

Different Biological Materials are Found in Neoplastic Glands with Pores at the Invading Edge of Sporadic Colonic Carcinomas

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Abstract. *Background:* Studying the invading edge of colorectal carcinomas it was previously noticed that the neoplastic glands with pores could contain mucin, inflammatory cells and/or necrotic material. *Aims:* To systematically record the type of intraglandular materials found in the neoplastic glands with pores at the invading edge of sporadic colonic carcinomas. *Materials and Methods:* Histological sections from 12 adenocarcinomas were selected when the following materials were predominantly found in the neoplastic glands with pores at the invading tumor edge: mucin (3 cases, group I), neutrophilic granulocytes (3 cases, group II), necrotic material (3 cases, group III) and invaginated stroma (3 cases, group IV). Surgical histological sections were stained with hematoxylin and eosin (H&E) and analyzed at 20 magnifications. The length of the invading edge was measured with a translucent millimetre ruler and the number of glands with pores per mm was calculated in each case. *Results:* Two hundred and ninety-five glands with pores were analyzed in the 12 cases. Mucus was found in 68% (n=55) of the 81 glands with pores in group I, granulocytes in 95% (n=41) of the 43 glands with pores in group II, necrotic material in 72% (n=84) of the 117 glands with pores in group III and stromal invaginations in 61% (n=33) of the 54 glands with pores in group IV. The mean number of glands with pores per mm was 1.02 (55 glands/54 mm) in group I, 0.93 (41 glands/44 mm) in group II, 1.22 (84 glands /69 mm) in group III and 0.62 (33 glands /53 mm) in group IV. *Discussion and Conclusion:* Neoplastic glands with pores at the invading front of colonic adenocarcinomas may

predominantly contain various biological materials. These materials may differ in different individuals. While tumor mucins harbour proteolytic enzymes able to digest the juxtaposed stroma of the host, the possible significance of the other intraglandular materials contributed by the host, such as neutrophils, necrotic material and/or stromal invaginations on tumor growth, is still poorly understood. Further studies are necessary to assess whether each one of these intraglandular materials has any bearing on the progression of colonic carcinomas.

Colorectal adenomas are foci of atypical cells with aberrant proliferation and the main source of colorectal invasive carcinomas (CRC) (1), the third most commonly diagnosed cancer in Europe and the US (2).

The mechanism(s) whereby colorectal tumor cells invade the host remains poorly understood. In attempts to disclose one or more histological parameters possibly involved in tumour invasion, several workers have studied the growing edge of the tumor. Some of the variables investigated have been the characteristics of the tumor growth (expansive vs. infiltrating (3)), the degree of tumour differentiation (3), the presence of tumour "budding" (*i.e.* foci of up to 5 cancer cells) at the invasive margin (4), tumor cell locomotion (5) and the immunologic host reaction, by assessing the presence of peritumoral lymphocytes (6), to name but a few.

Years ago, investigations of the growing edge of sporadic CRC histological parameters which were at variance with those previously published in the literature were reported (7-14). It was demonstrated that the neoplastic glands at the growing edge lacked a consecutive group of tumour cells. The latter were referred to as glandular pores (7, 12). A similar phenomenon as that for sporadic CRC was also recorded at the growing edge of hereditary CRC in patients with hereditary non-polyposis colorectal cancer (HNPCC) (9), in CRC evolving in inflammatory bowel disease (IBD) (10), in sporadic colonic carcinomas in baboons (15) and in experimentally induced colonic carcinomas in rats (16).

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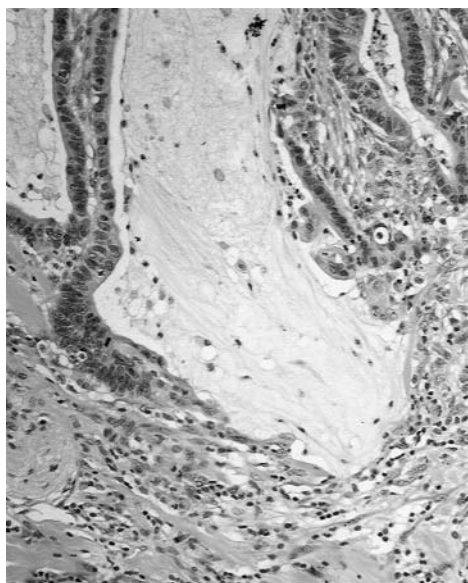


Figure 1. Colon adenocarcinoma. Neoplastic gland with pore at the invasion tumor edge, containing mucin that is being released through the pore into the juxtaposed stroma (H&E. $\times 20$).

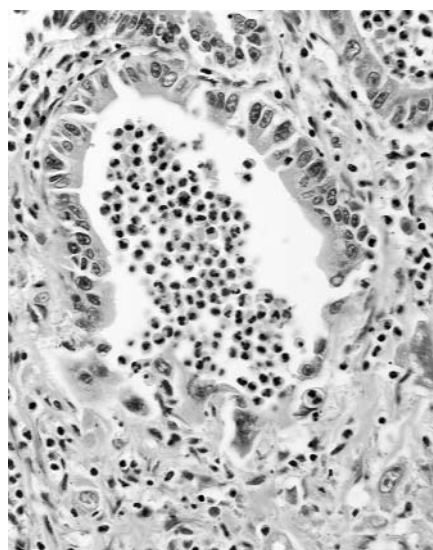


Figure 2. Colon adenocarcinoma. Neoplastic gland with pore at the invasion tumor edge, containing granulocytes that are being released through the pore into the juxtaposed stroma (H&E. $\times 20$).

During the course of these studies it was noticed that the neoplastic glands at the invading edge could contain mucin, inflammatory cells and/or necrotic material (8, 9). These intraglandular retained materials were seen released through the glandular pores into the juxtaposed matrix. In a survey of surgical specimens with CRC in inflammatory colonic disease (IBD) (10) it was found in 46 of the 65 specimens that the material discharged through the neoplastic glandular pores was either mucin, neutrophilic granulocytes and/or necrotic material.

The intraglandular accumulation of these materials seems to occur early during colonic carcinogenesis. In this respect it was found that neoplastic glands with pores were present in 49% of 47 colonic adenomas (17). The glands showed retained mucin, inflammatory cells and/or necrotic material, materials that were often seen released into the surrounding *lamina propria* through the glandular pores. In contrast, none of the 14 hyperplastic colonic polyps showed a similar phenomenon.

A recent review of 53 endoscopically removed colonic adenomas measuring ≥ 10 mm revealed that 45 of the 53 adenomas (85%) had dilated neoplastic glands. In 39 of the 45 adenomas, the dilated glands contained mucin, granulocytes in 5 adenomas and necrotic material in the remaining adenoma (Rubio, Lenander, in preparation). It would appear that in colonic adenomas many neoplastic glands retain different materials in an individual manner, before invasion ensues. Thus, dysplastic colonic glands may

already exhibit retained intraglandular materials. More recent observations showed that some of the neoplastic glands having none of the aforementioned materials, contained part of the surrounding stroma. The impression was gained that the stroma had "entered" the neoplastic gland through the pores.

It should be stressed that the systematic study of the number of neoplastic glands with pores containing either mucin, inflammatory cells and/or necrotic material was not carried out in any previous investigations.

The purpose of the present study was to systematically quantify the number of glands with pores containing each one of the aforementioned intraglandular materials, at the invading edge of sporadic colonic adenocarcinomas.

Materials and Methods

From a series of 102 surgically resected colonic carcinomas (18), histological sections from twelve patients were selected a priori when fulfilling the following prerequisites: Group I: Three cases with neoplastic glands with pores having predominantly intraglandular mucin at the invading edge (Figure 1), Group II: Three cases with neoplastic glands with pores having predominantly intraglandular neutrophilic granulocytes at the invading edge (Figure 2), Group III: Three cases with neoplastic glands with pores having predominantly intraglandular necrotic material at the invading edge (Figure 3), Group IV: Three cases with neoplastic glands with pores having predominantly intraglandular stroma at the invading edge (Figure 4).

Surgical histological sections were stained with hematoxylin and eosin (H&E) and analyzed at 20 magnifications. By measuring the

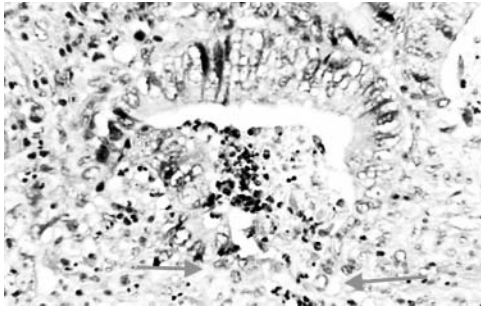


Figure 3. Colon adenocarcinoma. Neoplastic gland with pore at the invasion tumor edge, containing necrotic material that is being released through the pore into the juxtaposed stroma (H&E. $\times 20$).

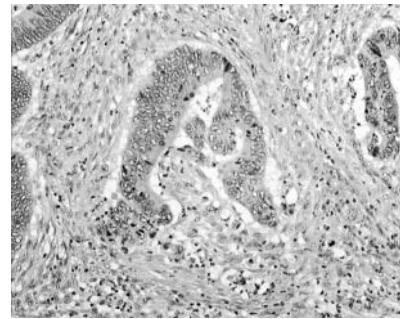


Figure 4. Colon adenocarcinoma. Neoplastic gland with pore at the invasion tumor edge, with stromal invagination "entering" the gland through the pore (H&E. $\times 20$).

length of the invasion front with a conventional translucent millimetre ruler, the number of glands with pores per mm in groups I to IV could be calculated.

The Ethical Committee of the Karolinska Institute, Stockholm, approved this study.

Results

Number of neoplastic glands with pores containing different materials at the invasion edge. The total number of glands with pores was 295 glands in the 12 cases. Table I shows that mucus was present in 68% (n=55) of the 81 glands with pores in group I, granulocytes in 95% (n=41) of the 43 glands with pores in group II, necrotic material in 72% (n=84) of the 117 glands with pores in group III and stromal invaginations in 61% (n=33) of the 54 glands with pores in group IV. On the other hand, different contents as those stipulated for a particular group were recorded in 32% (26/81) of the neoplastic glands in group I, in 5% (2/43) in group II, in 28% (33/117) in group III and in 29% (21/54) in group IV.

The number of neoplastic glands with pores containing different materials at the invasion edge per mm. The mean number of glands with pores per mm in group I was 1.02 (55 glands/54 mm), in group II 0.93 (41 glands/44 mm), in group III 1.22 (84 glands/69 mm), and in group IV 0.62 (33 glands/53 mm).

Discussion

The results of this study showed that four different types of biological materials might be predominantly found in neoplastic glands with pores at the invading edge of sporadic colonic adenocarcinomas. In the present series, the predominance of these materials varied from 61% in group

Table I. The predominant intraglandular content found in neoplastic glands with pores at the invading tumor edge of colonic carcinomas in 12 selected patients: in 3 having mucin (group I), 3 having granulocytes (group II), 3 with necrotic material (group III) and 3 with stromal invaginations.

	Mucus	Granulocytes	Necrotic material	Stromal invagination	All
Group I	55 (64%)	3	17	6	81
Group II	2	41 (89%)	0	0	43
Group III	14	2	84 (79%)	17	117
Group IV	15	0	6	33 (59%)	54
All	86	46	107	56	295

IV, to 95% in group II. The results also indicated that even if each group was selected because of a predominant intraglandular material, other types of intraglandular materials could be simultaneously found in the remaining neoplastic glands with pores in individual sections (from 5% in group II to 32% in group I).

The causes leading to the intraglandular accumulation of these materials remains elusive. Notwithstanding, since the 12 patients neither received preoperative irradiation nor cytostatic medication, these treatment modalities could not have induced the accumulation of these biological materials within the neoplastic glands in this study.

While tumor mucins carry proteolytic enzymes (19-21) able to digest the juxtaposed stroma of the host (8), the possible significance of intraglandular materials contributed by the host (*v.gr.* neutrophils, necrotic material and/or stromal invaginations) on tumor growth, is still poorly understood.

Against that background, several relevant questions arise: i) Which molecular signals bring forth the accumulation of

materials (other than mucins) within the neoplastic glands with pores in colonic carcinomas? ii) Why is a particular material found predominantly in the neoplastic glands of some individuals but not in the neoplastic glands of others? iii) Are these materials necessary for the progression of the tumor? Further studies are necessary to assess whether each one of these intraglandular materials has any bearing on the progression of colonic carcinomas. Only then, some of the aforementioned questions might be elucidated.

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