Table 3 Der p 1 y Der f 1 levels in 10 classroom dust samples

Der p 1	% > 0 μg	50
	% > 2 μg	30 sample size
	% > 10 μg	20
	Geom. mean	6.7
	Range	3.4–77.7
Der f 1	% > 0 μg	50
	% > 2 μg	40 sample size
	% > 10 μg	10
	Geom. mean	5.9
	Range	1.6–79.5

of dust allergens were reported by North American authors.[5,6]

In conclusion, there is a considerable proportion of dust mite allergic school children in Quito. Dust mite allergens may be present in schools in Quito in concentrations to induce allergic/asthmatic symptoms. The humidity of 75% in spite of the height of 2,800 m above sea level may explain the presence of dust mites.

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R. Valdivieso[a,*], V. Iraola[b], M. Estupiñan[a], E. Correa[a], M.del C. Romero[a]

[a]*Allergy and Dermatology Center, Av Mariana de Jesus Oe8 y Occidental, Meditropoli Medical Center, Ofice 113, Quito-Ecuador*
[b]*Leti Laboratories SL, Tres Cantos, Madrid, España*

*Corresponding author.
E-mail address: rommelva@andinanet.net
(R. Valdivieso).

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Immunophenotypic abnormalities of CD8$^+$ T-cell subsets in a patient with unusual Good's Syndrome

To the Editor:

Good's syndrome is characterised by hypogammaglobulinemia associated with thymoma. It accounts for 7% to 13% of adult onset cases of hypogammaglobulinemia and patients usually present over 4th or 5th decade of life.[1,2] These patients are at high risk for recurrent infections, and the most common are sinopulmonary infections by encapsulated bacteria. Chronic diarrhoea is also a very common manifestation although pathogens are not always isolated.[2] Patients develop opportunistic fungal and viral infections more frequently than in common variable immunodeficiency (CVID) patients. The cause and pathogenesis of Good's Syndrome are still unknown. The data on abnormalities in T cell immunophenotype and function are limited[2]. We here report on immunophenotypic T-cell abnormalities in an unusual case of Good's Syndrome.

In 1999, a 45-year-old woman was discovered to have panhypogammaglobulinemia. She was previously diagnosed of inflammatory bowel disease (Crohn's disease) with chronic diarrhoea, neutropenia secondary to 5-aminosalicylic acid, sulfamides allergy, iron deficiency anaemia and renal lithiasis. At that moment she had no history of recurrent infections. Peripheral blood tests showed mild anaemia (Hb: 10.9 g/dl), and confirmed panhypogammaglobulinemia by nephelometry (IgG: 348 mg/dL, IgA: 70 mg/dL, IgM: $<$7 mg/dl, IgE $<$2 mg/dL). Lymphocyte surface markers (performed by flow cytometry) in peripheral blood revealed no circulating B cells (CD19$^+$: 0%), normal CD3$^+$ T-cell counts and inverted CD4$^+$/CD8$^+$ ratio. There was no specific antibody production after tetanus immunisation. Specific antibody production to pneumococcus polysaccharide vaccination was abnormally low. Agglutinins to Candida and serum complement levels were normal. Antinuclear antibodies were negative and she had no acute phase reactants. Serologic testing for HIV, HBV and HCV were negative. Biochemical parameters were within normal ranges. Intravenous immunoglobulin (IVIG) infusions were started at substitutive doses of 400 mg/kg every 4 weeks.

During follow-up she developed episodes of upper respiratory tract infections and two episodes of pneumonia (2002 and 2004), which responded to intravenous antibiotic treatment. Although diarrhoea due to inflammatory bowel disease diminished in frequency, by 2004 she had developed intestinal infection due to *Campylobacter jejuni* and was

Table 1 Immunologic characteristics in a patient with Good's Syndrome

	1999 Hypogammaglobulinemia	2007 Thymoma
Serum immunoglobulins (mg/dL)		
IgG	348	814
IgA	70	55
IgM	<7	<7
IgE	<2	<2
Specific antibodies (mg/dl)		
Basal TT IgG	0.153	2.31
Basal TT IgG1	0.08	N.D.
Basal PPS IgG	3.8	15.69
Basal PPS IgG2	2.3	N.D.
Complement (mg/dL)		
C3	94	99
C4	21	23
White blood count (cells/μl)	9100	9300
Neutrophiles % (cells/μl)	64 (5800)	64.5 (6000)
Lymphocytes % (cells/μl)	27.9 (2500)	26.9 (2500)

Immunological characteristics at the time of detection of panhipogammaglobulinemia and at the time of diagnosis of Good's Syndrome (the patient was receiving IVIG replacement therapy at this time). TT: tetanus toxoid; PPS: pneumococcal polysaccharyde. N.D.: Not done.

treated with oral metronidazole (500 mg for 10 days). She began developing painful ulcers in oral and vaginal mucosa. One year later, oral ulcers (OU) became recurrent. She received different treatments for this complication: bicarbonate mouthwashes, sodium perborate monohydrate, local salicylic acid, local antiseptics such as carbenoxolone sodium 2%, oral colchicine and oral famcyclovir with no answer. As maintenance therapy for Crohn's disease, the patient was being treated with oral corticosteroids (5–20 mg daily) at the time she had OU. From time to time she suffered from oral candidiasis and recurrent urinary tract infections due to *Proteus* which were treated with specific antimicrobial therapy. In June 2007, at the age of 52, she underwent programmed surgery for bilateral inguinal hernia. Ruled chest X-rays revealed a mediastinic mass and CT-scan confirmed an anterior mediastinic mass of 2.8. × 3.0 cm. Thymectomy was performed by videothoracoscopy. A malignant thymoma (type AB/B1, Masaoka stage IIa) was found and confirmed by biopsy. The patient received adjuvant external radiotherapy with success. Based on the association of panhypogammaglobulinemia, thymoma and absence of peripheral blood B-cells, the patient was diagnosed with Good's syndrome. Comparative humoral immunity data at the time of detection of hypogammaglobulinemia and when Good's Syndrome was diagnosed are presented in Table 1.

In July 2007, one of the main patient's complaints was recurrent and painful OU. She was not able to eat and had marked weight loss. The patient was given topical granulocyte colony-stimulating factor G-CSF (filgrastim) for OU as mouthwashes. A dose of 300 micrograms of G-CSF in 250 cc of water for 30 min of mouth-washing one-three times a day was prescribed. The procedure started once OU were detected. Patient was evaluated for OU, pain, and weight loss. Blood tests were taken during G-CSF administration and followed-up for 18 months after treatment. The patient said she felt less pain during the G-CSF treatment. Since then OU became less frequent. In July 2008, we could delay cycles of G-CSF for the following months. In September 2008, the patient had diarrhoea relapse by *Campylobacter jejuni*, requiring treatment with fluid replacement and antibiotics. Further investigations of diarrhoea revealed cytomegalovirus (CMV) which was confirmed by polymerase chain reaction from the biopsy. Oral valgancilovir (5 mg/Kg) was started. No dissemination of CMV disease was observed during follow-up.

At the time of diagnosis of Good's syndrome an extended immunophenotypic analysis of peripheral blood was performed. When this study was performed the patient did not have active infections, was not using corticosteroids and had not received recent immunisations. Cell surface staining was drawn using different combinations of fluorochromes conjugated to monoclonal antibodies (MoAbs). The following MoAbs were used: fluorescein isothiocyanate (FITC) conjugated anti-CD45RA, HLA-DR; phycoerythrin (PE) conjugated anti-CCR7, CD38, CD25; peridinin chlorophyll protein (PerCP) conjugated anti-CD4, CD3; allophycocyanin (APC) conjugated anti-CD8, CD19. IgG1 and IgG2a isotypes were used as controls. All, but CCR7-PE MoAbs were obtained from Becton Dickinson, San Jose, CA. The results are shown in Table 2. The patient was found to have higher percentages (above one-standard deviation of mean of 35 healthy controls) of distinct $CD4^+$ and $CD8^+$ activation T-cell subsets. In addition she disclosed lower percentages (below one-standard deviation of mean of 35 healthy controls) of central memory (CCR7+CD45RA-) and effector memory (CCR7-CD45RA-) $CD8^+$ T-cells as well as increased percentages of final effector $CD8^+$ T-cells (CCR7-CD45RA+).

Reported disease complications associated to Good's syndrome include haematological disorders (such as red cell pure aplasia, haemolytic anaemia or pernicious anaemia), liquen planus, or immunoglobulin G gammopathy.[2] The coexistence of malignant thymoma in Good's Syndrome is an

Table 2 Distribution of lymphocyte subpopulations at the time of diagnosis of Good's Syndrome and in healthy controls

Lymphocyte subpopulations:	Percentages ± SD	
	Patient	HC (n=35)
Maturation of $CD4^+$ T-cells		
Naive ($CCR7^+$ $CD45RA^+$)	32	37 ± 14
Central Memory ($CCR7^+$ $CD45RA^-$)	52	47 ± 13
Effector memory T-cells ($CCR7^-$ $CD45RA^-$)	14	15 ± 9
Final effector T-cells ($CCR7^-$ $CD45RA^+$)	2	2 ± 3
Maturation of $CD8^+$ T-cells		
Naive ($CCR7^+CD45RA^+$)	40	33 ± 15
Central Memory ($CCR7^+CD45RA^-$)	4	15 ± 8
Effector memory T-cells ($CCR7^-CD45RA^-$)	13	25 ± 11
Final effector T-cells ($CCR7^-CD45RA^+$)	42	28 ± 12
Activated T-cells		
$CD4^+$ HLA-DR^+ $CD38^+/CD4^+$	6	3 ± 2
$CD8^+HLADR^+/CD8^+$	39	20 ± 12
$CD8^+CD38^+/CD8^+$	83	52 ± 14
$CD8^+$ HLA-DR^+ $CD38^+/CD8^+$	34	11 ± 6
$CD4^+$ $CD25^+/CD4^+$	48	59 ± 11
$CD8^+$ $CD25^+/CD8^+$	6	14 ± 10

SD: standard deviation. HC: healthy controls.

uncommon finding. Most thymomas in these patients are benign, well encapsulated tumours and spindle cell type.[3] Our patient developed malignant thymoma which was discovered years after hypogammaglobulinemia was detected.

Thymoma, hypogammaglobulinemia and oral/genital ulcers have been rarely described in the literature.[4] Our patient had OU before the diagnosis of thymoma and receiving radiotherapy, and differential diagnosis of mucositis post radiotherapy could be discharged. Several treatments proved unsuccessful for OU, including local and oral steroids, remaining recurrent. They did not regress after surgical resection of thymoma. To our knowledge, this is the first published report of development of OU in the context of Good's syndrome that was treated with topically applied G-CSF. Recent studies suggest that G-CSF stimulates not only the stem cells but also the proliferation and differentiation of cells which participate in acute and chronic inflammation and immune responses including mature leucocytes, macrophages, dendritic cells in dermis and submucosa, keratinocytes, fibroblast and synovial cells.[5] It also might have a role in promoting angiogenic factors and stimulates mechanically wounded endothelial monolayers.[6,7] In other studies, topically applied G-CSF, decreased the healing time and pain of OU and genital ulcers of patients with Behçet disease.[8] Our patient had transient improvement with this treatment after an 18-month follow-up.

Cytomegalovirus (CMV) disease is a frequent complication of Good's Syndrome and can be fatal in these patients.[2] Our patient did not develop a severe or disseminated CMV disease which might be explained, at least in part, by the fact that she was receiving IVIG replacement therapy and maintaining normal IgG levels and antibody responses.[9]

As in CVID, several defects in cell–mediated immunity have been described in Good's Syndrome including $CD4^+$ T-cell lymphopenia, inverted $CD4^+/CD8^+$ ratio and depression of in vitro lymphocyte prolipherative responses.[2–4] Marked reduced numbers of B cells on peripheral blood is more frequent in Good's Syndrome than in CVID patients.[2] In addition to these immunological features, this is the first report of a patient with Good's Syndrome that describes decreased percentages of central memory and effector memory $CD8^+$ T-cells as well as increased percentages of final effector $CD8^+$ T-cells. Increased $CD4^+$ and $CD8^+$ T-cell activation marker expression was similar than that described in CVID patients.[10] Unlike HLA-DR, CD25 (interleukin 2 receptor alpha) is an early activation marker which is down-expressed in our patient. As far as we know, down-expression of CD25 on $CD4^+$ and $CD8^+$ T-cells coexisting with increased levels of other activation markers has not been previously documented in Good's Syndrome. We have previously described such discordant activation pattern in CVID patients.[10]

Immunophenotyping might be important in detecting and monitoring immunodeficiency in patients with immunodeficiency and thymoma. IVIG therapy did not prevent development of malignant thymoma in this patient. Central memory and effector memory CD8+ T-cells have a role in the control of viral infections and malignancy, which are frequent complications of Good's Syndrome. We propose that extended immunophenotypic evaluation of T-cells should be performed in patients with Good's Syndrome to improve the knowledge of this disease. This is also necessary to assess if there is association between immunophenotypic characteristics and clinical findings (i.e. higher incidence of viral infections or malignancy) in this unusual combined immunodeficiency, as we have previously suggested in CVID patients.[10]

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N. Del Pozo[a], E. Sarmiento[a], N. Lanio[a], A. Gallego[a], J. Largo[b], J. Carbone[a,*]

[a]*Transplant Immunology Group, Clinical Immunology Unit, Hospital General Universitario Gregorio Marañón, Madrid, Spain*
[b]*Servicio de Medicina Interna, Complejo Hospitalario de Toledo, Toledo, Spain*

*Corresponding author.
E-mail address: jcarbone.hgugm@salud.madrid.org (J. Carbone).