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EXPRESSION OF THE BREAST CANCER-ASSOCIATED PROTEIN pS2 IN ADENOSQUAMOUS CARCINOMAS OF THE GASTROINTESTINAL TRACT

CORNELIUS WELTER, GERHARD SEITZ AND NIKOLAUS BLIN

The breast cancer-associated protein pS2 is also present in many human gastrointestinal tumors. In contrast to breast carcinomas, gastrointestinal tumors do not express estrogen receptors, indicating that the expression of pS2 is not estrogen-dependent. The pS2 expression was analyzed in 14 adenosquamous tumors of the human gastrointestinal tract. The aim was to investigate if the cell type specific localization of pS2 was limited to the glandular part. The data clearly confirm such a specific compartmentation of the pS2 expression, suggesting pS2 to be a secreted protein. Due to the specific expression, pS2 may become a new and useful diagnostic marker of adenocarcinoma.

The human pS2 gene isolated from the breast carcinoma cell line MCF7 and shown to be under estrogen transcriptional control in a subclass of breast cancer cells was reported to be expressed in normal stomach surface epithelial cells whereas additional gastrointestinal tissues like pancreas and colon do not produce pS2 at all (1, 2). On RNA (northern blots) and protein (immunohistochemistry) level we have demonstrated pS2 expression in various tumors of the human gastrointestinal tract like carcinoma of the stomach, colon, pancreas and biliary tract (3–6). These tumors did not show any estrogen receptors by immunohistochemistry or by receptor assays (7). Thus, the transcription of pS2 is estrogen-independent in these tumors in contrast to breast carcinomas. The mechanism of pS2 gene activation and the biological function of its protein remain to be elucidated.

Adenocarcinoma is the most common type of malignant tumors found in the gastrointestinal tract. Squamous differentiation in gastrointestinal adenocarcinoma is a rare phenomenon. According to the World Health Organization histologic classification, an adenosquamous carcinoma is an adenocarcinoma containing unequivocal squamous cell elements. Such squamous cell differentiation is found in 3–12% of gallbladder carcinomas, whereas it is rarely detected in colorectal and stomach cancer. In the present investigation pS2 expression in this particular tumor type was analyzed and the possible tissue specific activity of the gene elucidated.

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From the Institut für Humangenetik (C. Welter, N. Blin) and Pathologisches Institut (G. Seitz) Universität des Saarlandes, Homburg/Saar, Germany.
Correspondence to: Dr C. Welter, Institut für Humangenetik, Universität des Saarlandes, D-6650 Homburg/Saar, Germany.

Material and Methods

Fourteen advanced adenosquamous carcinomas of the gastrointestinal tract (10 gallbladder, 3 colon, and 1 stomach cancer) were classified and graded according to WHO (8). Specimens were dissected for histopathological investigations. Samples for immunohistochemical staining were fixed in formalin for pS2 detection and embedded in paraffin (for technical details, see 5, 7, 9 and references within).

Immunostaining of pS2 in tissue sections was essentially performed as described previously (2) using the indirect three-step-peroxidase-antiperoxidase (PAP) or the alkaline phosphatase-antialkaline phosphatase (APAAP) method. The monoclonal antibody against the pS2 protein (p2802) (9) was applied at a 1:2 000 dilution for PAP and 1:6 000 for APAAP. For better demonstration of squamous cell differentiation in the carcinomas a monoclonal antibody against cytokeratins (No. 1, 5, 10, and 14 in the Moll catalogue) of stratum corneum (clone 34βE12, Enzo Diagnostics) was used (PAP method).

Results and Discussion

Following our previous observations of pS2 expression in various carcinomas of the gastrointestinal tract the goal of the present study was to analyze the expression of pS2

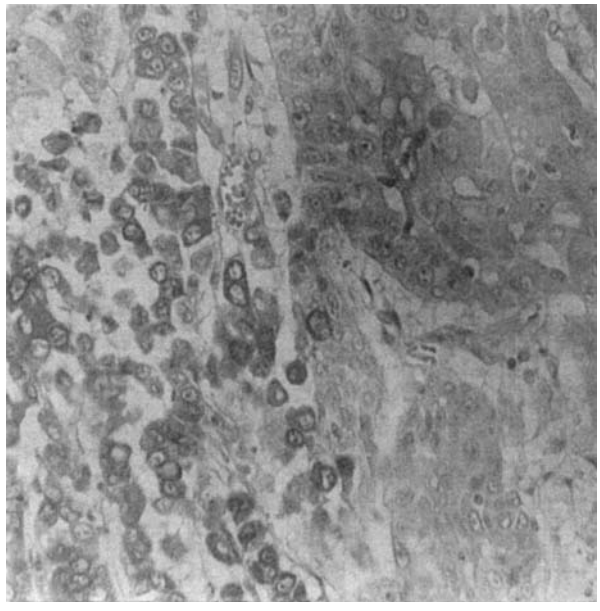

Figure. Immunostaining pf pS2 in a colon adenosquamous carcinoma. The squamous part of the neoplastic tissue remains negative (right part), the glandular part (left) shows a distinctive immunostaining.

in adenosquamous carcinomas of the gastrointestinal tract. Since this type of neoplasm is composed of two uniquely differentiated parts (squamous and glandular) it offers an apt possiblity to address the question whether the pS2 activity is limited to some tissues differentiated in a specific way. To investigate this in more detail, 14 adenosquamous carcinomas of the human gastrointestinal tract were screened for pS2 expression using the p2802 monoclonal antibody. This antibody was previously shown to be devoid of unspecific crossreactivity by extensive studies of various human tissues (2). Our immunostaining experiment (Figure) showed persistently in all 14 tumor samples that only the glandular component of the tumor was (partly) positive whereas the squamous component remained constantly negative (Table).

As in breast tumor cells, the pS2 immonostaining in stomach mucosa cells was predominantly cytoplasmic with an uneven distribution and often perinuclear localisation corresponding to the Golgi apparatus was noted (2). This result may indicate that the pS2 gene encodes a secretory protein. The data from the adenosquamous tumors also support the conclusion that the pS2 protein is being secreted due to its exclusive localisation in the glandular tissue of this carcinoma type. In squamous cells of the portio and in cells of squamous carcinoma, the pS2 expression remains negative (unpublished data). The function of the pS2 protein presently remains unknown but the first structural evidence suggests that pS2 might be a growth factor or an enzyme secreted to the extracellular matrix (10). However, growth-stimulating properties have not been demonstrated until now, although pS2 has a sequence similarity with the insulin-like growth factor and porcine pancreatic spasmolytic polypeptide and shares immunological properties with epidermal growth factor (11). Luqmani et al. (12) concluded in their immunohistochemical study of pS2 in various human epithelial tumors that this antigen may become a useful marker of adenocarcinomas in human neoplasms apart from those in the breast. Our previous study of the significance of pS2 expression in gastrointestinal tumors and our present data in human adenosquamous carcinomas support this view and indicate divergent differentiation of this tumor type into two entities.

Table

Results of pS2 immunostaining in all gastrointestinal adenosquamous carcinomas investigated

No. of samples	Tumor type	Glandular part	Squamous part
1	stomach cancer	+	−
3	colon cancer	+	−
10	gall bladder cancer	++ or +	−

+ = focal pS2 immunoreactions
++ = extensive pS2 immunoreactions
− = no pS2 immunoreactions

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