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OTITIS (DP SKONER, SECTION EDITOR)

# Vitamin D and Otitis Media

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**Abstract**

**Purpose of Review** To examine the relationship between vitamin D and otitis media.

**Recent Findings** Vitamin D deficiency has been associated with several respiratory diseases, including otitis media. Vitamin D supplementation may reduce the risk of otitis media. This relationship may be explained by vitamin D supporting the immune system by upregulating antimicrobial peptides which are effective against otopathogens and biofilm formation, supporting a less inflammatory immune response, or promoting beneficial commensal bacteria.

**Summary** This review will explore risk factors of both otitis media and vitamin D deficiency, the evidence of vitamin D being beneficial for various forms of otitis media, and possible mechanisms of action.

**Keywords** Otitis media · Vitamin D · Viral infections · Respiratory microbiota · Antimicrobial peptides · Cytokines · Biofilms · Upper respiratory tract infections

## Introduction

Otitis media (OM) is an inflammatory condition characterised by the accumulation of fluid or effusion in the middle ear space. In OM with effusion (OME), this middle ear effusion is present without signs or symptoms of acute infection. OME may persist as chronic OM with effusion (COME) if the fluid remains for 3 months or longer. In acute OM (AOM), acute infection accompanies the effusion [1, 2]. AOM may reoccur as recurrent acute OM (RAOM), and if the tympanic membrane is perforated and there is ongoing discharge of purulent effusion, it is considered to be chronic suppurative OM (CSOM) [3].

The incidence rate of AOM in children under 4 years of age is 61 new episodes per 100 individuals per people year [4]. The estimated prevalence of OME in preschool children is 10–30% [5–9]. COME has an estimated prevalence rate of between 5 and 9% [10–12]. CSOM has an incidence rate of 4.8 new episodes per 1000 people per year [4]. OM is the leading cause of hearing loss in childhood, with a prevalence of OM-related hearing loss estimated at 30 per 10,000 individuals [4]. OM is also a very common reason for doctors' visits, antibiotic prescriptions, and surgery [13–17].

The management/treatment approach for OM is dependent on a child's age and whether they are in an at-risk group. Children considered to be at risk include those with craniofacial abnormalities such as down syndrome and cleft palate, autism spectrum disorder, permanent hearing loss that is not related to OM, blindness, and developmental delays [18]. While watchful waiting is indicated in some cases, AOM and CSOM are most often treated with antibiotics, and COME and RAOM are sometimes treated with tympanostomy tube placement or adenoidectomy [18].

Vitamin D was long regarded as primarily relevant to bone health, but it is now recognised to also play important roles in the immune system, including increasing mucociliary clearance, influencing the production of antimicrobial factors, regulating epithelial cell production, modulating inflammatory pathways, and influencing the microbial communities [19–21]. The active form of vitamin D is 1,25-dihydroxyvitamin D ($1,25(OH)_2D$), which activates the vitamin D receptor (VDR) expressed in many immune cells including monocytes and epithelial cells. With this changing

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perspective on the role of vitamin D in general health, vitamin D deficiency has been investigated and found to be a risk factor for a range of respiratory conditions, including OM [22–25]. Vitamin D supplementation has been considered as both a preventative measure and a treatment [18, 26].

Vitamin D is largely obtained from sunlight exposure, and a small amount is also absorbed from certain foods such as oily fish. Deficiency of vitamin D (usually defined as < 50 nmol/L serum concentration of 25-hydroxyvitamin D (25(OH)D)) is relatively common, with an estimated prevalence of 30–40% [27, 28]. Serum 25(OH)D concentration usually decreases in winter.

This review summarises our current understanding of the pathogenesis of OM, examines risk factors shared between vitamin D and OM, explores the evidence regarding vitamin D supplementation as a treatment option for OM, and discusses the possible mechanisms by which vitamin D may be beneficial in children with OM.

## Pathogenesis of Otitis Media

The various types of OM form a continuum, sharing many risk factors and similar causal pathways. The pathogenesis is multifactorial and may involve a cascade starting with viral upper respiratory infection (URI), which stimulates overgrowth and spread of bacterial pathogens from the nasal passages to the middle ear, resulting in histopathological changes. In response to the invasion, the healthy middle ear epithelium changes from being mostly simple cubical squamous cells to a pseudostratified epithelium [29]. Characteristic changes include mucosal hyperplasia and the proliferation of goblet cells, leading to an increase in mucin production, which creates a middle ear effusion. The mucosal hyperplasia and goblet cell proliferation are more pronounced in the chronic forms of OM than in the acute forms [30]. Mucous production continues until the infection has cleared; however, the additional goblet cells remain months after the infection and may never return to pre-infection levels [31].

Bacterial pathogens are ubiquitous in the middle ear during OM and are central to its pathogenesis. The microbiota of the nasal passages has also been reported to be associated with OM. Although these otopathogens are present in the nasal passages of healthy children, their abundance is greater in children with OM [32]. The three pathogens most commonly found in middle ear effusion are *Haemophilus influenzae*, *Streptococcus pneumoniae*, and *Moraxella catarrhalis*. *Staphylococcus aureus*, *Streptococcus pyogenes* and *Pseudomonas aeruginosa* are also found sometimes, especially in CSOM [33, 34].

Effusion production is the body's natural response to infection and is seen in all forms of OM. Mucins bind to the outer cell of pathogenic bacteria, so that the mucociliary system can flush them out of the middle ear. AOM, RAOM, and CSOM involve an acute inflammatory immune response to bacteria in the middle ear, often characterised by redness of the tympanic membrane, ear pain, and fever. The effusion may also become purulent. The effusion is rich in mucins, which increase its viscosity and contain innate immune cells including lysozymes and beta-defensins [35, 36]. White blood cells including macrophages, neutrophils, and lymphocytes are also present, producing cytokines that regulate the inflammatory response to pathogens [37]. Levels of TNFα, IL-10, and IL-1β in effusion and serum have been found to correlate with tympanostomy tube placement [38, 39]. Mucoid effusion tends to have higher levels of TNFα, IL-8, IL-2, and IL-10. TNFα, IL-10, and IL-6 have been associated with OM during URI. Elevated levels of IL-10, TNFα, and IL-8 have been implicated in goblet cell production [40, 41]. IL-4, IL-5, and TNFα have been reported to be higher in atopic children with OME, while a higher level of IFNγ has been reported in non-atopic children suggesting a protective effect [42, 43]. These cytokine findings suggest a possible role for allergy in the pathogenesis or persistence of OM.

Chronic or persistent forms of OM have also been explained by the presence of live bacteria in the middle ear intracellularly and in biofilm state [44, 45]. Children with COME have been reported to have bacteria in their middle ear living inside mucous vacuoles of epithelial cells, and live biofilms have been observed inside neutrophil extracellular traps. Bacteria in biofilm state or within cells are less likely to be affected by antibiotic treatment [46].

## Risk Factors Shared Between Vitamin D Deficiency and Otitis Media

Levels of 25(OH)D are lower in winter and in preschool children. Similarly, the best-established risk factors for OM include winter season, younger age, and URI. The lower vitamin D status seen in winter, and in younger children, may increase the risk of precursors for OM, such as URI and bacterial overgrowth, while also impeding the immune response to early infection. Daycare contributes to exposure to infections, and has also been found to be a risk factor for OM.

Serum 25(OH)D concentration of 75 nmol/L or more has been reported to be associated with a reduced risk of aeroallergen sensitisation and allergic rhinitis (AR) in a systematic review and meta-analysis [47]. Findings regarding a possible association between atopy or AR and OM have been mixed. We have previously reported nasal obstruction, an important symptom of AR, to be associated with COME independent of colds and atopic diseases [48]. We proposed that bacterial overgrowth in the nose and nasopharynx might be a common factor in both nasal obstruction and OM.

Children with higher levels of the skin pigment melanin synthesise less vitamin D from sunlight exposure. Although it has been proposed that ethnic variation in the carrier protein of 25(OH)D (vitamin D binding protein) may affect the proportion of 25(OH)D that is free or bioavailable [49], it has more recently been shown that total serum 25(OH)D does correlate closely to free 25(OH)D independently of ethnicity [50]. Asian ethnicity has been reported as being protective against OM [48], despite the lower levels of 25(OH)D often found in these groups. We have reported higher concentration of 25(OH)D to be protective against COME but only after adjusting for ethnicity. We reasoned that this was due to a dampening effect of Asian ethnicity, which was inversely associated with COME [25•].

## Vitamin D Supplementation for OM

At this time, only one randomised controlled trial (RCT) has been reported that examined the effectiveness of vitamin D supplementation for OM. Marchisio and colleagues recruited 116 Italian children aged under 5 years with a recent history of RAOM [51••]. Half of the children were administered 1000 IU of vitamin $D_3$ per day orally for 4 months, while the remainder were administered an oral placebo, with double blinding. The children in the intervention group had a mean serum 25(OH)D concentration of 66.1 nmol/L before the trial commenced, just before the start of winter. At the end of the study, the mean serum concentration of vitamin D in the intervention group was 91.9 nmol/L, 38% higher than the baseline. By contrast, the mean serum concentration of vitamin D in the controls had dropped by 27% as a result of reduced sunlight exposure during winter, from 64.4 to 46.7 nmol/L. Among the children who received vitamin D, 44.8% had at least one episode of AOM, significantly fewer than the 65.5% of the control group who had at least one episode ($P = 0.03$).

Vitamin D supplementation was not observed to have any effect on the likelihood of AOM with otorrhea in the subjects. The authors speculated that vitamin D supplementation may increase production of the antimicrobial peptide LL-37, which may in turn increase the virulence of *S. pyogenes*, a bacteria associated with otorrhea. Usually LL-37 suppresses bacterial infection; however, it has been reported to have the opposite effect on *S. pyogenes* when studied in vitro [52].

There is growing evidence that daily and weekly supplementation with vitamin D may be the most effective against acute respiratory infection (as compared with bolus doses), with the greatest benefit seen in patients who are more deficient [23]. In Marchisio and colleagues' study, supplementation was taken daily, and the mean serum concentration of 25(OH)D in the controls was at deficiency levels by the end of the trial.

## Low Vitamin D Status as a Risk Factor for OM

Lower serum concentration of 25(OH)D has been associated with increased risk of COME [25•, 53•], AOM [54•], and RAOM [55•] (Table 1). Vitamin D deficiency (< 50 nmol/L) has also been reported to increase the risk of earache or discharge in the presence of fever [61]. Linday and colleagues examined 16 children undergoing tympanostomy tube placement and found 50% of the children had serum 25(OH)D levels less than 50 nmol/L (20 ng/mL), and another 31% less than 75 nmol/L (30 ng/mL) [56]. Hosseini et al. found that mean level of 25(OH)D in 40 children with OME was 65.1 nmol/L, compared with 73.6 nmol/L in the 80 controls; however, the difference was not statistically significant ($P = 0.27$) [59].

No association has been reported between low 25(OH)D concentration and CSOM. Elemraid and colleagues conducted a case-control study of 149 Yemeni children with CSOM, and found no association after adjustment for albumin and age [57]. Park and colleagues investigated 16,063 subjects aged 20 years or older in Korea. In a multivariable analysis, no association was found between blood 25(OH)D concentration and chronic OM (COM). Diagnosis of COM was based on a questionnaire and findings of tympanic membrane perforation and/or cholesteatoma, and a retraction pocket and/or OME or a tympanostomy tube present [58]. This definition may include a variety of conditions and etiologies, and it is unclear whether the finding is generalisable to more specific forms of OM or to children.

A 2016 meta-analysis and systematic review by Li and colleagues included five studies and concluded that blood level of 25(OH)D was not associated with OM (OR 0.80; 95% CI 0.47–1.38; $P = 0.43$) [60•]. However, the authors noted that low 25(OH)D concentration was associated with AOM in a sub-analysis of the three studies that focused on that condition in children. The other two studies in the meta-analysis were the abovementioned Park et al. study on COM and the Elemraid et al. study on CSOM, which included older subjects. The high weighting given to the latter studies in the meta-analysis may help to explain the overall null finding.

## Possible Mechanisms of Action

There are several mechanisms by which 25(OH)D and the vitamin D receptors (VDRs) may support the functioning and maturation of the immune system, and thereby help to reduce the risk of OM.

One of the first defences against infection in the airways is the mucociliary blanket that lines the airway epithelium. The mucus captures pathogens, and the rhythmic movement of the cilia forces the secretions to the nasopharynx where it passes into the oropharynx and is swallowed. Akcan and colleagues

**Table 1** Studies of vitamin D and otitis media

| Study | Year | Design | Size | Age | Condition | Results |
|---|---|---|---|---|---|---|
| Linday et al. [56] | 2008 | Cohort | 16 | 1–8 years | Tympanostomy tube placement | In children having tympanostomy tube placement, 50% had 25(OH)D level < 49.2 nmol/L, a level considered deficient |
| Elemraid et al. [57] | 2011 | Case-control | 149 | 6month–15 years | CSOM | Yemeni children with CSOM not associated with lower 25(OH)D after adjusted for albumin ($P < 0.05$)<br>In CSOM, duration of discharge negatively correlated with 25(OH)D ($p < 0.05$) No longer significant when adjusting for age |
| Cayir et al. [55•] | 2014 | Case-control | 192 | 1–5 years | RAOM | Serum 25(OH)D level in the children with RAOM were 28.5 ± 24.5 nmol/L and in the controls 72.9 ± 34.7 nmol/L ($P < 0.05$) |
| Marchisio et al. [51••] | 2013 | RCT | 116 | 1–5 years | RAOM | Vitamin D supplementation reduced the risk of RAOM ($P = 0.03$) |
| Cayir et al. [54•] | 2014 | Case-control | 169 | 1–13 years | AOM | Serum 25(OH)D level in the children with AOM were 51.4 ± 25.5 nmol/L and in the controls 59.4 ± 25.7 nmol/L (P < 0.05) |
| Park et al. [58] | 2015 | Cohort | 16,063 | > 20 years | COM | No association between vitamin D and COM |
| Hosseini et al. [59] | 2016 | Case-control | 120 | 3–10 years | OME | Mean level of 25(OH)D in children with OME 65.1 nmol/L and in the controls 73.6 nmol/L (P = 0.27) |
| Li et al. [60•] | 2016 | Meta-analysis and systematic review | 5 studies | 6months–20 years | CSOM, RAOM | Vitamin D not associated with OM OR 0.80: 95% CI (0.47–1.38) ($P = 0.43$) |
| Walker et al. [25•] | 2017 | Case-control | 178 | 3–5 years | COME | Higher 25(OH)D associated with lower risk of COME aOR 0.86 per 10 nmol/L: 95% CI (0.77–0.97) ($P = 0.01$) |
| Akcan et al. [53•] | 2018 | Case-control | 146 | 1–13 years | COME | Higher 25(OH)D associated with lower risk of COME ($P < 0.001$) |

reported that subjects deficient in vitamin D had less effective mucociliary clearance than controls. After 3 months of supplementation, the mucociliary clearance rates improved. The authors hypothesised that the decreased mucociliary clearance rate could be due to increased squamous metaplasia in the respiratory tract of subjects deficient in vitamin D, or decreased bactericidal nitric oxide which also affects mucociliary function [19].

Vitamin D also upregulates the antimicrobial peptide cathelicidin, an endogenous antibiotic [20]. Cathelicidin is upregulated on exposure to pathogens via an increase in vitamin D receptors and $1,25(OH)_2D$ [62, 63]. Higher 25(OH)D concentration has been reported to correspond to greater circulating cathelicidin concentration [64], although not all researchers have found this association [65]. In animal models, treatment with cathelicidin is effective against the otopathogens *Haemophilus influenzae*, *Streptococcus pneumoniae*, and *Moraxella catarrhalis* [66], and suppresses the formation of *Pseudomonas sinusitis* and *Pseudomonas aeruginosa* biofilms [67, 68]. Cathelicidin has been shown in-vitro to break down *Staphylococcus aureus* biofilms [67–69]. High levels of mucins in middle ear effusion may suppress the antimicrobial activity of cathelicidin [70]. High concentration of 25(OH)D could therefore both promote cathelicidin and make it more effective by suppressing antagonistic mucin production.

Vitamin D status may also affect the respiratory microbiome. Serum 25(OH)D concentration is inversely correlated with the risk of *Staphylococcus aureus* carriage in the nose [71]. In subjects with cystic fibrosis, vitamin D treatment was associated with a reduction of airway *Staphylococcus aureus* abundance compared with placebo [72]. The VDR allows the active form of vitamin D to bind. Animal models have compared VDR knockout mice with healthy controls and found the latter had a greater abundance of bacteria belonging to the Lactobacillaceae taxa, which are usually commensals [21, 73], and mice that lack the VDR had increased gut bacterial load [74], and a different microbiome profile from wild type controls [21, 73, 74]. Wang and colleagues found that the VDR gene was associated with overall variation of the gut microbiota [75]. These findings may indicate that a lack of vitamin D or VDR could cause a disrupted microbiome. Disrupted microbiota may be a risk factor for OM, as children with COME [32] and RAOM [76] have been observed to have different nasal microbiomes compared to healthy children.

Sufficient 25(OH)D levels may help to decrease the risk of AR [47]. AR has sometimes been found to be associated with OME, particularly in children 6 years of age or older [77]. AR

may cause inflammation of the nasal mucosa and Eustachian tube, which may lead to OM, perhaps through increased bacterial and viral load or Eustachian tube dysfunction. Biopsy of the middle ear mucosa in children with COME reported increased inflammatory cells and IL-5 [78]. Alternatively, children with AR may be more predisposed to developing OME due to increased risk of URI or bacterial infections.

Frequent URI is often found to be associated with all forms of OM [48]. Viral infections are thought to precede the development of OM, possibly by disrupting bacterial communities in the nasal passages. Viruses have also been shown to promote bacterial ascension from the nasal passages to the middle ear [79]. URI resulted in the release of bacteria from a biofilm that were more virulent than typical planktonic bacteria and were disseminated to other areas such as the middle ear [80]. Vitamin D has been shown to be protective against influenza A carriage [81] and rhinovirus infection by influencing chemokine synthesis and altering the growth and differentiation of airway epithelial cells [82]. Respiratory syncytial virus and rhinovirus have been reported to suppress VDR in vitro, which may act to limit the antiviral activity of 1,25(OH)D. Despite this, adding 1,25(OH)D in vitro was found to reduce viral replication [23, 83]. A systematic review and meta-analysis concluded that vitamin D supplementation reduced the risk of acute respiratory infections [23].

Vitamin D may also promote a less inflammatory response that it still effective against pathogens [84]. It suppresses the pro-inflammatory cytokines TNFα [85, 86] and IL-10 [87] and the inflammatory NF-κB pathway [84]. NF-κB has been associated with increased mucin gene expression and local TNFα production in the ME of mice exposed to cigarette smoke condensate [88, 89]. Rats with experimentally induced OM on a vitamin D deficient diet had thicker mucosa, and higher expression of IL-6 and TNFα, but lower IL-10 [90]. In a mouse model of asthma, 1,25-$(OH)_2D_3$ injection suppressed goblet cell hyperplasia, NF-κB expression, inflammation, and thickening of the epithelia of the lungs [91]. It is possible that when pathogens make their way into the middle ear, higher concentration of serum 25(OH)D may help to protect against an exaggerated immune response by reducing NF-κB, TNFα, goblet cell hyperplasia, and mucin production.

## Clinical Implications

OM is one of several conditions that are likely worsened by vitamin D deficiency, especially during winter or in climates with low sunlight exposure. Recurrent viral URI, chronic nasal obstruction, and bacterial superinfection are all syndromic with OM, and may all be similarly exacerbated by compromised immune function as a result of vitamin D deficiency. Clinical and public awareness of this pattern may benefit patients struggling with recurrent or chronic OM, by highlighting the importance of serum 25(OH)D testing and the ready availability of ways to increase vitamin D status. Sunlight exposure and diet may be beneficial, but are unlikely to raise vitamin D levels to those seen in summer. In patients with recurrent or chronic conditions, or a history of them, supplementation to ensure sufficient vitamin D levels may be beneficial.

## Topics for Further Research

We recommend further investigation of the effect of vitamin D supplementation on OM, as there has only been one RCT to date. Variations include intervention as a prophylactic or treatment measure, different doses and intervals, and effects on the different varieties of OM. The pathways by which vitamin D may be effective may be further clarified by measurement of serum 25(OH)D, URI, nasal obstruction, and immune markers including cytokine concentrations, respiratory microbiota, and symptoms of AR and asthma before and after supplementation.

It would also be instructive to measure the abundance of otopathogens before and after treatment. Vitamin D supplementation may be more effective at suppressing some bacterial species or strains, and certain viruses, than others. It may even contribute to overgrowth of some bacteria, as has been proposed in relation to *S. pyrogenes* via the mechanism of LL-37 [52]. More research is needed regarding whether vitamin D concentration influences respiratory commensal bacteria, and whether supplementation with vitamin D can modify the community composition.

We acknowledge that reverse causality could explain the retrospective studies reporting an association between vitamin D status and OM, as systemic inflammation may cause a decrease in 25(OH)D levels [92]. Growing RCT evidence for the benefit of daily/weekly vitamin D supplementation on acute respiratory infections argues against this explanation. Similarly, RCTs on the effect of vitamin D supplementation on OM are preferable for advancing the science.

## Conclusion

Several studies have found that low concentrations of serum 25(OH)D are a risk factor for COME, AOM, and RAOM. However, studies to date do not indicate that low 25(OH)D concentration is a risk factor for CSOM. There has only been one RCT of vitamin D supplementation, which reported that it may be beneficial for AOM, but did not help reduce the risk of suppuration.

These findings place vitamin D as having a promising, but far from definitive, link with OM. In particular, more RCTs on the effects of vitamin D supplementation on the risk of the

various forms of OM are needed, especially in at-risk children. Vitamin D supplementation alone is unlikely to present a "cure" for OM, but it could form part of a programme of treatment to improve the immune response against URI and pathogen overgrowth, in addition to existing interventions such as antibiotics, vaccines, and surgery.

## Compliance with Ethical Standards

**Conflict of Interest** The authors declare that they have no conflict of interest.

**Human and Animal Rights** All reported studies/experiments with human or animal subjects performed by the authors have been previously published and complied with all applicable ethical standards (including the Helsinki declaration and its amendments, institutional/national research committee standards, and international/national/institutional guidelines).

**Informed Consent** No information or photographs identifying particular individuals are included in this article.

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- • Of importance
- •• Of major importance


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