Response letter

Dear Reviewers,

Thank you for your comments. On behalf of my co-authors, we very much appreciate the time and effort you have put into your comments on our manuscript (NO.35562). Your reviews are most helpful. We have carefully reviewed the comments and thoroughly revised the manuscript accordingly. Our responses are given in a point-by-point manner below. We have submitted a revised version of our manuscript. All the changes are marked in red color.

We are looking forward to your reply on revision. We are more than happy to make any further changes that will improve the paper. Thank you again for your attention and consideration.

Sincerely yours,
Ying Huang MD, PhD,

Reviewer Comments:

Reviewer #1: Wang et collaborators presented a retrospective experience with thalidomide to treat children with Crohn's Disease (CD) who had laboratory evidence of infection from Mycobacterium Tuberculosis. The main drawbacks of the study are its retrospective nature and the lack of a control group. Indeed, it is clinical research and it can offer useful data for the development of future studies. Even if several questions stay unanswered about the relationships and the differential diagnosis between CD and Intestinal Tuberculosis (TB), the results of the study may be relevant for clinicians dealing with CD in countries with high prevalence of TB. The authors adopted practical criteria to differentiate CD from IT, arguing that CD can be diagnosed when intestinal compliant fail to recover after a proper treatment against TB (ATT). In the discussion, they argue that we cannot exclude a role of MTB in triggering CD in such patients and this could be of importance when considering conventional antinflammatory treatments, such as anti-TNFalpha biological therapies, which may affect the response to mycobacteria. I have some comments and

questions: - The efficacy of thalidomide in children with CD has already been described. The main information added by Wang at al. regard the safety of the drug in subjects with CD and evidence of latent TB. I suggest to change the title accordingly. For example: “Safety of thalidomide therapy in children with Crohn’s Disease and evidence of latent tuberculosis”. - A better description of the histological finding would be welcome: in particular, were caseous granulomata found in any patient at any time? - As concern TB, were other signs of TB present, before ATT? - The authors should report if any change in TB status (either AFB or tuberculin skin testing) occurred after long term treatment with thalidomide. - The authors must state if any of the cases described in their 2011 manuscript has been included in the present series. - As concerns the patient with monogenic CD (IL10RA deficiency), how the mutations have been judged as causative? I wasn’t able to find ref 12 on the web. - Even if this is a pediatric series, in methods, it could be worth to specify if females in reproductive age were included or not in the research and if contraception was prescribed and controlled in these cases. There are some errors that need correction. For example, “one patient were received nutritional treatment”, or “conditions could be coexist in countries”. Moreover, I recommend not using the name of the name of the disease as an adjective for the subjects with that disease. Not CD patients, but patients with CD. Not “CD who has been treated for tuberculosis”, but “children with CD who have …”.

Response:

1) Thank you for pointing out the inappropriate title. We have revised it according to your suggestion. Limited by the rules of no more than 12 words, it is changed to “Efficacy of thalidomide therapy in pediatric Crohn’s disease with evidence of tuberculosis”.
2) As for the histological findings, case 5 was found with caseous granulomata before ATT treatment. Granuloma were found in case 1 and case 7. The biopsy histology from case 9 was shown with multinucleated giant cells.
3) We showed the TB status in the results section of the manuscript. It is in the second paragraph “All patients have evidence of tuberculosis existence; AFB positive was observed in eight cases and positive tuberculin skin testing in two. One patient suffered from spleen TB and cell culture of Mycobacterium was

positive". We also added the signs of TB present in each case in the revised Table 1.

4) We could track the TB status in case 3 and case 8. The positive AFB findings were turned to negative after 16 months and 10 months of treatment, respectively. These data are added in the outcome section.
5) In the present series, two cases (case 1 and case 5) described in 2011 were incorporated into the study. However, two of them were followed up for a long time, 38 and 36 months, respectively.
6) Sorry, we apologize for the wrong information. The ref. 12 has been accepted by the journal of *Inflammatory Bowel Disease*, but still not online. In that paper, it has been reported that IL10RA c.301C.T (p.R101W) and c.537 G.A (p.T179T) mutation are the most common mutations in Chinese patients with very early onset inflammatory bowel disease. Moreover, the functional analysis of the variants has been performed in several patients with IL10RA mutations, which was previously described by our group. We add this paper as the new No.13 reference (Huang ZH, Peng KY, Li XQ, et al. Mutations in Interleukin-10 Receptor and Clinical Phenotypes in Patients with Very Early Onset Inflammatory Bowel Disease: A Chinese VEO-IBD Collaboration Group Survey. Inflamm Bowel Dis. 2017; 23:578-590. [PMID:28267044]).
7) Changes has been made in the revised manuscript. We add the following sentence in the Methods section: "Contraception was controlled in the patients who were in reproduction age.".
8) We are very sorry for our incorrect writing. Necessary changes have been made in the revised manuscript which are highlighted in red color.

Reviewer #2: This study examined the efficacy of thalidomide in the treatment of children diagnosed with both Crohn's disease (CD) and tuberculosis. The authors concluded that thalidomide is an effective and safe drug in inducing remission for pediatric patients with CD who have been treated for tuberculosis. The same group in 2010 published a paper reporting the use of thalidomide to treat pediatric refractory CD, of which three children had TB. The current study reported the outcomes of thalidomide treatment in 10 younger children with CD coexist with tuberculosis. The patient numbers included in this study are small. However, this study still

provides some useful information for the management of pediatric CD, if more clinical data are included. Please see my comments below. 1. Of the 10 patients included in this study, two children were 2-month and 7-month old at the time of their diagnosis of intestinal TB and CD. CD is rare at this age, although it can occur. Please provide clinical evidence for the diagnosis of CD and TB for each individual child included in this study. 2. Table 1. The start dose varied in individual patients. For example, both patient 6 and patient 9 had ileocolonic CD and anal fissure, but the thalidomide starting doses were different. What was the base for choosing the starting dose? Table 1 provided final dose and treatment duration. However there were no details regarding how the doses were changed during the treatment. Please provide a table including detailed changes of doses corresponding to the clinical information of the patients at the time of dose changing including the time when the remission was achieved for each individual patient. Please also provide information whether the thalidomide was ceased in any of the patients, if yes, were other medications used for the maintenance of the remission? For example, there was no final dose for patient 1. Does this mean that this patient was off thalidomide and was free of any other CD treatments? 3. All patients received TB treatments. Was their TB cured at the time starting CD treatment using thalidomide?

Response:

1) Special thanks to you for your good comments. It is really helpful to provide the evidence of CD and TB. The evidence of TB in each individual child were listed in the revised Table 1. And the diagnosis of Crohn's disease was based on endoscopic and clinical symptoms and also defined as no improvement of clinical and endoscopic symptoms after anti-TB treatment for at least one year. All the patients were fulfilled with the criteria. Case 2 and case 3 were both diagnosed with CD in the early age, which could be also defined as very early onset inflammatory bowel disease (VEOIBD). Morever, both of them had perianal abscess. The case 2 was finally confirmed with IL10RA mutation. CD can be occurred in the small age group. Even if the population is small, we are seeing an increasingly number of IBD patients in recent years. We even report 38 cases with gene mutation in infantile-onset IBD (<2 years old) (ref No.12).
2) We have to clarify that we often choose the start dosage around 2mg/kg/d, based

on the severity of the disease to modify. However, in case 6, the reason we use the low dose to start is that he was attempted to administrate thalidomide once, but fever and abdominal pain occurred quickly within one day. Therefore, we chose to reduce the initial dose for that case.

3) The details of dose changes in each individual patient were listed in Table 2. However, given that it is a retrospective study, we could not get the whole data at the regular time point, such as 3 months, 6 months, 9 months. We try our best to list all the data that available on a case-by-case basis.

Table 2 Available data for the dose changes at different time-points after thalidomide treatment

Patient No.	Dose (mg/kg/d) at different follow-up time (months)
1	2.5(baseline) - 2.5(1m*) - 1.2(12m) - stop (34m)
2	2.5(baseline) - stop (7m)
3	2(baseline) - 1.1(12m) - 0.7(18m) - 0.4(21m) - 0.33(33m)
4	1.8(baseline) - 3 (5m) - 1 (12m) - 0.8(15m)
5	2(baseline) - 0.5(2m) - 1.8(9m) - stop (22m) - 0.5(36m)
6	1.2(baseline) - 0.6(12m)
7	2(baseline) - 1.8(1m) - 1.1(10m) - stop(44m)
8	2(baseline) – 2.4(10m) - 1.6(16m)
9	2.2(baseline) -1.4(5m) - 1.8(9m)
10	1.8(baseline) -1.3(9m)

*m for months.

4) In Table 1, we provide the final dose in each case. Three of them were off thalidomide, thalidomide was ceased in two patients (case 1 and case 7) because of clinical remission and in one with IL10 RA deficiency due to lack of response (case2). Case 1 and case 7 were free of other CD treatments until the end of follow-up timepoint. Case 2 was treated with umbilical cord blood transplantation. For case 5, he discontinued thalidomide after 22 months of treatment and then transfer to use acetazolamide (AZA) for maintenance remission, but the symptom of oral ulcers relapsed. Therefore, the patient later opted to restart thalidomide and responded quickly. He was in remission with a minimum dose of 0.5mg/kg/d.

5) As for the TB status, detailed information on TB status are provided in Table 2. And we also added this sentence in the outcome section: “With regard to TB

status, the positive AFB findings were turned to negative in case 3 and case 8 after 16 months and 10 months of treatment, respectively.”.