High Levels of Endogenous Tumor Necrosis Factor–Related Apoptosis–Inducing Ligand Expression Correlate With Increased Cell Death in Human Pancreas

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Objectives: Type 1 diabetes (T1D) has been characterized by the T cell–mediated destruction of pancreatic β cells. Although various members of the tumor necrosis factor (TNF) family, such as Fas ligand or TNF, have recently been implicated in the development of T1D, the lack of TNF-related apoptosis–inducing ligand (TRAIL) expression or function facilitates the onset of T1D. Thus, the goal of the present study was to investigate the expression profiles of TRAIL and its receptors in human pancreas.

Methods: Pancreata of 31 patients were analyzed by immunohistochemistry using antibodies developed against TRAIL and its receptors. Apoptosis was confirmed by Annexin V–fluorescein isothiocyanate binding and terminal deoxynucleotidyl transferase–mediated 2′-deoxyuridine 5′-triphosphate nick end labeling assays.

Results: Acinar cells displayed high levels of TRAIL and death receptor 4, but only low levels of death receptor 5. In contrast, only TRAIL and TRAIL decoy receptors (DcR1, DcR2) were detected in ductal cells. Similarly, Langerhans islets expressed only TRAIL and TRAIL decoy receptor. High levels of TRAIL expression in pancreas correlated with increased number of apoptotic cells.

Conclusions: Although the expression of TRAIL decoy receptors might be necessary for defense from TRAIL-induced apoptosis, high levels of TRAIL may provide protection for Langerhans islets from the immunological attack of cytotoxic T cells.

Key Words: TRAIL, death-decoy receptors, pancreas, Langerhans islets

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The pancreas consists of both exocrine and endocrine tissues that operate independently to regulate carbohydrate metabolism and digestion in the gastrointestinal tract. Langerhans islets are the discrete units of the endocrine compartment involved in the secretion of insulin and glucagon to control glucose homeostasis. The exocrine pancreas, by contrast, holds clusters of acinar cells full of zymogens (inactive pancreatic digestive enzymes), including trypsin, chymotrypsin, carboxypeptidase, amylase, and lipase. Zymogen activation typically takes place in the duodenum after its secretion through the pancreatic duct.

Type 1 diabetes (T1D) occurs as a result of the pancreatic β-cell destruction induced by an autoimmune reaction.[1,2] The inflammatory mediators that trigger the immune reaction in T1D are also responsible for type 2 diabetes and islet graft failure.[3,4] Various members of the tumor necrosis family, such as tumor necrosis factor (TNF) and Fas ligand (FasL), have recently been implicated in the development of T1D (Sanlioglu AD, PhD, unpublished data). Invading immune cells (activated mononuclear cells) release proinflammatory cytokines such as TNF, interleukin 1β (IL-1β), and interferon-γ to induce pancreatic β-cell death.[5] In addition, these inflammatory cytokines increase the vulnerability of islet cells to autoimmune destruction.[6] For example, TNF production by human islets induces postisolation cell death.[7] Similarly, patients with T1D demonstrate elevated levels of TNF production from islet-infiltrating macrophages and dendritic cells.[8] In addition, up-regulation of Fas in pancreatic islets via macrophage production of inflammatory cytokines is another means of causing β-cell death.[9] Fas expression can be up-regulated by streptozotocin (STZ), a diabetogenic agent which induces β-cell death.[10] To further explore the functional role of the Fas-FasL pathway in the pathogenesis of T1D, Lin et al[11] created cytomegalovirus–human FasL transgenic mice. Interestingly, transgenic mice were more sensitive to low doses of STZ-induced T1D than the control wild-type mice. In similar studies, Miwa et al[12] observed that FasL expression in β cells in rat insulin promoter–FasL–nonobese diabetic (NOD) mice resulted in the earlier onset of T1D because the FasL stimulated IL-1β production that facilitated neutrophil infiltration.[13,14]

The TNF-related apoptosis–inducing ligand (TRAIL) is another TNF family member that interacts with 4 different receptors: TRAIL-R1 (death receptor 4 [DR4]), TRAIL-R2 (death receptor 5 [DR5]), TRAIL-R3 (decoy receptor DcR1), and TRAIL-R4 (decoy receptor DcR2).[15] The DR4 and DR5 are the TRAIL receptors that signal for apoptosis, whereas DcR1

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and DcR2 are unable to induce such signaling because they lack the intracellular death domain.[16] In comparison with other members of the TNF family, TRAIL has distinct cytotoxic and immunologic properties. For example, unlike TNF, which initiates and exacerbates autoimmune diseases, TRAIL can down-regulate immune responses. The potential outcome of TRAIL blockade or TRAIL deficiency has been analyzed in 2 different animal models of autoimmune diabetes.[17] First, TRAIL function was counteracted by an injection of soluble TRAIL receptor into NOD mice, which enhanced the degree of autoimmune inflammation in pancreatic islets and facilitated the onset of diabetes. Second, the delivery of multiple, low doses of STZ into TRAIL-deficient mice resulted in a higher degree of islet inflammation and an earlier onset of diabetes. Although TNF and IFN-γ treatments also up-regulate TRAIL gene expression in pancreatic islets of NOD mice,[18] TRAIL does not induce apoptosis in freshly isolated pancreatic islets. Because of its connection to T1D, this study investigated the endogenous expression profile of TRAIL and its receptors in human pancreas.

MATERIALS AND METHODS

Patient Assessment

Pancreata of 31 patients admitted to the Akdeniz University Hospitals and Clinics were sectioned and immunostained as described below. Patients' written consents (Helsinki Declaration) were obtained before the operation, and the procedures were approved by the Akdeniz University Hospital Committee on Ethics. Retrospective analyses and immunostaining procedures were performed in the Pathology department. Patients' ages ranged from 36 to 74 years, with a median age of 54 years. Fourteen patients were men and 17 were women. The normal tissues were obtained from 19 Whipple resections of duodenal adenocarcinoma. In addition, 12 specimens were derived from patients who underwent resection for gastric adenocarcinoma in which the tumor extended close to the pancreas. Moreover, spleen sections prepared from 6 different patients (n = 6) were also stained using antibodies developed against TRAIL and its receptors. Formalin-fixed and paraffin-embedded tissue blocks from these cases were retrieved from the database of the Department of Pathology. Hematoxylin and eosin–stained sections were prepared from each block, and slides were reevaluated. In each case, there was no evidence of tumor involvement.

TRAIL/TRAIL Receptor Immunohistochemistry on Normal Pancreas

All pancreatic tissue sections were initially stained with hematoxylin and eosin. Immunohistochemistry for TRAIL and TRAIL receptors was carried out as described previously.[19,20] All primary antibodies were obtained from Alexis Biochemicals (Switzerland). The following primary antibodies were used at 1:300 dilution in the staining of pancreatic tissues: antihuman TRAIL monoclonal antibody (mAb) (III6F; ALX-804-326-C100), antihuman TRAIL-R1 mAb (HS101; ALX-804-297A-C100), antihuman TRAIL-R2 polyclonal antibody (ALX-210-743-C200), antihuman TRAIL-R3 polyclonal antibody (ALX-210-744-C200), and antihuman TRAIL-R4 mAb (HS402; ALX-804-299A-C100). Whereas lymph node staining was performed as a positive control, negative controls included pancreatic tissue samples that were stained only with the appropriate secondary Ab. All sections were counterstained with hematoxylin in Figures 1–4, 5B, and 6.

Immunohistochemical Scoring of TRAIL and TRAIL Death-Decoy Receptor Expression

One pathologist (O.E.) with no prior knowledge of the data and blinded to the names of the antibodies was charged with specimen analysis. Both intensity and marker distribution (percentage of the positively stained epithelial cells) were considered for the calculation of the final immunohistochemical staining scores in pancreatic tissues. The intensity of the pancreatic tissue staining was scored as: 0, negative; 1, weak; 2, moderate; and 3, strong. Similarly, the marker distribution was scored as 0, less than 10%; 1, 10% to 40%; 2, 40% to 70%; and 3, greater than 70% of the epithelial cells stained on the sections. The final immunostaining score was assigned by summing the scores of both the intensity and the marker distribution for a given patient.

Annexin V Binding Assay and the Quantification of Apoptosis

Paraffin-embedded blocks were sectioned at 4-μM thickness. After deparaffinization and dehydration, the antigen retrieval process was carried out by boiling samples in a solution containing 0.01 M of citrate buffer for 20 minutes. Proteinase K treatment was then performed for an additional 10 minutes. An Annexin V fluorescent microscopy kit (BD Pharmingen, cat no. 550911) was used for the detection of apoptotic cells in pancreas. To do this, pancreatic sections were washed with 1× phosphate-buffered solution (BD Pharmingen, cat no. 51-6635KC) then with 1× Annexin V binding buffer (BD Pharmingen, cat no. 51-66121E). Sections were then stained with Annexin V–fluorescein isothiocyanate (FITC) (BD Pharmingen, cat no. 51-8074KC) diluted 1:10 in 1× Annexin V binding buffer for 15 minutes at room temperature. After washing in Annexin V binding buffer, Annexin V–FITC–stained cells were analyzed under fluorescent microscopy. In each section, positive and negative cells were counted in randomly selected 200× high-power fields of acinar, ductal, or islet cells (area of each field, 0.06 mm^2). The apoptotic index refers to the percentage of Annexin V–positive cells and calculated from the ratio of Annexin V–stained cells to the total number of cells counted.

Terminal Deoxynucleotidyl Transferase–Mediated 2′-Deoxyuridine 5′-Triphosphate Nick End Labeling Assay

The in situ cell death detection kit POD (Cat no. 11 684 817 910) was used to confirm apoptosis according to the protocol described by the manufacturer (Roche Applied Science, Philadelphia, Pa). The kit is specifically designed for the immunohistochemical detection and quantification of apoptosis at single-cell level based on labeling of DNA strand breaks. In this assay, labeling of DNA strand breaks is accomplished via terminal deoxynucleotidyl transferase enzyme, which catalyzes polymerization of labeled nucleotides to free 3′-OH DNA ends in a template-independent manner.

Then, incorporated fluorescein is detected by antifluorescein antibody Fab fragments from sheep, conjugated with horseradish peroxidase (POD). After substrate reaction, stained cells were analyzed under light microscope. The apoptotic index was calculated as described previously.

Statistical Analysis

The Statistical Package for the Social Sciences 13.0 software for Windows (SPSS Inc, Chicago, Ill) was used to perform the statistical analyses as specifically stated in the results. Statistical significance was considered at 5% probability level ($P < 0.05$). Error bars for all data points in all figures display the ±SEM.

RESULTS

TRAIL and TRAIL Receptor Expression Profiles in Pancreatic Acinar Cells

Before the analyses of TRAIL and its receptor expression profiles in pancreas, specificities of these primary antibodies were confirmed on lymph node sections. As shown in Figure 1, primary antibodies developed against TRAIL and its receptors generate a strong staining pattern on lymph node sections. Conversely, incubation of lymph node sections with the secondary antibody alone (negative control) did not produce any detectable staining.

Normal acinar expression profiles of TRAIL and its receptors in 31 pancreata were revealed using immunohistochemistry as described in "Materials and Methods," and representative images are depicted in Figure 2A. Normality of the group was tested by the Shapiro-Wilk method. Because a Gaussian distribution was not observed, the Friedman test followed by the Wilcoxon signed rank test was applied to reveal the statistical differences in the group. Although DR4 and TRAIL expression levels were the highest in acinar cells compared with other death and decoy receptor expressions, no statistical difference in the expression levels was observed between DR4 and TRAIL (Fig. 2B). As both decoy receptors DcR1 and DcR2 exhibited equivalent levels of expression, DR5 expression was statistically the lowest. Spearman ρ correlation test was administered to determine a possible correlation among the markers. A

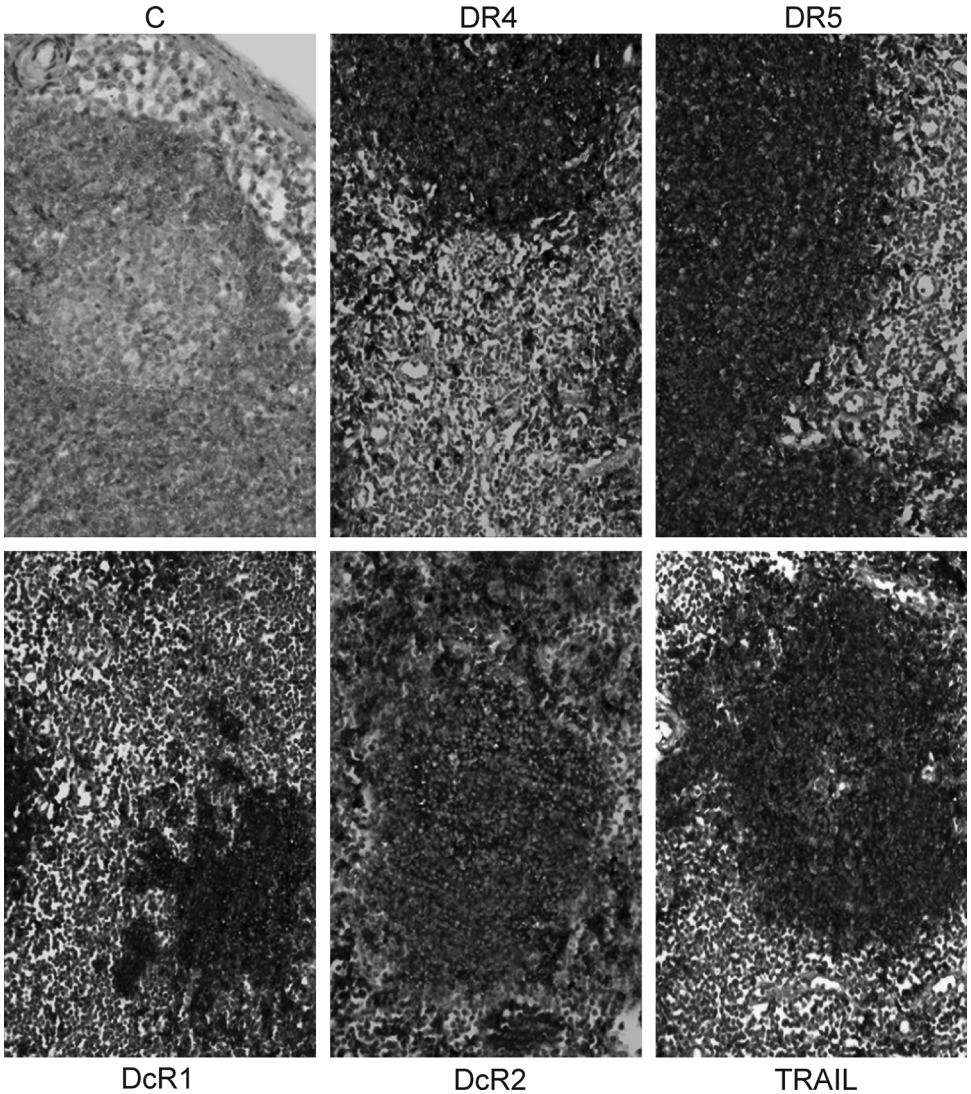

FIGURE 1. Lymph node staining of TRAIL and its receptors. All representative images are provided only from a single patient. C represents negative control staining using secondary antibody alone. Original magnification ×200.

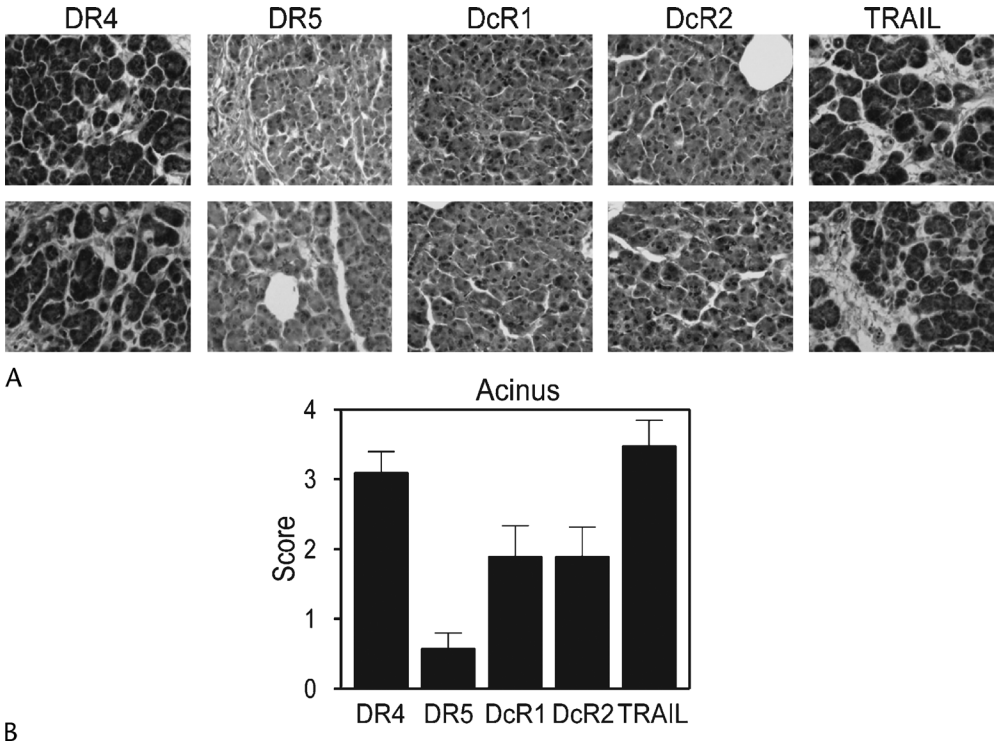

FIGURE 2. Acinar expression profile of TRAIL and TRAIL receptors in pancreas (n = 31). A, Representative images of immunohistochemical staining, with duplicate samples representing 2 different patients are provided. Original magnification ×200. B, Quantitative immunohistochemical scoring results. Error bars represent ±SEM.

positive correlation was detected between the decoy receptors DcR1 and DcR2, as well as between DcR1 and TRAIL (Table 1).

Pancreatic Ductal Staining Profiles of TRAIL and Its Receptors

Immunohistochemical staining of ductal region of pancreas was analyzed microscopically, and representative images from 2 patients are displayed in Figure 3A. Although both decoy receptors and TRAIL were readily detectable, no TRAIL death receptor (DR4 or DR5) expression was observed in ductal cells. As equivalent levels of expression from both decoy receptors were measured, TRAIL expression was statistically the highest (Fig. 3B). In addition, Spearman ρ correlation test demonstrated the presence of a positive correlation between DcR1 and TRAIL (Table 2).

TABLE 1. Spearman ρ Correlation Test Indicating Putative Correlations Detected Among Acinar Staining Profiles of TRAIL and TRAIL Receptors

Spearman ρ Correlation (Acinus)		DR4	DR5	DcR1	DcR2	TRAIL
DR4	Correlation coefficient	1.000	−0.098	−0.197	0.039	0.005
	Significance (2-tailed)		0.601	0.287	0.837	0.980
	N	31	31	31	31	31
DR5	Correlation coefficient	−0.098	1.000	0.182	0.133	0.269
	Significance (2-tailed)	0.601		0.327	0.477	0.143
	N	31	31	31	31	31
DcR1	Correlation coefficient	−0.197	0.182	1.000	0.465*	0.566*
	Significance (2-tailed)	0.287	0.327		0.008	0.001
	N	31	31	31	31	31
DcR2	Correlation coefficient	0.039	0.133	0.465*	1.000	0.240
	Significance (2-tailed)	0.837	0.477	0.008		0.193
	N	31	31	31	31	31
TRAIL	Correlation coefficient	.005	.269	.566*	.240	1.000
	Significance (2-tailed)	0.980	0.143	0.001	0.193	
	N	31	31	31	31	31

*Correlation is significant at the 0.01 level (2-tailed).

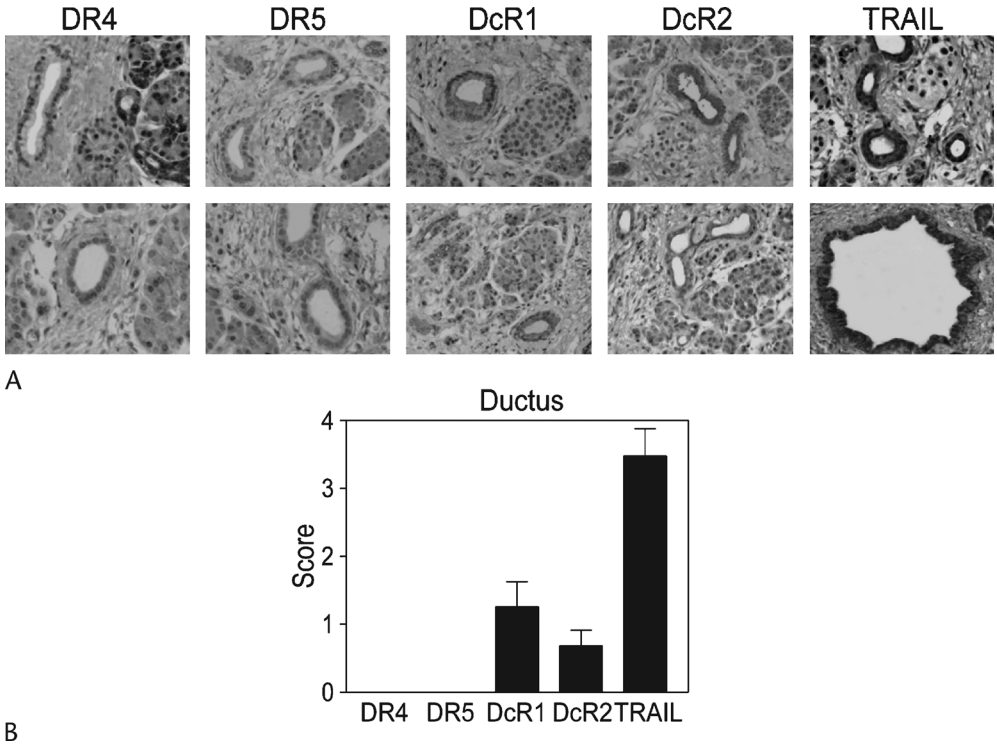

FIGURE 3. The TRAIL and TRAIL receptor expression profiles in ductal cells. A, Representative images of immunohistochemical staining. Original magnification ×200. B, Quantitative assessment of ductal staining patterns of 31 pancreata. Error bars represent ±SEM.

Correlation coefficients for DR4 and DR5 were not assessable because of the lack of expression in ductal cells.

Distinctive Expression Profiles of TRAIL and Its Receptors in Langerhans Islets

Representative images of the immunohistochemical staining of the Langerhans islets using specific antibodies against TRAIL and the TRAIL receptors are provided in Figure 4A. Similar to the finding in ductal cells, the Langerhans islets primarily expressed the TRAIL decoy receptors DcR1 and DcR2 and TRAIL. Although very low levels of TRAIL death receptors were detectable, TRAIL expression was statistically the highest (Fig. 4B). Langerhans islets displayed equivalent levels of DcR1 and DcR2 expressions. Only DcR1 displayed a positive correlation with TRAIL as shown by Spearman ρ correlation test (Table 3).

TABLE 2. Correlation Coefficients Among TRAIL and TRAIL Receptors in Pancreatic Ducts as Revealed by Spearman ρ Correlation Test

Spearman ρ Correlation (Ductus)		DcR1	DcR2	TRAIL
DcR1	Correlation coefficient	1.000	0.151	0.550*
	Significance (2-tailed)		.417	0.001
	N	31	31	31
DcR2	Correlation coefficient	0.151	1.000	−0.060
	Significance (2-tailed)	0.417		0.747
	N	31	31	31
TRAIL	Correlation coefficient	0.550*	−0.060	1.000
	Significance (2-tailed)	0.001	0.747	
	N	31	31	31

*Correlation is significant at the 0.01 level (2-tailed).

Presence of Apoptotic Cells in Pancreas and the Connection to TRAIL and TRAIL Receptor Expression Profiles

Because acinar, ductal, and islet cells in the pancreas displayed differential amounts of TRAIL and TRAIL receptor expression, an Annexin V binding assay was performed on pancreatic sections to determine the level of apoptosis. Whereas fluorescent microscopic views display Annexin V–FITC–stained cells (Fig. 5A), quantitative assessments regarding the apoptotic index are provided below each panel. No difference in the degree of apoptosis was observed among acinar, ductal, and islet cells in pancreas. These results were further confirmed by terminal deoxynucleotidyl transferase–mediated 2′-deoxyuridine 5′-triphosphate nick end labeling (TUNEL) assay on the same sections (Fig. 5B). Quantitative analyses of TUNEL assay did not reveal any difference in the degree of apoptosis among the cell types tested, as shown below each panel in Figure 5B. Moreover, both Annexin V binding assay and TUNEL method detected equivalent levels of cell death for each cell type. However, high levels of TRAIL expression did correlate with increased cell death as revealed by Spearman ρ correlation test (Table 4). Intriguingly, some degree of correlation was also evident

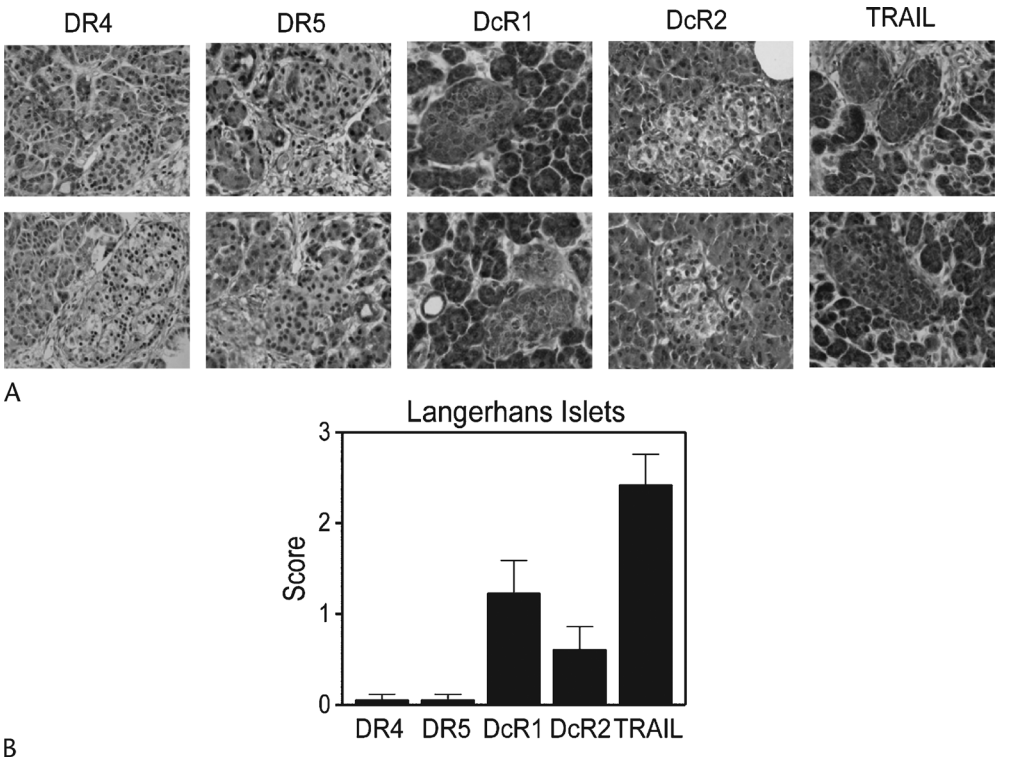

FIGURE 4. The TRAIL and TRAIL receptor expression profile in Langerhans islets. A, Representative images of immunohistochemical staining. Original magnification ×200. B, Quantitative analysis of scoring. Error bars represent ±SEM.

between apoptotic cells and DcR1 expression in acinar versus islets cells.

TRAIL and TRAIL Receptor Expression Profiles in Spleen

Because TRAIL is preferentially expressed by immune cells, we also analyzed TRAIL and TRAIL receptor expression profiles in the spleen using immunohistochemistry for comparison with those observed in the pancreas. Death and decoy receptors, as well as TRAIL, were all expressed in the spleen (Fig. 6A). Statistical analysis of the normality of the group was tested by the Shapiro-Wilk method. Because a Gaussian distribution was not observed, the Friedman test followed by the Wilcoxon signed rank test was applied to reveal the statistical differences in the group. Equivalently high levels of staining were observed in the

TABLE 3. Differential Expression Profiles of TRAIL and TRAIL Receptors in Langerhans Islets as Illustrated by Spearman ρ Correlation Test

Spearman ρ Correlation (Islets)		DR4	DR5	DcR1	DcR2	TRAIL
DR4	Correlation coefficient	1.000	−0.033	−0.115	−0.089	−0.220
	Significance (2-tailed)		0.859	0.538	0.634	0.234
	N	31	31	31	31	31
DR5	Correlation coefficient	−0.033	1.000	0.332	−0.089	0.262
	Significance (2-tailed)	0.859		0.068	0.634	0.154
	N	31	31	31	31	31
DcR1	Correlation coefficient	−0.115	0.332	1.000	0.061	0.450*
	Significance (2-tailed)	0.538	0.068		0.743	0.011
	N	31	31	31	31	31
DcR2	Correlation coefficient	−0.089	−0.089	0.061	1.000	−0.256
	Significance (2-tailed)	0.634	0.634	0.743		0.165
	N	31	31	31	31	31
TRAIL	Correlation coefficient	−0.220	0.262	0.450*	−0.256	1.000
	Significance (2-tailed)	0.234	0.154	0.011	0.165	
	N	31	31	31	31	31

*Correlation is significant at the 0.05 level (2-tailed).

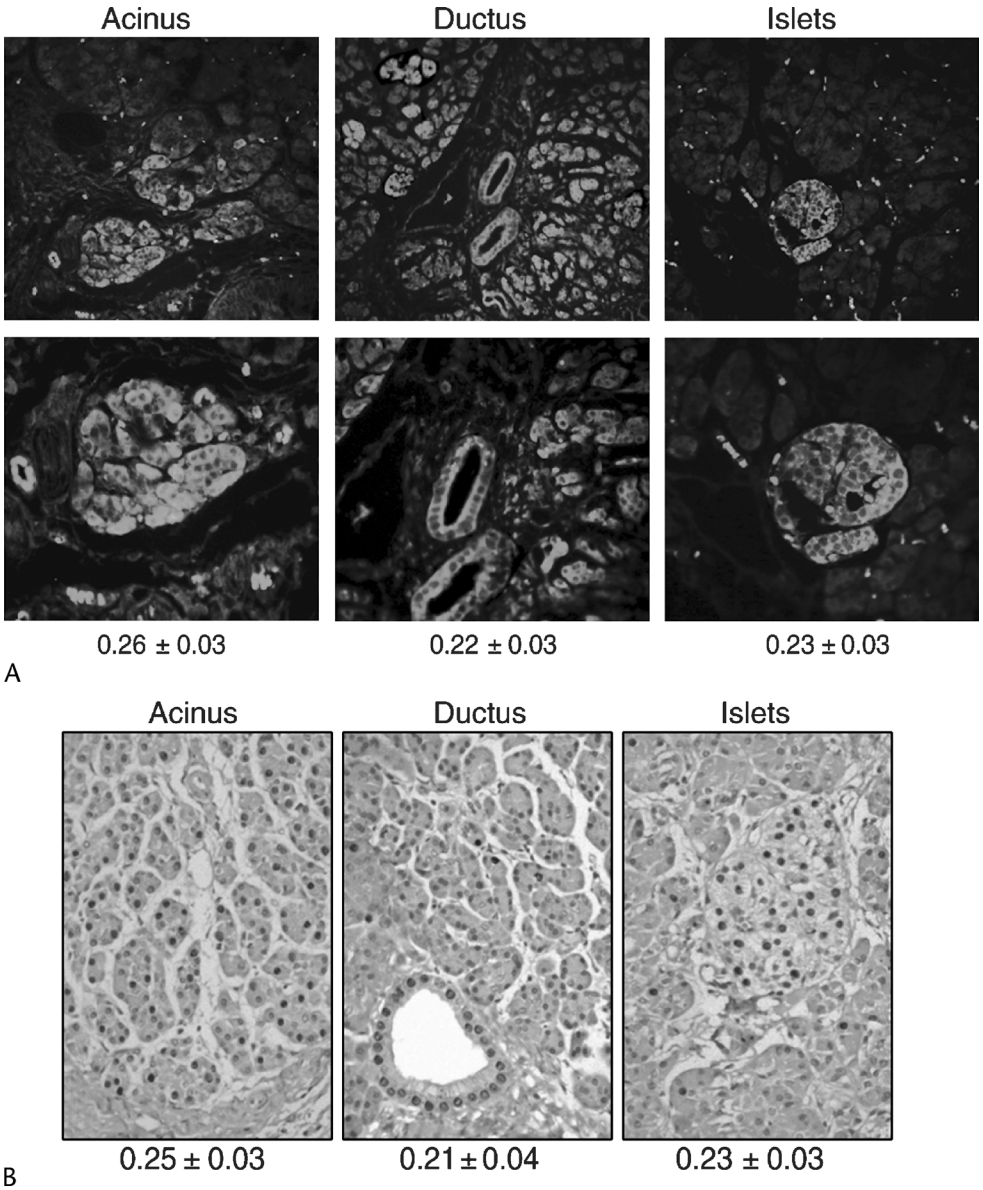

FIGURE 5. A, Annexin V–FITC binding assay on pancreas. Apoptotic cell staining and the quantifications were performed as described in "Materials and Methods." Upper row shows representative views of acinar (left), ductal (middle), and islet cell (right) at 200× magnification; lower row shows high-power magnifications (400×) of the same fields. Quantitative analysis of apoptotic scoring is given below each panel as a percentage of Annexin V–stained cells. B, TUNEL assay on pancreas. Only 1 representative image (views of acinar [left], ductal [middle], and islet cells [right]) for each condition is shown for clarity. The numbers below each panel represent quantitative analysis of TUNEL assay results in percentages (apoptotic index). Original magnification ×200. Error bars represent ±SEM in both panels.

spleen for all the TRAIL markers tested (Fig. 6B). Despite the fact that the mean DcR2 expression level was low in comparison with other markers, the difference was not statistically significant. Nonetheless, our results demonstrated that the amount of TRAIL and its receptor expression on average were much higher in the spleen than those of the pancreas (Fig. 6B).

DISCUSSION

Immunostaining approaches, either by immunohistochemistry or flow cytometry, have become valuable tools to analyze TRAIL and TRAIL receptor expression profiles in tissues such as prostate, lung, breast, and synoviocytes.[19–22] Despite its anticancer properties,[23–25] TRAIL has also recently been implicated in the destruction of pancreatic β cells because the analysis of activated T cell lines derived from 29 children with new-onset T1D showed an increase in TRAIL expression in the infiltrating β cell–specific T cells ($CD56^+$).[26] The fact that TRAIL induced much stronger cytotoxicity to the human β cell lines (CM and HP62) than did TNF and FasL further strengthened the argument of TRAIL mediating β-cell destruction.[27]

TABLE 4. Correlation of Apoptotic Cells With TRAIL and TRAIL Receptor Profiles in Pancreas

Apoptotic Cell Staining		Acinus	Ductus	Islets
DR4	Correlation coefficient	−0.044	NA	−0.218
	Significance (2-tailed)	0.813		0.239
	N	31		31
DR5	Correlation coefficient	0.135	NA	0.177
	Significance (2-tailed)	0.468		0.342
	N	31		31
DcR1	Correlation coefficient	0.383*	0.310	0.433*
	Significance (2-tailed)	0.034	0.090	0.015
	N	31	31	31
DcR2	Correlation coefficient	0.001	0.012	−0.168
	Significance (2-tailed)	0.994	0.947	0.367
	N	31	31	31
TRAIL	Correlation coefficient	0.872†	0.693†	0.978†
	Significance (2-tailed)	0.000	0.000	0.000
	N	31	31	31

*Correlation is significant at the 0.05 level (2-tailed).
†Correlation is significant at the 0.01 level (2-tailed).
NA indicates not applicable.

Because of its presumed role in the development of T1D, our study assessed TRAIL and TRAIL receptor expression profiles using immunohistochemistry of 31 human pancreata specimens. Our investigation revealed varying degrees of TRAIL and TRAIL receptor expression in the acinar cells. Although DR4 and TRAIL expressions were the highest, there were substantial levels of TRAIL decoy receptor expression in the acinar cells. Ductal cells, in contrast, exclusively expressed TRAIL and TRAIL decoy receptors. Similar observations were made for the Langerhans islets, despite the presence of very low, but detectable, amounts of TRAIL death receptor expression. Thus, the absence of TRAIL death receptor expression was clearly evident in ductal cells and in Langerhans islets. Despite equivalent levels of apoptosis observed among the cell types tested, high levels of TRAIL expression were correlated with the increased amount of cell death in all 3 cell types (acinar, ductal, and islet cells) analyzed. This may strengthen the arguments implicating TRAIL as an apoptosis-inducing agent in pancreas.[26,27] Some degree of correlation was also observed between Annexin V–FITC–stained cells and DcR1 expression in acinar versus islet cells, but not in ductal cells. Although the biologic basis of this finding is not clear and cannot be explained with our current knowledge, our studies on prostate cancer cells suggest that DcR1 expression by itself is not sufficient to prevent TRAIL-induced cytotoxicity.[24,28] Yet, the removal of DcR1 from the membrane increased the susceptibility of primary islet cells to TRAIL-induced apoptosis,[27] further suggesting the potential protective roles of TRAIL decoy receptors in protecting Langerhans islets from the death ligand–mediated apoptosis.

The expression profiles of TRAIL and TRAIL receptors on fetal pancreas were recently analyzed using laser scanning confocal microscopy.[29] In this study, TRAIL-positive cells were primarily positioned on the periphery of the pancreatic islets. Although DcR1 and DcR2 expressions were noticeable on a few cells, no DR4 or DR5 expression was detected in the pancreatic islets. Our study however revealed uniform but substantial levels of TRAIL and TRAIL decoy receptor expressions in the Langerhans islets. Differences between these

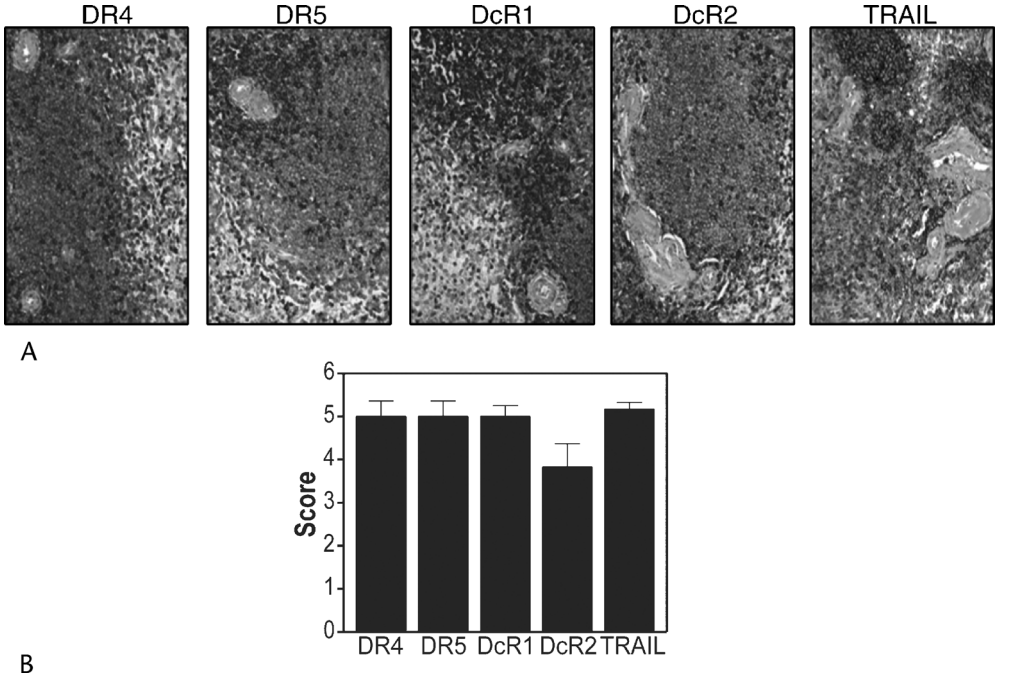

FIGURE 6. Expression profile of TRAIL and TRAIL receptors in spleen (n = 6). A, Examples of immunohistochemical staining representing a single patient are provided. Original magnification ×200. B, Quantitative immunohistochemical scoring results. Error bars represent ±SEM.

2 studies can be attributed to the use of fetal versus adult pancreata, in addition to the differences in antibodies used. Nevertheless, the presence of substantial levels of TRAIL decoy receptor expression in the absence (or trivial levels) of TRAIL death receptor expression may suggest that Langerhans islets are naturally resistant to the cytotoxic effects of apoptotic ligands. Not surprisingly, certain differences in the TRAIL sensitivity of cancerous versus normal islets were also reported.[30] Although β cell lines were sensitive to TRAIL treatment, normal primary islet cells isolated from most donors displayed resistance to TRAIL-mediated cytotoxicity.[27] Moreover, because TRAIL-transduced dendritic cells protected mice from acute graft-versus-host disease and leukemia relapse through the suppression of antigen-specific T-cell activity,[31] it is reasonable to assume that high levels of TRAIL expression may provide immune protection to Langerhans islets. In summary, our study revealed differential expression profiles of TRAIL and TRAIL receptors in the pancreas with potential implications in T1D. More importantly, this study demonstrated that high levels of TRAIL expression correlated with increased amount of cell death in the pancreas.

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