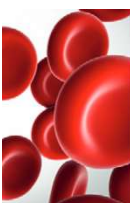


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IMMUNOBIOLOGY AND IMMUNOTHERAPY

Key diagnostic markers for autoimmune lymphoproliferative syndrome with molecular genetic diagnosis

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KEY POINTS

- In patients with lymphadenopathy and/or splenomegaly with elevated DNTs, ALPS must be suspected; genetics and biomarkers can confirm this.

Autoimmune lymphoproliferative syndrome (ALPS) is a rare immunodeficiency caused by mutations in genes affecting the extrinsic apoptotic pathway (*FAS*, *FASL*, *CASP10*). This study evaluated the clinical manifestations, laboratory findings, and molecular genetic results of 215 patients referred as possibly having ALPS. Double-negative T-cell (DNT) percentage and in vitro apoptosis functional tests were evaluated by fluorescence-activated cell sorting; interleukin 10 (IL-10) and IL-18 and soluble FAS ligand (sFASL) were measured by enzyme-linked immunosorbent assay. Genetic analysis was performed by next-generation sequencing. Clinical background data were collected from patients' records. Patients were categorized into definite, suspected, or unlikely ALPS groups, and laboratory parameters were compared

among these groups. Of 215 patients, 38 met the criteria for definite ALPS and 17 for suspected ALPS. The definite and suspected ALPS patient populations showed higher DNT percentages than unlikely ALPS and had higher rates of lymphoproliferation. Definite ALPS patients had a significantly more abnormal in vitro apoptosis function, with lower annexin, than patients with suspected ALPS ($P = .002$) and patients not meeting ALPS criteria ($P < .001$). The combination of elevated DNTs and an abnormal in vitro apoptosis functional test was the most useful in identifying all types of ALPS patients; the combination of an abnormal in vitro apoptosis functional test and elevated sFASLs was a predictive marker for ALPS-FAS group identification. Lymphoproliferation, apoptosis functional test, and DNTs are the most sensitive markers; elevated IL-10 and IL-18 are additional indicators for ALPS. The combination of elevated sFASLs and abnormal apoptosis function was the most valuable prognosticator for patients with *FAS* mutations. (*Blood*. 2020;136(17):1933-1945)

Introduction

Autoimmune lymphoproliferative syndrome (ALPS; also called Canale-Smith syndrome) is a rare immunodeficiency with several pathognomonic hallmarks, such as nonmalignant chronic lymphoproliferation, unexplained lymphadenopathy, splenomegaly, immune-mediated cytopenia, and hypergammaglobulinemia caused by an abnormal extrinsic FAS-mediated apoptotic pathway.¹⁻³ Defective lymphocyte apoptosis could principally cause autoimmune manifestations, mainly multilineage cytopenias and less frequently nephritis, hepatitis, uveitis, arthritis, or colitis. Furthermore, patients with ALPS as well as genetically affected siblings without an ALPS phenotype are predisposed to several malignancies, such as solid tumors (eg, thyroid, breast, and liver) and leukemia; the risk of non-Hodgkin lymphoma is up to 50 times greater compared with the general population.⁴⁻⁶

In ALPS, the defective extrinsic apoptotic signaling is caused by germline and somatic mutations in *FAS* (*TNFRSF6*, *CD95*, *AP01*), *FASLG*, and *CASP10* genes, encoding Fas cell surface death receptor protein, FAS ligand (FASL) protein, and caspase 10.⁷ A majority of ALPS patients present with germline heterozygous or somatically acquired *FAS* mutations. Rarely, *FAS* mutations are inherited in an autosomal recessive manner.^{2,3,8,9} The *FAS* gene encodes a protein receptor expressed on B and T lymphocyte lineages. *FASLG* gene mutations are identified in extremely rare cases of ALPS.¹⁰ On the basis of the underlying pathogenic genetic variants, ALPS is classified into 4 categories according to the affected genes, including ALPS-FAS, ALPS-FASLG, and ALPS-CASP10^{2,7}; in ALPS patients without a defined mutation in these genes (20% to 30% of cases), the condition is classified as ALPS-U (unknown).^{2,6,8,11-17} Patients with pathogenic caspase-8