BMJ Global Health

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

Fasil Wagnew ,[1,2] Kefyalew Addis Alene,[3,4] Setegn Eshetie,[5,6] Tom Wingfield,[7,8] Matthew Kelly ,[2] Darren Gray[2]

To cite: Wagnew F, Alene KA, Eshetie S, *et al*. Effects of zinc and vitamin A supplementation on prognostic markers and treatment outcomes of adults with pulmonary tuberculosis: a systematic review and meta-analysis. *BMJ Global Health* 2022;**7**:e008625. doi:10.1136/bmjgh-2022-008625

Handling editor Seye Abimbola

► Additional supplemental material is published online only. To view, please visit the journal online (http://dx.doi.org/10.1136/bmjgh-2022-008625).

Received 12 February 2022
Accepted 7 August 2022

For numbered affiliations see end of article.

Correspondence to
Fasil Wagnew;
fasilw.n@gmail.com

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 sputum-positive 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.

PROSPERO registration number CRD42021248548.

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.

HOW THIS STUDY MIGHT AFFECT RESEARCH, PRACTICE OR POLICY

⇒ Supplementing standard TB treatment with zinc and vitamin A for adults with sputum-positive pulmonary TB may improve TB prognostic markers such as sputum smear conversion.

⇒ To optimise dosing and to evaluate the impact on TB treatment success, high-quality, well-powered multicentric clinical trials of zinc and vitamin A supplementation for adults with TB stratified by nutrition and comorbid status are required.

INTRODUCTION

Tuberculosis (TB) is caused by the bacterium *Mycobacterium tuberculosis* (*M. tuberculosis*).[1] 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

deaths among HIV-negative individuals and an additional 214 000 deaths among HIV-positive people.[2] Nearly one-quarter of the global population is infected with *M. tuberculosis*, and 5%–10% of this population may develop TB during their lifetime. The risk of progression from TB infection to TB disease is significantly higher in immunosuppressed and malnourished individuals.[3]

Geographically, the burden of TB remains substantially high in the South-East Asian (44%) and African (25%)[4] regions where it overlaps with poverty, malnutrition and other comorbidities such as HIV infection and diabetes mellitus (DM).[5] Poor nutritional status and food insecurity are significant contributors to the burden of TB, since undernutrition increases the risk of progression from TB infection to TB disease[6–8] and was estimated to be responsible for 1.9 million cases globally in 2020.[2] Micronutrient deficiencies, such as vitamin A and zinc deficiencies, can also be common among people with TB,[9–11] and the effects of these deficiencies on TB treatment or whether supplementation is useful to help improve TB treatment outcomes are unknown.

Vitamin A and zinc play a pivotal role in the immune system, specifically regarding the growth and function of B and T cells, the maintenance of mucosal epithelia and antibody responses,[12 13] which may aid bacterial clearance. Vitamin A, in the form of retinol in plasma,[14] plays a vital role in the development of Th1 and Th2 lymphocyte subsets and the activation of lymphocytic proliferation.[15] 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,[16 17] a prognostic marker for TB treatment outcomes.[18] Although the mechanism remains unclear, vitamin A is crucial for the protection of mucosal immunity.[19] In addition, zinc is a trace element crucial for immune cell function.[20] It also has a role in vitamin A metabolism.[21] Sufficient serum zinc level can also decrease free radical membrane damage during inflammation,[22] and increase the numbers of T and natural killer cells.[23] The bacterial load has been found to be considerably reduced through administering zinc supplementation.[24] 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.[25]

The effectiveness of TB treatment relies on a competent cell-mediated immune system and the appropriate use and duration of antibiotics.[26] Successful chemotherapy causes a rapid return to protective immune responses.[27] 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.[28] In some trials, micronutrient supplementation has been linked to improved sputum smear and culture conversion rates, and lower mortality rates compared with a placebo.[29 30] However, evidence from earlier trials has been contradictory, showing no therapeutic effects of micronutrient supplementation on TB treatment outcomes.[31–33] 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.[34–36] 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.[37 38] 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.[39]

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.[40] 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.[41] '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'.[42]

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.[43] Discrepancies were resolved by discussion, and when needed, a third reviewer acted as mediator (KAA). Studies were considered as a low risk of bias when all-important contents were judged and found to be at low risk.[43] 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.[44] 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 I^2 statistic with corresponding p values and qualitatively by visually examining forest plots.[45] 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

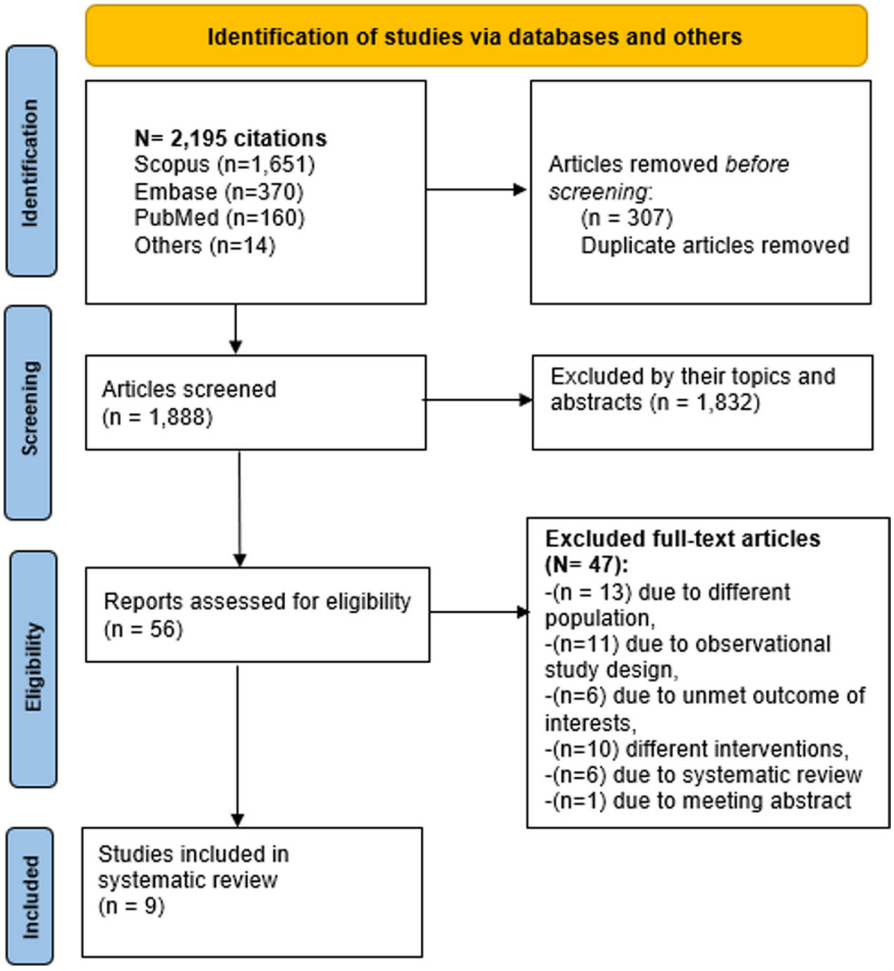

Figure 1 Flowchart diagram describing selection of studies for a meta-analysis of the effect of zinc and vitamin A supplementation on tuberculosis treatment outcomes and its prognostic markers.

full-text reviews. Then, nine studies[32 33 46–52] comprising a total of 1375 participants met the eligibility criteria and were incorporated in the systematic review (figure 1).

Of the included studies, three[46 50 51] were published in 2010, while all studies were published between 2002 and 2018. Study-specific sample sizes ranged from 26[47] to 499[32] 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,[33] Nigeria,[50] India,[47 48 52] Tanzania,[32] Indonesia[49 51] and Mexico.[46] 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[33 48–51] 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[32 33] 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[33 46–49] 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

Authors	Year of publication	Country	Total number of participants	Intervention arm	Comparator arm	Follow-up months	Primary outcomes
Karyadi *et al*[29]	2002	Indonesia	110	Daily dose of retinol (1500) equivalents vitamin A (5000 IU) as retinyl acetate and zinc (15 mg) as zinc sulfate	The placebo contained lactose alone with anti-TB drugs	6 months	The primary outcomes of the study were clinical responses and nutritional status.
Lawson *et al*[50]	2010	Nigeria	233	Zinc (90 mg) five times and daily dose of vitamin A (5000 IU)	Placebo with Anti-TB drugs	6 months	The primary outcomes of the study were sputum smear conversion and resolution of radiographic abnormalities.
Pakasi *et al*[51]	2010	Indonesia	152	Daily dose of zinc only (15 mg), vitamin A only (5000 IU), and combined.	Placebo and anti-TB drugs	6 months	The primary outcome of the study was sputum smear conversion.
Visser *et al*[33]	2011	South Africa	154	Daily dose of retinyl palmitate (200 000 IU) equivalent to retinol (5000 IU) plus zinc (5 mg) for 8 weeks durations	Standard anti-TB drugs	6 months	The primary outcomes of interest were sputum smear and culture conversion.
Ginawi *et al*[48]	2013	India	82	Daily dose of Zinc (15 mg), Vitamin A (5000 IU) and combined	Placebo and anti-TB drugs	6 months	The primary outcomes of the study were tuberculosis treatment outcomes and biochemical indexes.
Range *et al*[32]	2005	Tanzania	499	Daily dose of zinc tablets contained 45 mg of elementary zinc	Placebo tablets which were identical in colour, shape and size.	2 months	The primary outcome of interest was sputum culture conversion.
Armijos *et al*[46]	2010	Mexico	39	Daily dose of vitamin A (5000 IU/day) as retinyl acetate and zinc (50 mg) as zinc chelate for 4 months	Placebo group subjects received organoleptically identical, matched placebos	2 months	The primary outcomes of interest were sputum smear conversion and nutritional status.
Singh *et al*[47]	2013	India	26	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	Anti-TB drugs only	2 months	The primary outcome of the study was sputum smear conversion.
Kumar *et al*[52]	2018	India	80	Daily dose of zinc tablets (15 mg)	Anti-TB drugs only	2 months	The primary outcomes were sputum smear conversion and serum zinc levels.

TB, tuberculosis.

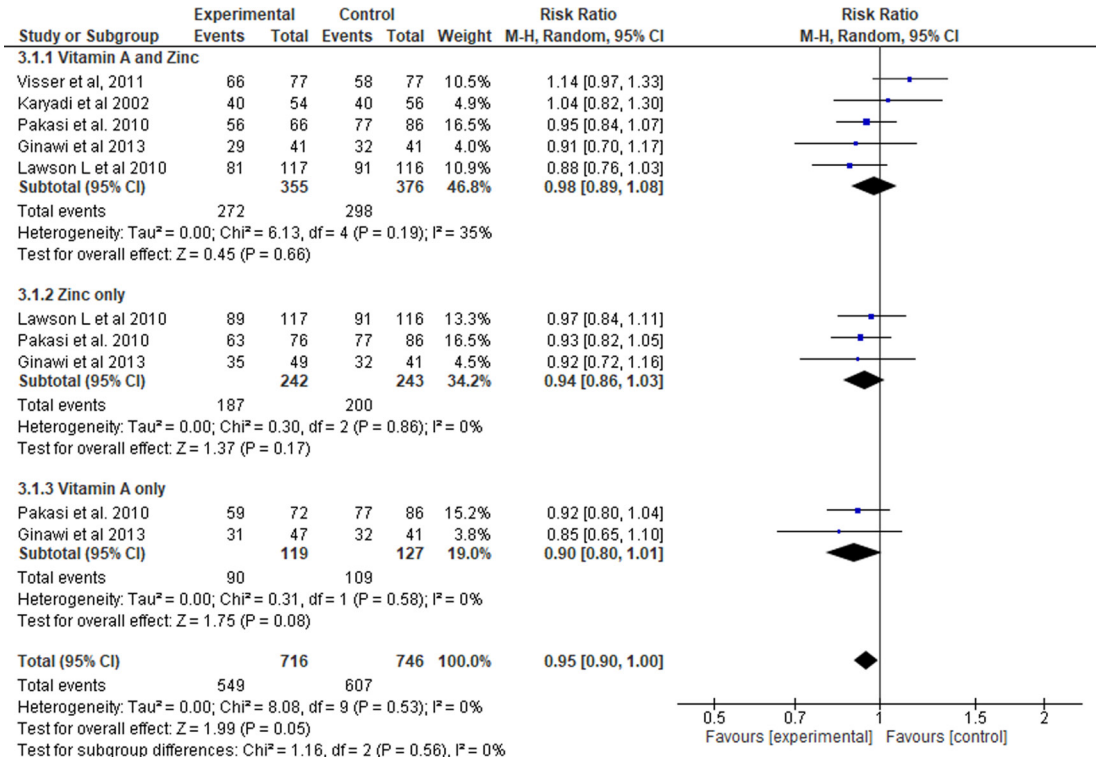

Figure 2 Effect of vitamin A, zinc and combined vitamin A and zinc supplementation on successful TB treatment outcomes. TB, tuberculosis.

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

Three studies[46 48 49] 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,[46 48 49 51] 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

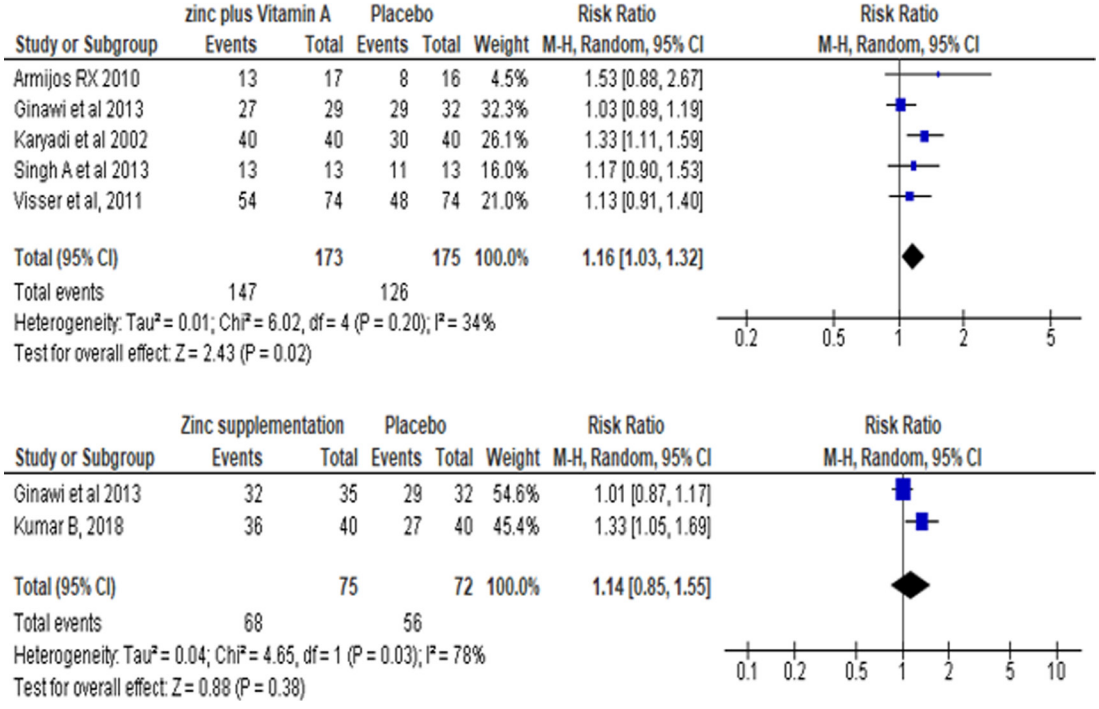

Figure 3 The effect of zinc and combined zinc and vitamin A supplementation on sputum conversion rate among adults with pulmonary TB. TB, tuberculosis.

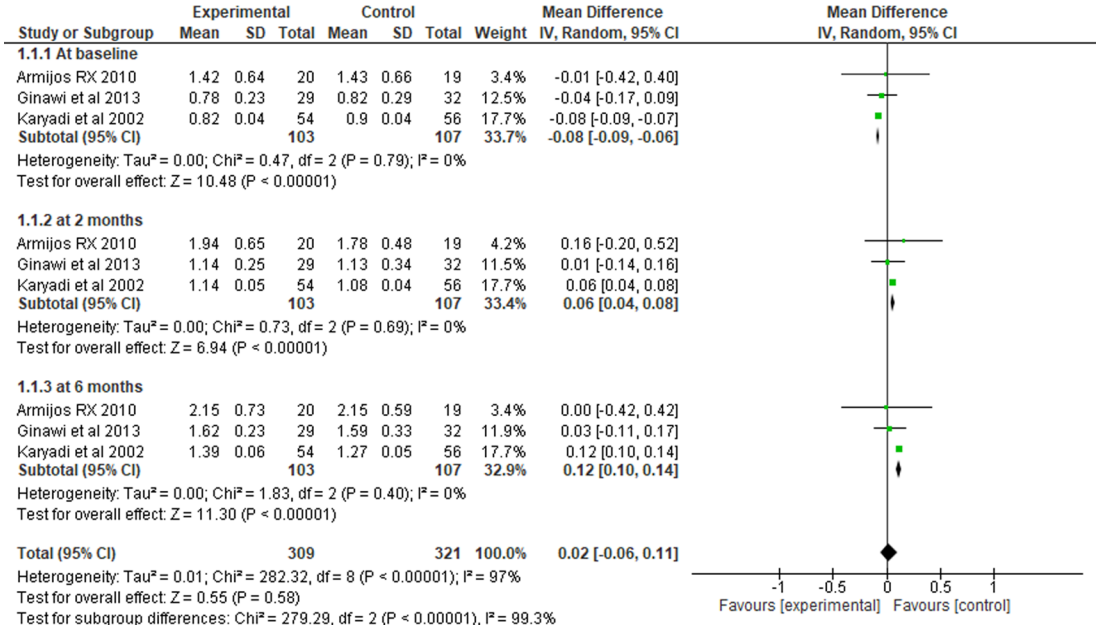

Figure 4 Effect of combined zinc and vitamin A supplementation on serum retinol levels at baseline, 2 months and 6 months.

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,[47 52] where this was not explicitly indicated. Five trials[32 33 48–50] 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

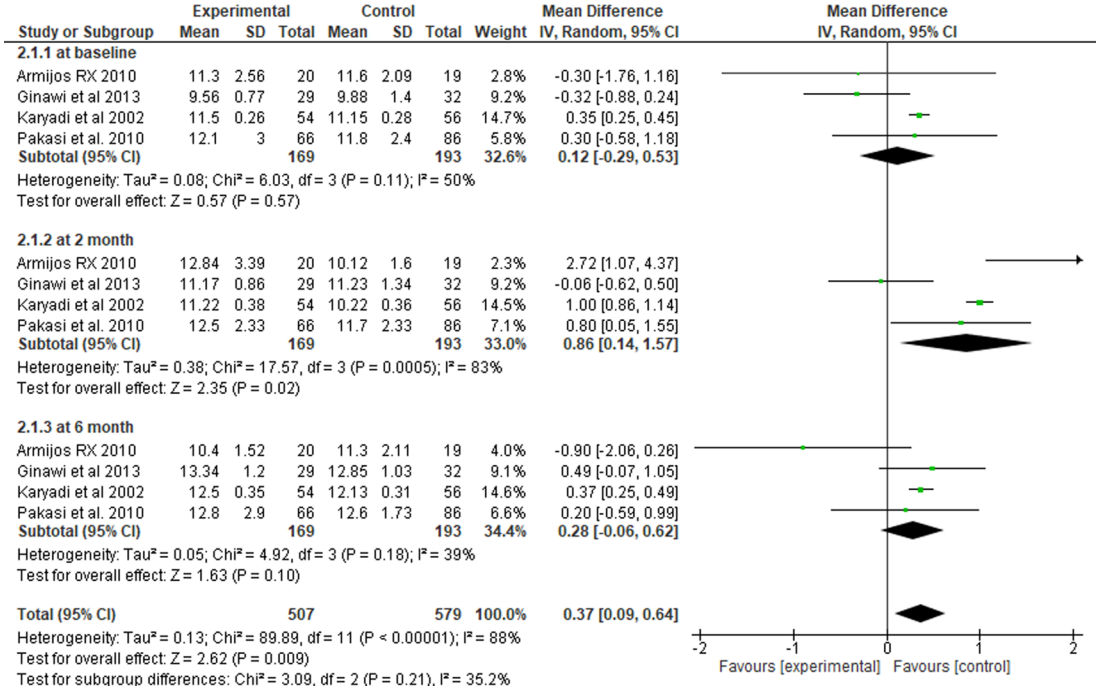

Figure 5 Effect of combined vitamin A and zinc on serum zinc levels at baseline, 2 months and 6 months.

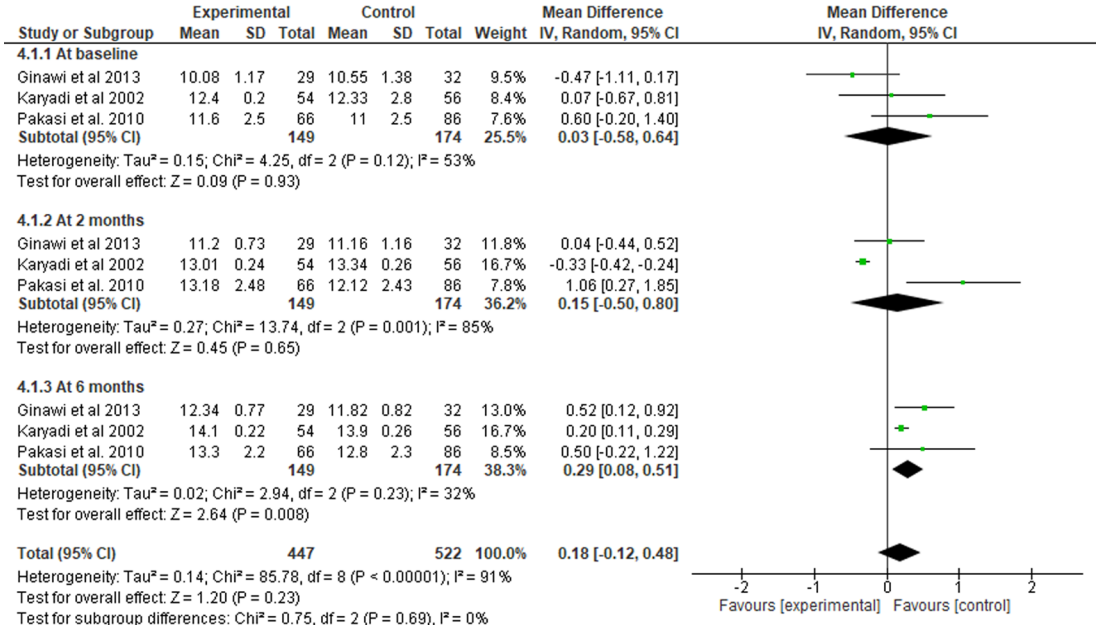

Figure 6 Effect of combined zinc and vitamin A on serum haemoglobin levels at baseline, 2 months and 6 months.

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

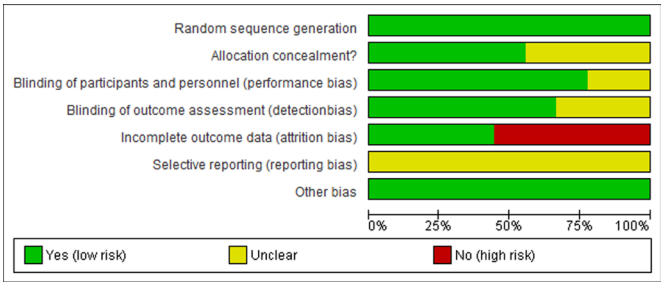

Figure 7 Risk of bias graph: review authors' judgements about each risk of bias item presented as percentages across all included studies.

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.[36] 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.

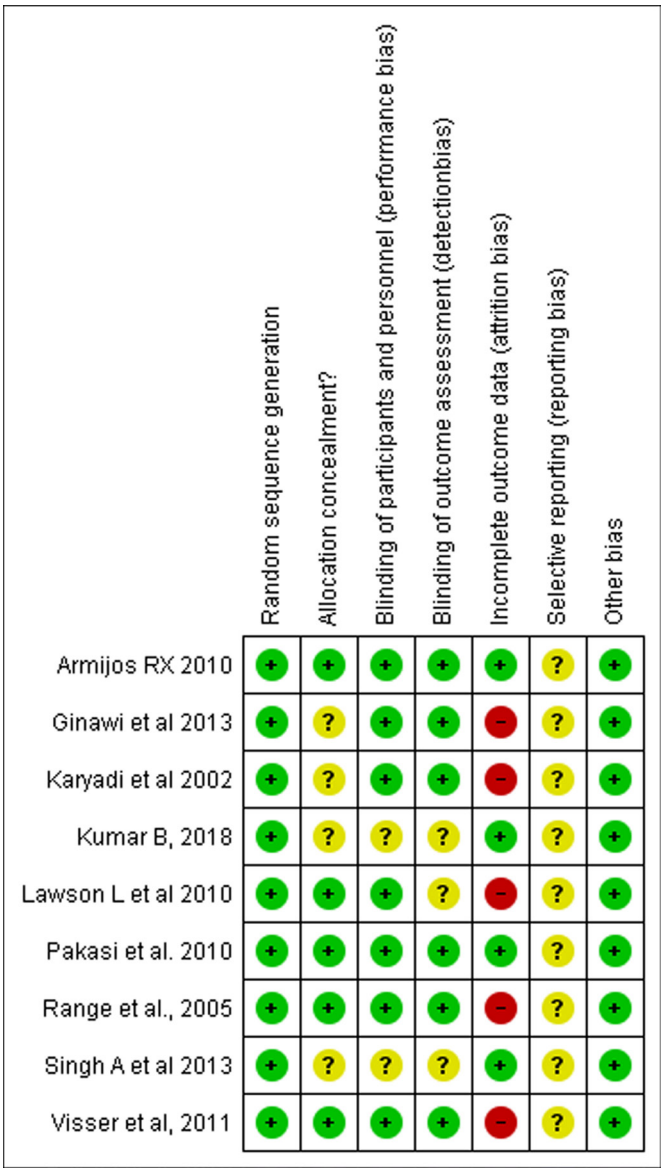

Figure 8 Risk of bias summary: review authors' judgements about each risk of bias item for each included study.

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 month 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 supplementations had a positive effect on sputum smear conversion.[63] Another meta-analysis showed that nutritional support considerably improved sputum smear conversion.[36] 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 for the observed variations may include variations in drug absorption and adverse reactions,[68 69] and development of drug resistance[70] 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

6 months.[80] Also, this finding is similar to a single study which reported that the intervention group had a significantly elevated mean serum zinc value.[81] 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[82 83] and can increase vulnerability to TB infection, and is thought to decrease phagocytosis and reduce tuberculin reactivity.[84] This is confirmed by the fact that zinc has various biological functions in enzymatic catalysis, cellular signal transduction and the immune system.[85] As well, zinc contributes to the production of IL-1a from alveolar macrophages in people with pulmonary TB.[86] High zinc levels in phagosomes are expected to kill mycobacteria through the mechanism of metal poisoning,[87] and are fundamental in thymus function modulating T lymphocyte maturation.[88]

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.[89 90] This could be due to various mechanisms, including the fact that zinc increases the lifespan of red blood cells,[91] zinc-dependent enzymes are involved in haemoglobin synthesis,[92] 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.[92 93] Similarly, vitamin A appears to mobilise iron stores, enhances hematopoiesis through increasing circulating erythropoietin and prevents anaemia related to infections.[94 95]

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 multimicronutrient 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).[48]

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.

Author affiliations
[1]College of Health Science, Debre Markos University, Debre Markos, Ethiopia
[2]National Centre for Epidemiology and Population Health, College of Health and Medicine, Australian National University, Canberra, Australian Capital Territory, Australia
[3]Faculty of Health Sciences, Curtin University, Perth, Western Australia, Australia
[4]Geospatial Health and Development, Telethon Kids Institute, Nedlands, Western Australia, Australia
[5]College of Health Science, University of Gondar, Gondar, Ethiopia
[6]Allied health performance Academic Unit, University of South Australia, Adelaide, South Australia, Australia
[7]Clinical Sciences and International Public Health, Liverpool School of Tropical Medicine, Liverpool, UK
[8]WHO Collaborating Centre for Social Medicine and Tuberculosis, Department of Global Public Health, Karolinska Institute, Stockholm, Sweden

Acknowledgements We thank the Australian National University library for offering us a wide range of available online databases. The authors additionally acknowledge Dr Kerri Viney, of the Global Tuberculosis Programme, World Health Organization (WHO), Switzerland, who provided constructive and helpful comments during the development of this manuscript.

Contributors FW contributed to conception of the research protocol, study design, quality assessment, data extraction, data analysis, interpretation and draft of the manuscript. SE and KAA contributed to study design, data extraction, quality assessment, data analysis and reviewing and editing the manuscript. DG, TW and MK contributed to interpretation of the data, reviewing and editing the manuscript. The manuscript has been read and approved by all the authors. FW is the guarantor for this publication.

Funding The authors have not declared a specific grant for this research from any funding agency in the public, commercial or not-for-profit sectors.

Competing interests None declared.

Patient and public involvement Patients and/or the public were not involved in the design, or conduct, or reporting, or dissemination plans of this research.

Patient consent for publication Not applicable.

Provenance and peer review Not commissioned; externally peer reviewed.

Data availability statement All data relevant to the study are included in the article or uploaded as supplementary information. All data generated or analysed during this study are included in this published article (and its supplementary information files).

ORCID iDs
Fasil Wagnew http://orcid.org/0000-0003-3087-2312
Matthew Kelly http://orcid.org/0000-0001-7963-2139

REFERENCES

1 Gutierrez MC, Brisse S, Brosch R, *et al*. Ancient origin and gene mosaicism of the progenitor of Mycobacterium tuberculosis. *PLoS Pathog* 2005;1:e5.
2 WHO. Global tuberculosis report 2021.
3 Yuniastuti A. The role and characteristics of antioxidant for redox homeostasis control system in Mycobacterium tuberculosis. *Int Res J Microbiol* 2012;3:416–22.
4 WHO. Global tuberculosis report 2020 2020.
5 WHO. WHO report 2008, global tuberculosis control; surveillance, planning, financing, WHO, Geneva, Switzerland 2008.
6 Cegielski JP, McMurray DN. The relationship between malnutrition and tuberculosis: evidence from studies in humans and experimental animals. *Int J Tuberc Lung Dis* 2004;8:286–98.
7 Lönnroth K, Castro KG, Chakaya JM, *et al*. Tuberculosis control and elimination 2010–50: cure, care, and social development. *Lancet* 2010;375:1814–29.
8 Lönnroth K, Williams BG, Cegielski P, *et al*. A consistent log-linear relationship between tuberculosis incidence and body mass index. *Int J Epidemiol* 2010;39:149–55.
9 Karyadi E, Schultink W, Nelwan RH, *et al*. Poor micronutrient status of active pulmonary tuberculosis patients in Indonesia. *J Nutr* 2000;130:2953–8.
10 McMurray DN, Bartow RA, Mintzer CL, *et al*. Micronutrient status and immune function in tuberculosis. *Ann N Y Acad Sci* 1990;587:59–69.
11 van Lettow M, West CE, van der Meer JWM, *et al*. Low plasma selenium concentrations, high plasma human immunodeficiency virus load and high interleukin-6 concentrations are risk factors associated with anemia in adults presenting with pulmonary tuberculosis in Zomba district, Malawi. *Eur J Clin Nutr* 2005;59:526–32.
12 Semba RD. Vitamin A, immunity, and infection. *Clin Infect Dis* 1994;19:489–99.
13 Rink L, Gabriel P. Zinc and the immune system. *Proc Nutr Soc* 2000;59:541–52.
14 Wolf G. Multiple functions of vitamin A. *Physiol Rev* 1984;64:873–937.
15 Halevy O, Arazi Y, Melamed D, *et al*. Retinoic acid receptor-alpha gene expression is modulated by dietary vitamin A and by retinoic acid in chicken T lymphocytes. *J Nutr* 1994;124:2139–46.
16 Anand PK, Kaul D, Sharma M. Synergistic action of vitamin D and retinoic acid restricts invasion of macrophages by pathogenic mycobacteria. *J Microbiol Immunol Infect* 2008;41:17.
17 Gogulamudi VR, Dubey ML, Kaul D, *et al*. Downregulation of host tryptophan-aspartate containing coat (TACO) gene restricts the entry and survival of Leishmania donovani in human macrophage model. *Front Microbiol* 2015;6:946.
18 Crowle AJ, Ross EJ. Inhibition by retinoic acid of multiplication of virulent tubercle bacilli in cultured human macrophages. *Infect Immun* 1989;57:840.
19 Semba RD. Vitamin A. In: *Diet and human immune function*. Springer, 2004: 105–31.
20 Sobha Kumari B, Chandra RK, Kumari BS. Overnutrition and immune responses. *Nutr Res* 1993;13:S3–18.
21 Rahman MM, Wahed MA, Fuchs GJ, *et al*. Synergistic effect of zinc and vitamin A on the biochemical indexes of vitamin A nutrition in children. *Am J Clin Nutr* 2002;75:92–8.
22 Taylor CG, Bray TM. Effect of hyperoxia on oxygen free radical defense enzymes in the lung of zinc-deficient rats. *J Nutr* 1991;121:460–6.
23 Chandra RK. Effect of vitamin and trace-element supplementation on immune responses and infection in elderly subjects. *Lancet* 1992;340:1124–7.
24 Botella H, Peyron P, Levillain F, *et al*. Mycobacterial p(1)-type ATPases mediate resistance to zinc poisoning in human macrophages. *Cell Host Microbe* 2011;10:248–59.
25 Prasad AS, Meftah S, Abdallah J, *et al*. Serum thymulin in human zinc deficiency. *J Clin Invest* 1988;82:1202–10.
26 Gutiérrez-González LH, Juárez E, Carranza C, *et al*. Immunological aspects of diagnosis and management of childhood tuberculosis. *Infect Drug Resist* 2021;14:929.
27 Mazlan MKN, Mohd Tazizi MHD, Ahmad R, *et al*. Antituberculosis targeted drug delivery as a potential future treatment approach. *Antibiotics* 2021;10:908.
28 Karyadi E, Dolmans WMV, West CE, *et al*. Cytokines related to nutritional status in patients with untreated pulmonary tuberculosis in Indonesia. *Asia Pac J Clin Nutr* 2007;16:218–26.
29 Karyadi E, West CE, Schultink W, *et al*. A double-blind, placebo-controlled study of vitamin A and zinc supplementation in persons with tuberculosis in Indonesia: effects on clinical response and nutritional status. *Am J Clin Nutr* 2002;75:720–7.
30 Range N, Changalucha J, Krarup H, *et al*. The effect of multi-vitamin/mineral supplementation on mortality during treatment of pulmonary tuberculosis: a randomised two-by-two factorial trial in Mwanza, Tanzania. *Br J Nutr* 2006;95:762–70.
31 Villamor E, Mugusi F, Urassa W, *et al*. A trial of the effect of micronutrient supplementation on treatment outcome, T cell counts, morbidity, and mortality in adults with pulmonary tuberculosis. *J Infect Dis* 2008;197:1499–505.
32 Range N, Andersen AB, Magnussen P, *et al*. The effect of micronutrient supplementation on treatment outcome in patients with pulmonary tuberculosis: a randomized controlled trial in Mwanza, Tanzania. *Trop Med Int Health* 2005;10:826–32.
33 Visser ME, Grewal HM, Swart EC, *et al*. The effect of vitamin A and zinc supplementation on treatment outcomes in pulmonary tuberculosis: a randomized controlled trial. *Am J Clin Nutr* 2011;93:93–100.
34 Cabrera Andrade BK, Garcia-Perdomo HA, Andrade BKC. Effectiveness of micronutrients supplement in patients with active tuberculosis on treatment: systematic review/Meta-analysis. *Complement Ther Med* 2020;48:102268.
35 Chen L, Yin C, Cai H. The effect of micro-nutrients on malnutrition, immunity and therapeutic effect in patients with pulmonary tuberculosis: a systematic review and meta-analysis of randomised controlled trials. *Tuberculosis* 2020;125:101994.
36 Si Z-L, Kang L-L, Shen X-B, *et al*. Adjuvant efficacy of nutrition support during pulmonary tuberculosis treating course: systematic review and meta-analysis. *Chin Med J* 2015;128:3219.
37 Higgins J. Cochrane handbook for systematic reviews of interventions. Version 5.1. 0 [updated March 2011], 2011. Available: www.cochrane-handbook.org
38 Moher D, Liberati A, Tetzlaff J, *et al*. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *Int J Surg* 2010;8:336–41.
39 Schardt C, Adams MB, Owens T, *et al*. Utilization of the PICO framework to improve searching PubMed for clinical questions. *BMC Med Inform Decis Mak* 2007;7:16.
40 WHO. *Global tuberculosis report who 2014*. Geneva, Switzerland: World Health Organization, 2014.
41 Guyatt GH, Oxman AD, Vist GE. Schünemann HJ: rating quality of evidence and strength of recommendations: grade: an

emerging consensus on rating quality of evidence and strength of recommendations. *BMJ* 2008;336:924.
42 Grade working group. GRADEpro GDT: GRADEpro guideline development tool [Software]. McMaster University, (developed by Evidence Prime, Inc.) McMaster University, 2015 (developed by Evidence Prime, Inc.) [online], 2015. Available: gradepro.org [Accessed 10 Mar 2020].
43 Jüni P, Altman DG, Egger M. Systematic reviews in health care: assessing the quality of controlled clinical trials. *BMJ* 2001;323:42.
44 RevMan.5.41 download. Available: /online/learning/coresoftwarecochranereviews/revman/revman [Accessed 11 Oct 2020].
45 Higgins JPT, Thompson SG, Deeks JJ, *et al*. Measuring inconsistency in meta-analyses. *BMJ* 2003;327:557.
46 Armijos RX, Weigel MM, Chacon R, *et al*. Adjunctive micronutrient supplementation for pulmonary tuberculosis. *Salud Pública de México* 2010;52:185–9.
47 Singh AK, Gogoi JB, Pant NC, *et al*. A study on the role of vitamins and minerals supplementation in the treatment of tuberculosis. *Ind. Jour. of Publ. Health Rese. & Develop.* 2013;4:26–30.
48 Ginawi IA, Ahmed MQ, Ahmad I. Al-Hazimi am: effect of zinc and vitamin A supplementation along with inter-tubercular treatment in pulmonary tuberculosis in North Indian patients. *Int J Pharm Sci Res* 2013;4:3426–31.
49 Karyadi E, West CE, Schultink W, *et al*. A double-blind, placebo-controlled study of vitamin A and zinc supplementation in persons with tuberculosis in Indonesia: effects on clinical response and nutritional status. *Am J Clin Nutr* 2002;75:720–7.
50 Lawson L, Thacher TD, Yassin MA. Cuevas LE: Randomized controlled trial of zinc and vitamin A as co-adjuvants for the treatment of pulmonary tuberculosis. [French] Essai randomise controle de l'utilisation du zinc et de vitamine A comme co-adjuvants dans le traitement de la tuberculose pulmonaire. *Trop Med Int Health* 2010;15:1481–90.
51 Pakasi TA, Karyadi E, Suratih NMD, *et al*. Zinc and vitamin A supplementation fails to reduce sputum conversion time in severely malnourished pulmonary tuberculosis patients in Indonesia. *Nutr J* 2010;9:41.
52 Kumar BPD. Vijey M a prospective single-blinded study on the safety and efficacy of zinc supplementation in pulmonary tuberculosis. *Asian J Pharm Clin Res* 2018;11:90–4.
53 Lutge EE, Knight SE, Volmink J. Incentives for improving patient adherence to anti-tuberculosis treatment. *Cochrane Database Syst Rev* 2009;3.
54 WHO. *Guideline: nutritional care and support for patients with tuberculosis*. Geneva, Switzerland, 2013.
55 Maggini S, Wintergerst ES, Beveridge S, *et al*. Selected vitamins and trace elements support immune function by strengthening epithelial barriers and cellular and humoral immune responses. *Br J Nutr* 2007;98:S29–35.
56 Sinclair D, Abba K, Grobler L, *et al*. Nutritional supplements for people being treated for active tuberculosis. *Cochrane Database Syst Rev* 2011;11:CD006086.
57 Benn CS, Friis H, Wejse C. Should micronutrient supplementation be integrated into the case management of tuberculosis?. *J Infect Dis* 2008;197:1487–9.
58 Cabrera Andrade BK, Garcia-Perdomo HA. Effectiveness of micronutrients supplement in patients with active tuberculosis on treatment: systematic review/Meta-analysis. *Complement Ther Med* 2020;48:102268.
59 WHO. *Nutritional care and support for patients with tuberculosis*. Guideline: World Health Organization, 2013.
60 Kurbatova EV, Taylor A, Gammino VM, *et al*. Predictors of poor outcomes among patients treated for multidrug-resistant tuberculosis at DOTS-plus projects. *Tuberculosis* 2012;92:397–403.
61 World Health Organization. *Compendium of indicators for monitoring and evaluating national tuberculosis programs*. World Health Organization, 2004.
62 Bouti K, Aharmim M, Marc K, *et al*. Factors influencing sputum conversion among smear-positive pulmonary tuberculosis patients in Morocco. *ISRN Pulmonol* 2013;2013:1–5.
63 Machmud PB, Djuwita R, Gayatri D, *et al*. Influence of micronutrient consumption by tuberculosis patients on the sputum conversion rate: a systematic review and meta-analysis study. *Acta Med Indones* 2020;52:118–24.
64 Organization WH, Initiative ST. *Treatment of tuberculosis: guidelines*. World Health Organization, 2010.
65 Asemahagn MA, Alene GD, Yimer SA. Tuberculosis infectious pool and associated factors in East Gojjam zone, Northwest Ethiopia. *BMC Pulm Med* 2019;19:1–11.
66 MA SA, Salmiah M, Saliluddin S. Lim P: factors delaying sputum conversion in smear positive pulmonary tuberculosis: a systematic review. *Int J Public Health Clin Sci* 2018;5:56–61.
67 Mokti K, Isa Z, Manaf MRA. Abd Rahim SSS: nutritional burden in tuberculosis and inter-sectoral nutritional management for tuberculosis patients in Malaysia. *Türkiye Klinikleri Tip Bilimleri Dergisi* 2020;40:83–95.
68 Pereira MA, Schmaltz CA, Arriaga MB. Adverse drug reactions related to treatment of drug-susceptible tuberculosis in Brazil: a prospective cohort study 2022.
69 Wen S, Yin J, Sun Q. Impacts of social support on the treatment outcomes of drug-resistant tuberculosis: a systematic review and meta-analysis. *BMJ Open* 2020;10:e036985.
70 Warner DF. Mycobacterium tuberculosis metabolism. *Cold Spring Harb Perspect Med* 2015;5:a021121.
71 Ahn J, Koo SI. Intraduodenal phosphatidylcholine infusion restores the lymphatic absorption of vitamin A and oleic acid in zinc-deficient rats. *J Nutr Biochem* 1995;6:604–12.
72 Mathur ML. Role of vitamin A supplementation in the treatment of tuberculosis. *Natl Med J India* 2007;20:16.
73 Christian P, West KP. Interactions between zinc and vitamin A: an update. *Am J Clin Nutr* 1998;68:435S–41.
74 Wheelwright M, Kim EW, Inkeles MS, *et al*. All-trans retinoic acid-triggered antimicrobial activity against Mycobacterium tuberculosis is dependent on NPC2. *J Immunol* 2014;192:2280–90.
75 Getz HR, Long ER, Henderson HJ. A study of the relation of nutrition to the development of tuberculosis; influence of ascorbic acid and vitamin A. *Am Rev Tuberc* 1951;64:381–93.
76 Wiid I, Seaman T, Hoal EG, *et al*. Total antioxidant levels are low during active TB and rise with anti-tuberculosis therapy. *IUBMB Life* 2004;56:101–6.
77 Stephensen CB. Vitamin A, infection, and immune function. *Annu Rev Nutr* 2001;21:167–92.
78 Chandra RK. Increased bacterial binding to respiratory epithelial cells in vitamin A deficiency. *BMJ* 1988;297:834.
79 McMurray DN, Carlomagno MA, Cumberland PA. Respiratory infection with attenuated Mycobacterium tuberculosis H37Ra in malnourished guinea pigs. *Infect Immun* 1983;39:793–9.
80 Feillet-Coudray C, Meunier N, Rambeau M, *et al*. Long-Term moderate zinc supplementation increases exchangeable zinc pool masses in late-middle-aged men: the Zenith study. *Am J Clin Nutr* 2005;82:103–10.
81 Nenni V, Nataprawira HM, Yuniati T. Role of combined zinc, vitamin A, and fish oil supplementation in childhood tuberculosis. *Southeast Asian J Trop Med Public Health* 2013;44:854–61.
82 de Benoist B, Darnton-Hill I, Davidsson L, *et al*. Conclusions of the joint WHO/UNICEF/IAEA/IZiNCG Interagency meeting on zinc status indicators. *Food Nutr Bull* 2007;28:S480–4.
83 Tang AM, Graham NM, Semba RD, *et al*. Association between serum vitamin A and E levels and HIV-1 disease progression. *AIDS* 1997;11:613–20.
84 McMurray D. Micronutrient status and immune function in tuberculosis. *Ann NY Acad Sci* 1980;587:238–309.
85 Miao X, Sun W, Fu Y, *et al*. Zinc homeostasis in the metabolic syndrome and diabetes. *Front Med* 2013;7:31–52.
86 Abul HT, Abul AT, al-Athary EA, *et al*. Interleukin-1 alpha (IL-1 alpha) production by alveolar macrophages in patients with acute lung diseases: the influence of zinc supplementation. *Mol Cell Biochem* 1995;146:139–45.
87 Chandrasekaran P, Saravanan N, Bethunaickan R, *et al*. Malnutrition: modulator of immune responses in tuberculosis. *Front Immunol* 2017;8:1316.
88 Jaganath D, Mupere E. Childhood tuberculosis and malnutrition. *J Infect Dis* 2012;206:1809–15.
89 Allen LH, Peerson JM, Olney DK. Provision of multiple rather than two or fewer micronutrients more effectively improves growth and other outcomes in micronutrient-deficient children and adults. *J Nutr* 2009;139:1022–30.
90 Mehta S, Mugusi FM, Bosch RJ, *et al*. A randomized trial of multivitamin supplementation in children with tuberculosis in Tanzania. *Nutr J* 2011;10:1–9.
91 Powell SR. The antioxidant properties of zinc. *J Nutr* 2000;130:1447S–54.
92 Garnica AD. Trace metals and hemoglobin metabolism. *Ann Clin Lab Sci* 1981;11:220–8.
93 Labbaye C, Valtieri M, Barberi T, *et al*. Differential expression and functional role of GATA-2, NF-E2, and GATA-1 in normal adult hematopoiesis. *J Clin Invest* 1995;95:2346–58.
94 Semba RD, Bloem MW. The anemia of vitamin A deficiency: epidemiology and pathogenesis. *Eur J Clin Nutr* 2002;56:271–81.

95 Mazza JJ. Insights into Global Nutritional Anemia: Nutritional Anemia. Kraemer K, Zimmermann M, eds. 399 pp. $35 donation to the UN World Food Programme "school feeding" is requested. ISBN 978-3-906412-33-4. Basel, Switzerland: Sight and Life Press, 2007; * The Guidebook: Nutritional Anemia. Badham J, Zimmermann MB, Kraemer, K, eds. 50 pp. Free of charge. ISBN 978-3-906412-35-1. Basel, Switzerland: Sight and Life Press, 2007. *Clin Med Res* 2007;5:209.