Theophylline as a precision therapy in a young girl with PIK3R1 immunodeficiency

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Clinical Implications

- Based on its phosphatidylinositol 3-kinase-delta (PI3Kδ) inhibitory properties, theophylline was administered to a young girl with activated PI3Kδ syndrome (APDS). We report reduced frequency of infections, decreased lymphoproliferation, and noticeable changes in immunophenotype, encouraging further trials with theophylline in children with APDS.

TO THE EDITOR:

Activated phosphatidylinositol 3-kinase-delta (PI3Kδ) syndrome (APDS), also named PASLI (p110δ-activating mutation causing senescent T cells, lymphadenopathy and immunodeficiency), is a primary immunodeficiency associated with increased susceptibility to respiratory infections, bronchiectasis, and lymphoproliferation. The disease is caused by hyperactivation of PI3Kδ, due either to gain-of-function mutations in the *PIK3CD* gene, encoding for the catalytic subunit p110δ (APDS1), or to the splice site mutations c.1425+1 G>a or G>c of *PIK3R1* resulting in skipping of the exon 11, encoding for the regulatory subunit p85α and a consequential reduced inhibition of p110δ (APDS2). Immunological features include class switch recombination deficiency with hypogammaglobulinemia mirroring a hyper-IgM syndrome, together with a reduction of recent thymic emigrants, inversion of the CD4/CD8 ratio due to a reduction of naïve CD4, and expansion of senescent CD8+ CD57+ T cells.[1]

Ex vivo treatment with p110δ inhibitors can counteract most of the clinical and immunological anomalies typical of APDS, providing a proof of concept for the experimental use of such inhibitors into the clinics. Indeed, clinical trials with p110δ inhibitors are currently ongoing in patients with APDS/PASLI (EudraCT Number: 2014-003876-22 12 years; NCT02435173 16 years); however, children below 12 years of age or patients who have already experienced a malignant lymphoproliferative disorder cannot be recruited.[2] However, a search in the current literature showed that theophylline also has a good inhibitory action on PI3Kδ at concentrations in the therapeutic range for other indications, such as asthma or apneas.[3] For these reasons, we conducted a clinical trial of theophylline in a young girl with PIK3R1 deficiency.

We administered theophylline to an 11-year-old girl with APDS, due to the exon skipping mutation c.1425+1G>A in *PIK3R1*, and observed clinical and laboratory changes. Patient clinical history was characterized by growth retardation, multiple and symptomatic lymphadenopathies, particularly mediastinal adenopathy causing median lobe syndrome, cervical lymphoid enlargement, and lymphoid infiltration of laryngeal mucosa inducing permanent aphonia. In view of clinical progressive deterioration of the symptomatic lymphoproliferation, she started an off-label treatment with Sirolimus, which led to a partial reduction of lymphadenopathies but without any improvement of vocal function. On the basis of the report of Fukas about the inhibitory action of theophylline on p110δ and with the aim of obtaining a more complete control of the disease, we considered an off-label use of this drug, administered at 200 mg in a single daily dose orally (7 mg/kg), with the hematic level 15 to 20 mg/L. During a year of treatment, a significant reduction of lymphadenopathies was noticed, with a decline of mediastinal and laterocervical involvement (Figure 1, *A-F*) and a decrease of lingual and pharyngeal enlargement, resulting in voice restoration (Figure 1, *G* and *H*). Laboratory data showed a reduction of acute-phase reactants, mild increase in hemoglobin concentration, and mild improvement of immunological anomalies, particularly a slight increase of recent thymic emigrants and CD27+ B cells, and a reduction of the IgM level (Figure 2). Furthermore, a striking reduction of infections requiring antibiotic therapy (9 episodes per year vs 2) and substantial improvement of scholastic performance were reported.

Precision therapy for primary immunodeficiency is ongoing, particularly in patients with APDS1. Achieving an efficient inhibition of p110δ can reduce the hyperactivation of the PI3K/AKT (protein kinase B)/mammalian target of rapamycin pathway and correct the immunological phenotype, as recently reported by Rao et al.[2] Hyperactivation of this pathway inhibits FOXO1 causing altered development in early B cells, homing to lymph nodes and class switch mutation, resulting in a permanent alteration and progressive enlargement of the lymph node structure.[1] Theophylline is a methylxanthine drug, working as a phosphodiesterase inhibitor that increases cyclic adenosine monophosphate levels, and it has been widely used to treat asthma and neonatal apnoeas.[3] In addition, theophylline, as well as caffeine, was shown to inhibit the isoform p110δ of PI3K, which is expressed only in lymphocytes and melanocytes.[4] Thus, theophylline could enter the group of p110δ inhibitors that have been proposed as precision drugs for APDS, having the advantage of a well-known safety profile and a low cost compared with new synthetic products.[3] Alimentary intake of methylxanthines can contribute to the effect of treatment, but in our case, the girl did not take coffee and seldom drank tea.

Sirolimus has also been proposed to treat lymphoproliferation in APDS, but in our case, similar to what was reported by other authors, we observed only a slight improvement in lymphoproliferation with no benefit on infectious recurrence.[5,6]

In APDS, both immune deficiency and dysregulation are present: for this reason, frequent infections, autoimmune diseases, and progressive and possibly life-treating lymphadenopathy could occur, so patients often need courses of

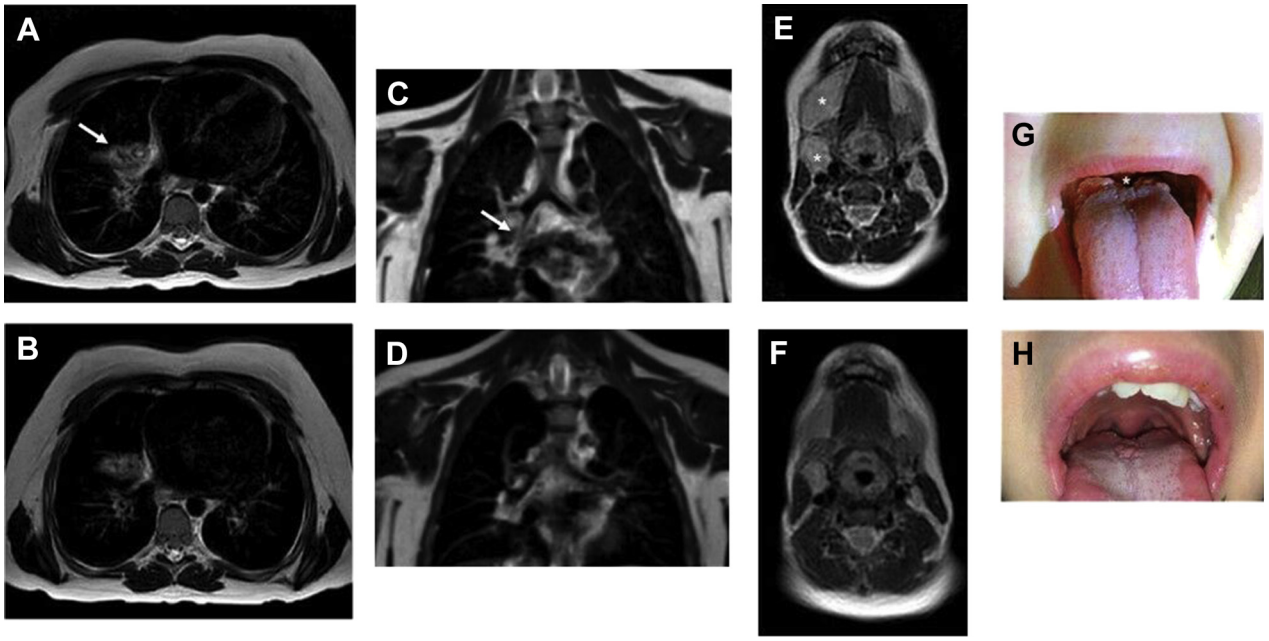

FIGURE 1. Comparison of lymphadenopathies (**A**, **C**, **E**, **G**) before and (**B**, **D**, **F**, **H**) after 1 year of theophylline treatment. Thoracic Turbo Spin Echo MRI-T2 weighted scans showing mediastinal lymphoid enlargement causing median lobe obstruction (white arrows in **A** and **C**), and submandibular lymphadenopathies (white asterisks in **E**). (**B**, **D**, **F**) After 1-year theophylline treatment, a reduction of lymphadenopathies and patency of medium bronchus is evident. The same effect has been observed for the hypertrophic lingual tonsils (asterisk in **G** and **H**).

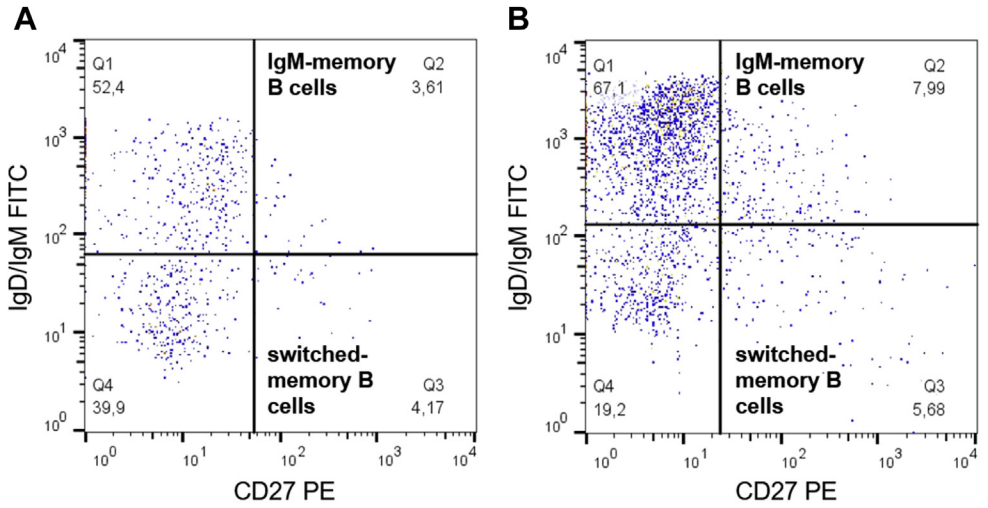

FIGURE 2. Phenotype of peripheral B cells evaluated by means of flow cytometry. The coexpression of CD27 and IgD/IgM on B cells identifies mature IgM-memory B cells, whereas expression of CD27 but not IgD/IgM identifies switched-memory B cells. Both populations, mature IgM-memory and switched-memory B cell, increase after theophylline administration (**A,** dot plot vs **B,** dot plot).

prophylactic antibiotics, glucocorticoids, and immunoglobulin. During theophylline therapy, we observed a substantial reduction of prednisone and antibiotic requirements.

Moreover, after a temporary suspension of the drug, we observed a clinical deterioration, consisting of a relapse of inspiratory dyspnea and vocal cord dysfunction that required symptomatic therapy. Finally, we would like to stress that theophylline therapy does not lead to any important side effects with only occasional mild headache.

Although the description of a single case makes it difficult to judge the clinical and immunological efficacy of the treatment, our results suggest that theophylline could have played a role in the observed changes in our patient. For safety reasons, we did not investigate whether a higher dosage of theophylline could

lead to better results, because the inhibitor effect of p110δ is achievable within the therapeutic concentration range for asthma.[3] Moreover, the experimental treatment was started in a grown-up patient although it is logical to expect that any drug acting on the immune development could have its maximal potential in the first years of life. For this reason, we hypothesized that changes in immunological features could be enhanced by early treatment that could also prevent complications due to recurrence of infection as bronchiectasis.

In conclusion, we believe that theophylline may be worth a trial in children with APDS since the first years of life, while awaiting completion of trials for authorization of novel specific molecules. Because theophylline acts as an ATP-competitive p110d inhibitor and the mutant form of this protein is targetable by other inhibitors acting on the same site, we propose that a trial of this drug may be worthwhile both in APDS2 and in APDS1. We thus believe that this experience may be the basis for the development of a controlled clinical trial of theophylline in children who cannot be enrolled in trials with other p110d inhibitors.

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REFERENCES

1. Lucas CL, Chandra A, Nejentsev S, Condliffe AM, Okkenhaug K. PI3Kδ and primary immunodeficiencies. Nat Rev Immunol 2016;16:702-14.
2. Rao VK, Webster S, Dalm VASH, Šedivá A, van Hagen PM, Holland S, et al. Effective 'Activated PI3Kδ Syndrome'-targeted therapy with the PI3Kδ inhibitor leniolisib. Blood 2017;130:2307-16.
3. Hendeles L, Weinberger M, Szefler S, Ellis E. Safety and efficacy of theophylline in children with asthma. J Pediatr 1992;120:177-83.
4. Foukas LC, Daniele N, Ktori C, Anderson KE, Jensen J, Shepherd PR. Direct effects of caffeine and theophylline on p110 delta and other phosphoinositide 3-kinases. Differential effects on lipid kinase and protein kinase activities. J Biol Chem 2002;277:37124-30.
5. Lucas CL, Kuehn HS, Zhao F, Niemela JE, Deenick EK, Palendira U, et al. Dominant-activating germline mutations in the gene encoding the PI(3)K catalytic subunit p110δ result in T cell senescence and human immunodeficiency. Nat Immunol 2014;15:88-97.
6. Elkaim E, Neven B, Bruneau J, Mitsui-Sekinaka K, Stanislas A, Heurtier L, et al. Clinical and immunologic phenotype associated with activated phosphoinositide 3-kinase δ syndrome 2: a cohort study. J Allergy Clin Immunol 2016;138:210-8.