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Le^x and Le^y Antigen Expression in Human Pancreatic Cancer[1]

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ABSTRACT

Carbohydrate antigens are useful markers for the serological detection of pancreatic cancer. However, data concerning the expression of structurally well-defined carbohydrate antigens in normal and malignant pancreatic tissue is quite limited. The Le^x and Le^y antigens are closely related carbohydrate antigens synthesized on type 2 blood group oligosaccharide side chains of glycolipids and glycoproteins. Monoclonal antibodies anti-SSEA-1 and AH6 recognize "simple" Le^x and Le^y epitopes, respectively, regardless of the length of the carrier carbohydrate. Other monoclonal antibodies recognize Le^x (FH4), sialyl Le^x (FH6, IB9) or Le^y (KH1, CC-1, CC-2) carried only by elongated type 2 side chains with or without internal α1,3 fucosyl substitution. The present comparative immunohistochemical study used tissues of normal pancreas, chronic pancreatitis, and pancreatic cancer to determine the normal expression of Le^x and Le^y antigens in the pancreas and to elucidate any cancer-associated alterations. Le^x-related antigens were not expressed in normal pancreas, expressed in only 10–20% of chronic pancreatitis tissues, but expressed in 50–70% of pancreatic cancer tissues. The frequency of Le^x-related antigen expression in pancreatic cancer tissues was lowest in poorly differentiated cancers. Within a given specimen, at least three or all four of the Le^x recognizing monoclonal antibodies were simultaneously expressed. Unlike Le^x antigens, Le^y-related antigens were expressed in 32–77% of specimens of normal pancreas, with similar frequencies in specimens of chronic pancreatitis and pancreatic cancer. In normal pancreas, simple Le^y was expressed by both ductal and acinar cells, but extended Le^y antigens were expressed only by acinar cells. In pancreatic cancer, extended Le^y antigen expression was found in less than 10% of poorly differentiated tumors. Coexpression among the Le^y-related antigens was less common than with the Le^x-related antigens. Also in cancer specimens, simple Le^x and simple Le^y antigens were often concordantly expressed, whereas extended Le^x and extended Le^y antigen expression was often discordant. Hyperplastic ducts and ductules associated with pancreatic cancer expressed Le^x-related antigens more frequently than morphologically similar lesions associated with chronic pancreatitis. These results demonstrate that Le^x-related antigens are cancer-associated determinants in the human pancreas. The discrepant expression between Le^x and Le^y antigens in these tissues implies altered regulation of fucosyltransferase activity associated with the malignant state.

INTRODUCTION

Cancer-associated antigens possessing carbohydrate immunodeterminants are assuming importance as markers of gastrointestinal neoplasia (1). For example, the CA 19-9 antigen is a sialylated Le^a blood group antigen which, despite its presence in several normal tissues and secretions, exhibits reasonable sensitivity and specificity for the serological detection of pancreatic cancer (2). The DU-PAN-2 antigen is another promising serological marker for patients with pancreatic cancer (3), and although the exact structure of the epitope has not yet been determined, it resides on high molecular weight mucin-type glycoproteins and requires sialic acid for antigenicity (4).

Le^x and Le^y[3] antigens are carbohydrate structures found on type 2 blood group chains of glycoproteins and glycolipids. The carrier carbohydrate chain bearing these epitopes may vary in length, and MAbs[4] that recognize short (simple) or extended Le^x and Le^y antigens have been developed (Table 1). This difference in the length of the oligosaccharide side chain has important implications because certain cancer cells, unlike normal cells, have the rather unique ability to synthesize extended type 2 chain antigens (6, 12). In recent immunohistochemical studies with these MAbs, we observed that in human colorectal tissues, the extended forms of Le^x and Le^y antigens were preferentially expressed by malignant and premalignant tissues, and that antigen expression in premalignant (adenomatous) polyps correlated with histological criteria for malignant potential (13–15).

A few studies have examined the expression of Le^x and Le^y antigens in the sera of patients with pancreatic cancer, noting sensitivities of 22% for Le^x antigens (16), 38–66% for sialylated Le^x antigens (17, 18), and 41% for Le^y antigens (16). However, the expression of these antigens in pancreatic tissues has received very little attention. One study compared the expression of Le^x and Le^y antigens in normal pancreas only (19), and others have examined either Le^x or Le^y antigens in a handful of tissues derived from fetal pancreas (18, 20–22), adult pancreas (21–23), or pancreatic cancer (24). The present investigation was therefore performed to simultaneously characterize the expression of simple and extended Le^x and Le^y antigens in tissues of normal pancreas, chronic pancreatitis, and pancreatic carcinoma with an aim toward identifying cancer-associated alterations.

MATERIALS AND METHODS

Tissues. The tissues used in this study were accumulated from patients in the U. S. and Japan, and included both surgical and autopsy specimens. Care was taken to exclude specimens exhibiting autolysis. Neither the nation of origin nor the method of tissue acquisition influenced the staining results so these tissue groups have been combined.

Twenty-three specimens of normal pancreatic tissue were obtained from 12 surgical resections and 11 autopsies. The surgical specimens represented histologically normal tissue at the resection margin of endocrine or exocrine pancreatic cancers. Most of the autopsy tissues

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[3] The nomenclature Le^x and Le^y is adopted for the structures previously designated X and Y, respectively. The X-Y designation may cause confusion between the antigens related to X and Y chromosomes. The adoption of Le^a and Le^y is based on the fact that they are positional isomers of blood group Le^a and Le^b. However, Le^x and Le^y have been identified as being unrelated to Lewis blood group antigens, *i.e.*, these structures do not represent a subtype of Lewis antigens, which are designated using a, b, c, and d. This statement was previously published (Ref. 28).

[4] The abbreviations used are: MAb, monoclonal antibody; TZ, transitional zone.

Table 1 *Le^x and Le^y antigens evaluated in this study*

Antigenic determinant[a]	Monoclonal antibody	Reference
Le^x-related antigens		
Gal β1,4 → GlcNAc-R; GlcNAc ↑α1,3 Fuc	anti-SSEA-1	5
Gal β1,4 → GlcNAc ↑α1,3 Fuc β1,3 Gal β1,4 → GlcNAc-R ↑α1,3 Fuc	FH4	6
NeuAc α2,3 → Gal β1,4 → GlcNAc ↑α1,3 Fuc β1,3 Gal β1,4 → GlcNAc-R ↑α1,3 Fuc	FH6	7
NeuAc α2,6 → Gal β1,4 → GlcNAc β1,3 Gal β1,4 → GlcNAc-R ↑α1,3 Fuc	IB9	8
Le^y-related antigens		
Gal ↑α1,2 Fuc; Gal β1,4 → GlcNAc-R ↑α1,3 Fuc	AH6	9
Gal ↑α1,2 Fuc β1,4 → GlcNAc ↑α1,3 Fuc β1,3 Gal β1,4 → GlcNAc-R	CC1, CC2	10
Gal ↑α1,2 Fuc β1,4 → GlcNAc ↑α1,3 Fuc β1,3 Gal β1,4 → GlcNAc-R ↑α1,3 Fuc	KH1	11

[a] Gal, galactose; GlcNAc, *N*-acetylglucosamine; Fuc, fucose; NeuAc, *N*-acetylneuraminic acid (sialic acid); *R*, core glycolipid or glycoprotein.

Table 2 *Expression of Le^x and Le^y antigens in human pancreatic tissues*

	Normal pancreas ($N = 23$)				Chronic pancreatitis ($N = 10$)				Pancreatic cancer ($N = 35$)
Monoclonal antibody	Total positive cases	Ducts	Ductules	Acini	Total positive cases	Ducts	Ductules	Acini	Total positive cases
Anti-SSEA-1	0/20	0	0	0	0	0	0	0	25 (71)[a]
FH4	0/20	0	0	0	1 (10)	1	1	1	19 (54)
FH6	0/20	0	0	0	2 (20)	0	0	2	22 (63)
IB9	0/20	0	0	0	2 (20)	1	1	2	20 (57)
AH6	17/22 (77)	16	10	14	10 (100)	10	10	9	30 (86)
KH1	7/22 (32)	0	0	7	4 (40)	3	3	4	11 (31)
CC1	12/22 (55)	0	0	12	3 (30)	2	2	3	17 (49)
CC2	16/22 (73)	0	0	16	5 (50)	3	3	5	13 (37)

[a] Numbers in parentheses, percentage.

were obtained from kidney donors immediately following accidental death according to the protocol of the University of Maryland Department of Pathology. Chronic pancreatitis tissues were obtained from 10 surgically removed specimens.

Primary pancreatic carcinoma tissues were obtained from 35 patients: 28 surgical resections and seven early autopsies. Histological examination revealed that 14 cases were well-differentiated, 10 were moderately differentiated, and 11 were poorly differentiated adenocarcinomas. One cancer was a case of well-differentiated adenoacanthoma, and one was a poorly differentiated pleomorphic carcinoma. Among the cancer specimens, 11 contained normal-appearing pancreatic tissue immediately adjacent to the cancer. This region is referred to as TZ. In all 11 cases, the TZ exhibited certain hyperplastic alterations of ducts and ductules. Because antigen expression in hyperplastic ducts and ductules as well as acinar cells of TZ differed from that of normal pancreas, the results of TZ are presented separately.

All tissues were fixed in formalin, embedded in paraffin, and cut into 5-μm serial sections.

Antibodies. Table 1 lists the MAbs used in this study and their antigenic specificities. The anti-SSEA-1 and AH6 MAbs recognize "simple" Le^x and Le^y antigen, respectively, regardless of the length of the carrier carbohydrate side chain. The other MAbs recognize these antigens only when synthesized on extended oligosaccharide side chains. All MAbs are of the IgM isotype except for FH4 which is IgG_3.

Undiluted hybridoma supernatant was used in the case of CC-1 and CC-2, but for all other MAbs working concentrations were obtained by diluting ascites as follows: anti-SSEA-1 1:1000; FH4 1:200; FH6 1:300; IB9 1:400; AH6 1:400; KH1 1:400.

Staining Methods and Controls. The avidin-biotin complex technique of immunoperoxidase histochemistry was performed as described previously (13). Substitution of PBS for either primary or secondary antibody completely abolished any staining. Other negative controls were prepared by substituting normal mouse IgM (or IgG) for primary antibodies. As positive control, antigen-positive colon cancer specimens were stained simultaneously.

Scoring. A case was considered "positive" if at least 5% of the specimen exhibited antigen expression. This was accomplished by sequentially examining all of the low power (10X objective) optical fields on the slide.

RESULTS

Normal Pancreas. Normal pancreatic tissues did not express any of the Le^x antigens (Table 2, Fig. 1). In marked contrast, however, Le^y antigens were expressed in the majority of these specimens (Table 2). Of the cases that expressed Le^y antigens, an analysis of the cellular distribution revealed that MAb AH6,

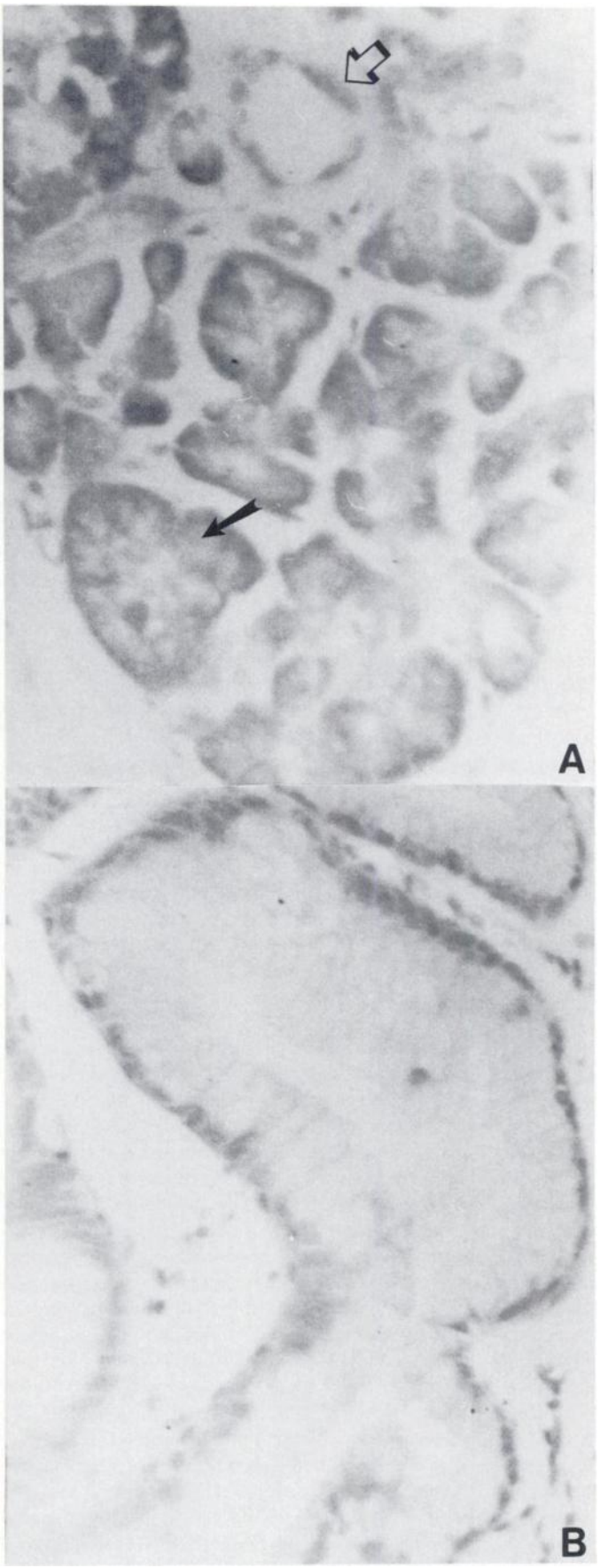

Fig. 1. Le^x expression in normal pancreas. Specimen stained with anti-SSEA-1 demonstrating absence of antigen in: acinar cells (*A*) (*solid arrow*) and ductular cells (*open arrow*), as well as cells of a large pancreatic duct (*B*). (× 250) An identical pattern was noted with MAbs FH4, FH6, and IB9.

which recognizes both short and extended Le^y structures, stained both pancreatic ducts and acini, (Fig. 2*A*), but MAbs KH1, CC-1, and CC-2, which preferentially recognize extended Le^y epitopes, stained only acinar cells (Fig. 2, *B* and *C*). At the subcellular level, both ductal and ductular cells expressed AH6 on cell apical membranes and diffusely in cytoplasm. In acinar cells, all four Le^y MAbs bound to the supranuclear (Golgi) region of the cells. Centroacinar cells and pancreatic islet cells did not express any of the Le^x and Le^y antigens.

Chronic Pancreatitis. In general, the expression of the various Le^x and Le^y antigens in chronic pancreatitis tissue closely resembled that of normal pancreatic tissue in terms of percentage of positive cases, cellular location, and subcellular distribution (Table 2). In the one or two cases that were positive for extended Le^x expression, the staining intensity was very faint. In 20–30% of chronic pancreatitis tissues, extended Le^y antigens were expressed in the ductal and ductular cells, whereas these cells in normal pancreatic tissue were devoid of extended Le^y expression.

Pancreatic Cancer. All four MAbs directed against Le^x-related determinants stained the majority (54–71%) of pancreatic cancer tissues (Table 2, Fig. 3*A*). Thus, these antigens may be considered cancer-associated antigens in the pancreas because of their absence from normal pancreatic tissue. The pancreatic cancer tissues that did not stain with Le^x antibodies tended to be the poorly differentiated specimens, although some of the moderately differentiated and well-differentiated tumors were also negative (Table 3).

Of the four MAbs directed against Le^y-related antigens, only AH-6 stained the majority of pancreatic cancers whereas the MAbs recognizing extended Le^y determinants bound to fewer than one-half of the cases (Table 2). When the degree of differentiation was considered, AH-6 bound to cancers regardless of degree of differentiation, but the other three MAbs rarely if ever stained poorly differentiated tumors (Table 3, Fig. 3*B*).

When simultaneous staining with the four Le^x MAbs were examined on a case-by-case basis, most of the specimens bound either all four MAbs or at least three of the four (Table 4). In contrast, it was uncommon for all four Le^y MAbs to be bound simultaneously in a given tumor, and more often only one or two of these MAbs bound. Table 5 demonstrates the coexpression of Le^x and Le^y antigens on a case-by-case basis in pancreatic cancers. Two phenotypic patterns are presented: (*a*) the coexpression of simple Le^x with simple Le^y antigens; and (*b*) the coexpression of extended Le^x with extended Le^y antigens. The simple Le^x and Le^y antigens (pattern a) were usually expressed in a concordant fashion, and rarely were both antigens absent. However, in pattern *b*, extended Le^y antigen expression was often absent resulting in a greater frequency of discordant expression between extended Le^x and extended Le^y antigens. Considering that the normal pancreas phenotype is Le^x-/Le^y + the patterns observed in cancer tissues are a marked departure from normal.

Lesions Associated with Pancreatic Cancer and Chronic Pancreatitis. Several histological alterations of pancreatic ducts that can accompany pancreatic cancer have been described. Under the general term of "pancreatic ductal hyperplasia," these lesions include nonpapillary hyperplasia (mucous cell hypertrophy), papillary hyperplasia, and atypical hyperplasia (25). The latter is believed to be a premalignant lesion by some authors (26). As noted by Kloppel (26), the various hyperplastic changes of pancreatic ducts may overlap and can also be accompanied by focal proliferation of smaller ducts, known as "adenomatous duct hyperplasia" (27). All of these alterations of duct mor-

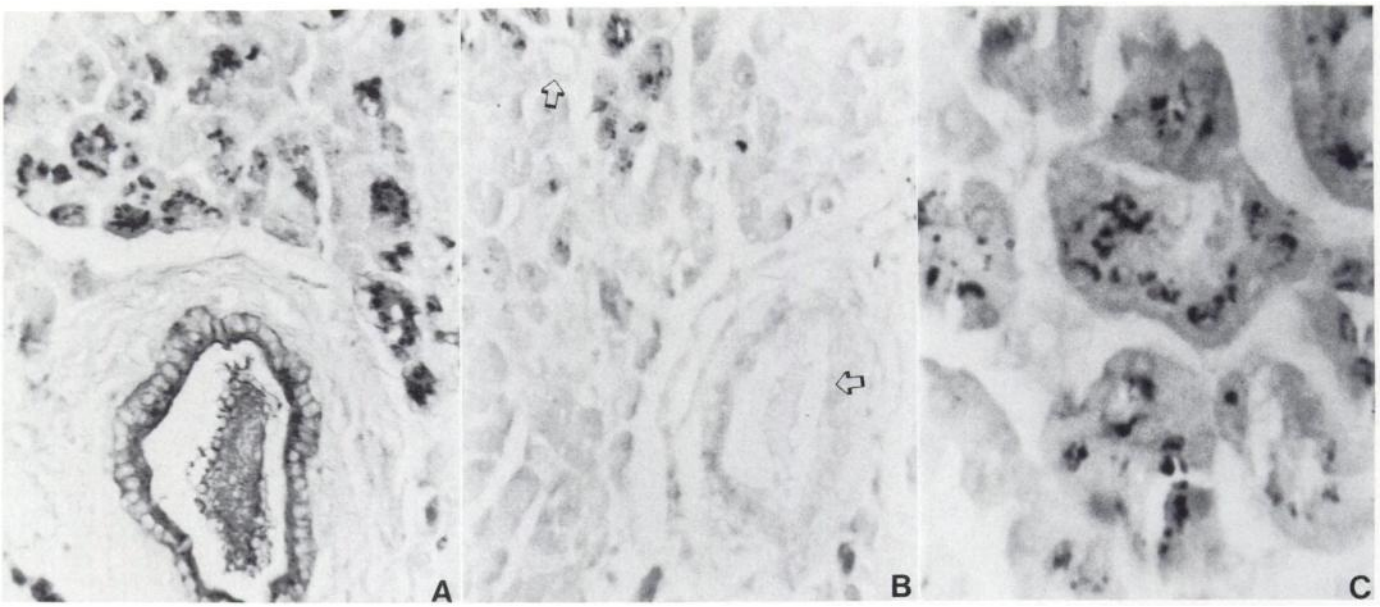

Fig. 2. Le^y expression in normal pancreas. *A*, stained with AH6. Antigen is expressed in ductal cells (apical cytoplasm and apical membrane) and acinar cells (supranuclear region) (× 100); *B*, stained with CC-1. Antigen is expressed only in acinar cells and not in cells of ductules or ducts (*arrows*) (× 100). MAbs KH-1 and CC-2 demonstrated a similar pattern; *C*, stained with CC-1. High power view demonstrating supranuclear staining of acinar cells (× 250).

phology can be observed in association with pancreatic cancer, but (with the exception of atypical hyperplasia) can also be found in the setting of chronic pancreatitis (26). Thus, there is some controversy about whether these morphological changes are precursors of neoplasia or merely reflect a reaction to ductal obstruction.

To address this issue, the present study analyzed specimens of chronic pancreatitis and the TZ of pancreatic cancers for pancreatic ductal hyperplasia (Table 6). There were no examples of atypical hyperplasia but all of the 11 cases of TZ and nine of the 10 cases of chronic pancreatitis demonstrated papillary or nonpapillary hyperplasia (which we term "hyperplastic ducts"). In addition, all cases with hyperplastic ducts also contained some areas of adenomatous proliferation of smaller ducts (termed "hyperplastic ductules").

Perhaps the most striking observation was that the Le^x and Le^y antigens which were completely absent from normal ductal and ductular cells (Table 2), were frequently expressed by cells of hyperplastic ducts and ductules in the TZ adjacent to cancer (Fig. 4). Moreover, the frequency of Le^x-related antigen expression in hyperplastic ducts and ductules of TZ was greater than that of morphologically similar duct lesions in chronic pancreatitis.

Another intriguing observation was the enhanced expression of Le^x and Le^y antigens in normal appearing acinar cells immediately adjacent to cancer (Table 6; Fig. 5). In fact, in several cancer specimens, there was a "gradient" of Le^x antigen expression such that acinar cells remote from the cancer were negative, whereas those in the TZ were positive. Further, Le^x and Le^y antigen expression in acinar cells of TZ was diffusely cytoplasmic (Fig. 5), whereas in normal acinar cells, only Le^y antigens were expressed and expression was limited to the supranuclear region of the cytoplasm (Fig. 2).

DISCUSSION

Extensive biochemical studies have demonstrated that various human adenocarcinomas accumulate a series of novel fucolipids having fucosylated type 2 chain (12). From this work, it has been determined that adenocarcinoma cells, unlike their normal counterparts, have the ability to synthesize the prototype Le^x antigen on elongated carbohydrate side chains (12), and the biosynthetic scheme for chain elongation and fucosylation has been elucidated (28). The availability of MAbs to recognize various Le^x and Le^y antigens has facilitated the investigation of these substances in tissues. In recent comparative immunohistochemical studies of colorectal tissues, we noted that the Le^x and Le^y antigens behave similarly to each other in that the extended forms of these antigens (with or without sialylation) were cancer associated. That is, these antigens were (*a*) not expressed in normal tissues but frequently expressed in cancer specimens, and (*b*) selectively expressed by the premalignant type of colorectal polyps, with antigen expression correlating with malignant potential (13–15).

The present investigation furthers our understanding of fucosylated type 2 chain antigen expression in adenocarcinomas by examining pancreatic exocrine cancers; the second most prevalent type of gastrointestinal malignancy in the U. S. today. These results differ from the previous observations in colorectal tissues in two important ways. First, in the colon, the expression of extended Le^x (FH4, FH6, IB9) and extended Le^y (KH-1, CC-1, CC-2) antigens were similar in that both antigen groups were absent from normal mucosa but were present in the majority of colorectal cancers. However, in the pancreas, while extended Le^x antigen expression displayed this same type of cancer-associated expression, extended Le^y antigens were frequently found in normal pancreatic tissue (localized to acinar cells) and chronic pancreatitis, thereby decreasing the specificity of these antigens for cancer tissues (Table 2). Second, in the colon, the SSEA-1 and AH6 MAbs, which recognize simple Le^x and Le^y epitopes, respectively, exhibited a more "promiscuous" distribution than MAbs recognizing only extended Le^x and Le^y antigens in that the former MAbs stained tissues irrespective of malignant potential. In the pancreas, while AH6 displayed a similar promiscuity in normal pancreas, chronic pancreatitis and pancreatic cancer, SSEA-1 was highly selective and was found only in cancer tissues. Therefore, our results indicate a dissociated processing of two closely related type 2 chain anti-

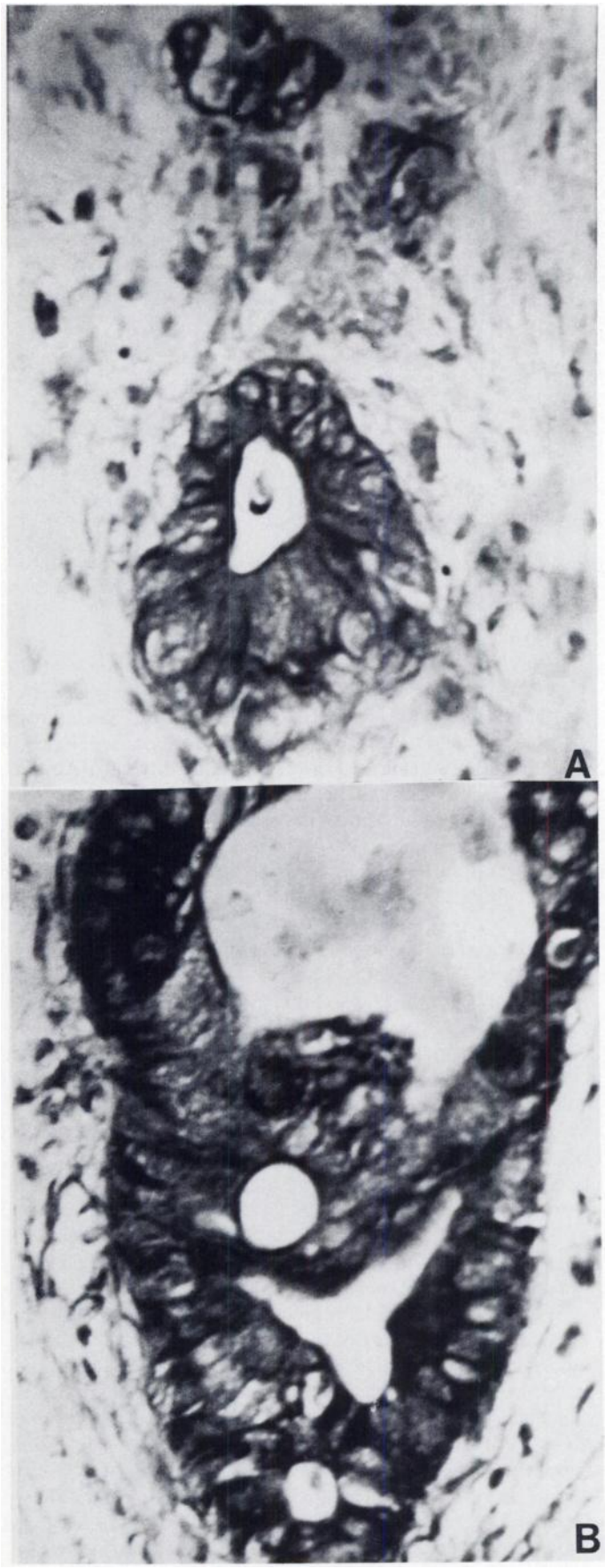

Fig. 3. Well-differentiated pancreatic cancer stained with FH4 (*A*) and CC-2 (*B*). Both antigens are expressed diffusely in cell cytoplasm and on apical cell membranes (× 250).

Table 3 *Expression of Le^x and Le^y antigens in pancreatic cancer*

Antigen	Degree of differentiation: Well (N = 14)	Moderate (N = 10)	Poor (N = 11)
Le^x			
SSEA-1	11 (79%)	9 (90%)	5 (45%)
FH4	9 (64%)	6 (60%)	4 (36%)
FH6	9 (64%)	7 (70%)	6 (55%)
IB9	10 (71%)	8 (80%)	2 (18%)
Le^y			
AH6	11 (79%)	9 (90%)	10 (91%)
KH1	6 (43%)	5 (50%)	0
CC1	10 (71%)	6 (60%)	1 (9%)
CC2	9 (64%)	3 (30%)	1 (9%)

Table 4 *Simultaneous expression of either Le^x or Le^y antigens in individual cases of pancreatic cancer*

Staining pattern	Le^x-related (SSEA-1, FH4, FH6, IB9)	Le^y-related (AH6, KH1, CC1, CC2)
All four MAbs bound	10[a]	4
Any three MAbs bound	10	10
Any two MAbs bound	5	7
Only one MAb bound	6	11
No MAb bound	4	3
Total number of cases	35	35

[a] Number of cases positive.

Table 5 *Coexpression of Le^x and Le^y-related antigens in individual cases of pancreatic cancer*

Pattern	Concordant expression of Le^x and Le^y: + and +	− and −	Discordant expression of Le^x and Le^y: + and −	− and +
Simple Le^x and simple Le^y				
SSEA-1 and AH6	22[a]	2	3	8
Extended Le^x and extended Le^y				
FH4 and KH1	9	14	10	2
FH4 and CC1	13	12	6	4
FH4 and CC2	9	12	10	4
Sialylated-extended Le^x and extended Le^y				
FH6 and KH1	8	10	14	3
FH6 and CC1	13	9	9	4
FH6 and CC2	9	9	13	4
IB9 and KH1	9	14	10	2
IB9 and CC1	15	13	5	2
IB9 and CC2	12	14	8	1

[a] Number of cases. Total number in each row is 35.

Table 6 *Lesions associated with pancreatic cancer and chronic pancreatitis*

Monoclonal antibody	Hyperplastic ducts[a]: Transitional zone (N = 11)	Chronic pancreatitis (N = 10)	Hyperplastic ductules[b]: Transitional zone (N = 11)	Chronic pancreatitis (N = 10)	Acinar cells (Transitional zone) (N = 11)
Anti-SSEA-1	4/9 (44)[c]	0	4/9 (44)	1/9 (11)	5/10 (50)
FH4	5/9 (56)	3/9 (33)	7/11 (64)	3/9 (33)	6/11 (55)
FH6	6/6 (100)	0	7/8 (88)	2/9 (22)	8/10 (80)
IB9	6/10 (60)	2/9 (22)	6/11 (55)	3/9 (33)	6/11 (55)
AH6	10/11 (91)	7/9 (78)	9/11 (82)	9/9 (100)	8/11 (73)
KH1	3/5 (60)	2/9 (22)	2/9 (22)	3/9 (33)	7/11 (64)
CC-1	1/8 (13)	4/9 (44)	1/8 (13)	3/9 (33)	6/9 (67)
CC-2	5/11 (45)	3/9 (33)	1/10 (10)	2/9 (22)	8/11 (73)

[a] Papillary and nonpapillary hyperplasia (25).
[b] Adenomatous duct hyperplasia (27).
[c] Number of positive cases/total number of cases examined. Numbers in parentheses, percentage. Note: the total number of cases available for staining with each Mab differs because ductal lesions are focal and may not be found in all of the serial sections.

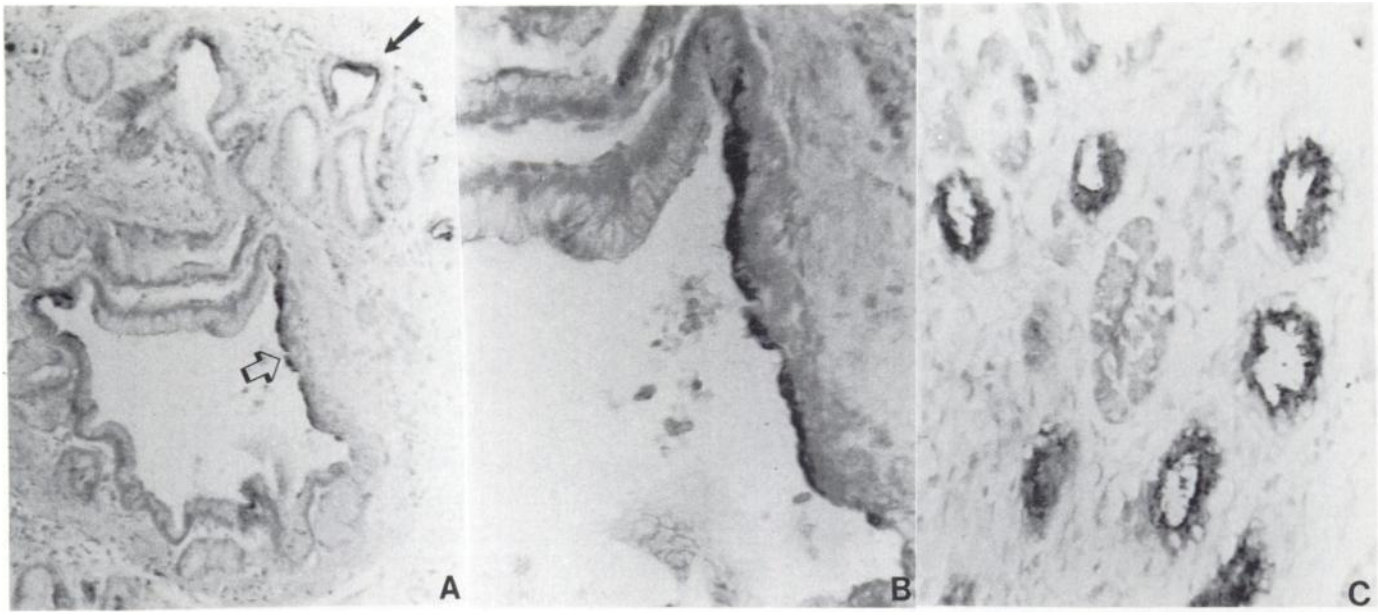

Fig. 4. Le^x expression in cells of hyperplastic ducts and ductules in the transitional zone adjacent to cancer. Stained with anti-SSEA-1. *A*, low power view demonstrating antigen expression in a large hyperplastic duct (*open arrow*) as well as several hyperplastic ductules (*solid arrow*) (× 100). In *B*, higher magnification of *A* showing intense staining of cell apical membrane and apical cytoplasm (× 250). In *C*, several hyperplastic ductules demonstrating SSEA-1 expression diffusely in the cell cytoplasm. Acinar cells in the center are negative in this field (× 100).

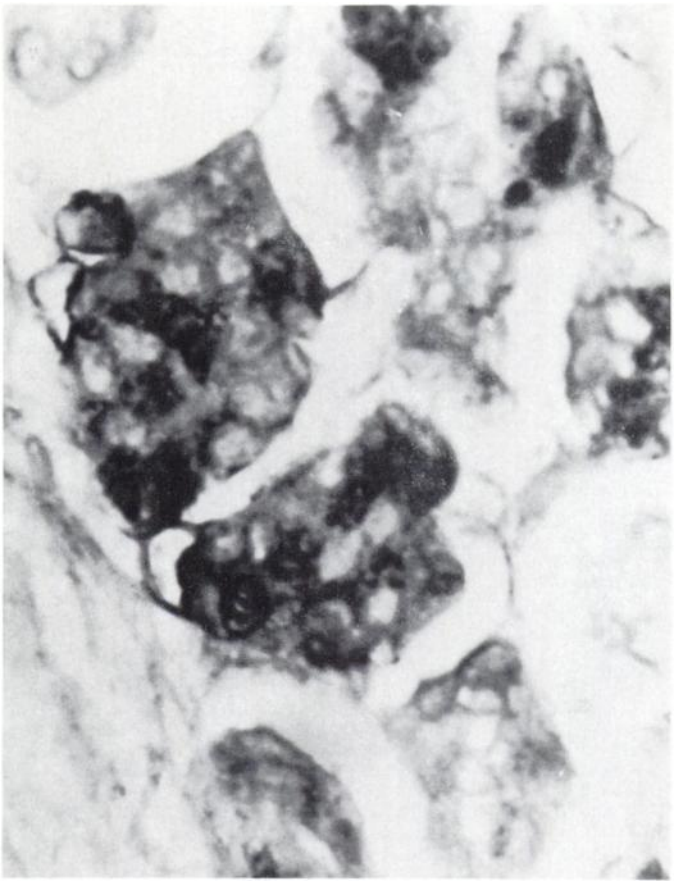

Fig. 5. Le^x expression by acinar cells in the transitional zone adjacent to cancer. Stained with anti-SSEA-1. Antigen is expressed diffusely in the cytoplasm of these acinar cells, in sharp contrast to negatively stained acinar cells of normal pancreas (Fig. 1*A*). (× 400).

gens in normal pancreatic tissue, with all of the Le^x-related antigens behaving as highly specific cancer-associated antigens in the pancreas.

Because the Le^x and Le^y epitopes differ by only one α1,2 fucose residue, the mechanism to explain the differential processing of these two antigens is likely to be a tight regulation of specific α1,2 fucosyltransferase activity by normal pancreatic cells which then becomes altered upon malignant transformation. In the normal pancreas, the considerable expression of Le^y in the complete absence of Le^x suggests that an α1,2 fucosyltransferase is active. Our data suggest that in pancreatic cancer cells, Le^x expression becomes "unmasked," perhaps due to a cancer-associated loss or repression of the α1,2 fucosyltransferase activity (although an α1,2 fucosidase activity in cancer cells which might also explain this observation cannot be ruled out). This raises the possibility that these antigens might be under the control of the *H* or secretor (*Se*) genes which are responsible for α1,2 fucosyltransferase activity in hematopoietic and epithelial cells (29–31). In this retrospective study secretor status was not known so we cannot accurately define the role of the *H* or *Se* gene functions. Another study in which secretor status was not determined also confirmed the predominance of Le^y over Le^x in normal pancreatic tissue (19). Brown *et al.* (23) reported that Le^y was expressed in normal pancreatic tissue regardless of the individual's secretor status, suggesting that the *H* gene-encoded fucosyltransferase may play the major role in Le^y expression in the pancreas.

The process of cellular differentiation has been associated with selective regulation of α1,2 fucosyltransferase activity. For example, in normal cells of both stomach and duodenum (two organs which share a common developmental origin with the pancreas), the blood group antigen phenotype that accompanies upward differentiation (to the surface epithelium) or downward differentiation (to the deep mucosal glands) is governed by *Se* and *H* gene functions, respectively (32). Therefore, altered fucosyltransferase activity in pancreatic cancer might represent a manifestation of aberrant cellular differentiation.

Le^x and Le^y are oncodevelopmental antigens in the colon (13, 15, 33), but whether this applies to the pancreas has not been elucidated. It appears that Le^x antigens are present in fetal pancreas (18, 20–22, 22) and then absent or considerably di-

minished in adult pancreas (18, 20–22, present study). The results herein demonstrate that all of the Le^x and sialylated Le^x antigens are expressed in pancreatic cancer and therefore can be considered as oncodevelopmental antigens in this organ. With Le^y, no information is available concerning antigen expression by fetal pancreas, so the oncodevelopmental nature of this antigen in the pancreas is not yet known.

Most pancreatic adenocarcinomas are believed to have a duct cell origin, with fewer than 5% of cases arising from acinar cells (26). Investigators have compared antigen expression on cancer cells with that of adult or fetal pancreatic cells to draw conclusions regarding the cellular origin of pancreatic cancer. In fact, immunohistochemical studies using MAbs that have been raised against human pancreatic duct cells (34), pancreatic ductal mucin (35), and acinar cells (36), suggest that most pancreatic cancers are of duct cell origin. However, another study demonstrated that all pancreatic adenocarcinomas reacted with an antibody which recognizes antigens associated with acinar differentiation in fetal pancreas (37). Whether Le^x or Le^y antigen expression can help define the cellular origin of pancreatic adenocarcinoma is uncertain. Curiously, the extended Le^y antigens which were only expressed by acinar cells in normal adult pancreas, were also only expressed in moderate or well-differentiated pancreatic cancers.

In saliva, there is a rather strict coexpression of Le^a and Le^x antigens in nonsecretor individuals, and coexpression of Le^b with Le^y in secretors (38). A similar pattern has also been noted on the surface epithelium of stomach and duodenum (32). However, this situation may not apply to all tissues. So, for example, Le^a and Le^x expression in urothelium as well as distal colonic mucosa occurs in both nonsecretors and secretors (39, 40). Thus, different organs manifest individualistic expression of these blood group antigens. In pancreas, based on our present and previous results using the same specimens (41), Le^a expression is similar to Le^x expression in that both antigens are absent from acinar cells; however unlike Le^x, Le^a can be found in centroacinar and ductular cells. The Le^b and Le^y antigens are usually coexpressed in acinar and duct cells. However, we did note five cases in which Le^y was decreased compared to Le^b. It has been observed that in gastric, pancreatic (42), and colorectal cancers (43, 44) there is enhanced Le^b expression, suggesting that an α1,2 fucosyltransferase is activated in malignant gastrointestinal cells. The present results with Le^y antigen, however, suggest a repression of α1,2 fucosyltransferase activity in pancreatic cancer. Thus, it would be important to determine whether the α1,2 fucosyltransferases responsible for Le^b and Le^y synthesis are similar.

In the course of this study, we noted lesions of pancreatic ductal hyperplasia in both pancreatic cancer and chronic pancreatitis specimens. Although hyperplastic ducts and ductules in both of these disease states were morphologically identical, the hyperplastic ducts and ductules in cancer specimens expressed Le^x antigens much more frequently than similar lesions in chronic pancreatitis tissues. This enhanced expression of cancer-associated antigens would support the concept that ductal hyperplasia is a premalignant lesion. However, we would caution that Le^x expression was also greatly enhanced in acinar cells adjacent to cancer, and therefore, increased Le^x expression by hyperplastic ducts, ductules or acini in TZ may simply reflect a reactive response to the nearby cancer, as described with other antigens (37).

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