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REVIEW ARTICLE

The role of circulating 25 hydroxyvitamin D in asthma: a systematic review

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Abstract

Asthma is a major public health issue. The co-occurrence of the high prevalence of asthma and vitamin D deficiency documented globally in recent decades has prompted several investigations into a possible association between the two conditions. The objective of this paper was to synthesize the evidence from studies that have measured the association between serum vitamin D and asthma incidence, prevalence, severity and exacerbations. A systematic search of the literature was performed in PubMed, and the available evidence was summarized both qualitatively and by meta-analysis. Only English language, observational studies measuring serum levels of 25(OH)D as the exposure were included, as this is the most robust measure of vitamin D levels. The search identified 23 manuscripts: two case–control, 12 cohort and nine cross-sectional studies. Collectively, the evidence suggests that higher serum levels of 25(OH)D are associated with a reduced risk of asthma exacerbations, but there was little evidence to suggest an association with asthma incidence, prevalence or severity. A significant amount of heterogeneity between study methodology and results restricted the scope for meta-analysis. These results suggest that vitamin D supplementation may be effective for the prevention of asthma exacerbations, but the findings need to be confirmed by clinical trials.

Asthma is a major public health issue globally due to the high and increasing prevalence of the condition, as well as the considerable adverse impact it has on the community with regard to reduced quality of life, productivity and extensive healthcare costs (1–3). While the aetiology of asthma has been the focus of a substantial amount of research, no effective preventive strategies have been identified, and evidence concerning modifiable risk factors is inconsistent (1, 3).

Given the presence of vitamin D receptors on a variety of tissues and immune cells including the airways, the potential role of vitamin D as a modifiable factor in asthma has received much interest in the recent past. Furthermore, vitamin D status, which is assessed by serum 25(OH)D levels, is easily modified by sun exposure or diet and therefore makes an attractive target biomarker.

The biological mechanisms through which vitamin D may play a role in asthma are briefly outlined below. Airway epithelial cells express the activating enzyme 1α-hydroxylase, which causes inactive 25(OH)D to be hydroxylated to the active form calcitrol (1, 25(OH)2-D) (4–6). Calcitrol then interacts with various immune cell types both directly, and indirectly through vitamin D receptors (VDR) (7, 8). VDRs are nuclear steroid receptors, which regulate the transcription of numerous genes associated with inflammation and immunomodulation (7, 9, 10). In addition to this, vitamin D can suppress the pro-inflammatory cytokines IL-17 (7, 11) and the IL-4-mediated expression of IL-13 (7). Vitamin D may also act directly on CD4+ T cells to promote T-regulatory cells (Tregs) that secrete the anti-inflammatory cytokine IL-10 (7, 8, 11, 12). In doing so, vitamin D enhances glucocorticoid responsiveness in severe asthmatics (8, 9, 13) and therefore may mediate control of asthma exacerbations and severity of asthma symptoms. Furthermore, 25(OH)D may shift the dominance of a T-regulatory lymphocyte response from the TH1 to the less inflammatory TH2 phenotype (12, 14, 15). Additionally, vitamin D possesses antiviral properties such as the upregulation of antimicrobial peptide production and reduction of NF-kB signalling in response to respiratory

syncytial virus (RSV) (6). RSV is associated with asthma inception and is an important cause of asthma exacerbations (16, 17). Through the aforementioned mechanisms, vitamin D may plausibly play a role in the aetiology of asthma and asthma exacerbation (12).

To date, multiple observational studies have been conducted to examine the relationship between serum 25(OH)D and asthma. Reviews synthesizing the current evidence on the role of vitamin D in asthma have also been performed; however, these were either not systematic (11, 14, 18–23), or did not specifically look at the association between serum 25 (OH)D and asthma (7–9, 12, 24–27). A recently published systematic review by Rajabbik et al. (28) investigated the association between low serum vitamin D levels and the diagnosis of asthma in children, but their review was limited to cohort studies, only including a total of three, and did not include meta-analysis.

Hence, we aimed to conduct a systematic review and meta-analysis to specifically investigate the existing evidence of the relationship between serum 25(OH)D and asthma in both children and adults. This review attempts to present an overview of all the available evidence from observational studies on the association between serum vitamin D levels and asthma regardless of study design or population type.

The review aimed to address two research questions: Are serum 25(OH)D levels associated with (i) the incidence of asthma, and (ii) the prevalence, severity and exacerbations for asthma?

Methods

Search strategy

A systematic search was performed using the PubMed database for articles published from inception to 28 September 2014. Search terms used for the exposure variable were as follows: "vitamin d'[MeSH Terms], 'ergocalciferols'[MeSH Terms], 'cholecalciferol'[MeSH Terms], 'alfacalcidol', 'serum 25-hydroxyvitamin D'. For the outcome variable, the following search terms were used: 'asthma'[MeSH Terms], 'wheeze', 'asthma symptoms'. Reference lists were also manually checked to identify any articles that may have been missed by the search.

Inclusion/Exclusion criteria

Inclusion and exclusion criteria were predefined. Only English language, epidemiological studies that measured 25(OH) D concentrations in the sera of participants and correlated the measurements to any measure of subsequent or persistent asthma were included. Articles were excluded if they used 25 (OH)D estimates obtained from food frequency questionnaires and dietary and sun exposure assessments, if participants were supplemented with vitamin D or if the vitamin D exposure was combined with other exposures, if they were *in vitro* or animal model studies, or if they did not focus specifically on the exposure or outcome of interest. Only studies with a measure of 25(OH)D were included because this is the most robust measurement of vitamin D status (7, 29, 30). Additionally, existing reviews, letters to the editor and other comments were excluded if they did not contain data.

The selection criteria were checked by two independent reviewers (RC and JK). Any discrepancies were resolved by SD.

Data extraction from primary studies

The following information was extracted from each included study: author, country, year of publication, number of participants included in the analysis, study design, method of measurement and categorization of the 25(OH)D exposure and whether or not an association was found. We also extracted information on the type of asthma outcomes assessed in each included study (e.g. asthma incidence, prevalence, asthma severity or exacerbations) and the reported measure of association (i.e. odds ratios and 95% confidence intervals or mean difference and standard deviation).

To include all relevant studies in the review, an attempt was made to contact authors of two articles (31, 32) where results were not presented as odds ratios and 95% confidence intervals; however, no responses were received.

The data were tabulated separately according to each aim. Within these tables, the studies were grouped by age of participant, study design and asthma outcome, as follows: (i) association of serum 25(OH)D with the incidence of asthma in previously asthma-free populations, (ii) the association between current 25(OH)D level and the presence of prevalent asthma symptoms, (iii) the association of serum 25(OH)D with asthma severity and (iv) the association between serum 25(OH)D and asthma exacerbations. Results of the individual studies were displayed on forest plots that correspond with the tables, where appropriate.

Meta-analysis methods

Studies were included in a meta-analysis if they were of the same study design in a similar population, if they assessed comparable outcomes and if they measured and analysed the exposure variable in a like manner.

Both fixed and random effect models were used for the meta-analysis. The P statistic and I^2 statistic were utilized to assess heterogeneity between studies, and if the I^2 was significantly higher than 70%, results were not pooled. Results are presented graphically as forest plots in Fig. 7. All analyses were conducted using Stata 12 (StataCorp, College Station, TX, USA).

Results

Search strategy

A total of 340 articles were identified by the search (Fig. 1). Of these articles, 295 articles were eliminated after an initial screening of their titles and abstracts. The full texts of the remaining 45 articles were assessed in detail, and a further 22 publications were excluded as they did not meet the inclusion criteria. Hence, 23 articles were included in the final review.

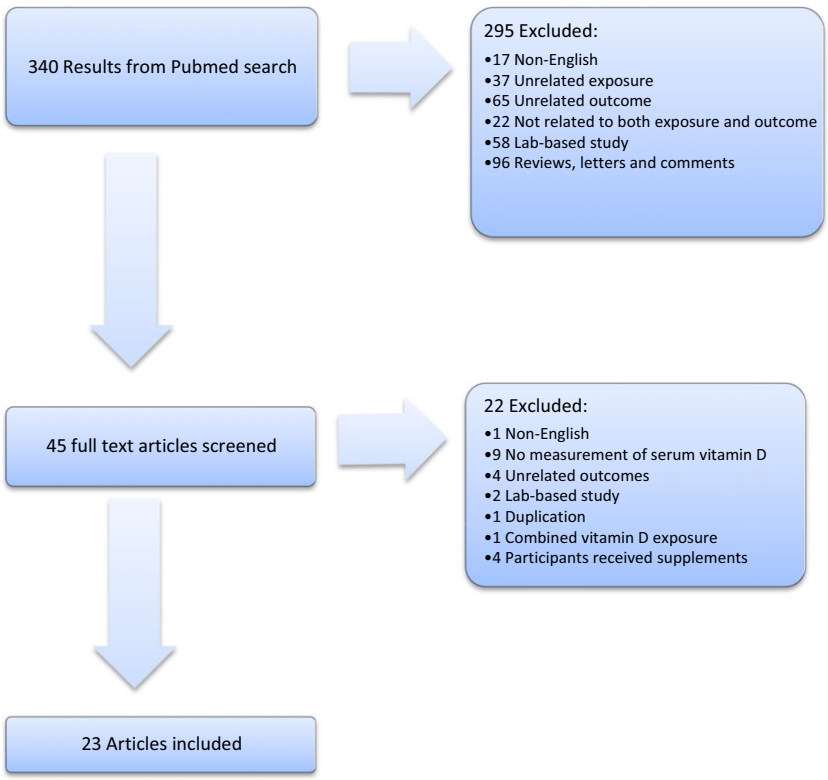

Figure 1 Flow chart of studies included in the systematic review of the association between serum 25(OH)D and asthma.

Exposure measurement

The method of measuring 25(OH)D concentration varied among the studies. Nine studies used liquid chromatography (33–41), seven used radioimmunoassay (42–47), four used a chemiluminescence assay (31, 32, 48, 49), and one used an enzyme immunoassay (50). One manuscript reported results on two distinct cohorts and utilized liquid chromatography to quantify serum 25(OH)D concentrations in one cohort and radioimmunoassay in the other (51). The measurement tool used by Beigelman et al. (52) was not clearly stated in the text or the available supporting documents.

Additionally, the definitions and categorization thresholds of 25(OH)D sufficiency, insufficiency and deficiency varied across the studies. The most commonly used categorical levels to define serum 25(OH)D sufficiency, insufficiency and deficiency were ≥30 ng/ml (≥75 nmol/l), 20–29.9 ng/ml (50–74.9 nmol/l) and <20 ng/ml (50 nmol/l), respectively (31, 33–36, 42–44, 46, 49). More than half of the included studies analysed 25(OH)D as a continuous variable (31, 32, 34–36, 39, 40, 43, 47, 48, 51).

Outcomes

As with the exposure, studies assessed and defined asthma differently, employing different guidelines and using various measurement tools.

Many of the studies that assessed asthma incidence relied on questionnaires (34, 46, 49, 51) and parental reports of physician-diagnosed asthma in their offspring (31, 37, 38, 41–43, 45, 47, 48). Commonly, asthma was defined as wheezing (37, 39, 47, 48), use of inhaler medication for asthma (35, 38), parental reports (39, 50) or physician diagnosis of asthma (32, 33, 36, 40, 43, 46, 51, 53). Asthma control and severity studies used questionnaires such as the Asthma Control Test (ACT) (31, 35, 51) or assessed control and exacerbations based on the frequency and dosage of asthma controller medications (32, 50, 52) or hospital or emergency department visits (33, 43, 44, 51) as a result of asthma.

Confounding variables

Age, sex, BMI, season of blood draw for 25(OH)D measurement, ethnicity and socio-economic status (SES) were identified from the literature as important potential confounders of the association between 25(OH)D and asthma. Of the 23 included studies, 19 (83%) adjusted or matched for sex (31, 33–37, 39–44, 46–51, 53) and 16 (70%) for age (31–35, 39–44, 46, 48, 49, 51). More than half (61%) of the included studies adjusted for season of 25(OH)D measurement (31, 34, 36, 38–41, 44, 46, 48–51), 48% adjusted for BMI (32, 35, 37, 39, 40, 43, 44, 46, 47, 49, 51) and 52% adjusted for ethnicity (32–35, 38, 39, 41, 42, 44, 46, 48, 51). Only two studies adjusted for socio-economic status (41, 49). In addition to these variables, it

is possible that unreported and undocumented steroid use may also have affected the results of the included studies by improving asthma outcomes in participants.

Studies in child populations

There were 18 individual studies conducted on children and adolescents, whose ages ranged from birth to 20 years. Two of these studies presented both prospective and cross-sectional analyses in one manuscript (50, 51); hence, there are twenty analyses in total.

Incident asthma

Six cohort studies measured asthma incidence in children after having measured either maternal serum 25(OH)D during pregnancy (37, 41, 45, 47) or serum 25(OH)D in cord blood at birth (38, 48) (Table 1). Four studies prospectively assessed the relationship between 25(OH)D levels in childhood and subsequent asthma (36, 39, 50, 51) (Table 1). All of these cohort studies included a large number of participants (n = 226–3323), except the study by van Oeffelen et al. (50), who followed only 14 children from the age of 4–8 years. All studies followed participants for at least 4 years except Gergen et al. (51) who followed 226 asthmatics over 46 weeks. This was a short period of observation compared with the other prospective studies which followed children for four (44, 50), six (39) and eight (36, 41) years.

Five of the six studies that measured 25(OH)D from cord blood or during pregnancy did not find any association between 25(OH)D and asthma (37, 38, 41, 47, 48); only Gale et al. (45) found that increasing maternal serum 25(OH)D during pregnancy increased the risk of asthma in offspring at age nine (Fig. 2).

A further four studies investigated incident asthma in children with serum vitamin D measured in childhood (36, 39, 50, 51) (Fig. 3). With the exception of Hollams et al. (36), these studies found no association. Gergen et al. (51) was excluded from Fig. 3 as they presented mean serum 25(OH)D concentrations instead of odds ratios.

Prevalent asthma

One large case–control study investigated the association in 966 asthmatic and age-matched healthy children and found that 25(H)D deficiency was associated with increased asthma (AOR = 4.82, 2.41-8.63) (42).

Severe asthma

There were two small (31, 35) cross-sectional studies conducted in Italy and the UK (n = 75 and n = 86, respectively) and three larger cross-sectional studies (32, 50, 51) investigating the relationship between serum 25(OH)D and asthma severity in cohorts of children aged between 2 and 20 years (Table 2).

One of the five studies that investigated asthma severity in children was a population-based study (50), and the remaining four were conducted in clinical cohorts of asthmatic children (31, 32, 35, 51). Three studies measured asthma severity based on the Asthma Control Test (ACT) (31, 35, 51). Both Menon et al. (32) and van Oeffelen et al. (50) defined severity as the use of controller medication.

Menon et al. (32) presented means and standard deviations for five steps of asthma severity (Table 2). Chinellato et al. (31) found the correlation between serum 25(OH)D and asthma severity, as measured by the Childhood Asthma Control Test (C-ACT), to be 0.28 (0.06–0.49). Gupta et al. (35) found that the B coefficient for the ACT was 0.15 (0.09–0.20) per 1 nmol/l increase in serum 25(OH)D concentration (Table 2). These three studies were excluded from Fig. 4 due to the measures of association that they presented. The non-African American analysis by Gergen et al. and the 8-year Van Oeffelen et al. analysis found no associations, while the African American analysis by Gergen et al. (51) and the 4-year Van Oeffelen et al. (50) analysis found that high serum 25(OH)D was associated with decreased asthma severity (Fig. 4).

Asthma exacerbations

Three cohort and three cross-sectional studies investigated the association between serum 25(OH)D and asthma exacerbations measured in terms of requirement for hospitalization and treatment with oral steroids (33, 35, 43, 44, 51, 52) (Table 2, Fig. 5). One large cross-sectional study, a *post hoc* analysis from the MIST randomized controlled trial, examined the association between serum 25(OH)D and rate of exacerbations in 264 children aged 12–53 months with a history of severe intermittent wheezing and a positive modified Asthma Predictive Index (52). Gupta et al. (35) had a relatively small sample size of 86 children compared to the remaining studies whose sample sizes ranged from 226 to 1024 asthmatic children. Gergen et al. (51) had a short follow-up period of 46 weeks and the number of cases of hospitalization was very small, resulting in a large odds ratio with a wide confidence interval, while the study by Brehm et al. (44) followed children for a period of 4 years. With the exception of the study by Gergen et al. that found an increased risk of exacerbations with high serum vitamin D levels, all of the included studies which measured asthma exacerbations in children found that low serum vitamin D was associated with an increased risk of asthma exacerbations requiring hospitalization or medicinal treatment (33, 35, 43, 44, 52).

Given the inclusion criteria, we were able to conduct a meta-analysis of four studies, which measured asthma exacerbations in children. Brehm et al. (33), Brehm et al. (44), Gergen et al. (51) and Beigelman et al. (52) measured asthma exacerbation outcomes in asthmatic children. In addition, all measured serum 25(OH)D as a categorical variable. Overall, high serum 25(OH)D concentrations were associated with reduced asthma exacerbations (RR = 0.64, 0.5–0.81) using the fixed effects model; however, the random effects model suggested that there may not be an association (RR = 0.67, 0.39–1.17) (Fig. 6).

Studies in adult populations

Five of the 23 studies included were conducted on adults aged 18 years and over.

Incident asthma

Mai et al. (49) conducted a nested case–control study on 2542 participants to investigate the association between

Table 1 Study characteristics of studies measuring serum 25(OH)D and asthma incidence and prevalent symptoms

Author, year, location	Number included in the analysis population type and Study design	Analytical unit of measurement Time of exposure Measurement scale	Outcomes measured (Definition)	Adjusted results	Confounding variables
Maternal serum 25(OH)D and outcomes in offspring					
Morales (2011), Spain (37)	1233 followed to age 6. Population-based cohort study	Baseline: 12.6 ± 2.5 weeks gestation (ng/ml) Categorical (by season of blood draw: Q1 = winter, Q2 = spring, Q3 = autumn, Q4 = summer)	Current asthma (asthma diagnosis, treatment at 4–6 years or wheezing since age 4). Wheeze at 1 year and annually thereafter	Asthma age 4–6: Q1: 1.00 (ref) Q2: 0.88(0.59–1.29) Q3: 0.81(0.54–1.20) Q4: 0.89(0.59–1.32) AOR wheeze at 1 year: Q1: 1.00 (ref) Q2: 1.04(0.78–1.40) Q3: 0.96(0.71–1.29) Q4: 0.91(0.67–1.23) AOR wheeze at 4 year: Q1: 1.00 (ref) Q2: 0.89(0.59–1.34) Q3: 0.82(0.54–1.25) Q4: 0.94(0.62–1.43)	Child's gender and prepregnancy BMI. Six additional variables
Gale (2008), UK (45)	178 followed until age 9. Population-based cohort study	Baseline: Median 32.6 weeks gestation (nmol/l) Categorical (<30, 30–50, 50–75, >75)	Maternal reports of asthma at 9 years	Asthma at 9 years by 25(OH)D category: <30 nmol/l = (ref) 30–50 nmol/l = 2.05(0.36–11.80) 50–75 nmol/l = 2.05(0.36–11.80) 75+ nmol/l = 5.40(1.09–26.65)	Did not adjust for any variables
Pike (2012), UK (47)	860 followed until age 6. Population-based cohort study	Baseline: 34 weeks gestation (nmol/l) Continuous	Current asthma (6-year olds diagnosed with asthma and who had symptoms or treatment in the last year). Transient *vs* persistent/ late-onset wheeze	Per 10 nmol/l change in 25(OH)D Asthma at 6 years ARR = 0.98 (0.92–1.04) Wheeze at 6 years ARR = 0.99 (0.94–1.05) Wheeze at or before 6 years ARR = 1.00(0.98–1.02) Transient wheeze at 6 years ARR = 1.00(0.98–1.02) Persistent/late wheeze at 6 years = 0.98(0.94–1.03).	Child's gender and maternal BMI. Seven additional variables
Wills (2013), UK (41)	Followed to age 7.5 years Population-based cohort study	Baseline: collected at any stage of pregnancy (nmol/l) Categorical (<38, 38–52, 52–67, 67 –82, ≥82)	Maternal report of doctor-diagnosed asthma and wheezing or asthma in the past 12 months when aged 7.5 years	Asthma at 7.5 years by 25(OH)D category <38: (ref) 38–52: 0.94 (0.71, 1.26) 52–67: 0.86 (0.64, 1.14) 67–82: 82: 0.86 (0.64, 1.15) 82 +: 1.00 (0.75, 1.33)	Maternal age and offspring sex, season of measurement, SES, ethnicity. Three additional variables

Table 1 (continued)

Author, year, location	Number included in the analysis population type and Study design	Analytical unit of measurement Time of exposure Measurement scale	Outcomes measured (Definition)	Adjusted results	Confounding variables
Cord blood 25(OH)D and asthma outcomes in the child					
Camargo (2011), New Zealand (48)	823 followed to age 5. Population-based cohort study	Cord blood (nmol/l) Continuous	Wheeze at 15 months, 3 years and 5 years. Asthma diagnosis by age 5	25(OH)D levels (per 10 nmol/l increase) and: Asthma age 5 years AOR = 1.03 (0.97–1.10) Wheeze age 15 months AOR = 0.98 (0.93–1.02) Wheeze age 3 years AOR = 0.96 (0.91–1.00) Wheeze age 5 years AOR = 0.95 (0.91–0.99)	Child's gender, season of birth, maternal age at birth, gestational age and ethnicity. Eight additional variables
Rothers (2011), USA (38)	219 followed to age 5. Population-based cohort study	Cord blood (nmol/l) Categorical (<50, 50–74.9, 75–99.9, >100)	Asthma (doctor-diagnosed asthma at age 1, 2, 3 or 5 and symptoms or meds since age 4 years)	Asthma age 5 by cord blood category: <50 AOR = 0.5 (0.2–1.6) 50–74.9 AOR = (ref) 75–99.9 AOR = 1.1 (0.4–3.1) >100 AOR = 1.4 (0.4–5.4)	Maternal ethnicity and birth season. One additional variable
Serum 25(OH)D measured during childhood and adolescence					
Hollams (2011), Australia (36)	689 followed to age 14. Population-based cohort study	Baseline: 6 years (nmol/l) Continuous	Current asthma at age 14 (wheeze and medication in the last 12 months, diagnosed asthmatics)	Univariate logistic regression with deseasonalized 25(OH)D at age 6 and asthma age 14 $\log_{10}$-transformed in: Whole population OR = 0.11 (0.02–0.84) Males OR = 0.03 (0.00–0.6) Females OR = 0.4 (0.02–7.64)	Sex and collection month. Then stratified by sex
Tolppanen (2013), UK (39)	3323 followed to age 16. Population-based cohort study	Baseline: 9.8 ± 1.18 years (ng/ml) Continuous 25(OH)D2 and 25(OH)D3	Incident asthma. Wheezing at least 1 year after phlebotomy when aged 15–16 years	Per doubling of 25(OH)D2: Asthma AOR = 0.89 (0.77–1.01) Wheezing AOR = 0.83 (0.69–1.02) Per doubling of 25(OH)D3: Asthma AOR = 1.03 (0.94–1.12) Wheezing AOR = 1.15 (1.03–1.27)	Sex and age; 25(OH) D3 (from UVB) adjusted for season, ethnicity and BMI. Five additional variables
Van Oeffelen (2011),	372 at 4 years and 328 at 8 years	4 and 8 years (nm)	Asthma (ISAAC)	25(OH)D (tertile 2 *vs* 1 and 3 *vs* 1)	

Table 1 (continued)

Author, year, location	Number included in the analysis population type and Study design	Analytical unit of measurement Time of exposure Measurement scale	Outcomes measured (Definition)	Adjusted results	Confounding variables
Netherlands (50)	in PIAMA study). Population-based cohort and cross-sectional study	Categorical (At 4 years: tertile 1 = 23.1–60.2, tertile 2 = 60.7–78.8, tertile 3 = 79.0–303.8. At 8 years: tertile 1 = 28.7–65.1, tertile 2 = 65.2–79.6, tertile 3 = 80.0–226.0)		at 4 years: AOR1 = reference AOR[2] = 0.54(0.29–1.00) AOR3 = 0.65(0.36–1.16) 25(OH)D at 8 years: AOR1 = reference AOR2 = 1.62(0.63–4.20) AOR3 = 2.21(0.88–5.57)	Season and gender. Four additional variables
Gergen (2013), US (51)	6487 adolescents NHANES population study cohort and cross-sectional study	12–20 years (ng/ml) Continuous	Asthma (prior diagnosis asthma and if still have asthma)	Mean (±SD) 25(OH)D concentrations: Non-asthmatic African Americans = 14.9 ± 6.4 ng/ml. Asthmatic African Americans = 15.0 ± 6.6 ng/ml. Non-asthmatic non-African Americans = 23.0 ± 8.5 ng/ml. Asthmatic non-African Americans = 23.6 ± 8.2 ng/ml	Stratified by race
Bener (2012), Qatar (42)	483 asthmatic and 483 controls. Case–control study	≤16 years (ng/ml) Categorical (<20 = deficient, 20–29 = insufficient, ≥30 = sufficient)	Doctor-diagnosed asthma	25(OH)D deficiency and asthma AOR = 4.82 (2.41–8.63)	Matched for age, gender and ethnicity
Serum 25(OH)D measured in adulthood					
Mai (2012), Norway (49)	2542 adults Nested case–control	19–65 years (nmol/l) Categorical (<50 = deficient, 50–74.9 = insufficient, ≥75 = sufficient)	Incident asthma (questionnaire at 2 time points 11–13 years apart)	25(OH)D and asthma in women ≥75: reference 50–74.9 AOR = 0.8 (0.57–1.13) <50: AOR = 0.94 (0.67–1.32) 25(OH)D and asthma in men ≥75: reference 50–74.9 AOR = 1.5 (0.95–2.38) <50: AOR = 1.47 (0.93–2.32)	Age, SES, season blood collection, BMI. Four additional variables. Stratified by sex, allergic rhinitis
Carroll (2011), USA (34)	340 women Cross-sectional study	(ng/ml) Continuous	Self-reported 'ever' or current asthma	Among white women, a 14 ng/ml increase in 25(OH) D AOR = 0.54 (0.33–0.86). No AOR reported for African American women	Race, age, year and season of enrolment. Eight additional variables. Stratified by race

Table 1 (continued)

Author, year, location	Number included in the analysis population type and Study design	Analytical unit of measurement Time of exposure Measurement scale	Outcomes measured (Definition)	Adjusted results	Confounding variables
Devereux (2010), UK (40)	160 adults Case–control	18–50 years (μg/l) Continuous	Physician confirmed mild/moderate asthma	25(OH)D and asthma AOR: 0.98 (0.91–1.06)	Age, gender matched, BMI, season. Two additional variables
Cheng (2014), Korea (53)	15 212 adults KNHANES population study Cross-sectional	≥19 years (ng/ml) Categorical (<12 = deficient, 12–19.9 = inadequate, ≥20 = adequate)	Physician-diagnosed asthma	25(OH)D and asthma: ≥20 = reference 12–19.9 AOR = 1.19 (0.92–1.53) <12 AOR = 1.29 (0.91–1.82)	Age, sex, season of blood sampling. Four additional variables

serum 25(OH)D and incident asthma and found no association (49) (Table 1).

Prevalent asthma

Two cross-sectional studies by Cheng et al. (53) and Carroll et al. (34) examined the relationship between 25(OH)D and prevalence of asthma in 15 212 Korean adults and 340 American women, respectively. Similarly, Devereux et al. (40) investigated this association in a case–control study (Table 1). Carroll et al. (34) found that low serum 25(OH)D levels were associated with increased asthma, but both the case–control and cross-sectional studies found no association between 25(OH)D levels and asthma (40, 53) (Fig. 7). A meta-analysis was not possible for this outcome due to the small number, and the methodological differences between the studies that investigated severity in adults.

Asthma severity and exacerbations

A cross-sectional analysis with a sample size of 280 participants investigated the relationship between serum 25(OH)D and asthma severity (46) (Table 2). This study investigated two separate outcomes of 'uncontrolled' and 'severe' asthma in adults. Both analyses found that low serum 25(OH)D was associated with increased asthma severity (46). No studies investigating the association between serum 25(OH)D and asthma exacerbations in adult populations were identified. A graphical presentation has not been included for these results due to a lack of sufficient studies.

Discussion

This systematic review of 23 articles investigating the relationship between serum 25(OH)D and asthma suggested the existence of a signal for the role of 25(OH)D in asthma exacerbations in children, but little evidence for a role in asthma incidence whether the exposure was measured prenatally or postnatally. The majority of the studies that investigated the role of 25(OH)D status in asthma prevalence and severity in both children and adults indicated that there was no association between low serum 25(OH)D levels and asthma; however, the majority of these studies were cross-sectional and results were conflicting.

The studies by van Oeffelen et al. (50) and Gergen et al. (51) had inconsistent results. Van Oeffelen et al. (50) measured both incidence and severity, although the number of children in this cohort was very small ($n = 14$) and the number of children with and without asthma at baseline was unclear. Additionally, Gergen et al. (51) measured asthma exacerbations in terms of hospitalization over the 46-week follow-up period, but again, the number of cases in their study was small. This small number resulted in a large odds ratio and a wide confidence interval (OR = 13.4, 95% CI: 1.6–110.2). Therefore, the conclusions drawn by these two studies were thought to be less accurate.

The first randomized controlled trials on this topic have been published on the effect of vitamin D supplementation on asthma. These three trials were excluded from this systematic review as 25(OH)D levels were measured after supple-

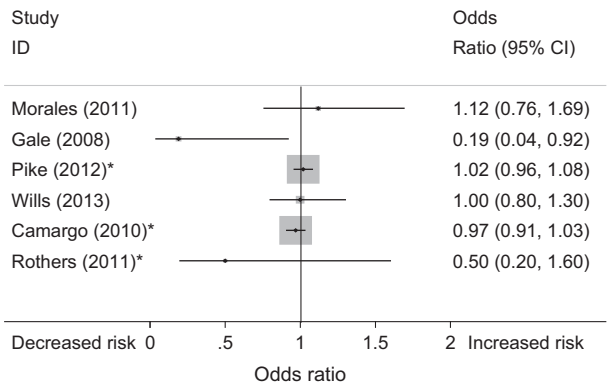

Figure 2 Forest plots of results for those studies measuring serum 25(OH)D during pregnancy or from cord blood at birth and asthma incidence. ORs for lowest *vs* highest serum 25(OH)D level. *OR per 10 nmol decrease in serum 25(OH)D level.

mentation with vitamin D for 28 weeks or 6 months (54–56). While all three trials were double-blinded and randomized trials, the Yadav trial was performed on 100 children aged between 3 and 14 years with moderate to severe asthma (54), compared to 48 children aged 5–18 years with newly diagnosed asthma in the Majak study (55). In addition, the intervention group in the Yadav study received oral vitamin D3 (cholecalciferol) 60 000 IU per month for 6 months and the control group received placebo (54). In contrast, in the Majak study, the control group received budesonide 800 μg/d and vitamin D placebo, while the intervention group received budesonide 800 μg/d and vitamin D (500 IU cholecalciferol) (55). The Yadav trial examined the effect of the vitamin D on measures of asthma severity, such as number of exacerbations during treatment period, level of asthma control and number of emergency visits (54). The outcome of interest in the Majak trial was asthma control assessed by number of exacerbations (55). Both Majak and Yadav trials, in support of the evidence from observation studies found in the current systematic review, found evidence for a role of vitamin D in the prevention of asthma exacerbations (54, 55). In contrast to our findings and those of the aforementioned trials, the VIDA study that randomized 408 adult asthmatics with symptomatic asthma and low serum 25-hydroxyvitamin D (<30 ng/ml) and supplemented those in the intervention group with an initial dose of 100 000 IU of oral vitamin D3, then 4000 IU per day for 280 weeks, found that vitamin D3 supplementation did not significantly reduce the rate of first exacerbations (adjusted HR 0.7, 0.4–1.2 $P = 0.21$) or overall rate of exacerbations (adjusted HR 0.63, 0.39–1.01 $P = 0.05$) in vitamin D insufficient adults with persistent asthma compared with the placebo group (56).

Our findings on the link between low 25(OH)D levels and asthma exacerbations are supported by the current biological knowledge. Asthma treatment involves the administration of glucocorticoids, which stimulate CD4+ T cells to produce IL-10 (13, 57). IL-10, a potent anti-inflammatory and immunosuppressive cytokine, can block allergic airway inflammation, thereby playing a role in asthma control (13). In their paper, Xystrakis et al. (13) demonstrated that in the presence of 25 (OH)D3, human CD4+ Tregs secreted high levels of IL-10. Additionally, 25(OH)D3 was found to enhance responsiveness to dexamethasone for the induction of IL-10 synthesis in steroid resistant asthmatics (13). Through these mechanisms, calcitrol (vitamin D3) can improve the control and reduce the severity of asthma symptoms and exacerbations. For this reason, participant use of corticosteroids within the window of observation of studies may potentially modify the observed association between 25(OH)D concentrations and asthma symptoms, severity and exacerbations, and therefore, future analyses should check for interaction.

Furthermore, 1,25 dihydroxyvitamin D has been linked to the stimulation of alveolar type II cell DNA synthesis (58) and surfactant production (59), which suggests that offspring born to 25(OH)D-deficient females may be predisposed to asthma. However, the results of the present review do not support this argument, as four of the five studies that measured 25(OH)D status in pregnant mothers and cord blood

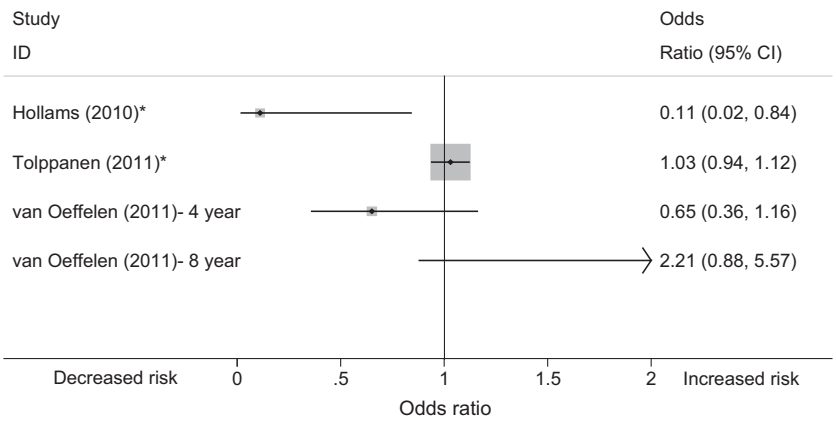

Figure 3 Forest plots of results for asthma incidence studies in children. ORs are for high *vs* low serum 25(OH)D level and risk of asthma. *Hollams per $\log_{10}$ increase; Tolppanen OR per doubling of 25(OH)D level.

Table 2 Study characteristics of studies measuring serum 25(OH)D and asthma severity and exacerbations

Author, year, location	Number included in the analysis Population type and study design	Time of exposure Analytical unit of measurement Measurement scale	Outcomes measured (definition)	Adjusted results	Confounding variables
Serum 25(OH)D and existing asthma in children					
Menon (2012), USA (32)	263 asthmatics. Cross-sectional study	2–19 years (ng/ml) Continuous	Asthma severity based on (controller medications and NHLBI's guidelines)	Mean (±SD) for five steps of asthma severity 1. 28.30 ± 9.83 2. 30.26 ± 10.64 3. 28.25 ± 11.43 4. 26.60 ± 8.38 5. 25.00 ± 6.70	Age matched. Stratified by BMI and then by race
Chinellato (2011), Italy (31)	75 physician-diagnosed asthmatics. Cross-sectional study	5–11 years (ng/ml) Continuous	Asthma control (Childhood Asthma Control Test [C-ACT])	25(OH)D and C-ACT correlation: $r = 0.28$ (0.06–0.49)	Sex, age, season of 25 (OH) D measurements was obtained. One additional variable
Van Oeffelen (2011), Netherlands (50)	372 at 4 years and 328 at 8 years in PIAMA study). Population-based cohort and cross-sectional study	4 and 8 years (nm) Categorical (At 4 years: tertile 1 = 23.1–60.2, tertile 2 = 60.7 –78.8, tertile 3 = 79.0–303.8. At 8 years: tertile 1 = 28.7–65.1, tertile 2 = 65.2–79.6, tertile 3 = 80.0 –226.0)	Severe asthma. Inhalation of steroids Four or more attacks of wheeze/dyspnoea in past 12 months Kept awake at night or limited speech or activities due to wheeze/dyspnoea	25(OH)D (tertile 2 *vs* 1 and 3 *vs* 1) at 4 years: AOR1 = reference AOR2 = 0.37(0.18–0.77) AOR3 = 0.49(0.25–0.95) 25(OH)D at 8 years: AOR1 = reference AOR2 = 1.29(0.38–4.39) AOR3 = 2.14(0.67–6.82)	Season and gender. Four additional variables
Gupta (2011), UK (35)	86 asthmatics [inc. severe, therapy-resistant asthma (STRA)]. Cross-sectional study	6–16 years (nmol/l) Continuous	Asthma Control Test, Acute exacerbations (episodes in the previous 6 months requiring steroids for at least 3 days)	For each 1 nmol/l increase in serum 25(OH)D levels: ACT B coefficient (95% CI) = 0.15 (0.09–0.20). Exacerbation AOR = 0.79 (0.64–0.97)	Age, sex, BMI and ethnicity. One additional variable
Brehm (2010), North America (44)	1024 mild/moderate persistent asthmatics followed for 4 years of Childhood Asthma Management Program (CAMP) study. Cohort study	Baseline: 7–11 years. (ng/ml) Categorical (≤30 = insufficient, >30 = sufficient)	Severe asthma exacerbations (hospitalization)	Insufficient 25(OH)D and hospitalization AOR = 1.4 (1.0–1.9)	Age, sex, BMI, season of 25(OH)D draw and race. Three additional variables. Then stratified by treatment group
Gergen (2013), US (51)	226 inner-city asthmatics followed for 46 weeks of ACE study.	Baseline: 12–20 years (ng/ml)	Exacerbations (systemic corticosteroids or hospitalization) Asthma Control Test	25(OH)D and hospitalizations in African Americans:	Age, sex, season of measurement, BMI at baseline.

Table 2 (continued)

Author, year, location	Number included in the analysis Population type and study design	Time of exposure Analytical unit of measurement Measurement scale	Outcomes measured (definition)	Adjusted results	Confounding variables
	Cohort and cross-sectional study	Categorical <20 = low, ≥20 = high)	Composite Asthma Severity Index (CASI) Maximum number of days with asthma symptoms	AOR = 13.4 (1.6–110.2). 25(OH)D and ACT in: African Americans: AOR = 0.18 (−1.00–1.35) Non-African Americans AOR = 1.36 (0.39–2.32) 25(OH)D and CASI in: African Americans: AOR = 0.36 (−0.72–1.43) Non-African Americans AOR = −0.72 (−1.58–0.14) 25(OH)D and Max number day with symptoms in: African Americans: AOR = 0.03 (−0.82–0.88) Non-African Americans AOR = −0.42 (−1.14–0.30)	One additional variable. Stratified by race
Brehm (2009), Costa Rica (43)	616 physician-diagnosed asthmatics. Cross-sectional study	6–14 years. (ng/ml) Continuous	Asthma exacerbations (hospitalization)	For each $\log_{10}$ increase in 25(OH)D level, odds of hospitalization for asthma: AOR = 0.05(0.004–0.71)	Age, sex, BMI z-score. One additional variable
Brehm (2012), Puerto Rica (33)	287 physician-diagnosed asthmatics. Cross-sectional study	6–14 years (ng/ml) Categorical (<30 = insufficient, ≥30 = sufficient)	Severe asthma exacerbations (hospitalization that lead to treatment)	Cases with one or more severe exacerbation: 25(OH)D (<30 ng/ml) and asthma AOR = 2.6 (1.5–4.7)	Age, sex, African ancestry. Four additional variables. Stratified by atopy
Beigelman (2014), (52)	264 children with history of wheeze and risk factors for asthma MIST study Randomized Controlled Trial (Cross-sectional analysis)	12–53 months (ng/ml) Categorical <20 = deficient, ≥20 = sufficient)	Rate of exacerbations requiring oral corticosteroids (OCS) over the course of 1 year	Rate of exacerbations for deficient compared to sufficient 25(OH) D levels: ARR = 1.68 (1.09–2.58)	Race
Serum 25(OH)D and asthma severity in adults					
Korn (2013), Germany (46)	280 asthmatics. Cross-sectional study	≥18 years. (ng/ml) Categorical <30 = insufficient, ≥30 = sufficient)	Asthma control Asthma severity	Insufficient 25(OH)D and: uncontrolled asthma AOR = 2.1 (1.3–3.5). severe asthma AOR = 1.9 (1.2–3.2)	Age, sex, BMI and seasonality

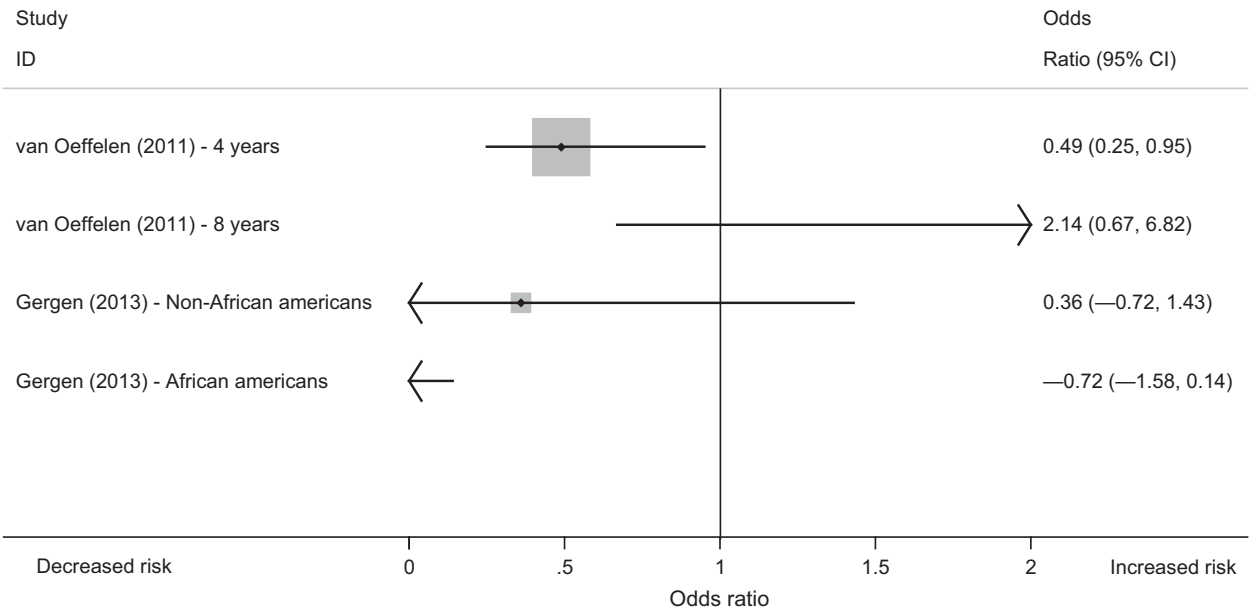

Figure 4 Forest plot of results for asthma severity studies in children. Odds ratios for high compared to low 25(OH)D.

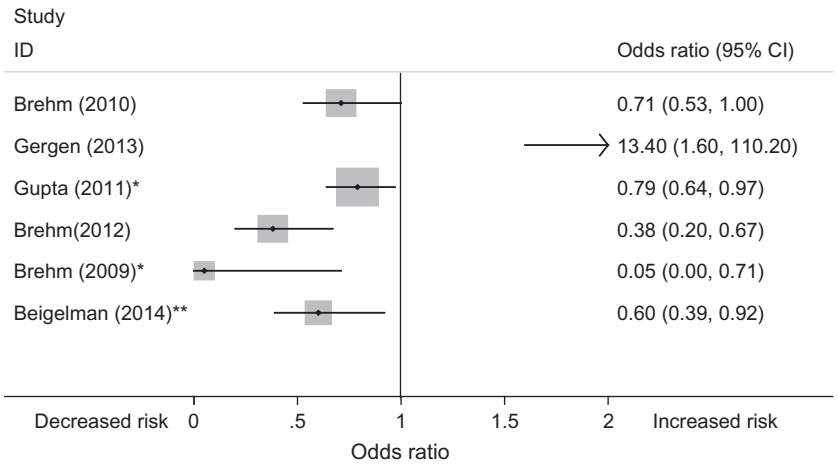

Figure 5 Forest plots of results for asthma exacerbation studies in children. Odds ratios are for high compared to low 25(OH)D levels. *Gupta per 1 nmol/l increase; Brehm per $\log_{10}$ increase. **Beigelman result was rate ratio.

found no associations with asthma in childhood. Vitamin D supplementation in pregnant women and the asthma and allergic outcomes of their offspring is currently being tested in an on-going randomized, double-blinded, placebo-controlled trial (VDAART) (60).

The principal strength of this review lies in the evaluation of studies that measured serum 25(OH)D in their participants. Serum 25-hydroxyvitamin D (25(OH)D) reflects contributions from both the cutaneous absorption of ultraviolet B radiation and the various dietary sources of 25(OH)D, and although it has a relatively short half-life, it is considered to be the best circulating biomarker of 25(OH)D (43, 61, 62). Despite the limitations that arise from utilizing a single measurement of serum 25(OH)D in a longitudinal study, it remains an accurate, albeit short term, measure of 25(OH)D status as it does not rely on food frequency and sun exposure questionnaires that are subject to error and misclassification. Additionally, the inclusion of studies in both adults and children enabled the investigation and comparison of the role of 25(OH)D in asthma in both of these populations.

The available evidence was difficult to summarize due to a lack of uniform and objective measures of asthma. The studies varied greatly in the way that asthma was both defined

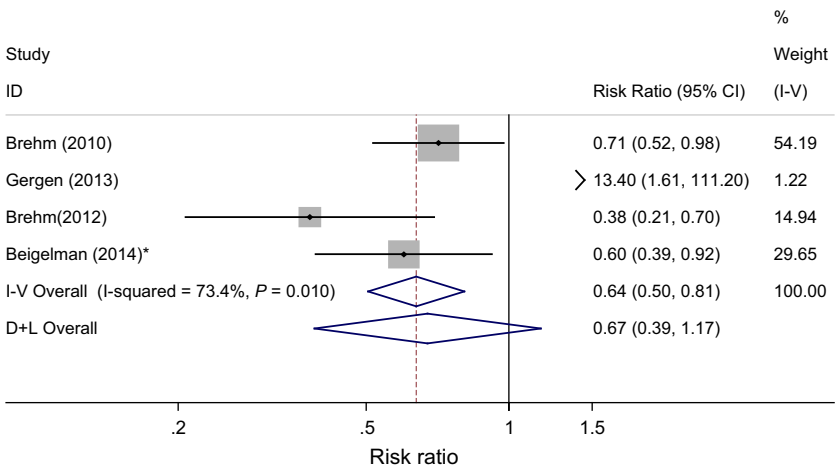

Figure 6 Forest plot of results from the fixed and random effects meta-analysis of the association between high *vs* low 25(OH)D levels and asthma exacerbations in children. Where I–V is the inverse variance/fixed effect method and D+L is the DerSimonian and Laird/random effects method. *Beigelman result was Rate Ratio.

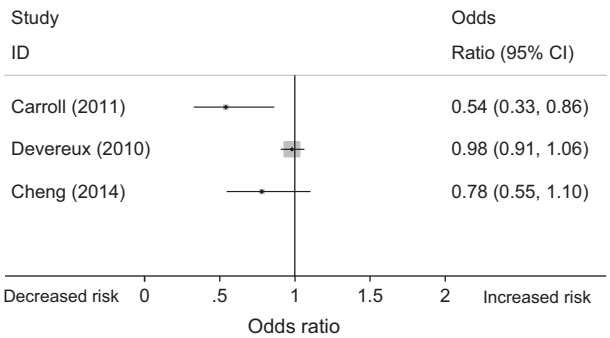

Figure 7 Forest plots of results for asthma prevalence studies in adults. Odds ratios are for high compared to low 25(OH)D levels.

and measured. For example, the asthma outcome measures ranged from parental reports of asthma, which may be subject to error, to more objective measures such as hospitalization for severe asthma. Equivalently, analysis of 25(OH)D was not standardized across studies, and some relevant articles were excluded as they measured 25(OH)D dietary and supplementary intake but did not include a measure of serum 25(OH)D. The lack of standardization of measurement of 25 (OH)D levels as well as the variability in the definitions between the studies also restricted the options for the meta-analysis. Furthermore, we were unable to identify a suitable tool to formally assess the quality of the articles. A formal analysis of potential publication bias, including constructing funnel plots, was not possible due to the limited number of comparable studies.

A major challenge in analysing the results of studies in this area is that optimal 25(OH)D concentrations for overall health have not been defined (62–64) and a consensus has not been reached on which threshold values constitute 25(OH)D deficiency, insufficiency and sufficiency (10, 43, 65). Historically, 'normal' 25(OH)D levels were derived from samples obtained from nondiseased individuals in the population and plotting the Gaussian distribution (66). However, several potentially confounding factors such as sunscreen use, lifestyle, latitude and race suggest that these values are inaccurate (43, 61, 62). Categorization of 25 (OH)D sufficiency, insufficiency and deficiency is generally based on the values that are commonly accepted for bone health (63, 65); in a recent report, the Institute of Medicine concluded that 20 ng/ml is sufficient for bone health and that there was insufficient evidence to inform nutritional requirements for extra-skeletal outcomes (67, 68). It has been suggested, however, that these estimates may be too low for general health (65, 69).

Another important issue to consider is the inability to establish a temporal relationship between 25(OH)D and asthma due to the cross-sectional nature of many of the included studies. It is plausible that many asthmatics refrain from outdoor activities to avoid asthma symptoms and therefore have lower 25(OH)D levels. In addition, 25(OH)D acts as a negative acute phase reactant, whereby inflammation causes a decrease in serum 25(OH)D levels (70). Therefore, asthma may be causing reduced 25(OH)D levels (26, 70, 71). Hence, there remains a need for longitudinal data to clarify the direction of the significant protective association on asthma outcomes.

In summary, despite the limitations of weak epidemiological study designs and the inconsistencies between the included studies, the current review suggests that 25(OH)D may play a role in asthma exacerbations among asthmatic children, but there is no evidence for a role in the incidence, prevalence or severity of asthma. The conclusions drawn from this review largely support those of previous reviews that suggest a beneficial association is evident between 25 (OH)D and asthma, although past reviews do not always differentiate between asthma incidence, prevalence, severity and

exacerbations (7, 11, 14). The review by Paul et al. (11) made this distinction and found that 25(OH)D had beneficial effects on asthma morbidity, but that there was insufficient evidence to suggest a preventive role for 25(OH)D in asthma. In contrast, Nurmatov et al. (25) concluded that there was weak supportive evidence for 25(OH)D, preventing childhood wheeze and possibly asthma.

In future, objective measures of asthma such as FEV_1/FVC or airway hyper-responsiveness (AHR) may improve the specificity of the outcomes and therefore the findings. Additionally, a standardized measurement of outcomes would enable future reviews to directly compare studies in their evaluation of the evidence. Longitudinal data on the role of 25(OH)D in asthma severity will help to strengthen the case for randomized controlled trial designs such as those currently in progress, which are better able to clarify causality and optimal dosage of 25(OH)D necessary for good control of asthma symptoms.

Although recommendations for the optimal 25(OH)D concentration have been published (65, 69), these may not be applicable for the control of asthma symptoms. Given the significant burden of asthma on the community and the lack of modifiable risk factors, further research is needed into the potential role of vitamin D in asthma control. If established as an effective treatment in acute or severe asthma, this may have significant public health implications.

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Author contributions

Justification for the inclusion of six contributing authors: This work was designed, conducted and subsequently written up by lead author R Cassim for her Master's degree. J Koplin conducted the side by side review (data search, analysis and extraction) to ensure validity of the review, and provided input to the writing. Given this review required multiple expertise (Biology, Epidemiology, Statistics, Respiratory Medicine), the other four authors (C Lodge, A Lowe, M Russell and S Dharmage) were involved in supervising all three stages of the review, that is design, analysis and write-up.

R. Cassim, M. Russell, C. Lodge, A. Lowe, J. Koplin and S. Dharmage contributed to the study conception, data collection and analysis; writing and revising article; and final approval of version. All of the listed authors contributed to the review from conception through to editing and revising.

Conflicts of interest

The authors declare that they have no conflicts of interest.

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Appendix

Search strategy used in PubMed database

((("vitamin d"[MeSH Terms] OR "vitamin d"[All Fields] OR "ergocalciferols"[MeSH Terms] OR "ergocalciferols"[All Fields] OR "cholecalciferol"[MeSH Terms] OR "cholecalciferol"[All Fields] OR "alfacalcidol"[All Fields] OR "serum 25-hydroxyvitamin d "[All Fields]) AND ("asthma"[MeSH Terms] OR "asthma"[All Fields] OR "wheeze"[All Fields] OR "asthma symptoms"[All Fields]))) AND ((Clinical Trial [ptyp] OR Controlled Clinical Trial[ptyp] OR Journal Article [ptyp] OR Letter[ptyp] OR Randomized Controlled Trial [ptyp]) AND ("1950/01/01"[PDat]: "2014/09/28 "[PDat]) AND Humans[Mesh].