Effects of zinc and vitamin A supplementation on prognostic markers and treatment outcomes of adults with pulmonary tuberculosis: a systematic review and meta-analysis

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ABSTRACT

Introduction Undernutrition is a major risk factor for tuberculosis (TB), which is estimated to be responsible for 1.9 million TB cases per year globally. The effectiveness of micronutrient supplementation on TB treatment outcomes and its prognostic markers (sputum conversion, serum zinc, retinol and haemoglobin levels) has been poorly understood. This study aimed to determine the effect of zinc and vitamin A supplementation on prognostic markers and TB treatment outcomes among adults with pulmonary TB.

Methods A systematic literature search for randomised controlled trials (RCTs) was performed in PubMed, Embase and Scopus databases. Meta-analysis with a random effect model was performed to estimate risk ratio (RR) and mean difference (MD), with a 95% CI, for dichotomous and continuous outcomes, respectively.

Results Our search identified 2195 records. Of these, nine RCTs consisting of 1375 participants were included in the final analyses. Among adults with pulmonary TB, zinc (RR: 0.94, 95% CI: 0.86 to 1.03), vitamin A (RR: 0.90, 95% CI: 0.80 to 1.01) and combined zinc and vitamin A (RR: 0.98, 95% CI: 0.89 to 1.08) supplementation were not significantly associated with TB treatment success. Combined zinc and vitamin A supplementation was significantly associated with increased sputum smear conversion at 2 months (RR: 1.16, 95% CI: 1.03 to 1.32), serum zinc levels at 2 months (MD: 0.86 μmol/L, 95% CI: 0.14 to 1.57), serum retinol levels at 2 months (MD: 0.06 μmol/L, 95% CI: 0.04 to 0.08) and 6 months (MD: 0.12 μmol/L, 95% CI: 0.10 to 0.14) and serum haemoglobin level at 6 months (MD: 0.29 μg/dL, 95% CI: 0.08 to 0.51), among adults with pulmonary TB.

Conclusions Providing zinc and vitamin A supplementation to adults with sputum-positive pulmonary TB during treatment may increase early sputum smear conversion, serum zinc, retinol and haemoglobin levels. However, the use of zinc, vitamin A or both was not associated with TB treatment success.

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WHAT IS ALREADY KNOWN ON THIS TOPIC

⇒ According to WHO recommendations, all individuals living with tuberculosis (TB) should be evaluated for their nutritional status and provided with adequate interventions.
⇒ Previous evidence has looked at the effect of multi-micronutrient (high heterogeneous) supplementation on mortality, with a non-significant effect being identified.
⇒ However, the evidence concerning the putative effects of adjunctive zinc and vitamin A supplementation to enhance TB treatment outcomes and its prognostic markers have not been well studied.

WHAT THIS STUDY ADDS

⇒ Results suggest that zinc and vitamin A supplementation was significantly associated with improved sputum smear conversion at 2 months and higher serum retinol, zinc and haemoglobin levels at different durations of follow-up among adults with pulmonary TB.
⇒ However, zinc and/or vitamin A supplementation during TB treatment had no effect on a successful TB treatment outcome.

INTRODUCTION

Tuberculosis (TB) is caused by the bacterium Mycobacterium tuberculosis (M. tuberculosis). It is currently second to COVID-19 as a leading infectious cause of death globally, with an estimated 9.9 million new cases, and 1.3 million...
and antibody responses, which may aid bacterial supplementation is useful to help improve TB treatment effects of these deficiencies on TB treatment or whether clearance. Vitamin A, in the form of retinol in plasma, is crucial for the protection of mucosal immunity. In vitro studies have shown that retinoic acid (a metabolite of vitamin A) can hinder mycobacterial multiplication in macrophages and is able to down-regulate tryptophan aspartate containing coat protein transcription, a prognostic marker for TB treatment outcomes. Although the mechanism remains unclear, vitamin A is crucial for the protection of mucosal immunity. In addition, zinc is a trace element crucial for immune cell function. It also has a role in vitamin A metabolism. Sufficient serum zinc level can also decrease free radical membrane damage during inflammation, and increase the numbers of T and natural killer cells. The bacterial load has been found to be considerably reduced through administering zinc supplementation. As well, individuals with zinc deficiency have been shown to have decreased levels of thymulin, with a resultant decrease in T cell-mediated immunity and an increased vulnerability to infectious diseases.

The effectiveness of TB treatment relies on a competent cell-mediated immune system and the appropriate use and duration of antibiotics. Successful chemotherapy causes a rapid return to protective immune responses. This suggests that improving the micronutrient status of adults with pulmonary TB on chemotherapy may lead to faster bacterial clearance and, thus, a faster clinical recovery process by enhancing the immune response. In some trials, micronutrient supplementation has been linked to improved sputum smear and culture conversion rates, and lower mortality rates compared with a placebo. However, evidence from earlier trials has been contradictory, showing no therapeutic effects of micronutrient supplementation on TB treatment outcomes. The evidence concerning the putative effects of adjunctive zinc and vitamin A supplementation to enhance TB treatment outcomes, and its prognostic markers including serum zinc, retinol and haemoglobin levels, as well as sputum smear conversions have not been well studied, with only a few randomised controlled trials (RCTs) having been carried out. Moreover, previous reviews have emphasised the effect of multi-micronutrient supplementation on mortality, weight gain, body mass index and degree of cavitation on chest radiography. Therefore, this systematic review and meta-analysis synthesised available evidence from RCTs to determine the association between zinc and vitamin A supplementation on TB treatment outcomes among adults with sputum-positive pulmonary TB as well as prognostic markers (ie, sputum smear conversion, serum zinc levels, serum retinol levels and serum haemoglobin levels).

METHODS
This systematic review followed the methods in the Cochrane handbook for the systematic review of interventions and was reported according to the Preferred Reporting Items for Systematic Reviews and Meta-analysis (PRISMA) guidelines. The PRISMA checklist is included in online supplemental file 1.

The protocol for this systematic review and meta-analysis was registered in the International Prospective Register of Systematic Reviews (PROSPERO: CRD42021248548).

Study selection and eligibility criteria
We included RCTs based on our research questions constructed using the PICOS (Population, Intervention, Comparator, Outcomes and Studies) format.

Population
Adults with pulmonary TB, aged 15 and above, with acid-fast bacilli sputum smear-positive, Xpert positive or culture-positive drug-susceptible TB, with or without comorbidities or risk factors associated with TB including undernutrition, HIV infection and DM.

Intervention
The intervention comprised zinc (15–90 mg/day) or vitamin A (2500–5000 IU/day) supplementation or both, with follow-up for 6 months to evaluate clinical (sputum smear and culture conversion) and laboratory (serum levels of zinc, retinol and haemoglobin) parameters and TB treatment outcomes. Studies with co-interventions (eg, food supplements and nutritional advice) were eligible on the condition that the co-intervention was comparable in both arms.

Comparator
The comparator groups were those who received a placebo or no intervention.
Outcomes
The primary outcome of interest was a successful TB treatment outcome which was defined according to WHO definitions. A successful treatment outcome was defined as the sum of those who completed treatment and who were cured. Unsuccessful treatment outcome was defined as the combination of those who died, were lost to follow-up or experienced treatment failure. The secondary outcomes were prognostic markers including sputum smear and culture conversion, and serum zinc, retinol and haemoglobin levels. Serum zinc, retinol and haemoglobin levels were assessed before treatment, and after 2 and 6 months of treatment.

Studies
RCTs were included.

Exclusion criteria
We excluded studies conducted in children aged less than 15 years, people with extra-pulmonary TB and people with multidrug-resistant TB or any other form of drug resistance. We excluded interventions that provided non-specific multivitamin related interventions. Conference and meeting abstracts and papers in languages other than English, animal studies and those with insufficient information on the primary outcomes of interest were excluded.

Search strategy
We searched PubMed, and Embase through OVID and Scopus for relevant RCTs. The search strategy combined terms for TB, zinc, vitamin A, treatment outcomes, sputum conversion and serum zinc, vitamin A and haemoglobin levels. The search strategies for all databases are provided in online supplemental file 1. We also searched www.clinicaltrials.gov for related ongoing RCTs. Reference lists of the included studies were searched for additional relevant articles. Corresponding authors were contacted by email in some instances when additional information was required. We performed our search between 15 January 2021 and 23 September 2021.

Quality of evidence
The quality of evidence was examined using the Grading of Recommendations, Assessment, Development and Evaluation method. The certainty rating of the evidence for each outcome was classified by an assessment of the available study data in terms of its risk of bias, inconsistency, indirectness, imprecision and publication bias. The quality of evidence was rated as high, moderate, low or very low. We used GRADEpro Guideline Development Tool software to generate summary of findings tables for each outcome.

Risk of bias
Two authors (FW and SE) independently reviewed the risk of bias of each included study as per the Cochrane risk of bias assessment tool. Discrepancies were resolved by discussion, and when needed, a third reviewer acted as a mediator (KAA). Studies were considered as a low risk of bias when all-important contents were judged and found to be at low risk. The risk of bias assessment tools is provided in online supplemental file 2.

Data extraction
A data extraction form was developed and pilot-tested with a subset of eligible studies. Two authors (FW and SE) independently extracted the required information from the relevant articles. When there were disagreements between the two authors, a third author (KAA) was consulted, and discrepancies were resolved by consensus. The following variables were extracted from each eligible study: primary author, publication year, country of study, study design, duration of follow-up, types of interventions and outcomes. We extracted the number of participants with the outcome and the total sample size for dichotomous data from tables or graphs. For continuous data, mean values with SD were extracted. Data were extracted into a Microsoft Excel spreadsheet.

Data synthesis and analysis
Data were imported to RevMan V.5 for analysis. The characteristics of the included RCTs were summarised in descriptive tables, and outcomes were presented in figures. Interventions were compared using risk ratios (RR) and 95% CIs for dichotomous outcomes (treatment success vs an unsuccessful treatment outcome, sputum smear conversion ‘yes’ vs ‘no’) and standardised mean differences (MD and 95% CIs) for the continuous variables (serum levels of vitamin A, zinc and haemoglobin levels).

An RR and corresponding 95% CI were computed using the total numbers of participants and events in each arm within each study.

Meta-analyses with a random-effects model were used to pool the results of the included studies. Funnel plots were used to test for publication bias and small-study effects. Heterogeneity across studies was examined quantitatively by calculating the I² statistic with corresponding p values and qualitatively by visually examining forest plots. Incomplete data were described under attrition bias, but no further analyses were applied to input these data. We did not perform sensitivity analyses to evaluate the influence of a single study on the outcomes by omitting others (eg, leave out one approach) as there were too few trials for comparison.

Patient and public involvement
Patients or the public were not engaged in the study design, conduct, reporting, or distribution plans owing to the nature of the study.

RESULTS
Study characteristics
We identified 2195 publications. After removing 307 duplicates, 1888 articles were screened by reviewing their titles and abstracts, resulting in 56 potential articles for...
full-text reviews. Then, nine studies comprising a total of 1375 participants met the eligibility criteria and were incorporated in the systematic review (figure 1).

Of the included studies, three were published in 2010, while all studies were published between 2002 and 2018. Study-specific sample sizes ranged from 267 to 499 participants. The follow-up duration for outcome evaluation ranged from 2 to 6 months (table 1). The trials were conducted in six countries: South Africa, Nigeria, India, Tanzania, Indonesia, and Mexico. Sputum smear conversion rates at 2 months were evaluated. The serum levels of zinc, retinol and haemoglobin at baseline, 2 and 6 months were measured in four of the nine trials (table 1).

Effect of zinc and vitamin A supplementation on TB treatment outcome of adults with sputum-positive pulmonary TB

A successful TB treatment outcome was the primary outcome of interest in five trials with a total of 731 adults with pulmonary TB. The pooled results from these trials indicated that combined zinc and vitamin A supplementation compared with placebo was not significantly associated with a successful TB treatment outcome (RR: 0.98, 95% CI: 0.89 to 1.08). Similarly, the results showed that zinc only supplementation (RR: 0.94, 95% CI: 0.86 to 1.03) or vitamin A only supplementation (RR: 0.90, 95% CI: 0.80 to 1.01) was not significantly associated with a successful treatment outcome compared with placebo (figure 2).

Subjective assessment of the funnel plots showed symmetric distribution, and there was no evidence of publication bias (online supplemental figure S1).

Effect of zinc and vitamin A supplementation on sputum smear and culture conversion

Regarding sputum culture conversion, two trials reported that supplementation with vitamin A and/or zinc alone had no significant effect on sputum culture conversion at 2 months.

The pooled effect from five studies showed that zinc and vitamin A supplementation was significantly associated with increased rates of sputum smear conversion (RR: 1.16, 95% CI: 1.03 to 1.32). In contrast, a non-significant difference was found in sputum smear conversion when comparing supplementation with zinc alone versus placebo (RR: 1.14, 95% CI: 0.85 to 1.55) (figure 3).
### Table 1  Summary description of included studies

<table>
<thead>
<tr>
<th>Authors</th>
<th>Year of publication</th>
<th>Country</th>
<th>Total number of participants</th>
<th>Intervention arm</th>
<th>Comparator arm</th>
<th>Follow-up months</th>
<th>Primary outcomes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Karyadi et al</td>
<td>2002</td>
<td>Indonesia</td>
<td>110</td>
<td>Daily dose of retinol (1500) equivalents vitamin A (5000 IU) as retinyl acetate and zinc (15 mg) as zinc sulfate</td>
<td>The placebo contained lactose alone with anti-TB drugs</td>
<td>6 months</td>
<td>The primary outcomes of the study were clinical responses and nutritional status.</td>
</tr>
<tr>
<td>Lawson et al</td>
<td>2010</td>
<td>Nigeria</td>
<td>233</td>
<td>Zinc (90 mg) five times and daily dose of vitamin A (5000 IU)</td>
<td>Placebo with Anti-TB drugs</td>
<td>6 months</td>
<td>The primary outcomes of the study were sputum smear conversion and resolution of radiographic abnormalities.</td>
</tr>
<tr>
<td>Pakasi et al</td>
<td>2010</td>
<td>Indonesia</td>
<td>152</td>
<td>Daily dose of zinc only (15 mg), vitamin A only (5000 IU), and combined.</td>
<td>Placebo and anti-TB drugs</td>
<td>6 months</td>
<td>The primary outcome of the study was sputum smear conversion.</td>
</tr>
<tr>
<td>Visser et al</td>
<td>2011</td>
<td>South Africa</td>
<td>154</td>
<td>Daily dose of retinyl palmitate (200 000 IU) equivalent to retinol (5000 IU) plus zinc (5 mg) for 8 weeks durations</td>
<td>Standard anti-TB drugs</td>
<td>6 months</td>
<td>The primary outcomes of interest were sputum smear and culture conversion.</td>
</tr>
<tr>
<td>Ginawi et al</td>
<td>2013</td>
<td>India</td>
<td>82</td>
<td>Daily dose of Zinc (15 mg), Vitamin A (5000 IU) and combined</td>
<td>Placebo and anti-TB drugs</td>
<td>6 months</td>
<td>The primary outcomes of the study were tuberculosis treatment outcomes and biochemical indexes.</td>
</tr>
<tr>
<td>Range et al</td>
<td>2005</td>
<td>Tanzania</td>
<td>499</td>
<td>Daily dose of zinc tablets contained 45 mg of elemental zinc</td>
<td>Placebo tablets which were identical in colour, shape and size.</td>
<td>2 months</td>
<td>The primary outcome of interest was sputum culture conversion.</td>
</tr>
<tr>
<td>Armijos et al</td>
<td>2010</td>
<td>Mexico</td>
<td>39</td>
<td>Daily dose of vitamin A (5000 IU/day) as retinyl acetate and zinc (50 mg) as zinc chelate for 4 months</td>
<td>Placebo group subjects received organoleptically identical, matched placebos</td>
<td>2 months</td>
<td>The primary outcomes of interest were sputum smear conversion and nutritional status.</td>
</tr>
<tr>
<td>Singh et al</td>
<td>2013</td>
<td>India</td>
<td>26</td>
<td>Daily dose of zinc capsules (50 mg) elemental Zinc as Zinc sulphate and vitamin A capsule (25 000 IU) of vitamin A as Vitamin A palmitate</td>
<td>Anti-TB drugs only</td>
<td>2 months</td>
<td>The primary outcome of the study was sputum smear conversion.</td>
</tr>
<tr>
<td>Kumar et al</td>
<td>2018</td>
<td>India</td>
<td>80</td>
<td>Daily dose of zinc tablets (15 mg)</td>
<td>Anti-TB drugs only</td>
<td>2 months</td>
<td>The primary outcomes were sputum smear conversion and serum zinc levels.</td>
</tr>
</tbody>
</table>

TB, tuberculosis.
Effect of zinc and vitamin A supplementation on serum zinc, vitamin A and haemoglobin levels

Three studies reported the effects of vitamin A and zinc supplementation on serum retinol levels. The pooled results of these trials showed that the mean difference (MD) of serum retinol values at 2 months comparing zinc plus vitamin A supplementation to placebo was 0.06 µmol/L (95% CI: 0.04 to 0.08), while at 6 months, the pooled MD was 0.12 µmol/L (95% CI: 0.10 to 0.14) (figure 4).

In four trials, the intervention group had elevated mean plasma zinc levels at 2 months (pooled MD of: 0.86 µmol/L (0.14 to 1.57)). However, there

Figure 2  Effect of vitamin A, zinc and combined vitamin A and zinc supplementation on successful TB treatment outcomes. TB, tuberculosis.

Figure 3  The effect of zinc and combined zinc and vitamin A supplementation on sputum conversion rate among adults with pulmonary TB. TB, tuberculosis.
was no significant difference between the intervention and comparison groups at 6 months (pooled MD: 0.28 μmol/L (−0.06 to 0.62)) (figure 5).

The estimated pooled MD in haemoglobin levels at 2 months comparing zinc plus vitamin A supplementation to placebo was 0.15 g/dL (95% CI: −0.05 to 0.80), and at 6 months, this difference was 0.29 g/dL (95% CI: 0.08 to 0.51) (figure 6).

Risk of bias and quality of evidence
All included trials were regarded as high quality, and the risk of bias was not thought to be significant. The trials described sufficiently a low-risk method of random sequence generation.

In addition, data collectors and researchers in seven trials remained blinded to participants and personnel, except for two trials, where this was not explicitly indicated. Five trials were evaluated to be at high risk of attrition bias owing to incomplete outcome data, and we considered all trials to be unclear for selection bias as this was not clearly explained (figures 7 and 8).

Overall, we judged the certainty of the evidence for each outcome to be low or moderate. We downgraded
the certainty of the evidence because the micronutrient status differed greatly among people living with sputum-positive pulmonary TB and may be difficult to generalise the findings of the trials to all populations or settings; and most of the included studies had a small sample size which may not be able to detect the true effect (online supplemental tables S1 and S2).

DISCUSSION

This systematic review and meta-analysis synthesised data from nine RCTs and showed that zinc and vitamin A supplementation was significantly associated with improved sputum smear conversion and higher serum retinol, zinc and haemoglobin levels at different durations of follow-up. However, zinc and/or vitamin A supplementation during TB treatment had no effect on a successful TB treatment outcome.

Nutritional supplementation may be provided for people with TB to promote treatment adherence, improve treatment outcomes or mitigate the financial cost of TB care.53 It was a cornerstone of TB management in the pre-antibiotic era. Currently, WHO recommends that all individuals living with TB should be evaluated for their nutritional status and provided with adequate counselling based on their nutritional status at the time of TB diagnosis and throughout the course of treatment.54 In addition, micronutrient supplementation with vitamins such as vitamin A has been found to boost the immune system or used for the adjunctive treatment of infectious diseases, including TB.55

The findings of our systematic review and meta-analysis are consistent with a previous meta-analysis that showed a non-significant effect on TB treatment success after 6 months of micronutrient supplementation.56 Similar studies have also demonstrated that multi-micronutrient supplementation does not significantly affect treatment outcomes among adults with pulmonary TB.57 58 Various factors may account for this non-significant finding, including a small number of included studies in the analysis. Moreover, the included trials did not categorise patients based on nutritional and comorbid status. For instance, people with TB who are malnourished and in a catabolic state may require a higher dosage of zinc and vitamin A supplementation to be effective than those who are well-nourished.

In contrast to our findings, another systematic review found that nutritional support (eg, protein, energy and micronutrient supplementation) improved TB treatment outcomes in the intervention group compared with the control group.50 The possible explanation for the variation between studies might be due to the different types of interventions given to each population. For instance, our study focused on zinc plus vitamin A supplementation only, rather than any nutritional support including multi-micronutrients, protein, fat, carbohydrates or other forms of nutritional support.
Nutritional support inclusive of locally available nutrient-rich or fortified supplementary foods is currently recommended by WHO for patients with TB with moderate undernutrition who fail to gain weight after 2 months of TB treatment or for those who lose weight during this time.59

Sputum conversion is generally a prognostic indicator of a positive TB treatment outcome,60 61 with sputum conversion a key indicator at the end of the initial phase of TB treatment.62 Our evidence synthesis demonstrated that a combination of zinc and vitamin A supplementation was associated with sputum smear conversion at the end of 2 months of TB treatment. This finding is consistent with a previous meta-analysis, which found that micronutrient supplemenations had a positive effect on sputum smear conversion.63 Another meta-analysis showed that nutritional support considerably improved sputum smear conversion.64 This has been shown that early sputum conversion has a positive impact on TB control more broadly by shortening the period of infectiousness and thereby decreasing transmission.64 Delayed sputum smear conversion has been associated with an increased length of the infectious period,65 a heightened risk of treatment failure and the subsequent development of drug resistance TB.66 Theoretically, it could also lead to an upsurge in treatment costs, non-adherence and ongoing stigma.67

The reason behind zinc and vitamin A supplementation being associated with sputum smear conversion at 2 months following treatment initiation but not with successful TB treatment outcomes merits further discussion. The included trials had a limited sample size and their primary outcome of interest was predominantly early bactericidal activity in the form of sputum conversion status 2 months following treatment initiation. The sample sizes were mostly underpowered to examine the comprehensive effects of zinc and vitamin A supplementation on final TB treatment outcomes, which are usually measured at 6 months following treatment initiation. Other explanations include the use of the Directly Observed Treatment Short-course (DOTS) strategy during the initial treatment phase (the first 2 months of treatment) in closely monitoring adherence to standard TB drugs and vitamin A and zinc supplementation. Other factors contributing to the observed variations may include variations in drug absorption and adverse reactions,68 69 and development of drug resistance70 in specific subpopulations of people with TB.

In this study, combined zinc and vitamin A is found to increase serum retinol levels in adults with sputum-positive pulmonary TB. It is known that zinc supplementation helps with vitamin A absorption in the intestine, as well as having a vital role in vitamin A transport.71 72 In addition, the oxidative conversion of retinol to retinal requires the zinc-dependent retinol dehydrogenase enzyme.73 This means that supplementation with vitamin A alone may not have a beneficial effect for adults with sputum-positive pulmonary TB, rather a combination with vitamin A and zinc is needed to correct it. Clinical studies have shown that high serum retinol levels enhance innate immunity, maintain the mucosal epithelium, and are connected with the function of T and B lymphocytes.16 74 Conversely, low serum retinol levels are associated with a significantly heightened risk of developing TB disease.75 76 Vitamin A deficiency distorts the development of neutrophils and macrophages, impairing macrophage function, making them unable to kill microorganisms.77 This leads to an upsurge in bacterial adherence to respiratory epithelial cells,78 a greater bacterial load and more severe lesions in the lung.79

Likewise, serum zinc levels were significantly higher in the zinc plus vitamin A supplementation group after 2 months of follow-up. This finding is in concordance with results from a previous trial, which showed a significant increase in serum zinc concentrations after supplementation with a lower dose of zinc of 15 mg/day over
Also, this finding is similar to a single study which reported that the intervention group had a significantly elevated mean serum zinc value. Adequate serum zinc levels are essential for immune system function, particularly T cell-mediated function, as zinc deficiency is thought to be one of the primary causes of morbidity in low-income countries and can increase vulnerability to TB infection, and is thought to decrease phagocytosis and reduce tuberculin reactivity. This is confirmed by the fact that zinc has various biological functions in enzymatic catalysis, cellular signal transduction and the immune system. As well, zinc contributes to the production of IL-1α from alveolar macrophages in people with pulmonary TB. High zinc levels in phagosomes are expected to kill mycobacteria through the mechanism of metal poisoning, and are fundamental in thymus function modulating T lymphocyte maturation.

Haemoglobin levels at baseline were comparable between the intervention and comparator groups in this analysis. In previous studies, zinc and vitamin A supplementation for adults living with pulmonary TB significantly increased serum haemoglobin levels which supports our finding. This could be due to various mechanisms, including the fact that zinc increases the lifespan of red blood cells, zinc-dependent enzymes are involved in haemoglobin synthesis, and an additional step mediated by aminolaevulinic acid dehydrase and erythropoiesis stimulation through its role in a zinc-finger transcription factor, GATA-binding protein 1 (globin transcription factor 1) during erythropoiesis. Similarly, vitamin A appears to mobilise iron stores, enhances hematopoiesis through increasing circulating erythropoietin and prevents anaemia related to infections.

Our systematic review and meta-analysis has some strengths and limitations. The included trials had a low risk of bias and were generally considered of moderate quality. Currently, the doses of micronutrients for people with TB are not standardised. We attempted to examine a specific micronutrient intervention, that is, supplementation with zinc and vitamin A only, rather than multi-micronutrient supplementation, which made our review question very specific. In addition, measurement bias is unlikely in our study as the included trials used homogeneous laboratory methods to measure serum haemoglobin, zinc and retinol levels. Spectrophotometry was used to estimate serum zinc, an automatic analyzer was used to estimate haemoglobin and High-Performance Liquid Chromatography was used to estimate vitamin A (except for one trial which used ELISA).

Important limitations included the inclusion of studies published only in the English language which may miss important studies published in other languages. Stratified and subgroup analyses were not carried out due to the small number of included trials. Lastly, owing to the unavailability of relevant reported data in the included trials, we were unable to evaluate the role of some potential confounders such as an individual’s nutritional status and the dietary source of vitamin A and zinc.

Implications for future research
More is to be done, notably on TB treatment outcome and important biomarkers, to adequately appraise the long-term effects of micronutrient supplementation such as re-infection and chronic sequelae. The WHO has guidelines on nutritional support for people with TB including micronutrient supplementation. These recommendations state that all essential macronutrients and micronutrients are required for the health and well-being of humans, including those with TB infection and TB disease. However, despite this, there are a number of research gaps related to nutritional support, micronutrient supplementation and TB prevention and treatment.

Rigorous, well-powered multicentric clinical trials to investigate the effect of zinc and vitamin A supplementation on successful TB treatment outcomes are warranted. Such trials should ideally stratify by nutritional and comorbid status (eg, comorbid HIV, diabetes) to examine differential dose-response effects in these key, vulnerable populations.

CONCLUSION
Our study found that the effect of zinc and vitamin A supplementation on TB treatment outcomes in adults with sputum-positive pulmonary TB was not significant although it improved sputum smear conversion, and serum zinc, serum retinol and serum haemoglobin levels. We recommend further trials to determine whether zinc with vitamin A supplementation is needed to improve treatment outcomes in adults with sputum-positive pulmonary TB.

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