

MOLECULAR ADVANCES IN GASTRIC CARCINOGENESIS AND GASTRIC ADENOCARCINOMAS

Fátima Carneiro, MD, PhD

Institute of Molecular Pathology and Immunology of the University of Porto (IPATIMUP) and Medical Faculty of Porto/Hospital S. João, Porto, Portugal

Carcinomas of the stomach are very heterogeneous from the morphologic standpoint. This heterogeneity, which is amply reflected in the diversity of histopathologic classifications on record, is based on different approaches: histologic profile, degree of differentiation, pattern of growth and histogenesis.

The classification of Laurén is one of the most widely used, recognizing two major types of gastric cancer: “intestinal” carcinoma and “diffuse” carcinoma, which display different clinicopathologic profiles and occur in distinct epidemiologic settings.

The large majority of gastric cancers are sporadic. However, familial aggregation of gastric cancer, both of the diffuse and the intestinal type, suggest the importance of genetic predisposition in these settings. Presently, it is calculated that about 1% of gastric cancers are hereditary, the majority corresponding to “Hereditary Diffuse Gastric Cancer” (HDGC).

In sporadic gastric cancer, the available evidence supports the existence of two main histogenetic pathways of carcinogenesis: one leading to “intestinal carcinoma” *via* chronic atrophic gastritis, incomplete intestinal metaplasia, namely type III¹ and adenomatous dysplasia,² and the other leading to diffuse carcinoma, either *de novo* or *via* hyperplastic changes.^{3,4} Both pathways appear to develop on the background of *Helicobacter pylori* associated gastritis.⁴

The initially suggested sequential evolution of intestinal metaplasia, from type I to type III, *via* a type II intermediate step, has been recently challenged by Reis *et al*⁵ who suggested that complete intestinal metaplasia and incomplete intestinal metaplasia represent, *ab initio*, divergent differentiation programs.

Ultrastructural studies of gastric carcinoma^{6,7} and the study of the expression of trefoil peptides and mucins in premalignant lesions and gastric carcinomas⁸⁻¹⁰ showed that “intestinal” carcinoma is heterogeneous regarding cell differentiation, encompassing gastric and non-gastric phenotypes. Without denying the validity of the histogenetic pathway *via* intestinal metaplasia to “intestinal” carcinoma, these data support that a sub-set of “intestinal” carcinomas may derive from gastric-type epithelium, either *de novo* or through hyperplastic changes.⁸ This concept is reinforced by the verification that “intestinal” carcinomas with gastric-type phenotype develop through the “mutator pathway”, characterized by Microsatellite Instability (MSI), whereas Chromosomal Instability (CIN) appears to be involved in the tumorigenesis of “intestinal” carcinomas displaying a non-gastric phenotype. These observations led us to claim that gastric carcinomas with gland formation should be designated as “glandular” carcinomas, instead of “intestinal carcinomas”, thus avoiding the confusion of mixing structural and cell differentiation concepts.¹¹

The histogenetic pathway of diffuse carcinoma is less well elucidated in the sporadic setting. It has been suggested that this type of gastric carcinoma may derive from a non-metaplastic dysplasia of gastric mucosa.¹² Further, different studies of our group¹³⁻¹⁵ provided enough evidence to claim that foveolar hyperplasia/hyperplastic polyps may serve as precursor lesions of the diffuse type of gastric carcinoma. At the molecular level, a large proportion of diffuse gastric carcinomas exhibit

E-cadherin mutations.^{16, 17} Recently, it was possible to demonstrate that hypermethylation of the promoter of E-cadherin gene constitutes the second hit of inactivation of this gene in sporadic diffuse gastric carcinomas.¹⁸

At this stage we remain with an unsolved problem regarding the histogenesis of the heterogeneous group of gastric carcinomas designated as “indeterminate” carcinomas according to Laurén’s classification. This group encompasses two major morphologic variants: solid and mixed carcinomas. Available evidence suggests that MSI may be involved in the tumorigenesis of solid carcinomas regardless of cell differentiation.¹⁹ Mixed carcinomas (characterized by the coexistence in the same tumour of distinct “intestinal” and diffuse histologic components) apparently derive from “intestinal” carcinomas with gastric phenotype *via* the occurrence of E-cadherin/*CDH1* expression/gene changes leading to the establishment of a diffuse carcinoma that coexists with the “intestinal” carcinoma.^{17, 20} We can not exclude, alternatively, that epigenetic alterations at the E-cadherin level in the superficial layer of the stomach may cause a diffuse carcinoma pattern that might evolve to an “intestinal” or mixed pattern together with the progression of the neoplastic cells and their invasion in the deep layers of the gastric wall.

Hereditary Diffuse Gastric Cancer (HDGC) was defined by the International Gastric Cancer Linkage Consortium.²¹ Various studies showed that E-cadherin/*CDH1* germline mutations occur in about 30% of HDGC families.^{22,23} However, failure to detect E-cadherin mutations in a relatively large proportion of families with HDGC suggests that genes other than E-cadherin/*CDH1* remain to be identified. In HDGC the vast majority of mutations lead to truncation of the encoded protein.²¹ This is at variance with the findings in sporadic diffuse gastric cancer showing that the majority of sequence changes result in either missense mutations or exon skipping.^{16, 17, 24} Putative missense mutations have been reported in families with HDGC, but their functional significance has not been elucidated yet. Hypermethylation of the *CDH1* promoter is likely to be a common cause of down-regulation or inactivation of the second *CDH1* allele in HDGC tumours.²⁵

Guidelines for genetic testing and counselling in HDGC and strategies for clinical management in families with high penetrance autosomal dominant predisposition to gastric cancer were established by the IGCLC.²¹ Such guidelines include intensive screening and prophylactic gastrectomy for asymptomatic carriers of E-cadherin/*CDH1* germline mutations.²¹ Subsequently, genetic screening, surgical management and pathologic findings in ten young E-cadherin/*CDH1* germline mutation carriers from three unrelated families with HDGC, submitted to prophylactic gastrectomy, were described.²⁶⁻²⁸ All ten cases had superficial diffuse gastric cancers, most lesions measuring less than 1mm in diameter. In seven cases, *in situ* lesions were also observed. HDGC constitutes a model of nature that may contribute decisively for a better understanding of the histogenetic and molecular pathways of diffuse gastric cancer.

References:

¹ Filipe MI, Munoz N, Matko I, Kato I, Pompe-Kirn V, Jutersek A, Teuchmann S, Benz M, Prijon T: Intestinal metaplasia types and the risk of gastric cancer: a cohort study in Slovenia. *Int J Cancer* 57: 324-329, 1994.

² Correa P: Human gastric carcinogenesis: a multistep and multifactorial process. *Cancer Res* 52: 6735-6740, 1992.

³ Carneiro F, David L, Seruca R, Castedo S, Nesland JM, Sobrinho-Simões M: Hyperplastic polyposis and diffuse carcinoma of the stomach. A study of a family. *Cancer* 72: 323-329, 1993.

- ⁴ Solcia E, Fiocca R, Luinetti O, Villani L, Padovan L, Calistri D, Ranzani GN, Chiaravalli A, Capella C: Intestinal and diffuse gastric cancers arise in a different background of *Helicobacter pylori* gastritis through different gene involvement. *Am J Surg Pathol* 20:S8-S22, 1996.
- ⁵ Reis C, David L, Correa P, Carneiro F, de Bolós C, Garcia E, Mandel U, Clausen H, Sobrinho-Simões M: Intestinal metaplasia of stomach displays distinct patterns of mucin (MUC1, MUC2, MUC5AC, and MUC6) expression. *Cancer Res* 59: 1003-1007, 1999.
- ⁶ Fiocca R, Villani L, Tenti P, Solcia E, Cornaggia M, Frigerio B, Capella C: Characterization of four main cell types in gastric cancer: foveolar, mucopeptic, intestinal columnar and goblet cells. An histopathologic, histochemical and ultrastructural study of “early” and “advanced” tumours. *Pathol Res Pract* 182: 308-325, 1987.
- ⁷ Carneiro F, Moutinho-Ribeiro M, David L, Seixas M, Sansonetty F, Soares P, Serrano A, Sobrinho-Simões M: Signet ring cell carcinoma of the stomach. A morphometric, ultrastructural and DNA cytometric study. *Ultrastruct Pathol* 16: 603-614 1992.
- ⁸ Machado JC, Carneiro F, Blin N, Sobrinho-Simões M: Pattern of pS2 protein expression in premalignant and malignant lesions of gastric mucosa. *Eur J Cancer Prevent* 5: 169-179, 1996.
- ⁹ Nogueira A, Machado JC, Carneiro F, Reis C, Gött P, Sobrinho-Simões M: Patterns of expression of trefoil peptides and mucins in gastric polyps with and without malignant transformation. *J Pathol* 187: 541-548, 1999.
- ¹⁰ Machado JC, Nogueira AMMF, Carneiro F, Reis CA, Sobrinho-Simões M: Gastric carcinoma exhibits distinct types of cell differentiation: an immunohistochemical study of trefoil peptides (TFF1 and TFF2) and mucins (MUC1, MUC2, MUC5AC and MUC6). *J Pathol* 190: 437-443, 2000.
- ¹¹ Carneiro F, Seixas M, Sobrinho-Simões M: New elements for an updated classification of the carcinomas of the stomach. *Pathol Res Pract* 191: 571-584, 1995.
- ¹² Ghandur-Mnaymneh L, Paz J, Roldan E, Cassady J: Dysplasia of nonmetaplastic gastric mucosa. A proposal for its classification and its possible relationship to diffuse-type gastric carcinoma. *Am J Surg Pathol* 12 :96-114, 1988.
- ¹³ Carneiro F, Taveira-Gomes A, Cabral-Correia A, Vasconcelos-Teixeira A, Barreira R, Cardoso-Oliveira M, Sobrinho-Simões M: Characteristics of the gastric mucosa of direct relatives of patients with sporadic gastric carcinoma. *Eur J Cancer Prevent* 2: 239-246, 1993.
- ¹⁴ Carneiro F, Santos L, Sobrinho-Simões M: Carcinoma arising in gastric hyperplastic polyps. *Gastrointest Endosc* 41: 178-179, 1995.
- ¹⁵ Carneiro F, Sobrinho-Simões M: Signet ring cell carcinoma in hyperplastic polyp. *Scand J Gastroenterol* 31: 95-96, 1996.
- ¹⁶ Becker KF, Atkinson MJ, Reich U, Becker I, Nekarda H, Siewert JR, Hofler H: E-cadherin gene mutations provide clues to diffuse type gastric carcinomas. *Cancer Res* 54: 3845-3852, 1994.
- ¹⁷ Machado JC, Soares P, Carneiro F, Rocha A, Beck S, Blin N, Berx G, Sobrinho-Simões M: E-cadherin gene mutations provide a genetic basis for the phenotypic divergence of mixed gastric carcinomas. *Lab Invest* 79: 459-465, 1999.

- ¹⁸ Machado JC, Oliveira C, Carvalho R, Soares P, Berx G, Caldas C, Seruca R, Carneiro F, Sobrinho-Simões M: E-cadherin gene (CDH1) promoter methylation as the second hit in sporadic diffuse gastric carcinoma. *Oncogene* 20: 1525-1528, 2001.
- ¹⁹ Nabais S, Carneiro F, Nogueira AMMF, Machado JC, Seruca R, Sobrinho-Simões M: Cellular phenotypes of differentiated-type adenocarcinomas and precancerous lesions of the stomach are dependent on genetic pathways (Letter-to-the-Editor). *J Pathol* (in press).
- ²⁰ Machado JC, Carneiro F, Beck S, Rossi S, Lopes J, Taveira-Gomes A, Cardoso-Oliveira M: E-cadherin expression is correlated with the isolated cell/diffuse histotype and with features of biological aggressiveness of gastric carcinoma. *Int J Surg Pathol* 6: 135-144, 1998.
- ²¹ Caldas C, Carneiro F, Lynch HT, Yokota J, Wiesner G, Powell S, Lewis FR, Huntsman DG, Pharoah P, Jankowski J, MacLeod P, Vogelsang H, Keller G, Park KGM, Richards F, Maher E, Gayther S, Oliveira C, Grehan N, Wight D, Seruca R, Roviello F, Ponder BAJ, Jackson CE: Familial gastric cancer: overview and guidelines for management. *J Med Genet* 36: 873-880, 1999.
- ²² Gayther SA, Goringe KL, Ramus SJ, Huntsman D, Roviello F, Grehan N, Machado JC, Pinto E, Seruca R, Halling K, Macleod P, Powell SM, Jackson CE, Ponder BA, Caldas C: Identification of germline E-cadherin mutations in gastric cancer families of European origin. *Cancer Res* 58: 4086-4089, 1998.
- ²³ Oliveira C, Bordin MC, Grehan N, Huntsman D, Suriano G, Machado JC, Kiviluoto T, Aaltonen L, Jackson CE, Seruca R, Caldas C: Screening E-cadherin in gastric cancer families reveals germline mutations only in hereditary diffuse gastric cancer kindred. *Hum Mutat* (in press).
- ²⁴ Berx G, Becker KF, Hofler H, van Roy F: Mutations of the human E-cadherin (*CDH1*) gene. *Hum Mutat* 12: 226-237, 1998.
- ²⁵ Grady WM, Willis J, Guilford PJ, Dunbier AK, Toro, TT, Lynch H, Wiesner G, Ferguson K, Eng C, Park JG, Kim SJ, Markowitz S: Methylation of the CDH1 promoter as the second genetic hit in hereditary diffuse gastric cancer. *Nat Genet* 26: 16-17, 2000.
- ²⁶ Huntsman DG, Carneiro F, Lewis FR, MacLeod PM, Hayashi A, Monaghan KG, Maung R, Seruca R, Jackson CE, Caldas C: Early gastric cancer in young, asymptomatic carriers of germ-line E-cadherin mutations. *N Engl J Med* 344:1904-1909, 2001.
- ²⁷ Lewis FR, Mellinger JD, Hayashi A, Lorelli D, Monaghan KG, Carneiro F, Huntsman DG, Jackson CE, Caldas C: Prophylactic total gastrectomy for familial gastric cancer. *Surgery* 130: 612-619, 2001.
- ²⁸ Chun YS, Lindor NM, Smyrk TC, Petersen BT, Burgart LJ, Guilford PJ, Donohue JH: Germline E-cadherin gene mutations. Is prophylactic gastrectomy indicated? *Cancer* 92: 181-187, 2001.