Intestinal T-cell and natural killer (NK)-
cell lymphomas are clinically aggressive
and can be challenging to diagnose in
small endoscopic biopsies. We describe
8 patients in whom atypical NK-cell lymph-
phoproliferative lesions mimicked NK- or
T-cell lymphoma. The patients (2 men; 6 women; ages 27-68 years) presented
with vague gastrointestinal symptoms
with lesions involving stomach, duode-
um, small intestine, and colon. At endos-
copy, the lesions exhibited superficial
ulceration, edema, and hemorrhage. Biop-
sies revealed a mucosal infiltrate of atypi-
cal cells with an NK-cell phenotype (CD56+/TIA-1+/Granzyne B+/cCD3+),
which displaced but did not invade the
glandular epithelium. Epstein-Barr virus–
coded RNA in situ hybridization was
negative, and T-cell receptor-γ gene re-
arrangement showed no evidence of a
clonal process. Based on an original diag-
nosis of lymphoma, 3 patients received
aggressive chemotherapy followed by au-
tologous bone marrow transplantation in
2. Five patients were followed without
treatment. However, no patient developed
progressive disease or died of lymphoma
(median follow-up, 30 months). Repeat
endoscopies in 6 of 8 patients showed
perfusion or recurrence of superficial
gastrointestinal lesions. This unique en-
tity mimics intestinal and NK-/T-cell lym-
phomas on endoscopic biopsies and can
result in erroneous diagnosis, leading to
aggressive chemotherapy. We propose
the term “NK-cell enteropathy” for this
syndrome of as yet unknown etiology.
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Methods

In each case, an endoscopic biopsy led to a pathologic consultation by the Hematopathology Section, National Cancer Institute, National Institutes of Health, Bethesda, MD between 2006 and 2010. Clinical information, endoscopic findings, and other relevant laboratory data were provided along with the biopsy material. The Institutional Review Board of the National Cancer Institute approved this study. All biopsy specimens were fixed in 10% buffered formalin, embedded in paraffin, sectioned (4 μm), and stained with hematoxylin and eosin. Additional studies were performed at the National Institutes of Health. Immunohistochemical stains were performed using an automated immunostainer (Ventana Medical Systems) according to the company’s protocols, as previously described.19 Tissue sections were incubated with antibodies specific for Ki-67 (MIB-1), CD20 (L26), CD3 (F7.2.38), CD8 (144B), CD30 (Ki-1), CD68 (KP1), CD5 (CD5/54/F6), CD7 (CBC.37), and EBV-LMP-1 (CS1-4) (Dako North America); CD4 (4B12) and CD56 (1B6) (Novocastra); PAX-5 (Transduction Labs); CD138 (B-B4) (Serotec); CD10 (56C6; Lab Vision); TIA-1 (2G9A10F5) (Beckman Coulter); Granzyme B (BnB7; Monosan); and CD43 (L60 (BD Biosciences). In situ hybridization for EBV-encoded RNA was conducted on formalin-fixed paraffin sections using a fluorescein isothiocyanate–labeled oligonucleotide probe supplied by Ventana on an automated stainer (Ventana-Benchmark).20 Visualization was achieved using the ISH iView system with alkaline phosphatase and nitroblue tetrazolium/5-bromo-4-chloro-3-indolyl-phosphate substrate, with Fast red as contrast. Multi-color flow cytometry was performed in selective patients, using a panel of T cell- and NK cell-associated fluorochrome-conjugated antibodies.21 Molecular analysis to assess rearrangement of the T-cell receptor-γ chain gene was performed using polymerase chain reaction on formalin-fixed, paraffin-embedded tissue, according to standard laboratory protocols in Clinical Laboratory Improvement Amendments-approved laboratories using the published methods of either McCarthy et al22 or Trainor et al.23 Both methods have a sensitivity of more than 90% for the detection of a clonal T-cell process.

Results

Clinical features

Clinical features are summarized in Table 1. The patients included 2 men and 6 women (male/female ratio = 1:3; between the ages of 27 and 68 years; mean, 47 years; median, 46 years). Seven patients...
were of white origin, whereas one patient had Hispanic ethnicity. Seven patients presented with vague gastrointestinal symptoms, including abdominal pain, constipation, diverticulosis, and reflux. One patient was asymptomatic and underwent elective colonoscopy because of a family history of colon cancer (case 1). There was no prior history of celiac disease, inflammatory bowel disease, or malabsorption. None of these patients had clinical evidence of any lymphadenopathy or organomegaly. Endoscopic examinations showed multiple small lesions (mucosal hemorrhages, target lesions, or superficial bleeding ulcers 1-2 cm; Figure 1). In 4 patients, the lesions were identified in a single gastrointestinal site (4 of 8; 50%) involving stomach (1) and colon (3), respectively. The other 4 had involvement of multiple gastrointestinal sites (stomach, duodenum, small intestine, and colon). An extensive imaging workup (including computed tomography scan, magnetic resonance imaging, positron emission tomography scan) was performed in all patients and showed no evidence of lymphadenopathy, organomegaly, or masses. Two patients had biopsies of tonsil and lymph node, respectively (patients 1, 5).

**Follow-up and clinical management**

All patients were closely followed by regular imaging studies and clinical evaluation. During the follow-up period (22-120 months; median, 30 months), continued endoscopic evaluation and repeat biopsies revealed persistence of lesions in the large bowel and other gastrointestinal sites in 6 patients. However, no progression was noted in terms of size or extent of involvement. One patient refused repeat endoscopy (patient 3) but has had persistence of vague gastrointestinal symptoms. One patient has not undergone repeat endoscopy after chemotherapy for an initial suspected diagnosis of lymphoma (patient 8). In 1 patient (patient 1), implementation of a gluten-free and lactose-free diet led to a significant improvement clinically, with the lesions reduced in number and size, but histologic features of celiac disease were not identified. Moreover, recurrence was noted on the restricted diet with persistent lesions observed during annual endoscopic evaluations and biopsies performed over a 7-year period.

In 3 patients (patients 5, 7, and 8), chemotherapy with the CHOP (cyclophosphamide, adriamycin, vincristine, and prednisone) regimen was initiated; and in 2 patients (patients 5, 8), this was followed by autologous bone marrow transplantation because of the presumed aggressive nature of the disease. These 3 patients are well and asymptomatic after 22 to 120 months of follow-up. However, 2 of the 3 who underwent repeat endoscopy have had recurrence of the gastrointestinal lesions confirmed by biopsy. No further therapy was instituted in these patients because of reassessment of the findings after pathologic consultation with the authors.

**Histologic findings**

The gastrointestinal biopsies showed similar morphologic features among all patients. There was expansion of the lamina propria by a relatively well-circumscribed but confluent infiltrate of intermediate to large-sized cells with irregular nuclei, inconspicuous nucleoli, finely clumped chromatin, and a moderate amount of pale cytoplasm (Figure 2). In early-phase lesions, the mucosal glands were displaced because of dense atypical cellular infiltrate; however, in advanced stages, sheets of atypical cells with destruction of mucosal glands were noted. There was in general an absence of epitheliotropism identified in glandular epithelium. No angiocentricity or angiodestructive pattern of growth was seen in any patient. Focal infiltration of the submucosa was seen rarely, but in most instances the muscularis mucosa, if observed, was intact. Apart from areas of mucosal ulceration, necrosis was absent, but focal apoptotic bodies were present. Focal superficial hemorrhage was observed in conjunction with some of the infiltrates. A rim of small mature lymphocytes (mainly B cells) and a polymorphous infiltrate of eosinophils, plasma cells, and histiocytes surrounded the atypical infiltrates or was present in the base. In some cases, the adjacent mucosa contained lymphoid follicles. No villous atrophy or crypt hyperplasia was identified in any sample. Peripheral blood and bone marrow specimens obtained (6 of 8 patients) around the time of the gastrointestinal biopsies showed neither atypical cells nor increase in large granular lymphocytes. In 2 patients, additional biopsies of tonsil and lymph node were performed (patients 1, 5). These were histologically unremarkable.

**Immunohistochemical and molecular results**

The atypical cells in the gastrointestinal biopsies expressed CD56, CD7, cytoplasmic CD3, TIA-1, and/or Granzyme B, but not CD5, CD4, CD8, CD10, CD20, CD30, EBV-latent membrane protein (LMP), PAX-5, CD138, or CD68 (Figure 3; Table 2). CD2 was negative in 3 cases and focally positive in one. This immunophenotype suggested an NK-cell origin of the atypical cells. The proliferative fraction as determined by MIB-1 or Ki-67 was low
EBV-encoded RNA was not detected in any patient, by in situ hybridization technique using EBV-encoded RNA probe. Patient 1 had a biopsy of tonsil showing few clusters of CD56+ CD3+ lymphoid cells in a background of reactive hyperplasia, not thought to be indicative of lymphoma. Flow cytometry was performed on lymphoid cells isolated from the biopsy specimens in cases 1 and 5. The cells failed to express surface CD3 but were positive for cytoplasmic CD3 and strongly positive for CD56 and CD7. The cells were negative for CD4, CD8, and CD5. Because of limited material available in these endoscopic biopsies, flow cytometry was not performed on additional cases. Polymerase chain reaction studies for clonal rearrangement of the T-cell receptor-γ gene were performed in each case and failed to identify evidence of a clonal process.

**Discussion**

In this report, we describe a unique series of 8 patients, who presented with vague abdominal symptoms resulting from persistent, multiple,
mucosal involvement of the gastrointestinal tract by an extensive atypical NK-cell lymphoproliferative process. Among all patients, a diagnosis of “NK-cell lymphoma” was suspected at initial review because of the strong expression of CD56 and cytoplasmic CD3, in the absence of clonal T-cell receptor-γ gene rearrangement by PCR analysis. Negative studies for EBV essentially eliminated the diagnosis of extranodal NK/T-cell lymphoma. All patients were extensively investigated by invasive procedures for an aggressive lymphoma, but lesions were limited to the gastrointestinal tract. Three patients ultimately received high-dose chemotherapy with or without bone marrow transplantation because of a presumptive diagnosis of lymphoma. The etiology of this process is unknown; but based on the protracted but indolent course in all patients, we think it is benign, although clonality cannot be determined in these NK cell-derived lesions. Cytogenetic studies were attempted in case 1 but were unsuccessful because of limited cell recovery and the absence of sufficient metaphases. In the future, additional cytogenetic or molecular studies would be of interest to investigate the presence of genetic aberrations. Based on our current data, we propose the term “NK-cell enteropathy” for this novel condition.

This series is unique in several aspects. Atypical but indolent NK-cell proliferations at extranodal sites have not been documented in the literature and are not recognized as a form of neoplasia in the new World Health Organization classification of hematopoietic and lymphoid tissue. We previously reported the first patient (patient 1) as an atypical NK-cell lymphoproliferative lesion in gastrointestinal tract. Since then, we have encountered 7 additional patients with a remarkably similar clinical syndrome and pathologic features. Recognition and detailed description of this entity are critically important to prevent misdiagnosis as an aggressive disease, such as NK-cell or T-cell lymphoma.

The differential diagnosis at the time of biopsy included EATL. Two forms of EATL are recognized in the World Health Organization classification, referred to as type I and type II, also termed the monomorphic variant. Both are characteristically associated with celiac disease and are of cytotoxic T-cell origin. However, the type II form also can occur sporadically and more closely mimics NK-cell enteropathy. The cells are CD56^+ and generally round and moderate in cell size. In addition, type II EATL is usually CD8^+, which was negative in all cases in the present series. In contrast to NK-cell enteropathy, the infiltrate shows marked epitheliotropism, which was largely absent. The absence of clonal T-cell gene rearrangement further precludes a diagnosis of EATL in the present cases.

Celiac disease is an intestinal inflammatory disorder induced by dietary gluten in genetically susceptible persons, with extensive expansion of intraepithelial cytotoxic T lymphocytes (CTLs). Genetic reprogramming of these CTLs leads first to oligoclonal expansion, gluten-independent tissue damage, and latter uncontrolled CTL proliferation leading to malignant lymphoma. It has been suggested that EATL is a stepwise transformation of CTLs into NK-like T cells by the underlying immunopathology. These observations might lead to speculation that NK-cell enteropathy may represent a precursor lesion of aggressive NK-cell lymphomas. Indolent NK-cell proliferations, progressing to aggressive NK-cell malignancies after a long follow-up, are rare but are described in 2 case reports. However, in our case series, persistence of the gastrointestinal lesions without evidence of clinical or pathologic progression argues against this eventuality.

NK cells (CD16/CD56^+) are a subset of lymphocytes associated with innate immunity and cytotoxic function against viruses and tumor cells, with primary distribution in peripheral blood, lymphoid tissue, and spleen. Less is known regarding the presence and function of NK cells in epithelial tissues. NK cells have been detected in the gut, where their prime function is to provide innate immunity. A subset of NK cells in gastrointestinal mucosa is devoid of cytotoxic function and has low levels of TIA-1 and Granzyme B. Because the NK-cell infiltrates in all of our patients had relatively high expression of Granzyme B and/or TIA-1, it appears that functionally the NK cells in these lesions were primed for the cytotoxic function, most probably responding to local inflammation, autoimmunity, or viruses.

It has been recently reported that NK cells can mediate hapten-specific recall responses, independent of B cells and T cells, in a model of contact hypersensitivity. In humans, NK cells have been shown to home to inflamed skin in various conditions, such as psoriasis, atopic dermatitis, and lichen planus. Among our series, the presence of persistent lesions in gastrointestinal tract with minimal or no involvement at other anatomic sites indicates that the NK-cell proliferation was primarily a localized phenomenon in the gastrointestinal tract, most probably secondary to local inflammation or immune reaction. The exact stimulus remains to be determined.

Since submission of this manuscript, Takeuchi et al described a virtually identical process to NK-cell enteropathy involving the stomach in 10 patients from Japan. In our cases, lymphoma was suspected histologically, but no patient had progressive disease, and spontaneous regression was observed in most cases. Two patients had gastrectomies for presumptive diagnoses of malignancy. Interestingly, 3 of the patients had a history of gastric cancer, which prompted gastroscopy. Takeuchi et al proposed the term “lymphomatoid gastropathy” for their cases and did not report lesions in the small or large bowel. However, it is not clear to what extent these patients were investigated for intestinal disease.

In intestinal immunology, it is important to distinguish between NK cells and natural killer T cells. NK T cells represent a minor subset of T lymphocytes that share cell-surface proteins with conventional T cells and NK cells. Being an essential component of intestinal mucosal immunity, NK T cells are present primarily as intraepithelial lymphocytes but also in lamina propria. The role of NK T cells in intestinal immunity is complex and paradoxical (promotion or suppression) probably because of the existence of functionally distinct subsets. These subsets can be CD4^+, CD8^+, CD4^+/CD8^+, and a minor subset (1%-2%) negative for both CD4 and CD8. Distinction by routine immunohistochemistry between true NK cells and this minor subset of NK T cells (CD4^-CD8^-) is currently not possible; however, expression of CD56 is unique to NK cells and has not been reported for NK T cells. This finding suggests that the cellular proliferation in our patients is of NK-cell derivation.

In conclusion, we describe a unique case series characterized by atypical NK-cell proliferative lesions involving the gastrointestinal mucosa in adult patients who presented clinically with vague or nonspecific abdominal symptoms. The pathologic features on endoscopic biopsies showed a diffuse and destructive atypical NK-cell proliferation, which was initially misdiagnosed as aggressive NK-cell lymphoma. This resulted in unnecessary extensive and invasive investigations, and in 3 patients led to treatment with combination chemotherapy with or without bone marrow transplantation. It is extremely critical to recognize this lesion as a distinctive clinicopathologic entity, to prevent misinterpretation, which can lead to harmful consequences for the patients. We propose the term “NK-cell enteropathy” for this condition to allow identification of other cases and investigation of its pathogenesis. A
close partnership between the pathologist and clinician is required to recognize this condition, with a careful clinical history and description of the endoscopic findings.

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Authorship

Contribution: E.S.J. described this entity and identified all patients; A.M. and E.S.J. collected data and wrote the manuscript; S.P. reviewed pathology material and contributed to the manuscript; and P.L.B., W.H.W., and J.A.F. provided pathologic and clinical information and contributed to the manuscript.

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References


NK-cell enteropathy: a benign NK-cell lymphoproliferative disease mimicking intestinal lymphoma: clinicopathologic features and follow-up in a unique case series

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