant therapy.

Results of genetic analysis of this patient has been documented recently (15) and DNA sequencing revealed a hitherto undescribed splice site mutation in exon 8 of *CDH1*, with the deletion of 8 bp and the insertion of 5 other bp (1135delACGGTAATinsTTAGA). This mutation causes a truncated protein with removal of the highly conserved cytoplasmic region (amino acid 732–879).

One year after surgery, there are no signs of recurrence or metastases.

DISCUSSION

This case demonstrates that *CDH1* mutations are associated with early-onset HDGC. The heterozygous germ-line *CDH1* mutation will most probably cause a 50% reduction in E-cadherin function, and, although this may be sufficient to develop HDGC, it is likely that inactivation of the wild-type allele is needed to enable tumor progression. The "second-hit" in most of the cases is due to hypermethylation of the *CDH1* gene promoter, (16) loss of heterozygosity, or somatic mutations, but none of these were detected in tumor material from our patient (15).

Although, the overwhelming majority (26 of 30) of germline mutations characterized, thus far, are associated with HDGC, (6) *CDH1* mutations have also been implicated in breast, colorectal, thyroid, and ovarian cancer (5). The consequences for the clinical follow-up of our patient with regard to the development of other malignant diseases, such as breast cancer, are still under discussion.

Present guidelines emphasize the importance of intensive clinical surveillance of germ-line *CDH1* mutation carriers. Regular endoscopy with multiple biopsies (every 6–12 months) has been recommended as the most optimal method for early detection of gastric cancer in HDGC families (17). However, mucosal abnormalities can be absent despite the presence of (sometimes) extensively spread malignancy. This calls for another screening modality, and as illustrated by our case report, the FDG-PET scan was able to detect HDGC in an early phase, leaving room for curation. This illustrates the high sensitivity of the screening technique, because histopathological examination demonstrated only a minor tumor load. In present gastroenterological practice, FDG-PET is most commonly used to stage esophageal carcinoma, to detect and stage recurrence of colorectal carcinoma, and to differentiate between benign and malignant pancreatic lesions (18–21). Experience with this technique in patients with gastric carcinoma is very limited. Most of the studies focused on recurrent gastric cancer or locally advanced disease (22–25). They all found a low sensitivity and specificity to detect recurrence or locally advanced disease, with figures around 70%. Most notably, in subgroup analysis, the sensitivity in signet-cell gastric carcinoma is even worse; it differs between 40 (23) and 60% (22). Moreover, there appears to be a significant lower uptake of FDG in signet-cell carcinoma compared with well- or moderate differentiated adenocarcinomas (24). These findings are in line with experimental data showing that, in gastric carcinoma, GLUT-1 is expressed late in carcinogenesis and that signet-ring-cell carcinomas rarely express GLUT-1 (26). This accords with the absence of GLUT-1 expression on immunohistochemical staining of gastric carcinoma specimens. This suggests that other mechanisms, such as elevated hexokinase levels or decreased activity of glucose-6-phosphatase, lead to

high FDG uptake in HDGC. However, apart from a late expression of GLUT-1, still little is known about FDG accumulation in gastric cancer. It is also possible that HDGC stands apart from isolated gastric cancer in that these signet cells might overexpress other GLUT receptors, such as GLUT-2 and -3, which facilitate the accumulation of FDG.

In view of the grave prognosis of HDGC, prophylactic gastrectomy has been recommended for asymptomatic *CDH1* mutation carriers. This probably leads to a better survival of these patients. For example, in two small series of five patients, foci of diffuse gastric cancer were identified in all, despite negative endoscopic screening (27, 28). However, there is a trade-off because surgery is accompanied by a mortality rate of 1 to 2% and a considerable morbidity such as persistent dumping symptoms (29). Furthermore, the timing of gastrectomy in mutation carriers is under debate. This case suggests that FDG-PET scanning can aid in the decision about the timing of surgery. In conclusion, HDGC is caused by mutations in *CDH1*, and most symptomatic patients present in an incurable stage. We present a case with some striking features: (*a*) early detection by endoscopic screening; (*b*) a novel *CDH1* mutation; and (*c*) identification of the tumor by FDG-PET scanning in a very early stage. Our data suggest that FDG-PET scanning can have a place as a screening tool in HDGC and/or might have a role in timing of the gastrectomy. Further investigation is warranted to determine the role FDG-PET in HDGC and to identify the mechanism of high FDG uptake in this specific syndrome.

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