

CORRESPONDENCE

Profound T-cell defects in Dubowitz syndrome

To the Editor,

Dubowitz syndrome is a rare autosomal recessive disorder characterized by short stature, microcephaly, low platelet counts, dysmorphic features, eczema, and mild to severe mental retardation. Less than 150 cases have been reported so far in the literature, and diagnosis is mainly based on key clinical findings (1–5). Clinical presentation is rather variable and the only core clinical feature seems to be a characteristic face (3). While the neurologic/behavioral features have been deeply investigated, the immunologic characterization of this syndrome remains rather incomplete, apart from anecdotal reports of selective IgA deficiency, hypogammaglobulinemia and hyper-IgE-like syndrome in index cases (6–9). We report on the first extensive immunologic evaluation of a male patient with Dubowitz syndrome revealing profound T-cell defects resembling combined immunodeficiency.

The patient was born to consanguineous parents of Pakistani origin. He came to our attention at the age of 8 for immunologic evaluation due to partial IgA deficiency. His clinical history included short stature (height: 120 cm <3°; weight: 18.5 kg <3°,

cranial circumference: 49 cm <3°), delay in mental development and speech, recurrent episodes of thrombocytopenia (with normal bone marrow aspirate). Key clinical features included high and receding forehead, sparse lateral eyebrows, blepharoptosis, abnormal ear conformation, broad and flat nose bridge, clinodactyly of the fifth finger of the hand and cutaneous syndactyly between the fourth and fifth fingers of the feet (Fig. 1a). Skin inspection revealed eczema in folds, keratosis pilaris on the back of arms and legs, and numerous molluscum contagiosum lesions (Fig. S1). Based on these clinical findings, diagnosis of Dubowitz syndrome was made. The patient's infectious history was particularly severe: sepsis at 6 yr of age, osteomyelitis at 8 yr of age and recurrent molluscum contagiosum skin infections from the age of 6. Asymptomatic low platelet counts were confirmed (ranging from 42,000 to 66,000/mm³), while the rest of the differential blood count was normal. Immunoglobulin serum levels were normal, with the exception of IgA serum levels at the lower range for age: IgG: 1370 mg/dl (normal range for age: 633–1916), IgA: 31 mg/dl (normal range for age: 41–315), IgM: 64 mg/dl (normal range for age: 56–261). Biochemical features such as cholesterol, LDL, HDL levels were

Figure 1 (a) Index patient's appearance and physical characteristics: The facial appearance was characteristic with a high and receding forehead, sparse lateral eyebrows, blepharoptosis, abnormal ear conformation, broad and flat nose bridge. The index patient had clinodactyly of the fifth finger of the hand and cutaneous syndactyly between the fourth and fifth fingers of the feet. (b) Peripheral T cell and T-cell blast proliferation-CFSE dilution assay: CFSE dilution was performed to define T-cell division upon stimulation. T-cell division from the index patient was extremely impaired, while the healthy controls' T cells divided in a normal manner.

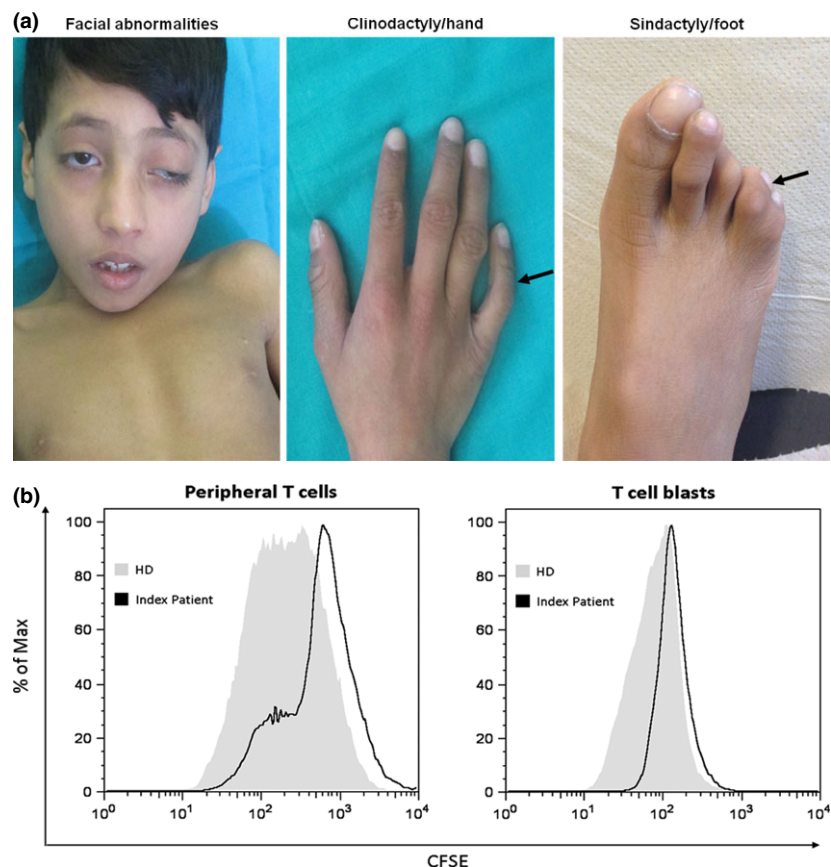


Table 1 Peripheral lymphocyte subsets from the index patient affected with Dubowitz syndrome.

	Index patient (%)	Normal range for age (%)
T cells (CD3 ⁺)	84.0	60.5–79.8
CD3 ⁺ CD4 ⁺	16.2	30.3–48.3
Naive (CD45RA ⁺ CCR7 ⁺)	10.4	34.3–74.6
RTE (CD45RA ⁺ CCR7 ⁺ CD31 ⁺)	6.5	21.1–63.5
Central memory (CD45RA ⁺ CCR7 ⁺)	24.6	13.0–43.5
Effector memory (CD45RA ⁺ CCR7 ⁺)	55.9	8.5–28.1
Terminally differentiated (CD45RA ⁺ CCR7 ⁺)	9.1	0.7–6.6
CD3 ⁺ CD8 ⁺	57.1	13.8–37.5
Naive (CD45RA ⁺ CCR7 ⁺)	3.1	26.7–72.9
Central memory (CD45RA ⁺ CCR7 ⁺)	5.8	1.2–11.6
Effector memory (CD45RA ⁺ CCR7 ⁺)	30.8	6.0–53.6
Terminally differentiated (CD45RA ⁺ CCR7 ⁺)	60.6	3.9–72.0
TCR γ/δ	14.9	0.5–21.5
B cells (CD19 ⁺)	13.5	5.7–19.7
RBE (CD38 ^{hi} CD21 ^{dim/lo} CD27 ⁺)	23.9	15.0–35.3
Naive (CD38 ^{dim/lo} CD21 ^{hi} CD27 ⁺)	56.4	33.8–79.6
CD19 ^{hi} CD21 ^{lo}	7.9	1.1–10
Switched memory (IgD ⁺ CD27 ⁺)	1.9	2.7–20.6
IgM memory (IgD ⁺ CD27 ⁺)	2.1	3.5–24.1
Terminally differentiated (CD38 ^{hi} CD27 ^{hi} CD21 ^{lo})	1.76	0.16–8.70
Plasma cells (CD38 ^{hi} CD20 ⁺ CD138 ⁺)	0.38	0.04–3.20
NK cells (CD3 ⁺ CD16 ⁺ CD56 ⁺)	1.8	4.6–27.8

within the normal range (cholesterol 160 mg/dl, LDL: 98 mg/dl, HDL 44 mg/dl). The patient carried a normal karyogram. No chromosomal instability, both spontaneous and bleomycin induced, was observed. Recently, the NSUN2 gene was reported to be mutated in three affected members of a multiplex consanguineous family (10). Direct gene sequencing of the NSUN2 gene was performed for the index patient, but no mutations were found.

Considering the infectious history, an extensive immunologic work-up was performed. Table 1 shows the distribution of peripheral lymphocyte subsets. CD4 T cells were particularly low (16.2%) and so were the naive and recent thymic emigrants (RTE) subsets of CD4 T cells (10.4% and 6.5%, respectively), suggesting a reduced thymic output, while the terminal states of CD4 maturation were particularly abundant (see Table 1). CD8 T cells were rather abundant, while naive CD8 T cells were extremely low (3.1%). T-cell receptor excision circles (TRECS) analysis confirmed the reduced thymic output (Fig. S2); T-cell receptor (TCR) repertoire was restricted (data not shown). Regarding B cells, although the memory subsets (IgM and switched) were at slightly lower percentages when compared to healthy controls, the remain-

ing B cell subsets were normal. Finally, NK cells were reduced (1.8%).

Considering the novelty of the T-cell abnormalities in Dubowitz syndrome here described, functional studies were undertaken to better define the severity of the T-cell defect. Peripheral T cells upon PHA stimulation and T-cell blasts were evaluated for their proliferative capacity both for the index patient and a healthy donor by means of CFSE dilution: patient's phytohemagglutinin (PHA) stimulated peripheral T cells and T-cell blasts showed a remarkably impaired proliferative response as compared to healthy controls (Fig. 1b).

Dubowitz syndrome is a rare autosomal recessive disorder typically characterized by neurologic, facial and skeletal abnormalities. The accompanying immunologic phenotype however is not well characterized, and the reported infectious complications are very limited (3, 4, 6–9). T-cell defects were previously reported on a single patient affected with Dubowitz syndrome who presented with reduction of T cells (CD3, CD4 and CD8 subsets), while the *in vitro* proliferative responses were normal (11). In the patient here described, the immunologic impairment is more profound and is characterized by reduction of naive CD4 and CD8 and RTE CD4 T cells, defective T cell *in vitro* proliferation upon stimulation, restricted TCR repertoire, and a significant reduction of thymic output as measured by T-cell excision circles (TRECS), mimicking combined immunodeficiency (12).

The fact that this patient presented recurrent molluscum contagiosum skin infections is also suggestive of profound T-cell defects, as seen in other combined immunodeficiencies. We suggest that extensive immunologic evaluation should be performed in patients affected with Dubowitz syndrome, as this may affect patient's clinical management and prognosis.

Disclosure of conflict of interests

The authors declare no conflict of interest.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Figure S1. Index patient's dermatologic alterations: Skin inspection revealed eczema in folds, keratosis pilaris on the back of arms and legs and numerous Molluscum contagiosum lesions.

Figure S2. T cell receptor excision circles (TRECS) evaluation was performed according to previously published protocols (*J Clin Immunol* 2011 Aug; **31**(4): 540–9). The value for healthy controls corresponds to the median value of pediatric healthy controls (3–16 yr of age).

Figure S1



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Figure S2

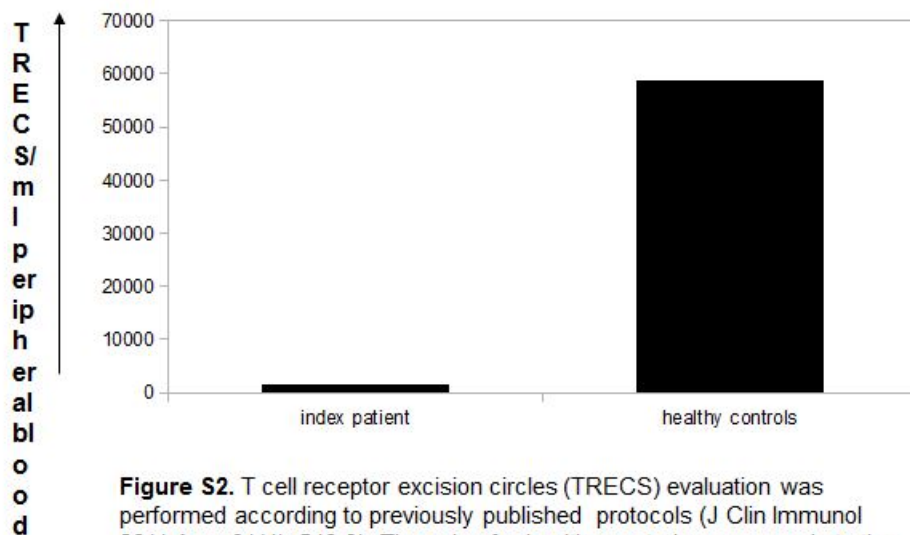


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